STATEMENT OF JUSTIN ANTHONY HOWES

I Justin Anthony Howes, of 39 Kessels Road, Coopers Plains in the State of Queensland, do solemnly and sincerely declare that:

Background

- 1. I am employed by Queensland Health and Forensic and Scientific Service (QHFSS).
- 2. I hold the position of Team Leader at QHFSS at Coopers Plains.
- 3. I hold a Master of Science in Forensic Science (Griffith University, conferred 2000), a Bachelor of Arts in Human Movement Science (University of Queensland, conferred 1997), and a Bachelor of Science in Molecular Biology (University of Queensland, conferred 1995). I also have a Diploma of Management (TAFE Queensland, conferred 2015) and a Certificate IV in Workplace Training and Assessment, conferred 2005.
- 4. On 19 September 2022, under s 5(1)(d) of of the Commissions of Inquiry Act 1950 (Qld), Commissioner Sofronoff KC issued Notice 2022/00199 (Notice) to me. I am required to provide a statement regarding my knowledge of the matters set out in paragraphs 1 to 64 of the Notice.
- 5. As part of my response, I have read the following:
 - (a) the Notice; and
 - (b) the documents exhibited to this statement.

Responses to paragraphs 1 to 64

Validations

Background

Question 1

Outline any specific qualifications, skills or experience you have that is relevant to performing or endorsing validations.

 I am one of two Team Leaders in the Forensic DNA Analysis team and have been employed in either temporary or permanent capacities within the work unit since 2006

(JH-1 RESUME). Within my Duty Statement (JH-2 Duty Statement) is information on my current duties including, but not limited to: '...reviewing documentation in QIS2 and within the Change Management Framework where required.' QIS2 is the Quality Information System that contains such things as Standard Operating Procedures (SOPs), Training Modules (TMs), Audit records, Performance Plans, and Opportunity for Quality Improvement (OQI) material. The Change Management Framework includes the process to raise project proposals and conduct process improvement studies, validations and verifications.

 My training and qualifications as a scientist and my experience working in the Forensic DNA Analysis Team are relevant to performing and endorsing validations.

Question 2

List all validations you have performed while employed at QHFSS.

8. I haven't performed any specific validations as the lead Project Officer since employment at QHFSS, where a 'validation' is the developmental process used to acquire necessary information to assess the ability of a procedure to obtain a reliable result, to determine the conditions under which such results can be obtained, and to determine the limitations of the procedure (National Association of Testing Authorities, 2020 – quoted in JH-5 23401v8- Validation guidelines SOP).

Question 3

List all validations you have endorsed while employed at QHFSS.

9. I am unable to confirm the list of validations as I do not have access to paper records at FSS at this point in time. The best assistance I can currently provide is the attached image of all projects that I may have been listed as an endorser of – I am unable to confirm at this point in time. (JH-3 Forensic DNA Analysis projects_JAH).

Endorsements

Question 4

Explain the purpose of endorsing a validation of an instrument or system in the DNA Analysis Unit.

10. The purpose of endorsing a validation is to provide feedback and support for the testing plan or results, and to support any recommendations that might be generated from the testing. This is prior to the approval of the plan or report.

Question 5

Outline the duties and responsibilities of staff when endorsing a validation proposal or report.

Attach any Standard Operating procedures or guidelines for the requirements of staff endorsing a validation report.

11. The duties and responsibilities of endorsers of validation proposals and/or reports is outlined in section 4.4, 4.5 and 4.6 of JH-4 22871v17 Change Mgt SOP, and section 4 of JH-5 23401v8- Validation guidelines SOP.

Question 6

Explain whether there are any internal or external audits or reviews of the QHFSS DNA Analysis Unit's validation proposals or reports.

12. In preparing for a scheduled NATA audit, the Forensic DNA Analysis' Quality Supervisor, Dr Kirsten Scott, prepares a standard Assessment Information Document (AID) that includes any instrument and software changes since the last NATA audit. This is provided to the Forensic and Scientific Services (FSS) Quality Advisor who provides it to NATA. For example, JH-6 NATA AID_2022 is an AID document from the NATA Audit in 2022 that describes (on page 6) the projects conducted since the last visit in December 2020. The attachment JH-7 Snip of Projects for NATA_2022 shows the Project Experimental Designs and Reports provided to NATA within the AID for the 2022 Audit.

- 13. I am aware of a Report by the Institute of Environmental Science and Research (ESR-NZ) into the Quantifiler procedure in 2005 and it was to 'examine issues relating to the validation and use of the Quantifiler Human DNA Quantification system' (JH-8 Report by ESR).
- 14. I am not aware of any internal audits of validation proposals or reports.

Explain how staff are chosen to validate and endorse a validation.

- 15. Staff can raise topics that could eventuate into Project Proposals initially by discussing with their Line Manager. Appendix 1 of JH-4 22871v17 Change Mgt SOP describes this workflow. Management Team members are endorsers of Proposals and Reports within the Change Management process.
- 16. It a staff member raises an Initial Request within the Change Management Procedure (JH-4 22871v17 Change Mgt SOP) it is usually, but not always the case that the staff member initiating the change is the lead or co-project officer.

Question 8

Explain who chooses the staff to validate and endorse a validation.

- 17. Staff who raise Project Proposals and lodge an Initial Request through the Change Management process are usually the staff members involved in the validations. This may not always be the case, as it depends on their availabilities to perform the work, considering leave and work priorities. Alternative and/or additional staff members could be identified through Career Success Plans (CSP) where staff discuss with their Line Managers any development opportunities that they may wish to pursue.
- 18. Endorsement of Proposals and Reports are performed by Management Team members (substantive or those performing Higher Duties at the time of project proposal/report due dates). I haven't known of any other staff outside of the Management Team performing endorsement duties, except if they are temporarily performing Higher Duties.

Explain if and how you became involved in endorsing the validations of:

- (a) PowerPlex21 (2012 and 2013);
- (b) STRMix (Project #105 and #151);
- (c) 3130xl B Genetic Analyzer;
- (d) Quantifiler Trio (Project #152);
- (e) Quant Studio 5 (Project #185);
- (f) QIAsymphony (project #192);
- (g) ProFlex (Project #199);
- (h) Hamilton STARlet A (Project #173);
- (i) 3500xl Genetic Analyzers (Project #182 and #186); and
- (j) any method for the cleaning of bone instruments.
- 19. As a Management Team member, I was an endorser of these documents.
- 20. Please note the bone cleaning project was #148 Cleaning bone processing equipment.

Question 10

Explain the extent of your involvement in the endorsement of the validations listed in point 9.

Attach any relevant documentation.

21. The extent of my involvement was to provide feedback on the plan (including risk assessments if applicable), proposal and final reports. After all feedback had been considered by the project team, I then signed the reports as an endorser.

Ouestion 11

State any timeframe you were given to provide feedback and your endorsement of the validations listed in point 9.

- 22. Of the projects in Point 9, the only timeframe that I have been able to find from the relevant project folder is for Project #152. The date for feedback was 29/01/2015 and I provided endorsement on 18/09/2015.
- I cannot determine from the other electronic project folders when the feedback was required.

Question 12

Explain any issues or concerns you had with any validations listed in point 9 and explain whether and how these issues were adequately addressed by the validator and/or other staff endorsing the validation. Attach any documentation evidencing the raising of any concerns and any response to your concerns.

24. I did not have any issues or concerns with the projects listed. I provided general feedback and I am confident it was considered. I understand that this does not necessarily mean it was always incorporated in the final report.

Question 13

Explain how you determined that each validation was completed successfully, appropriately and in accordance with international best practice.

25. In reviewing the work, I consider the methodology and that variables are considered. I consider these alongside my understanding of validation requirements from NATA documentation.

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Explain whether you were involved in any endorsement of subsequent validations of the validations listed in point 9.

26. There weren't any further validations of the ones mentioned in Point 9, other than for PowerPlex 21 where there was a second version. I was an endorser for both versions.

Feedback

Question 15

Explain whether any feedback, advice or direction from other staff impacted on your endorsement of the validations listed in point 9.

27. No.

Concerns

Question 16

Outline any concerns you have with the validation or endorsement process within the DNA Analysis Unit. Attach any documentation, if any, evidencing these concerns being raised.

- 28. I am aware some projects have not had feedback received by due dates, which is a concern and I would consider this an area that could improve. For example, in Project #184, the Project Proposal was distributed to the Management Team on 31 July 2017 (JH-9 FW_Proposal #184) and one staff member (Kylic Rika) did not meet the duedate of 17 August 2017. This was followed up and the review was completed on 30 August 2017 (JH-10 Microcon project KDR).
- 29. Attachment JH-11 Compiled presentation (slide 26) shows a point around projects at a Management Review in 2021 and being conscious to meet the targets that are set.

Sperm microscopy - past policies and procedures

Question 17

Explain when and how the suspension method was introduced for the preliminary testing of samples of suspected seminal fluid? Attach relevant validations for the new method.

- 30. I don't recall exactly when the suspension method was implemented as this is not recorded in the Change Register. It appears the first time it is described in the appropriate SOP is in 2010 (JH-75 17189v10 -Exam for sperm). This was after a comment was added to 17189v9 on 14 December 2009 (JH-13 Snip_comment to SOP).
- 31. I am unable to locate a distinct project validation document and testing outcomes, but the approved Part A Documentation- Change Proposal #31 describes the request for change and that the proposal is in conjunction with other change proposals (JH-14 Approved Part A #31; JH-15 Change Proposal #24; JH-16 Change Proposal #28; JH-33 Change Proposal #27).

Ouestion 18

Explain whether there were any subsequent changes to the suspension procedure between its implementation and 2016? If so, identify those changes, the reasons for them, and whether a validation was conducted for the changes.

32. I am unable to locate any documentation describing a change in process between the implementation (unknown exact date) and 2016. I have located Project #78 documentation that describes testing of a new ABA card p30 test kit for the detection of seminal fluid (JH-18 ABAcard p30 final report). This document describes the validation of the test kit using the same suspension procedure.

Witness —

Identify the Standard Operating Procedures (SOPs) (including version numbers) related to the detection and testing of spermatozoa that were in force as at January 2016.

17189v13 Examination for and of Spermatozoa – active from 29/07/2015 (JH-12 17189v13 Exam for sperm active 2016).

Question 20

Explain your understanding of the process and procedure in January 2016 for testing samples suspected to contain spermatozoa, including the use of preliminary and presumptive testing and policies concerning when the testing should cease.

- 34. The processes vary depending on the items being examined, should they be received eg. Sexual Assault Investigation Kits, or large pieces of fabric).
- 35. A general process for a Sexual Assault Investigation Kit (SAIK) is described. The process as per the SOP at the time (JH-12 17189v13 Exam for sperm active 2016 and JH-19 32106v3 Exam of Sexual Cases) describes that microscopy slides are prepared from a suspension of nanopure water of approximately 100-300uL. The material might either be a scraping, excised material or a swab. The sample is vortexed (agitated vigorously) and a drop is applied to a microscope slide, heat-fixed, stained and examined under a microscope for the possible presence of spermatozoa. A positive control slide is also prepared daily (and at other times) to ensure the process is operating correctly.
- 36. If spermatozoa is detected, the sample is submitted for DNA extraction. If spermatozoa is not detected, the suspension is tested for the possible presence of seminal fluid (ie with undetectable or absent sperm). The Acid Phosphatase (AP) test is one presumptive test for seminal fluid where a drop of the suspension (after centrifuge) is applied to filter paper with a drop of AP reagent. A colour reaction indicates the possible presence of seminal fluid. If AP negative, the sample is not submitted for testing unless it is an external swab (eg. vulval swab) which will be submitted for DNA profiling as per paragraph 37. If the AP test result is positive, the suspension undergoes a second

presumptive test for seminal fluid: p30 test. This operates similar to a COVID-19 rapid antigen test which develops a positive line if the p30 protein is detected. If a line is not detected, the sample does not progress to DNA profiling unless in situations described in paragraph 37.

37. Relevant policies include the following from SOP 32106v3 (JH-19 32106v3 Exam of Sexual Cases):

Female SAIKs which are semen negative, any external intimate swabs are submitted for cells (i.e. vulval and perianal). Where the complainant is a minor or has an intellectual impairment which may mean that the provided case history is unreliable, all possible offence scenarios are considered. Where the complainant is an adult who has lost consciousness, has impaired memory or has consumed alcohol or drugs prior to or during the offence which may impact on memory, all possible offence scenarios are considered. Consider previous intercourse with same or different partner, prior to the offence. For digital only female complainant cases with prior intercourse, submit external swabs for DLYS with no testing. For male SAIK swabs, consider submitting penile swabs for DLYS where previous intercourse with another partner has occurred.

Consider the number of offenders – for male SAIKs consider submitting penile swabs for DLYS (with no testing) to separate epithelial and spermatozoa. For child complainants, treat all vaginal swabs as external swabs for semen or cells. Samples taken from areas of biting, licking or kissing (or other oral contact) are submitted for CSUP. This does not include swabs taken from the mouth (internal or external), anal and vaginal areas which may give false positive results.

38. A policy that also exists relates to the outcomes of Project #92 (JH-20 Report – Proposal#92) where, based on data, a decision to not routinely perform DNA profiling on epithelial fractions from SAIKs was made. These are fractions from the DNA extraction process that are likely to contain biological material from the donor (eg. DNA from the female from whom the genital swabs were taken).

Sperm microscopy - 2016 and project 181

Question 21

Explain how you first became aware of the issue related to a discrepancy in the levels of spermatozoa detected during evidence recovery microscopy compared to the levels detected during differential lysis microscopy (the sperm microscopy issue).



39. I recall an email from Reporting Scientist Jacqui Wilson that was sent to Amanda Reeves (Jacqui's line manager at the time) and myself as carbon copy. This email was commented on by Amanda Reeves (JH-21 RE_599195993_AJR). I spoke to Luke Ryan who I understand was A/Team Leader Evidence Recovery & Quality at the time and replied to Jacqui and Amanda on the same date and forwarded to Luke Ryan (JH-22 FW:599195993 and JH-23 RE_599195993_JAH).

Question 22

Explain your understanding of the sperm microscopy issue at the time it was raised.

40. My understanding from the details in Jacqui Wilson's email is that sperm were not detected at the examination phase, but were detected with a grading of 3+ (ie. 'very easy to find') as per (JH-12 17189v13 Exam for sperm active 2016) in the slide prepared within the DNA extraction process. The risk was that a sample might have no sperm detected, and then test negative for AP and p30 (see paragraph 36) and potentially not be submitted for DNA profiling.

Question 23

Explain whether the management team at the DNA Laboratory was made aware of the issue. If yes, explain when and how.

 Yes. Jacqui Wilson sent an email to Amanda Reeves and carbon copied myself (see paragraph 38).

Question 24

Identify your role in responding to the sperm microscopy issue. Identify if any other person was also responsible for responding to or actioning the sperm microscopy issue.

42. As per paragraph 39, I made A/Team Leader Evidence Recovery and Quality Luke Ryan aware of the sample situation on the same date that Jacqui Wilson sent her email (JH-22 FW:599195993). The examination for spermatozoa, either in the examination

phase or the analytical phase, is a task for scientists within the Evidence Recovery and Analytical Team. The senior staff members in the team at that particular time were Dr Kirsten Scott (Quality and Projects Senior Scientist) and Allan McNevin (Evidence Recovery Senior Scientist).

Question 25

Identify the OQI or Adverse Event Entry, if any, raised in relation to the sperm microscopy issue. Attach any relevant documentation of either if raised.

43. I am not able to locate an Adverse Event entry, nor an OQI.

Question 26

Explain your role, if any, in the design, execution and reporting of results from each of the projects undertaken during or after 2016 in relation to the sperm microscopy issue.

- 44. My role was as a Management Team member was to review and comment where necessary on any relevant documentation.
- 45. I was also asked to look at some Data Analysis Reports that were prepared by Team Leader Paula Brisotto, Reporting Scientist Matthew Hunt and Senior Scientists Kylie Rika and Luke Ryan in August 2017 (JH-24 Data Analysis report, JH-25 Data Analysis report_draft1, JH-26 Data Analysis report_draft1 LBR_MOH_PMB and JH-27 Data Analysis report_draft1_LBR track changes). I was also asked to proofread an email and spreadsheet for Paula Brisotto in May 2017 (JH-28 Proof read).

Witness

Explain how Project 181 was proposed and how it commenced.

- At a Management Meeting on 12 May 2016, Allan McNevin raised the point on 'Sperm seen on Diff Lysis extraction slide vs ER suspension slide' (JH-29 Mgt Meeting 12052016). On that same day, Allan McNevin wrote an email to the Management Team with the information that there would be a Proposal (#181) called "Investigation into sensitivity of spermatozoa microscopy" (JH-30 FYI Project proposal#181). Also on that day, I sent Allan McNevin an email with some ideas from reporting staff that were shared with Amanda Reeves and then myself that could potentially assist with any projects (JH-77 RE_Diff lysis).
- 47. The Change Management and risk assessment documentation were sent to Management Team on 01 September 2016 (JH-31 project #181 – project plan and experimental design).

Question 28

What role did Allan McNevin take in responding to the sperm microscopy issue, and the reasons for his involvement?

48. Allan McNevin was the Senior Scientist of the Evidence Recovery Team at the time. In this team, scientists perform the task of preparing and examining microscope slides for the presence of spermatozoa. His role ended up raising the Change Proposal #181 and leading the work involved.

Question 29

If the issue was dealt with by way of developing project proposals and conducting projects, why was it dealt with in this way? Why was it not dealt with in a different way (for example, by use of the OQI process or Adverse Event Log)?



49. I can't recall the reasons why or conversations on the approach. I suspect that the decision was made within the Evidence Recovery and Quality Team. Having a higher reading of sperm on slides prepared during the differential lysis procedure, compared with the slides prepared at evidence recovery, is not an unexpected finding due to the concentrated sample at extraction. My assumption is that this in itself is not evidence of a systemic failure of the system that would be raised as an OQI.

Question 30

Explain whether any other staff members in the management team disagreed with your approach to handing the sperm microscopy issue. If so, identify who and what was communicated. Explain whether you consider their disagreements were reasonable and affected your proposed approach and why.

50. I am not aware of any staff members in the management team commenting on (including disagreeing with) my approach to handling the sperm microscopy issue.

Question 31

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Provide an explanation of the document entitled 'Data analysis of modified sexual assault process for zero spermatozoa detected at Evidence Recovery'. Identify:

- (a) the aim of this report;
- (b) who approved this report, if anyone;
- (c) your role and/or involvement, if any, in the formulation of the report, including drafts;
- (d) what was work was conducted pursuant to the report, and over what period the work was conducted; and
- (e) the results and conclusions of this report; and
- (f) whether the paper was finalised.

Witness



- 51. The aim of the project was to evaluate the set of samples, after the implementation of a modified protocol in August 2016, which had no spermatozoa or seminal fluid detected during the initial Evidence Recovery examination, and which were then submitted for differential lysis extraction. It was to evaluate these findings against what might have been obtained with the pre-August 2016 protocol to determine what, if any, impact there may have been on the DNA results reported for the case.
- 52. I am not aware if there was an official final report.
- I was emailed a short report from Paula Brisotto in February 2017 on some data that was obtained after the implementation of the modified protocol in August 2016 (JH-32 Microscopy stats FYI). I was sent a spreadsheet and an email to proofread before sending to Kylie Rika and Matthew Hunt (JH-28 Proof read). I was also asked to read a Data Analysis draft report, and other versions with feedback from Kylie Rika, Luke Ryan and Matthew Hunt (JH-24 Data Analysis report, JH-25 Data Analysis report_draft1, JH-26 Data Analysis report_draft1 LBR_MOH_PMB and JH-27 Data Analysis report_draft1_LBR track changes).
- 54. I believe the work was from approximately February to approximately August 2017 as determined by the dates of tracked changes/ comments.
- 55. The results found that there was not a demonstration of a systemic failure in the examination of exhibits when there were no sperm detected at Evidence Recovery phase, compared to slides prepared during Differential Lysis Extraction.
- 56. I am not aware if the final report was finalised; it appears to be in draft.

Provide an explanation of document entitled 'Project #181 Spermatozoa Microscopy Sensitivity'. Identify:

- (a) the aim of this project;
- (b) who approved this proposal, if anyone;
- (c) your role and/or involvement, if any, in the formulation of the report, including drafts;

Witness

- (d) what work was conducted pursuant to the project, and over what period the work was conducted; and
- (e) the results and conclusions of the final report.
- 57. The aim was to investigate the performance of the 'current' Evidence Recovery microscopy slide preparation process, and to compare to a newly 'proposed' method process.
- It appears the first Project Proposal was approved by Managing Scientist Cathie Allen on 13 October 2016 (JH-34 Signed Project Proposal).
- My role was as a Management Team member reviewing and endorsing plans and reports.
- 60. It appears work was conducted from the Initial Request in June 2016, through the submission of many Proposal parts, and then a final report that was approved on 5 August 2020.
- 61. The final report recommended that the 'proposed' method, developed and tested throughout the project, be implemented as a standard operating procedure. It also recommended the cessation of AP testing as a standard presumptive screening technique except in cases of screening large items for potential seminal stains. P30 testing alone was recommended as the standard presumptive screening technique.

Between 2016 and 2021, did you or another person determine why low levels of spermatozoa were detected on the Evidence Recovery slides even when significant amounts were detected and differential lysis:

- (a) if yes, what was the cause?
- (b) if no, was the attempt to find such a cause abandoned at some point? If so, advise when it was abandoned and explain the reasoning for that abandonment.
- 62. I didn't perform any data analysis of this type and I can't recall if there were more analyses other than the project itself, and the Data Analysis draft report from 2017.

Explain when and on what basis Project 181 concluded. Include any discoveries made from Project 181.

- 63. The report recommended that the 'proposed' method, developed and tested throughout the project, be implemented as a standard operating procedure. It also recommended the cessation of AP testing as a standard presumptive screening technique except in cases of screening large items for potential seminal stains. P30 testing alone was recommended as the standard presumptive screening technique.
- 64. The final report was endorsed by the Management Team and then approved by Cathie Allen on 5 August 2020.
- 65. The report was submitted for Ethics Approval with the view to publication. The findings were compiled and presented as a poster at the Australian and New Zealand Forensic Science Symposium in 2022.

Question 35

Explain whether you consider Project 181 adequately addressed the sperm microscopy issue.

66. I consider the project adequately addressed the sperm microscopy issue as it led to a method with improved sensitivity.

Question 36

Explain whether any other staff expressed concerns or disagreements with the approach taken to address the sperm microscopy issue during Project 181's completion. Identify each staff member and explain the nature of their concerns or disagreements.

67. I don't have a recollection of whether there have been concerns or disagreements raised with the approach.

##



Explain any changes to Standard Operating Procedures that occurred as a result of the sperm microscopy issue and/or any part of Project #181. For each change (if any), identify:

- (a) which SOP was changed;
- (b) how it was changed; and
- (c) when the change occurred.
- 68. QIS 34006v3 Procedure for the Release of Results Using the Forensic Register was updated with slight change to the Appendix for statements (JH-35 RE_Slight Appendix change). This was from a comment added 28/04/2021. The SOP was then updated with the comments and approved 22/07/2022 as QIS 34006v4.
- QIS 17189v17 Examination for and of Spermatozoa was updated from v16 to reflect the current processes.
- 70. QIS 33798v6 had extensive comments to change the SOP as per JH-36 Snip of comments to change SOP 33798v6. These were then incorporated into QIS 33789v7 which was activated 15/04/2021.
- 71. QIS 17186v14 had changes to the SOP due to no longer performing the test on substrates as per JH-37 Snip of comments to change SOP 17186. This was incorporated into v15 which was activated 27/06/2022.

Question 38

Explain whether there was any attempt to identify samples with suspected spermatozoa where DNA testing had been stopped prior to differential lysis, at least partly because of low levels of spermatozoa detected during microscopy of evidence recovery slides. If so, provide a list of cases reviewed or identified, and explain what action was taken in respect of those matters. If no, explain why not.

72. I am not aware of an attempt to find such samples. If low levels of spermatozoa were detected during microscopy of evidence recovery slides, the samples would have progressed to a differential lysis extraction.

Explain if any workplace culture/environment issues (for example, personality clashes or communication issues between individuals at FSS, favouritism, productivity etc.) impeded the efficient resolution of the sperm microscopy issue. If so, provide any examples or attach any relevant documentation.

73. The interpersonal difficulties made the environment challenging to work in, on top of the challenge for staff to balance the daily work commitments with project work. My view was that the interpersonal difficulties made it difficult to progress this project positively and affected the efficient resolution of the issue. This particular project also evolved to become an extraordinary amount of work as findings were discovered and worked upon, as evidenced by the project eventually become four experimental designs (and a second version to Part 4). Multiple staff from across the teams eg. Allan McNevin, Emma Caunt, Chelsea Savage, Matthew Hunt, Paula Brisotto and Cathie Allen all have their names listed on various Experimental Design parts (JH-38 Snip_Expntl designs 181).

Question 40

Explain your knowledge and involvement, if any, into procuring and engaging the New Zealand Institute of Environment and Science and Research ("ESR") to conduct an independent review, or provide an opinion about, the processing sexual assault investigation kits (SAIKS) at the QHFSS Forensic DNA Analysis Laboratory in 2016 and 2017, including:

- (a) who proposed the review;
- (b) the purpose of the review;
- (c) determining the scope of the review;
- (d) developing and finalising the Terms of Reference for the review sought;
- (e) the preparation of the documents and/or production of the documents considered to develop the Terms of Reference;

- (f) who determined what documents should be provided to ESR in relation to the review or opinion sought;
- (g) the documents provided to ESR relating to the review or opinion sought;
- (h) whether you saw the final report and the conclusions stated therein; and
- (i) were changes implemented at FSS in line with the conclusions stated in the final report. If so, identify and attach any relevant documentation evidencing the changes.
- 74. My only knowledge is that there was a desktop review of SOPs relevant to the examination of SAIKs and other items relating to alleged sexual assaults conducted by ESR. I didn't know about this until Paula Brisotto and I received an email with an attached report on 4 January 2018 (JH-39 Report and JH-40 ESR Report 11 April 2017 -admin release).
- 75. I don't know who proposed the review.
- 76. In being provided the report, I can ascertain that the purpose of the review was to determine if the protocols and methods were fit for purpose.
- 77. I had no involvement in determining the scope of the review.
- I had no involvement in developing and finalising the Terms of Reference for the review.
- 79. I had no involvement in the preparation of the documents and/or production of the documents considered to develop the Terms of Reference.
- 80. I had no involvement in determining what documents should be provided to ESR in relation to the review or opinion sought.
- I was provided the report on 4 January 2018 in an email from Cathie Allen (JH-39 Report and JH-40 ESR Report 11 April 2017 -admin release).
- 82. I am not aware if any changes were implemented at FSS after receipt of the report.

Explain and detail your knowledge and involvement, if any, in the decision that made to engage Livingstones to externally investigate the workplace allegations raised by Amanda Reeves, including:

- (a) your knowledge of who proposed the investigation;
- (b) your participation in and/or knowledge of any conversations in which the following was raised:
 - (i) the reasons for the investigation;
 - (ii) the scope of the investigation;
 - (iii) the intended or expected outcome from the investigation; and
 - (iv) why an external investigation was preferred instead of an internal process.
- 83. I have very limited recollection of Livingstones and what it entailed. I am not aware of who proposed the investigations.
- 84. I don't recall the exact reasons and scope of the investigation.
- 85. I don't recall the intended or expected outcome of the investigation.
- 86. I didn't propose the external investigation, but I agree that an external investigation is an acceptable course of action to take to investigate workplace issues.

Question 42

Explain and detail your knowledge and involvement, if any, in the decision that was made that Amanda Reeves should return from her leave of absence in March 2017 to undertake an alternate research role instead of her substantive role as a reporting scientist, including:

- (a) your knowledge of who proposed the arrangement;
- (b) your participation in and/or knowledge of any conversations in which the following was raised:

- the impact of Amanda Reeves' expressed concerns about the processing of spermatozoa samples to her suitability to perform the functions of her substantive role; and
- (ii) the impact of Amanda Reeves' expressed concerns about workplace issues to her suitability to perform the functions of her substantive role.
- 87. I have no knowledge of who proposed the arrangement. I was asked to inform the Management Team and Reporting Team 1 (that Amanda previously managed), and to personally inform a staff member (Jacqui Wilson) prior to Amanda's work at FSS (JH-76 Re_Next week).
- 88. I have no knowledge of conversations regarding (i) nor (ii).

DNAIQ

Question 43

Explain what DNAIQ is. Explain the way or ways in which the DNA laboratory used DNAIQ, and/or DNAIQ system(s), as at the start of 2008.

- Promega DNA IQ (DNAIQ) is DNA extraction chemistry that in 2008 was used to extract DNA from biological samples.
- 90. The chemistry was used within an automated DNA extraction of cell and blood samples on the PerkinElmer MultiPROBE® II PLUS HT EX with Gripper™ Integration platforms. There was also a manual method that was a back-up method should it be required (JH-41 24897v2_DNAIQ Extraction).

Question 44

Explain what problems with DNAIQ were experienced in approximately 2008. Explain, to the best of your knowledge, how these problems were first detected.

- 91. From viewing Opportunities for Quality Improvements (OQIs) and the Analytical Issues Log (JH-42 Analytical Issues Log Adverse event log worksheet), it appears to me that a mixed DNA profile in a Reference sample was identified on 11 February 2008 and was then recorded as OQI 19330.
- There were other instances of OQIs raised where unexpected DNA profiles were obtained in negative controls registered as OQI 19349 and OQI 19477.

Identify each OQI and adverse event that relates to DNAIQ problems at around this time, or has since been linked to DNAIQ problems from around this time.

93. From what I can obtain from searching electronic records, I have been able to locate a timeline that lists the OQIs as (JH-43 DNA IQ timeline 12-11-2008):

OQI 19330

OQI 19349

OQI 19477

OQI 19767

OQI 19768

OQI 20231

OQI 20351

OQI 20422

OQI 20437

OQI 20615

OQI 20617

OQI 20690

OQI 20925

OQI 21222

OQI 21309

- 94. In checking the list in paragraph 92 to QIS2, I was unable to access the record relating to OQI 20615. I have still included this number here as this was in the record I could find.
- 95. I located a document that appears incomplete (JH-79 OQI report v0.4). This report lists three additional OQIs as 22880, 22882 and 19703. I am unable to find the 22880 file in QIS2.

What actions did the management committee and/or staff at the DNA laboratory take in response to the discovery of the problem? Provide a clear timeline which covers the problems identified, the decisions taken in response and by whom, and how those decisions were implemented.

- 96. From the Management Meeting notes from 10 April 2008, there is a mention of an email in section 3.8 of (JH-44 10_04_2008_Mgt Meeting), that there was an email circulated. I am unable to locate this email from this time period. The meeting minutes describe information that will be retained in the Analytical Issues Log (JH-42 Analytical Issues Log Adverse event log worksheet).
- 97. It appears this then stayed on the agenda and expanded further as per 20 June 2008 minutes item 2.3 (JH-45 20_06_2008_Mgt Meeting). This is when the Quality Supervisor (Robyn Smith), Analytical Team Senior Scientist (Allan McNevin) and the Analytical Team had met to develop a plan to deal with mixture investigations, Quant/Profiles in neg controls. I believe these related to the issues experienced at the time.
- 98. An extraordinary Management Team Meeting on 14 July 2008 was held and actions devised; however, I am unable to locate the minutes. The details are written into the Action Details of JH-46 OQI 19477 Report. It is listed in JH-43 DNA IQ timeline 12-11-2008 (row 8) that a memo was sent to all DNA analysis staff outlining DNA IQ issues on 14 July 2008. I am unable to locate this memo to staff.
- Audit 8227 performed on 15 July 2008 by Iman Muharam, Peter Clausen and Amy Cheng (JH-47 Audit 8227 DNA IQ FINAL).
- Audit 8752 was performed on 28 July 2008 by Susan Brady (JH-78 Audit 8752 Report).
- 101. A summary of actions was provided in the OQI 20351 details as follows (JH-48 OQI 20351):

As a result of previous OQI's raised and concerns identified around the automated DNA IQ extraction process, the extraction of samples using the automated DNA IQ procedure was halted on the 28-7-2008. Prior to this, Audit 8227 had been commissioned and

carried out. A number of areas for improvement were identified through the audit, and these have been implemented or are under investigation as outlined in OQI's 20367, 20368 and 20369. After the cessation of the automated DNA IQ extraction protocol, a review of all batches processed through this protocol was carried out by a specially commissioned team. A number of potential contamination events were identified and each is to be investigated on batch-by-batch basis. Additionally, careful review of results obtained from samples processed through the automated DNA IQ extraction procedure prior to reporting will be carried out. Every DNA result obtained from these samples will be interpreted with caution. Modifications have been made to the automated DNA IQ extraction procedure (including the use of an alternative to the adhesive seal and an alternative resin mixing procedure). This modified procedure is undergoing extensive verification and approval from the DNA Analysis management team must be obtained prior to re-introduction. The contamination events and concerns and improvements etc. that surround the automated DNA IQ extraction procedure have been discussed at various departmental and team meetings.

- 102. Checklists were prepared it appears by Paula Brisotto (nee Taylor) and Emma Caunt in September 2008 (JH-49 Appendix 1- Checklist 1 and JH-50 Appendix 1- Checklist 2).
- 103. A Team was devised of Reporting Scientists (called Investigation Team) who worked with checklists to determine whether DNA profiles had passed Quality Control checks or not. The members of the team included Alicia Quartermain, Jacqui Wilson, Shannon Merrick, Julie Connell, Rhys Parry, Angelina Keller and Claire Gallagher (JH-51 Way forward team divisions_Oct2008 and line 208 of JH-52 Change Register). Later a process using a macro was developed to improve the checking process (JH-53 EB macro workflow).
- 104. External auditors were engaged by Senior Director Greg Shaw to review procedures pertaining to extraction. The report by Dr Theo Sloots and Dr David Whiley was dated 14 November 2008 (JH-54 External Auditors report Nov 2008).
- 105. In December 2008, the A/Managing Scientist of the DNA Analysis Unit and the Senior Director of Queensland Health Forensic and Scientific Services advised the Director of Public Prosecutions, Executive Director and two Principal Crown Prosecutors of DNA testing that had been conducted in a period where some results were the subject of an adverse event (JH-55 Cover Letter for all Statements).

- 106. Advice was sought from Crown Law by Managing Scientist Cathie Allen and Senior Director Greg Shaw and that advice (from Robert Hutchings) was received in December 2008 (JH-56 DNAIQ Crown Law Advice). This advice contained the joint opinion from Solicitor-General Walter Sofronoff QC and Peter Davis SC which related to advice on how to disclose the issues in court statements.
- 107. Further advice was received from Peter Davis SC in February 2009 with commentary on paragraphs to include in statements, and advice on where to position the explanatory paragraphs detailing the types of contamination and strategies used in the case. This advice also recommended a cover letter to be appended to all statements (JH-57 Adv Crown Law 23-2-09 and JH-55 Cover Letter for all Statements).
- 108. The Change Register shows on line 290 that the Extraction platform B had been implemented for the automated DNAIQ extractions of volume crime casework samples on 20 August 2009 (JH-52 Change Register).

Was the cause of the issues or problems relating to DNAIQ identified? If yes, what was it?

109. As a summary, I recall some of the likely causes of the detected contamination to have occurred during storage, during plate agitation on the robot, or by the removal of plate seals. I recall the sense that the major reason was an ineffective seal over a plate of samples used in the process, which appeared to cause seepage of DNA extract from one well/spot to another. Another reason described that it may have occurred by robotic dripping during elution, during capping/decapping, during storage or during the preparation of amplifications (JH-48 OQI 20351_report).

Question 48

What immediate action was taken after the cause of the issues or problems was identified?

110. Extensive testing commenced and resulted in a Change Proposal #56 for the reimplementation of automated DNAIQ extractions.

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Witness

111. The extensive testing regime included a number of update presentations, one example is JH-58 MPII Enhancements Update 20081113.

Question 49

Outline your role in responding to issues with DNAIQ, and any audits completed in relation to any OQI concerning DNAIQ. Provide an explanation of the findings of each issue and actions taken in response to those issues. When were the follow up actions finalised?

- 112. I did not do any audits in relation to the issues experienced. My role as a Management Team member was to review Change Management documentation when issued.
- 113. I am unable to locate any OQIs that I was listed as an approver for.
- 114. My role was in working with case managers to develop checklists and macros and to ensure appropriate paragraphs were included in statements.

Question 50

Identify any issues, if any, concerning the contamination of samples encountered in the R v Grant Westley Meredith case (reference: QP800109982). In doing so, explain:

- (a) Your involvement and the steps you took in respect of the matter;
- (b) The issues encountered;
- (c) How were the issues detected;
- (d) What was the cause of the issues;
- (e) What action did you take once the issues were identified.
- In answering this question, I have only accessed the electronic record in the Laboratory Information System at the time (AUSLAB), which contains copies of the statements (x3) issued. I have not been able to access the paper casefile.

- 116. I was the Reporting Scientist for this matter.
- 117. This case had an initial statement issued 2 September 2008. In reporting this case, the checklist was followed as per the specimen notes for Item 288908564 (among others), and the determination was that QC Checklist Failed ie. the sample did not pass the checklist criteria. The relevant OQI was 19477 (JH-46 OQI 19477 Report).
- 118. After the advice from Mr Peter Davis SC in February 2009 (JH-57 Adv Crown Law 23-2-09) in relation to the appropriate paragraph location to explain any issues, I issued a replacement statement on 26 May 2009 with the relevant paragraph added to the body of the statement (below). The affected samples did not have any remaining sample for retesting.

Testing for this case has been conducted in a period where some results were the subject of adverse events. An adverse event includes a deficient operation or function identified to have occurred within the automated extraction process utilised in testing during the period October 2007 and July 2008. Within this case, the adverse event is demonstrated to have affected a result or results and the integrity cannot be verified. There was no remaining sample for retesting to be conducted. These results have therefore been reported as follows 'these samples did not pass our Quality System requirements at the DNA analysis stage and therefore the DNA profiling results relating to these samples cannot be reported'.

- 119. According to the statement, some samples from Item 261634308 leaves and twigs (13), and Item 334050109 'Bloodstained leaves' '11' were reported as:
 - These samples did not pass our Quality System requirements at the DNA Analysis stage and therefore the DNA profiling results relating to these samples cannot be reported.
- 120. A third statement was issued for this matter on 13 November 2008 which detailed results that were not available at the time of issuing the initial statement (2 September 2008).
- 121. I reported the results that failed the checklist as 'Quality Control Failure Results not reportable'. I reissued the initial statement to insert the approved paragraphs. I provided evidence in court on 2 June 2009 and was asked a number of questions on the event under cross-examination.
- 122. This details of OQI 19477 describe a DNA profile was unexpectedly found in a negative control sample (which is not expected to have a DNA profile). This profile was searched against the batch and was found to match several other samples, including 288908564

Witness Witness

from this matter. There were some samples with the same DNA profile on the batch with high quantification values and one or more of these samples were the most likely sources of the contamination.

123. The cause of the issue, which was part of a system issue, was most likely related to an ineffective seal that caused sample to transfer from one location to another.

Question 51

Identify each staff member involved in detecting and responding to the problems with DNAIQ, and the nature of each person's involvement.

124. As per information from the Quality Information System (QIS), I have been able to locate the following information for the OQIs raised and mentioned in paragraph 93:

OQI	Raised by	Investigated by	Action by	Approved by
OQI 19330	Allan McNevin	Allan McNevin	Allan McNevin	Cathie Allen
OQI 19349	Allan McNevin	Quality Investigation System	Allan McNevin	Cathie Allen
OQI 19477	Amy Cheng	Quality Investigation System	Allan McNevin	Cathie Allen
OQI 19767	Maria Aguilera	Quality Investigation System	Allan McNevin	Cathie Allen
OQI 19768	Maria Aguilera	Quality Investigation System	Allan McNevin	Cathie Allen
OQI 20231	Chiron Weber	Quality Investigation System	Allan McNevin	Cathie Allen
OQI 20351	Kylie Rika (NB Helen Gregg performed	Quality Investigation System	Allan McNevin	Paula Brisotto

	followup due to Kylie's absence)			
OQI 20422	Justin Howes	Quality Investigation System	Allan McNevin	Paula Brisotto
OQI 20437	Amanda Reeves	Quality Investigation System	Allan McNevin	Paula Brisotto
OQI 20615	Unable to access in	QIS2		
OQI 20617	Rhys Parry	Quality Investigation System	Allan McNevin	Paula Brisotto
OQI 20690	Jacqui Wilson	Quality Investigation System	Allan McNevin	Paula Brisotto
OQI 20925	Justin Howes	Quality Investigation System	Allan McNevin	Paula Brisotto
OQI 21222	Julie Connell	Quality Investigation System, Thomas Nurthen	Thomas Nurthen	Paula Brisotto
OQI 21309	Thomas Nurthen	Quality Investigation System	Allan McNevin	Paula Brisotto
OQI 22880	Unable to find record in QIS2.			
OQI 22882	Adrian Pippia	Thomas Nurthen	Thomas Nurthen	Paula Brisotto
OQI 19703	Allan McNevin	Quality Investigation System	Allan McNevin	Vanessa Ientile

Identify whether any issue or problem with respect to DNAIQ was audited by an external agency? If yes, when did that occur and in respect of what particular issue or issues. Who decided that should occur? Provide:

- (a) instructions:
- (b) list of material; and
- (c) the report, including any drafts reports.
- 125. External auditors were engaged by Senior Director Greg Shaw to review procedures pertaining to extraction. The report by Dr Theo Sloots and Dr David Whiley was dated 14 November 2008 (JH-54 External Auditors report Nov 2008).

Question 53

How were the results of the audit by the external agency communicated to the DNA laboratory?

- 126. The external auditors provided a report on 14 November 2008 (JH-54 External Auditors report Nov 2008).
- 127. Details were provided by Iman Muharam to the Management Team on 14 November 2008 with details as follows:

Visit by External Auditors (12/11/08) – The Auditors looked at off deck lysis, storStar, programming end to end, platforms, OQI – what we did / processes, reporting, analysis, timelines. They identified no areas of risk, and complimented our staff. The auditors agreed with our actions taken and basic principles.

A report will be issued from the external Auditors. Some recommendations are – Locked batches – CJA and Iman to explore this

Reagents to be tested at 35

Strip Cap seals for PCR Plates - Iman will source these.

of QC plates per month (i.e. checkerboard)

Question 54

What permanent changes, or amendments to SOPs, were made as a result of identifying the problems related to DNAIQ?

128. QIS 17119v7 Release of Results SOP was updated to contain the paragraphs describing the different categories of samples to be added to statements as per legal advice. QIS

- 17119v8 was updated to include the Extraction Batch checking process used to detect possible contamination.
- 129. QIS 24897v5 DNAIQ Method of Extracting DNA from Casework and Reference Samples was updated to 'reflect changes in procedure as an outcome of internal and external audits'.

Explain what communications were made to external agencies, including the Queensland Police Service, the Office of the Director of Public Prosecutions, and the Queensland Courts, about the problems with DNAIQ and when the communications were made. Attach copies of any emails or letters sent to the external agencies.

- 130. I am not aware of how the information was communicated to the agencies.
- 131. I am aware that the Director of Public Prosecutions, Executive Director and two Principal Crown Prosecutors were advised of the adverse events. This is detailed in a cover letter that was drafted to accompany addendum statements that were issued to replace earlier statements that contained results processed during the period where issues were detected. The letter describes the Solicitor-General and Crown Law were consulted, and a recommendation was that the letter would be issued directly to the DPP (JH-55 Cover Letter for all Statements).

Question 56

Did the problems with DNAIQ lead to the retraction or amendment of results in these cases? If yes, by what means were the amendment and retraction of results communicated?

- 132. If a result had been reported and was then deemed to be a Quality Failure after a Quality check, the result would be marked as 'incorrect' in AUSLAB. This updated result would be then made available to QPS via an interface to the QPS Forensic Register.
- 133. If a result had been reported in a statement, then the statement would be retracted and replaced with the updated (Quality Control Failure) results.

134. After the development of explanatory paragraphs to explain the contamination, these were added to any new statement containing results processed during the period where issues were experienced. If statements had already been issued, addendum statements were issued to detail the explanatory paragraphs and were issued to replace the original.

Question 57

Has the DNA laboratory since returned to using DNAIQ processes, systems and/or products? Have there been any further problems with DNAIQ systems or products?

- 135. After a process of testing, the extraction platforms using DNAIQ chemistry were reimplemented, with Extraction Platform B implemented on 20 August 2009 and Extraction Platform A was reimplemented on 19 January 2010 until replaced by the QIASymphony for automated extractions in November 2016 (JH-52 Change Register).
- 136. DNAIQ chemistry was still used in manual processes and is used with the Maxwell instruments that are currently in the laboratory.

Interpretation of DNA profiles

Question 58

List all guidance, instructions or Standard Operating Procedures provided to reporting scientists about the interpretation of exhibit results and DNA profiles.

137. The following list is of active documents in QIS, noting some of these documents have next versions that are in review, or reviewed and yet to be approved.

QIS	Title
34112v8	STR Fragment analysis of PowerPlex 21 profiles using GeneMapper ID-X software
17117v21	Procedure for Case Management

Witness Witness

17168v14	Basics of DNA profile interpretation		
25303v12	Statistical Analysis for Paired Kinship and Paternity Trio/Missing Child Scenarios		
33773v3	Procedure for Profile Data Analysis using the Forensic Register		
34006v5	Procedure for the Release of Results Using the Forensic Register		
34229v3	Explanation of Exhibit Results for FR		
35007v4	Use of STRmix Software		
36061v1	Procedure for Resolving DNA Profile Interpretation Differences of Opinion		
33188v4	Introduction to DNA profile interpretation		
33193v6	Paternity presentation		
33538v4	Powerplex21 Case Management Presentation - Single source and complex mixed DNA profiles		
33539v3	Powerplex21 Case Management Presentation - Mixed DNA profiles		

Do you understand that there is consistency of approach between reporting scientists as to the interpretation and reporting of results?

138. I don't think there is absolute consistency of approach between reporting scientists as the interpretation of DNA profiles involves a degree of subjectivity. This does not mean the scientists don't strive for as much consistency as possible.

Question 60

How is consistency of approach amongst reporting scientists achieved?

139. Staff undergo the same training, following guidelines and standard operating procedures, have discussions with and review work of colleagues, and attempt to keep

abreast of the literature that is distributed by the FSS Information Service (Library) or through their own research. These elements contribute to assisting staff to strive for consistency.

Question 61

What difficulties, if any, are caused by differences in opinion between reporting scientists, including difficulties relating to:

- (a) laboratory processes; and
- (b) culture amongst scientists within the laboratory.
- 140. There are some differences of opinion between staff relating to laboratory processes. Some differences stem from the staff's level of understanding of the behaviour of DNA profiles. Some other difficulties relate to staff forming an opinion based on experience where others are more aligned with using empirical evidence to form an opinion.
- 141. In a large work unit, there are differences in personalities. This diversity can lead to difficulties between scientists in how they interact. These difficulties include staff feeling uncomfortable approaching others, and the reluctance to interact with others and the tendency to prefer to be surrounded by their group of friends. Another difficulty related to culture is the frustration that some staff feel in the widely disparate levels of output between scientists. I think these differences contribute to the willingness for staff to interact and work positively and productively together.

Question 62

Explain all difficulties created that you are aware of and what has been done to resolve them.

142. As an example that addressed difficulties with experience, and prior to implementing Powerplex 21 and STRmix, I developed a Statistics refresher training program where all reporting scientists worked with partners to develop powerpoint presentations to refresh staff on key concepts in interpretation. The program is as per JH-59 Project schedule 2012.

- 143. As an example to strive for consistency due to differences in opinion in determining a reasonable number of contributors to mixed DNA profiles, a project #149 Development of guidelines for number of contributors (JH-60 Project #149 Project Report v1.0).
- 144. An example of attempting to put some structure and accountability around differences of opinion between scientists, a workflow was developed with input from staff and ultimately it became a standard operating procedure: QIS 36061 (JH-61 Procedure for Difference of Opinion).
- 145. To enable targets to be set and measured, and facilitate weekly plans to be set by the senior scientists, I developed a spreadsheet to predict the number of samples expected to appear on worklists for staff the following week. This was in an attempt to prevent difficulties in knowing the work required of them in the following week.
- 146. As an example to address performance and to measure against realistic targets, I developed a KPI Predictor that can assist senior scientists set targets, track performance and identify bottlenecks (JH-62 KPI Data 2022). This is in progress at the moment (JH-63 RE Predictor Phase 2).
- 147. To address difficulties that staff have with differences interpreting DNA profiles that display either high stutter products, or displaying a mixed DNA profile, some staff got together and worked on guidelines that were added to SOP 17117 (JH-64 RE_SS High stutter guidelines).
- 148. As a forum for reporting scientists to meet and discuss DNA profile interpretations, a Profile Interpretation Meeting (PIM) was established as an outcome from a recent Cultural Change Program. The PIM was designed to be chaired by senior scientists and a place to discuss respectively and robustly, difficulties in profile interpretations. A recent agenda sent to me in August 2022 contains some information on what points of interpretation are wanting to be discussed (JH-65 RE_ PIM agenda meeting and actions). To my knowledge, this meeting has not been held as yet.
- 149. In 2020, I became aware that there was a thought that staff could interpret below the validated threshold for detection of DNA profiles on the Genetic Analyser instrument (LOD). This was brought to me as a report said to be authored by 'Kylie Rika, Reporting Scientists and Cathie Allen'. This caused much difficulty within the team to digest,

especially when there did not appear to be thorough consultation prior to the report. Through a series of discussions, I recommended to Kylie Rika that if this thought wanted to be explored, that an appropriate avenue would be a Change Management request (JH-66 Final summary of meeting 18 August 2020). The request to progress through a Change Management Process did not eventuate.

- A difference in opinion exists in interpreting profiles with high pull up peaks observed. A proof-of-concept change management request was initiated to address this. While there is support for the change request to be initiated, the timing to work on the idea is not ideal with commitments with the Commission of Inquiry and key Strategic Priorities (JH-67 FW_Initial request for new project).
- 151. A difficulty was raised that staff were preferentially reviewing each other's work, instead of reviewing a wide variety of scientist's work (the ideal state in order to prevent potential bias). This was discussed between senior scientists as a minor disagreement. I suggested a practical solution was to add an FR enhancement request to assist visibility of staff's work practices (JH-68 RE_ rep_rev pairings). I am not aware if the enhancement has been raised.
- 152. Difficulties can be created when staff write statements, and their wording is not ideal in the mind of the peer reviewer. Some of the experiences were very minor. To address this, all reporting scientists came together and developed wording for interpretations that ended up being added to a SOP (JH-69 Example Statement Wording Aug 2013).
- 153. Some difficulties were experienced between staff, including senior staff, on 'combined' or 'cumulative' stutter and how to consider these in the interpretations. This was raised as a point of difference between staff. I tasked Emma Caunt as our current StatsPWG representative to consult other jurisdictions to seek advice. Emma advised that the information was available in the STRmix manual and I asked that she update any SOPs to ensure we had the documentation available. I contacted the senior scientists to share with their teams (JH-70 RE_ taking into account combined stutter and JH-71 My email to seniors_08072020). Upon sharing, some views were gathered by the senior scientists and it appears in the way the information was shared between the seniors, the discussion broke down and another senior scientist shared their disappointment in this (JH-72 Thread of Info between seniors_09092020, JH-73 combined stutter 16 July

Witness

2020, JH-74 RT2 response combined stutter31Aug). This is with the senior scientists as part of the PIM agenda to come.

Question 63

In particular, explain the differences of opinion about the concept of 'double stutter' in the context of interpreting DNA profiles, how that mattes has been raised with you or with the management team, and what steps you or others have taken to resolve that difference of opinion.

154. I am not familiar with the term 'double stutter'. I will assume this is what FSS scientists term 'combined/cumulative' stutter which has been a matter where differences of opinion have been observed. The steps taken are described in paragraph 153.

Ouestion 64

Explain, to your knowledge, the extent to which 'double stutter' is interpreted by reporting scientists.

155. My understanding is that if a peak is in a position that could be a 'cumulative' stutter, it is considered as a possible stutter peak. I am not aware of further information on this but am aware that it is on the PIM agenda to discuss in the near future (JH-65 RE_PIM agenda meeting and actions).

All the facts and circumstances declared in my statement, are within my own knowledge and belief, except for the facts and circumstances declared from information only, and where applicable, my means of knowledge and sources of information are contained in this statement.

I make this solemn declaration conscientiously believing the same to be true and by virtue of the provisions of the *Oaths Act 1867*.

Witness

TAKEN AND DECLARED before me at Brisbane in the State of Queensland this sixth day of October 2022



Justin Howes

Witness TIMOTHY BOWLES, LAWYER

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Justin Anthony Howes

Education

Master of Science (Forensic Science): Griffith University (QLD)

■ 1999 – 2000 : Major: Forensic Biology (including Statistics)

Bachelor of Arts: University of Queensland

■ 1996 – 1997 : Major: Human Movement Science

Bachelor of Science: University of Queensland

■ 1993 – 1995 : Major: Biochemistry/ Molecular Biology

Diploma of Management - TAFE QLD

2015

Certificate IV (Workplace Training and Assessment)

2005

Professional experience

QLD Health Forensic & Scientific Services: Forensic DNA Analysis

Team Leader: Forensic Reporting and Intelligence Team (July 2012 – present) A/Team Leader: Forensic Reporting and Intelligence Team (July 2008 – 2012) A/Team Leader: Major Crime Team (May 2008 – July 2008)

- Including A/Managing Scientist (Forensic DNA Analysis) as 'Higher Duties' when required
- Provide leadership and scientific direction to all Reporting Scientists in Forensic DNA Analysis, and to Forensic DNA Analysis as a senior manager
- Develop, implement and monitor efficiency improvements primarily within own team, and to troubleshoot ideas and processes for other teams.
- Collaborative closely with the Team Leader Evidence Recovery & Quality on matters relating to both teams.
- Primary contact point for external enquiries (QPS, DPP, Defence lawyers) on matters of reporting, general DNA profiling, evidence collection, timeframe requests
- Biology Specialist Advisory Group (BSAG) delegate for Qld FSS since 2016
- Supervision of delegate to StatPWG (Statistics Project Working) national body of statistical DNA interpretation
- Training Co-ordinator for Forensic DNA Analysis
- Participate as Panel Chair or Panel Member in internal and FSS-wide recruitment processes when required
- Not current in last 12 months:
 - Lecturer: VIFM Forensic Nurse Course (2008, 2009); Griffith University Master of Forensic Science (Police Stream – scheduled October 2011)
 - Laboratory contact point for DNA Innocence Project
- All points per below

Senior/ Supervising Forensic Scientist (April 2006 – May 2008) A/ Senior/ Supervising Forensic Scientist (Oct 2005 – April 2006) Scientist (Oct 2005: Initial appointment)

- Including A/Team Leader (Major Crime Team) as 'Higher Duties' when required
- Reporting and Peer Reviewing case files and Court Statements
- Responsible for the examination, reporting and provision of Expert Evidence in matters of alleged Sexual Offences and other major crime cases
- Supervision, co-ordination, training and performance appraisal of team members
- Participation in the planning and decision-making process as part of the Forensic DNA Analysis Management team
- Provide scientific advice to courts, Qld Police Officers, medical officers, other scientists
- Conduct Team Meetings involving case discussions, communication of scientific advancement ideas, delegation of team roles when required
- Write weekly reports to line manager on team performance according to Key Performance Criteria
- Give scientific presentations and attend meetings pertaining to the direction of Forensic DNA Analysis
- Work in accordance with NATA guidelines for an accredited laboratory
- Participate in Collaborative Testing Services Proficiency Testing Program
- Participate as Panel Chair or Panel Member in internal and FSS-wide recruitment processes when required

Northern Territory Police, Fire and Emergency Services Forensic Services Section (Forensic Biology)

Forensic Scientist/ Biologist (December 2002 - October 2005)

- All scientific duties from item receipt to court presentation for both major and minor/volume crimes
- Reporting and Peer Reviewing case files and Court Statements
- Responsible for the examination, reporting and provision of Expert Evidence in all Forensic Biology matters
- Technical duties: screening, DNA extraction, amplification, capillary electrophoresis (using ABI 310 and 3100), analysis (using Genescan, Genotyper and GenemapperID)
- Lecturing Police recruits, Detective-training members
- Crime scene attendance
- Additional responsibilities: Acting Team Leader, Training Officer, Chair of panel for Technical
 Officer position in Biology, involved in validation of Identifiler, ABI 3100 and 7000, and
 GenemapperID, panel member for Authorisation Boards for Biology, Fingerprints and Crime
 Scene members
- Co-ordinated Operation Geensweep 'sweep' of Major and Volume crime cases in dedicated areas eq. Alice Springs.
- Work in accordance with NATA guidelines for an accredited laboratory

Forensic Science Service (UK: London Laboratory)

Forensic DNA Analyst/ Assistant Forensic Scientist (Violent Crime DNA Unit) (May 2001 – May 2002)

- Technical duties: DNA extraction, amplification, electrophoresis (ABI 377), analysis (using Genescan and Genotyper)
- Workflow co-ordinator and Mentor for new team members daily allocation of tasks, training new members in methods and procedures
- Work in accordance with UKAS guidelines for an accredited laboratory

Professional Activities

Australian and NZ Forensic Science Society (ANZFSS) member – initial membership 1999.

Secondment to the Australian Federal Police (AFP) for Operation CAWDOR – Tsunami Disaster Victim Identification Biology Team, Phuket, Thailand (May 2005)

QLD representative to National Institute of Forensic Science meeting on 'Closed Set DNA Typing' (January 2007)

QLD representative at EDNAP (European DNA Profiling Workshop) and ENFSI (European Network of Forensic Science Institutions) in Prague, 2008

Biology Specialist Advisory Group (BSAG) delegate for Qld FSS - since 2016

Referees

Cathie Allen

Managing Scientist (Police Services Stream)

David Keatinge

Inspector – Scientific Section Qld Police Service

Queensland Health

Forensic and Scientific Services



Forensic Reporting and Intelligence Team – Duty Statements

1 Purpose

To provide a framework for duties performed by members of the Forensic Reporting and Intelligence Team (FRIT) within Forensic DNA Analysis

2 Scope

This document applies to all members of the FRIT within Forensic DNA Analysis. Duties may vary from this document according to business requirements.

3 Definitions

DMU	DNA Management Unit
OQI	Opportunity for Quality Improvement (Queensland Health's equivalent of Non-Conformances and Corrective Actions)
CSP	Career Success Plan
FRIT	Forensic Reporting and Intelligence Team
KPI	Key Performance Indicator
NCIDD	National Criminal Investigation DNA Database
NIFA	NCIDD-Integrated Forensic Application
QPS	Queensland Police Service
SMU	Sample Management Unit
SOP	Standard Operating Procedure
SSDU	Scientific Skills Development Unit
SSLU	Scientific Services Liaison Unit
TAT	Turnaround Time
TM	Training Module

4 Team Leader – Forensic Reporting and Intelligence Team

4.1 Reporting structure

The Team Leader (HP6) of the FRIT reports to the Managing Scientist – Police Services Stream.

4.2 Duties

- Manage and develop the FRIT within Forensic DNA Analysis.
- Assist Managing Scientist (Police Services) in setting strategic direction for Forensic DNA Analysis.



- Monitor training and resource levels in the team in line with QIS <u>31010</u>, and coordinate the use of resources according to dynamic workloads.
- Continue to develop training programs within FRIT in collaboration with Senior Scientists, especially concerning the training and development of new court Reporting Scientists.
- Participate in moot court evaluations on staff to evaluate competence in providing court evidence.
- Conduct team meetings and lead discussions on profile interpretation, method development and court reports where necessary.
- Present FRIT progress reports to senior management.
- Provide scientific advice and support to the FRIT sub-team leaders, and court Reporting Scientists and Scientists within, and outside of FRIT.
- Provide support to Quality and Projects team, including undertaking Proficiency Tests when allocated.
- Participate in FSS Training Co-ordinator's meetings as directed by SSDU where necessary.
- Provide advice and assistance to FRIT sub-team leaders at case conferences when required.
- Provide scientific advice to QPS officers or legal parties on matters of collection, sampling, testing and explanations of DNA profile interpretations when required.
- Provide expert testimony on reported cases, and on cases peer reviewed when required. Provide expert testimony on cases when the Reporting Scientist and Peer Reviewer are both absent when required.
- Liaise with SSLU and QPS DMU when necessary to co-ordinate the allocation of priority cases to case scientists.
- Keep abreast of current literature as it relates to current and emerging technologies.
- Participate in Quality Program including using QIS2 appropriately to maintain a high standard of quality in the laboratory, completing TMs, following (and writing and updating where appropriate) SOPs, and reviewing documentation in QIS2 and within the Change Management Framework where required.
- Record tallies according to KPIs and collate for FRIT.
- Conduct tours for external parties where required.
- Attend meetings with QPS or other external agencies as a representative of Forensic DNA Analysis where required.
- Deliver presentations to clients, or other external groups, and internal groups where required.
- Conduct CSPs on sub-team leaders.

5 Senior Scientist – Forensic Reporting and Intelligence Team

5.1 Reporting Structure

The Senior Scientist (HP5) within the FRIT reports to the Team Leader - FRIT.

5.2 Duties

- Manage and develop FRIT sub-team within Forensic DNA Analysis.
- Monitor training and resource level in the sub-team in line with QIS <u>31010</u> and coordinate the use of resources according to dynamic workloads.
- Assist Team Leader in setting strategic direction for FRIT.

- Undertake casework duties according to competencies including profile data analysis, preparation of statements and peer review of all case and sample type, upload profiles to NCIDD, link reporting and reviewing, and NIFA uploading and reporting.
- Co-ordinate workflow to enable a Quality Service of short TAT on casework (including case management and statement reporting, and link reporting).
- Continue to develop training programs within team, especially concerning the training and development of new court Reporting Scientists.
- Participate in moot court evaluations on staff to evaluate competence in providing court evidence.
- Conduct team meetings and lead discussions on profile interpretation, method development and court reports.
- Provide scientific advice, support and mentoring to court Reporting Scientists and Scientists performing case management and NCIDD and NIFA tasks.
- Liaise with SSLU, QPS DMU when necessary to co-ordinate the allocation and/or priority of cases, and to seek feedback on processes implemented.
- Provide advice and assistance at case conferences where required.
- Provide advice to QPS and legal parties on casework where required.
- Provide expert testimony on reported cases, and on cases peer reviewed when required. Provide expert testimony on cases when the Reporting Scientist and Peer Reviewer are both absent when required.
- Provide support to Quality and Projects team, including undertaking Proficiency Tests when allocated.
- Provide support to other FRIT sub-teams and promote cohesion between teams.
- Commit to and lead continuous improvement strategies.
- Lead research and development initiatives when required including completing actions as designated at the management meetings.
- Keep abreast of current literature as it relates to current and emerging technologies.
- Participate in Quality Program including using QIS2 appropriately to maintain a high standard of quality in the laboratory, completing TMs, following (and writing and updating where appropriate) SOPs, and reviewing documentation in QIS2 and within the Change Management Framework where required.
- Participate in and lead specific projects within the Change Management Framework when required.
- Participate in rostered tasks as directed by Team Leader (eg. Quality Flag checking)
- Monitor sub-team performance and provide Team Leader with progress reports of performance of sub-team.
- Record tallies according to KPIs and collate for sub-team.
- Conduct tours for external parties where required.
- Attend meetings with QPS or other external agencies as a representative of Forensic DNA Analysis where required.
- Deliver presentations to clients, or other external groups, and internal groups where required.
- Conduct CSPs on staff under line management when required.

6 Reporting Scientist – Forensic Reporting and Intelligence Team

6.1 Reporting Structure

The Reporting Scientist (HP4) within the FRIT reports to the Senior Scientist within FRIT.

6.2 Duties

- Prepare and peer review statements for court on all sample and case type, including Intelligence Reports.
- Undertake casework duties according to competencies including profile data analysis, preparation of statements and peer review of all case and sample type, upload profiles to NCIDD, link reporting and reviewing, and NIFA uploading and reporting.
- Assist Senior Scientist in continuous improvement of workflow arrangements.
- Provide expert testimony on reported cases, and on cases peer reviewed when required. Provide expert testimony on cases when the Reporting Scientist and Peer Reviewer are both absent when required.
- Provide advice and assistance at case conferences where required.
- Provide advice to QPS and legal parties on casework where required.
- Commit to continuous improvement strategies.
- Provide support to other teams and promote cohesion between teams.
- Participate in research and development initiatives and change management projects.
- Participate in Proficiency Tests when allocated.
- Participate in moot court evaluations on colleagues to evaluate competence in providing court evidence when required.
- Keep abreast of current literature as it relates to current and emerging technologies.
- Participate in Quality Program including using QIS2 appropriately to maintain a high standard of quality in the laboratory, completing TMs, following (and writing and updating where appropriate) SOPs, and reviewing documentation in QIS2 and within the Change Management Framework where required.
- Participate in and lead specific projects within the Change Management Framework when required.
- Participate in rostered tasks as directed by Senior Scientist or Team Leader
- Record tallies according to KPIs.
- Deliver presentations to clients, or other external groups, and internal groups where required.
- Conduct tours for external parties where required.

7 Scientist – Forensic Reporting and Intelligence Team (within Intelligence Team)

7.1 Reporting Structure

The Scientist (HP3) within the FRIT reports to the Senior Scientist of the Intelligence Team within FRIT.

7.2 Duties

- Perform DNA interpretations (profile data analysis) according to competencies.
- Use NCIDD and provide Intelligence to QPS via uploading and reporting via the link process.
- Write Intelligence Reports where appropriate.
- Participate in research and development initiatives and change management projects.
- Provide support to other teams and promote cohesion between teams.

- Participate in quality initiatives including following SOP's and undertaking Proficiency Tests when allocated.
- Participate in Quality Program including using QIS2 appropriately to maintain a high standard of quality in the laboratory, completing TMs, following (and writing and updating where appropriate) SOPs, and reviewing documentation in QIS2 and within the Change Management Framework where required.
- Participate in and lead specific projects within the Change Management Framework when required.
- Commit to continuous improvement strategies.
- Participate in rostered tasks as directed by Senior Scientist or Team Leader
- Record tallies according to KPIs.
- · Conduct tours for external parties where required.
- Deliver presentations to clients, or other external groups, and internal groups where required.

8 Associated Documentation

QIS: 31010 - Forensic DNA Analysis Capability Development Program

9 References

Nil

10 Amendment History

Version	Date	Updated By	Amendments
1	11 Oct 2006	K Weller/ R Smith	First version – Major Crime
2	17 Dec 2007	J Howes	Major Crime update
3	09 May 2011	J Howes	Major update to duties performed including Title change in document
4	17 Dec 2012	J Howes	General update including header
5	16 July 2014	J Howes	Changed PADs to PDPs and expanded into words, edited header
6	11 Jan 2016	J Howes	New template
7	25 July 2017	J Howes	Updated header, removed 'case manager' and replaced with PDA, replaced CTS with Proficiency
8	02 Jan 2019	J Howes	No change required
9	13 May 2022	J Howes	Updated template, included amendment history and format updated, removed duplicates where found.

JH-3

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Project number	Project name B Evaluation of DBLR v1 2	Implementation date	
	Reduction in Physical Case file creation	ongoing Sep-21	
	Exhibit Result Line Revision	Nov-21	
	5 2021 FR version upgrade	ongoing	
234	Process mapping of Interpretation and reporting	closed	
233	Bone sampling and demineralisation protocol	on-hold	
	Paternity calculations for mixed DNA samples	closed	
	Verification of STRmix v2.8	Jun-21	
	Implementation of 3500xL PP21 Casework	Feb-21	
	Paternity Index Distributions by Locus in PP21 Review of current baseline thresholds 3130xl using PP21	closed closed	
	Baseline method trial	ongoing	
	6 Collection of sperm from pubic hair	on-hold	
	Evaluation of DBLR	not implemented	
224	Evaluation of FaSTR DNA	not implemented	
223	DCS v4.0	Apr-22 (3500xL B)	
222	Profiling of Spermatozoa from microscopy slides	ongoing	
224	Impact of magnetic fingerprint powders on bead-based trace DNA		
	. extraction (collab with QPS) Verification of commercial H & E stains	closed	
	Verification of commercial H & Estains Verification STRmix 2.7 for 3500xL	Q1-21 Mar-21	
	3 Verification of BSD600 Ascent A2	Nov-20	
	Verification of Maxwell FSC Instruments	Jun-20	
	Validation of Ion Chef & S5	ongoing	
215	STRmix v2.7 - comparison of LRs for 5p mixtures	on-hold	
214	Validation of STRmix v2.7	Feb-20	
213	Verifiler Plus	ongoing	
	Storage Transition into FR	Aug-19	
	Streamlining of DNA profile result reporting workflow	on-hold	
) Verification of GeneMapper v1.6) Verification of SPEX 6775 Freezer Mill	Jan-20 Q4-19	
	3 Verification of STRmix v2.6.2	Д4-19 Jun-19	
	Verification of Pro K	Aug-19	
	y Filer Plus	ongoing	
205	Post implementation review of STRmix v2.6.0	nil	
204	Diamond dye collaboration with QPS	not implemented	
	Number of alleles for SS LR greater than 100billion with STRmix v2.6	closed	
	! Validation STRmix v2.6.0	Jan-19	
	QIAsymphony QSL3 Verification	Feb-19	
) Statement format and wording revision) Proflex	on-hold Jan-22	
155	Tronex	Juli 22	
198	Assessment of OSD reworking on Intelligence Reference samples	closed	
197	Interpretation of 4 person mixtures using STRmix v.2.0.6	Aug-18	
196	Verifiler Plus Trial	nil	
	Testing of Quant and Amp reagent stability at room temperature	Apr-18	
	PVerification of QIAsymphony SPAS PVerification of STRmix v2.5.11	Apr-18	
	! QIAsymphony Bone Teeth	not implemented Apr-18	
	Effects of HCl on DNA persistence and profiling	nil	
	Research Project - MPS	nil	
189	Y Filer plus implementation	ongoing	
188	S Verification of Maxwell for Retain Supernatant	Jun-18	
	Verfication of STRmix v2 0.6 for use with the 3500	closed	
	5 Analysis of Casework PP21 samples using 3500xL A	not implemented at this time	
	Validation of QS5	Feb-19	
	Evaluation of the efficacy of Microcons Implementation of NCIDD-IFA (bonaparte)	Feb-18 Q3-19	
	PP21 WEN CW 3500xL Validation	closed	
	Sperm microscopy sensitivity	Nov-20	
	Use of STRvalidator for validation or verification	not implemented	
	DNA sequencing at D18S51 locus	closed	
178	Verification and implementation of STRmix v2.4.03	not implemented	
	' 3500 CW PP21-WEN	closed	
	Investigation of ICMP protocol	closed	
175	Hamilton ID STARlet - CE	May-19 (Starlet C)	MAN/C introduced to laborate 1: 0000 500
17/	Varification of ARTEL (RCS and MVS)	rotrocastivo	MVS introduced to laboratory in 2009, PCS
	Properties of ARTEL (PCS and MVS) Hamilton ID STARlet - Pre PCR	retrospective Jan-17 (A) and Jul-17 (B)	introduced 2013
	! Phadebas testing from suspension in ERT	closed	
	PP21 Verification of new ILS and Matrix	Q3-16	
170	Reassessment of in-house stutter thresholds and stutter file	not implemented	
	Verification of swab suspension at RT	closed	
	3 Validation of QIAsymphony	Nov-16	
167	Verification and implementation of STRmix V2 3	closed	

Project number		Implementation date	
	Verification of TMB Screening Test for Blood Verification of Phadebas paper	retrospective retrospective	
	Case Management improvements	Jan-15	
20.	case management improvements	50.1.15	
163	Assessment of results obtained from auto-microcon samples	closed	
162	OSIRIS Freeware for Profile Viewing	closed	
	FTP processing on OSD plate	Mar-15	
	Verification of STRmix V2.0.6	Jan-15	
	M-VAC trial by QPS Statement in Table Format	not implemented closed	
	Quant Standard Data Mining	closed	
	Verification of 7500A after thermal cycling block change	Aug-14 returned to service	
	Verification of software for 3130s	closed	
154	Verfication of software for 7500	Did not proceed	
153	Verification of Trigene Advanced	retrospective	
153	Validation of Quantifiles Trip and V. Files Phys	Nov. 2015 (avantifilar)	Y-filer: additional work is required to
	Validation of Quantifiler Trio and Y-Filer Plus Verification and implementation of STRmix 2 0.1	Jul-14	complete validation prior to use
	Suitability of combining wet and dry swabs from SAIKs	closed	
	Development of guidelines for number of contributors	Mar-15	
148	Cleaning bone processing equipment	Jul-19	
	Quantifiler re-validation after manufacturing changes	Aug-14	
	Globalfiler validation	closed	
	3500 validation Christmas Tree Staining	Mar-15 (A), Jan-16 (B) closed	
	Foreign DNA on Semen Negative SAIK swabs	Jan-14	
	Concentration of large items	not implemented	
	PowerPlex optimisation program	closed	
140	GlobalFiler Express Kit FTA sensitivity study	closed	
	Extraction negative tube	Not implemented	
	Batch Case Management	Dec-13	
	Accepting partial Amel Frozen AP	Mar-14 Jul-14	
	Verification of an additional Thermalcycler	Mar-14	
	Number of alleles for SS LR greater than 100billion	Jul-14	
	QPFREG - AUSLAB upgrade	Closed	
132	Mantis verification	Nov-15	
	PP21 post implementation review	nil	
130	GlobalFiler_testing	Not implemented	
129	Reference profile interpretation (EXH lines)_Combined with #126	All information under #126	
	Trial of QIAGEN Investigator Quantiplex Kit	Not implemented	
127	Verification of GM-IDX software upgrade	Jun-15	
	DNA profiles without STRmix)	May-13	
	AUSLAB All Incomplete requests	Jan-14	
	Generic Instrument interface Verification of Maxwell DNA Extraction from Bone	Sep-13 Not implemented	
	Verification of Maxwell DNA Extraction from Tissue	Not implemented	
	Verification of new Pro K and DTT	Mar-13	
120	Verification of new Taq in Profiler Plus kits	Q4-12	
	Validation of Extracting DNA from Concrete	Not implemented	
	Validation of Extracting DNA from Soil	Not implemented	
	Creation of animal semen repository Verification of a New Size Std for GeneScan	Folder empty Mar-13	
	Verification of a new membrane for M'con	Jan-13	
	Change of SAIK booklet and kit	Aug-12	
112	Evaluation of continued competence	closed	
	Sexual assault reassessment	closed	
	AUSLAB hardware replacement cutover	Jun-12	
	Maxwell C & D verification Pipette disposal	Jun-12 Feb-12	
	PowerPlex 21 Implemen	Ref Sep-12, CW Dec-12	
	PowerPlex 21 NCIDD	Ref Sep-12, CW Dec-12	
105	PowerPlex 21 Reporting and STRmix	Ref Sep-12, CW Dec-12	
104	PowerPlex 21 Concordance	Ref Sep-12, CW Dec-12	
	PowerPlex 21 Mixture	Ref Sep-12, CW Dec-12	
	PowerPlay 21 Population	Ref Sep-12, CW Dec-12	
	PowerPlex21 Population PowerPlex 21 Sens	Ref Sep-12, CW Dec-12 Ref Sep-12, CW Dec-12	
	PowerPlex 21 program	Ref Sep-12, CW Dec-12	
	Page numbering of statements	Jan-12	
	BSD 200uL	Q3-12	
	Statement Appendix 5	Feb-12	
	P30 addendum	Not implemented	
	ESI Pro kit	Not implemented	
	European Loci AUSLAB changes Efractions in SAIKS	Not implemented Feb-12	
	Maxwell Pre lysis	Jul-13	
32	•	14. 20	

	Implementation date
90 Population dataset	closed
89 New Change management 88 European population dataset	May-12 closed
87 Autoclave use	Nov-11
86 Maxwell Hair and Fingernail	Jan-12
85 Maxwell Diff Lysis	Feb-12
84 Maxwell Paper & Gum	Sep-11
83 Maxwell Fabric	Sep-11
82 Capillary Regeneration	Mar-14
81 Volume Case management	Jul-11
80 Volume Undetermined	Jul-11
79 Maxwell Tapelifts	Sep-11
78 ABA Cards	Jul-11
77 GeneAmp 9700 B & D	Mar-11
76 Manual Staining	Dec-12
75 QA pend	Apr-11 Nov-11
74 Destructions 73 GeneAmp 9700 C & E	Mar-11
73 GeneAmp 9700 C & E	Feb/Mar-11
71 BSD Series II	Feb-11
70 Maxwell	Q2/3-11
69 Sensitivity Amp Vol Euro Loci	closed
68 New Loci	closed
67 Tube FBX testcodes	Jun-13
65 CAPIT-ALL decapper	Nov-10
64 Modified Off-deck lysis	Not implemented
63 England Finder	Q1/Q2-11
62 Re-implement of auto DNAIQ	Aug-09
61 Theta in Reporting Stats	Dec-12
60 Change to retention of receipt	Dec-09
59 Commercial cell line	Not implemented
58 Half vol P+ reactions	Not implemented
57 7500	Jun-10
56 Re-implement of Auto DNAIQ	Aug-09
55 2uL for CE	Oct-11
54 400HD ROX	Sep-12
53 Artel 52 New Software & interp	See project #174 Proposal not approved
51 Paperless in-tube cases	Q3/Q4-09
50 Nuc clean-up double elution	Not implemented
49 Recal of Quant control ranges	May-10
48 Fingerprint techniques & DNA	Not implemented
47 DNAIQ clean-up	Not implemented
46 Modified chelex from blood & cells	Mar-09
45 Kinship Stage 3	Jan-09
44 NCIDD Bulk upload	Feb-09
Assessing the success rate of buccal cell controls spot	ted on FTA
43 indicating paper	Jul-08
42 Kinship Stage 2	Jan-09
41 3130 upgrade	Jul-08
40 Batch uploading to NCIDD	Jul-09
39 Semen	Project abandoned
38 GM ID-x	Feb-09
37 Tapelift	Project abandoned
36 Hair	Project abandoned
35 Quant DUO	Not implemented Jan-09
34 Kinship Stage 1 33 Pk Ht RFU & Al	Jan-09 Feb-09
32 Off-deck DNAIQ Super retention	Mar-08
31 One tube testing	Not implemented
30 RSID Saliva	Not implemented
29 Swab Submission Improvement	Jul-08
28 RSID semenogelin	Not implemented
27 Additional PSA verification	In-house study
26 Barcodes on receipts	Jul-08
25 7500 verification	May-08
24 Supplier change PSA	Not implemented
23 FTA Destruction checklist	Dec-07
22 Implementation of the Crime-lite	May-07
21 Packaging destruction	Jun-08
20 Upgrade 3130	Feb-08
19 Tech Admin redesign	Closed
18 Statement preblurbs	Oct-06
	Closed
17 SAIK Improvement	
16 Cut off limit for statstics and Fst	Closed
	Closed Not implemented Oct-06

Duning 1 5		
Project number Pro 12 BSI	oject name D punching for chelex extraction	Implementation date Apr-08
	ood Clothing test code and list	Dec-06
	gital imaging	Closed
9 Sta	iff movements	Closed
	ove Genotyper Comments Field	Closed
	amination form updates	May-06
6 Sna	aking vs Twirling - Cell Extraction	Closed
	ovement of DVI Skeletal Analysis to Mortuary Environment	Mar-06
	lidation	Feb-06
	lusion/Review of all Profiles	Aug-05
	urt Notification List (AUSLAB) ange Management	Oct-05 Nov-05
	ojects without a project number	1407 05
Date of report	,,	
Jul-11 Adl	hesive DNA Collector Trial Report for QPS	Not implemented
		Information for workflow purposes
	al Swab Submissions - Detecting Foreign Particles	only
	IA Profiling of Hair Exhibits ferential Extraction using the Iprep ChargeSwitch Extraction	Jun-11
	emistry and Instrument	Not implemented
	A kits: Analysis of sensitivity and high-dose hook effect	Jun-09
	comparison of DNA recovery and profiling from 4N6 versus rayon	3311 05
	abs: using chelex, Nucleospin and DNAIQ DNA extraction	
May-09 tec		May-09
	ectiveness of Nucleospin clean-up where the 9PLEX profile is no	Information for workflow purposes
Oct-08 sizi	5	only
	adebas Supernatent Trial: TNE Extraction of Amylase	Mar-08
	nantifiler initial validation conducted	Jun-04
	00 - Validation of Casework samples on the ABI 3100 Genetic alyser	Validation approval date: Jan-03
	00 Validation (Reference Samples)	Validation approval date: Jan-04
	A PFTA Protocol	Validation approval date: Jan-04
377	7/3100 Low level DNA study	Validation approval date: Jan-04
STA	ATS - BRB Stats Validation	Validation approval date: Feb-04
	restigation into DNA quantitation using Quantifiler sustem (with	
	ibition data)	Validation approval date: Jan-05
	antifiler Report - Victorian Police	Validation approval date: Apr-05
Kev	view of Petricevic Report	Validation approval date: Jun-05
ST.	ATS - BRB Stats population Data (Caucasion, Aboriginal, Asian)	Validation approval date: 2005
	IDD Validation and Test protocols (4.0.0)	Validation approval date: Mar-06
	tended Internal Prospective Valiation of the ABI Prism	
	00/Quantifiler system (Forensic Biology) Including external reports	Validation approval date: Jul-06
	ended Internal Retrospective Validation of the ABI Prism 00/Quantifiler System (Forensic Biology)	Validation approval date: Aug-06
	ATS - BRBStats v1.23	Validation approval date: Aug-00 Validation approval date: Apr-07
	ATS - BRBStats v 1.26	Validation approval date: Apr-07
	A Validation	Validation approval date: Apr-07
13		Not implemented
	tomation Project	
	port on the Verification of automated Quantifiler™ Human DNA lantification Setup using the MultiPROBE® II PLUS HT EX with	
	ipper™ Integration Platform	Feb-07
Troject I dil	pper integration rationii	16507
Rer	port on the Verification of automated AmpF&STR® Profiler Plus®	
	d COfiler® amplification reaction setup using the MultiPROBE® II	
•	US HT EX with Gripper™ Integration Platform	Feb-07
	port on the Verification of Automated 3100 Setup using the	
	IltiPROBE® II PLUS HT EX Platform	Refer to #15 and #19
	lidation of AUSLAB for Analytical Workflow	Feb-07
Project 4 Val	,	
	plementation of AUSLAB enhancements (Batch functionality)	Feb-07
Project 5 Imp Project 6 Imp	plementation of AUSLAB enhancements (Batch functionality) plementation of Pre-PCR platform	
Project 5 Imp Project 6 Imp	plementation of AUSLAB enhancements (Batch functionality)	Feb-07
Project 5 Imp Project 6 Imp Project 7 Imp	plementation of AUSLAB enhancements (Batch functionality) plementation of Pre-PCR platform plementation of Post-PCR platform	Feb-07
Project 5 Imp Project 6 Imp Project 7 Imp Rep	plementation of AUSLAB enhancements (Batch functionality) plementation of Pre-PCR platform plementation of Post-PCR platform port on the Validation of automated FTA® Processing using the	Feb-07 Not implemented
Project 5 Imp Project 6 Imp Project 7 Imp Rep	plementation of AUSLAB enhancements (Batch functionality) plementation of Pre-PCR platform plementation of Post-PCR platform	Feb-07 Feb-07 Not implemented Not implemented
Project 5 Imp Project 6 Imp Project 7 Imp Rep Project 8 Mu	plementation of AUSLAB enhancements (Batch functionality) plementation of Pre-PCR platform plementation of Post-PCR platform port on the Validation of automated FTA® Processing using the ultiPROBE® II PLUS HT EX with Gripper™ Integration Platform	Feb-07 Not implemented Not implemented
Project 5 Imp Project 6 Imp Project 7 Imp Rep Project 8 Mu Project 9 Rep	plementation of AUSLAB enhancements (Batch functionality) plementation of Pre-PCR platform plementation of Post-PCR platform port on the Validation of automated FTA® Processing using the	Feb-07 Not implemented Not implemented Refer to #11 and #13
Project 5 Imp Project 6 Imp Project 7 Imp Rep Project 8 Mu Project 9 Rep Project 10 Imp	plementation of AUSLAB enhancements (Batch functionality) plementation of Pre-PCR platform plementation of Post-PCR platform port on the Validation of automated FTA® Processing using the ultiPROBE® II PLUS HT EX with Gripper™ Integration Platform port on the evaluation of commercial DNA extraction chemistries	Feb-07 Not implemented
Project 5 Imp Project 6 Imp Project 7 Imp Rep Project 8 Mu Project 9 Rep Project 10 Imp Rep	plementation of AUSLAB enhancements (Batch functionality) plementation of Pre-PCR platform plementation of Post-PCR platform port on the Validation of automated FTA® Processing using the ultiPROBE® II PLUS HT EX with Gripper™ Integration Platform port on the evaluation of commercial DNA extraction chemistries plementation of FTA Processing on Multiprobe II	Feb-07 Not implemented Not implemented Refer to #11 and #13

Project number Project name

Implementation date Comments

	Report on the Verification of an Automated DNA IQ™ Protocol using	
Project 13	the MultiPROBE® II PLUS ht ex with Gripper™ Integration Platform	Oct-07
Project 14	Implementation of Extraction Chemistries on the MultiPROBE II	Oct-07
	Report on the Verification of Automated Capillary Electrophoresis	
Project 15	Setup using the MultiPROBE® II PLUS HT EX Platform	Not implemented
0,000 20	Mock sample cleaning: Comparing TriGene™ and bleach and its	not implemented
Project 16	efficiency in removing DNA.	Nov-06
,	Report on Automated preparation and testing of Quantifiler	
Project 17	standards and controls	Feb-07
,	Report on the validation of automate.it STORstar system for	
Project 18	automated sequence checking of DNA extracts	Jun-08
.,	Report on the Validation of the Manual 9+1 Method for Capillary	
Project 19 (A)	Electrophoresis Setup	Feb-08
, , ,	Close of Post-PCR MultiPROBE® II PLUS ht ex and the Automated	
Project 19 (B)	Capillary Electrophoresis Setup Method	Not implemented
, , ,	Report on the Verification of the RECAP-96M™ Automated	
Project 20	Decapper/Recapper System	Jan-08
	A Modified DNA IQ™ Method Consisting of Off-Deck Lysis to Allow	
Project 21	Supernatant Retention for Presumptive Identification of $\alpha\text{-Amylase}$	Mar-08
	A Modified DNA IQ™ Method for Off-Deck Lysis Prior to Performing	
Project 22	Automated DNA Extraction	Feb-08
Project 23	Hair extractions	Not implemented
Project 24	Sperm Extraction	Not implemented
Project 25	Investigation and evaluation of tapelift materials and procedures	Not implemented
Project 26	Report on Whatman® FTA Concentrator PS™ Parasite Purification	Not implemented
Project 27	DNA IQ Recovery	Jun-09
Project 28	Semen DNA IQ Validation	Not implemented
Project 29	DNA IQ Clean-Up Protocol	Not implemented
Project 30	Rcovery of DNA from IQ Store Plates	Not implemented
-	Suitability of Lovell cuticle pushers as a substrate for fingernail	
Project 31	scrapings for DNA Analysis	Not implemented
Project 32	Identifiler	Not implemented

Queensland Health

Forensic and Scientific Services



1 Purpose and Scope

This document describes the change management procedure that is to be used within Forensic DNA Analysis, to ensure that all process changes and projects occur in a controlled and timely manner. This procedure applies to all process changes or projects that:

- involve the validation/verification of equipment
- involve the validation/verification of technical procedures
- are projects with external funding
- are internal projects (minor or major) which impact on sample reporting/processing
- involve major LIMS function/configuration changes
- impact on multiple stakeholders
- require staff training to be implemented
- significantly alter workflow procedures

This procedure does not apply to:

- routine document updates/alterations
- minor technical changes which do not impact on sample reporting/processing (e.g. changes in specimen type, storage configuration changes)

As an appendix to this document - is a checklist that can be used to guide staff on how they might approach a new idea/observation. It will assist in establishing if it should be recorded as an emerging/novel practice, as a minor change, or as a full project/change management.

2 Definitions and Abbreviations

For a comprehensive list of abbreviations refer to QIS <u>23849</u> Common Forensic DNA Analysis Terms and Acronyms.

e-sign Electronic signature **FR:** Forensic Register

FSS: Forensic Scientific Services **IT**: Information Technology

LIMS: Laboratory Information Management System used to record information and

track exhibits/case files.

NATA: National Association of Testing Authorities

3 Principle

Changes within Forensic DNA Analysis have the potential to impact on our clients, on stakeholders (internal/external to FSS) and may impact on compliance with NATA. As such changes which occur with Forensic DNA Analysis must be carefully considered and

Approver/s: Cathie ALLEN

documented. There are a number of types of changes that may occur within Forensic DNA Analysis; for the purpose of documentation - these are classified into five types: administrative change, IT/LIMS change, minor project, major project, and external projects.

Administrative changes: are restricted to changes in processes/workflows that impact on documentation or administration processes only. These changes will most likely occur within the Administrative team within Forensic DNA Analysis. It does not include any changes of a technical nature.

IT/LIMS change: An IT change would apply to the introduction of new software into Forensic DNA Analysis, in some instances for upgrades in software versions or the introduction of new hardware. This type of change would require collaboration with IT services. A LIMS project would include any alteration that required a change in the LIMS function, or major configuration changes. It would not include minor changes such as storage configurations, or minor changes to specimen types etc.

Minor Project: are generally defined as projects that have a duration of <6 weeks and a budget of <\$5,000. These projects have a minor impact on sample processing/reporting. Any project which major impact on workflow or sample reporting should be considered under major projects.

Major Project: are generally defined as projects that have a duration of >6 weeks and/or a budget of >\$5,000. Major projects require significant planning and detailed consideration of project impacts and implementation procedures.

External Projects: is to be used for all projects which have been externally funded. Where there are no documentation requirements for an externally funded project – standard change management document as described in this document apply. For RDAC projects, RDAC documentation requirements apply (QIS 33017) with the additional requirements of:

- A change management number will be assigned within Forensic DNA Analysis
- Management Team are to indicate that they have reviewed all RDAC proposals by adding their name to the Excel sheet included within the project folder

The change management procedure utilises a three step process:

- the initial request (Step 1)
- minor change (Step 2a) OR project plan (Step 2b)
- final report, approval/implementation (Step 3)

The utilisation of these steps is dependent on the type of change (administrative, IT/LIMS, minor, major and external) and on the progression of the change management process. Refer to Section 4 for details.

4 Actions

Prior to the preparation of any change management documentation it is recommended that ideas are discussed at the work unit level to determine the merit of each idea or proposal. If the process of change management is initiated it will need to follow the documentation requirements as listed in sections 4.1 to 4.8 and the workflow as shown in Appendix 1.

*An <u>exception</u> is made for projects that are a mandatory requirement for the laboratory e.g. validation/verification of a new process or equipment item. In these cases it is possible to proceed directly to a full project plan (section 4.3).

For large projects an overarching project number is allocated (by quality) to the work, and sub-projects may then be allocated "a letter" such that sections/parts of a project can be signed off separately. For example in validating an amplification kit a project number 1234

may be applied (for the overarching project), with sub-projects 1234a – referring the sensitivity testing, 1234b referring to concordance, 1234c referring to thresholds etc.

In cases where supplementary testing for a project is required (post-sign off), if the data is an extension of previous work - it may also be appropriate to allocate the supplementary work "a letter" ie. part b of the same work. If the supplementary work is substantially different in topic or content a new project number should be allocated.

All project documents are to e-signed and locked at completion. Refer to Appendix 5 for e-sign procedure.

Technical Review:

For major projects and for validations it is a requirement for the project to have a technical reviewer. The role of the technical reviewer is to 'peer view' critical technical aspects of the project (e.g. new instrument programs/settings, new analytical procedures) and/or to review data analysis with the project (e.g. Excel data transformations, formula's and calculations etc.). The technical reviewer/s are nominated by the team leader and/or management team at project proposal stage (section 4.3). The technical review is completed either during the project or at the completion of the laboratory work and data analysis - but prior to final report being presented to the management team. The technical review should provide to the Management Team as a written document that outlines the aspects of the project reviewed and general findings (Refer to Appendix 2 for template)

Communication:

- For large projects regular project updates should be given by the project leader (or delegate) to the management team. This will allow the management team to ensure that the project is meeting all requirements (NATA, internal needs etc), and that they have a full understanding of the project prior to final report preparation and sign off.
- When projects are complete presentations should be made at team meetings so that all staff have an awareness of new processes and technology as it is released.
- Appropriate communications should be made at time of implementation (emails to applicable staff, additions to minor change registers, records to quality etc).

4.1 Initial Request (Stage 1)

Change requests can be initiated by any staff member within Forensic DNA Analysis, and are to be recorded on an **Initial Request Form** (QIS <u>31543</u>). Submission of an initial request requires the following actions:

- Complete the Initial Request Form (QIS <u>31543</u>). The initiator is required to complete the blue sections of the form only.
- Initiator is to email the Quality Team and Line manager (of the person initiating the request) with the network location of the document so it can progress.
- · Quality will allocate the request a proposal number
- The Line Manager is to complete the red sections of the form, create a PDF of the request form and e-sign the document. Store/save the document to the appropriate project folder in I:\Change Management

The Line Manager will assess the initial request recommending either:

- Abandon process at Initial Request (Refer to section 4.7)
- Proceed to Step 2:
 - Minor Change (Refer to section 4.2)

or

Project Proposal (Refer to section 4.3)

If the line manager wants to recommend proceeding to a full project proposal – they will need to seek Management Team approval.

If the initial request is abandon - no further action or documentation will be required.

On completion of the initial request form (e-signed and locked), the line manager is to advise quality team

4.2 Minor Change (Stage 2a)

The minor change form is used to document the purpose, method and date of change. If the Line Manager recommends that the change management is to proceed as a minor change, the project initiator must complete the blue sections of the **Minor Change Form** (QIS <u>31548</u>) and submit it to their line manager. In some circumstances a small amount of experimental data may be included within a minor change – where the data is used for decision making purposes.

The Line Manager must then complete the following actions:

- E-sign the minor process change document (QIS <u>31548</u>). Store/save the document to the appropriate project folder in <u>I:\Change Management\Minor Change Forms</u> completed
- Add the change to the Minor Change and emerging or novel practices register located in: <u>I:\Change Management\Change Register - Minor Changes and emerging or novel practices.xls</u>
- Inform the quality team and all stakeholders of the change e.g. team meetings or email
- Update SOPs etc. if required
- Inform the Quality & Projects Senior Scientist to complete the process

The Quality & Projects Senior Scientist must:

- E-sign and lock the minor process change document (QIS 31548)
- Ensure all above actions have been completed by the line manager.

4.3 Project Plan (Stage 2b)

If the Management Team recommends that a change management should proceed as a full proposal (administrative, IT/LIMS, major change or external project) the project leader is required to complete the following project documents:

- Project Risk Assessment Document (QIS <u>22872</u>): A risk assessment must be completed documenting the risks of the project for each team.
- Change Management Project Proposal (experimental design) Document: This
 document should cover all aspects of what the project is proposing to do: It should
 include an introduction to the project (including literature review), purpose/background,
 methodology and experimental design (either laboratory experiments or data analysis
 as applicable) and a detailed materials and methods section.

Refer to QIS <u>23402</u> for writing guidelines and template for the project proposal. These project proposals will essentially constitute the introduction and materials and methods section of the projects final project report

This document must be prepared and submitted to the Forensic DNA Analysis Management Team along with the Project Risk Assessment Form (QIS 22872).

- Consider ethics requirements: QIS <u>33268</u> Police Services Human Ethics Review Checklist, it may impact on the projects methodology, and ethics approval maybe required before the project can start.
- (Optional) Project Budget (QIS <u>31052</u>): A budget can be prepared and submitted to the Forensic DNA Analysis Management Team - with the project proposal. A budget template is provided in QIS <u>31052</u>.

For a new piece of equipment, new chemical or new process a formal risk assessment (QIS 29106) will be needed in addition to the project risks that are outlined in QIS 22872 The formal risk assessment addresses workplace health and safety risks and the project risk assessment is in relation to business risks.

After all project documents have been prepared (as listed above); risk assessments (if applicable) and LIMS documentation completed (if applicable) email your Line Manager and Quality Team (FSS_BiologyQuality@health.qld.gov.au) and advise them of the location of the documents in I:\Change Management. The Line Manager/Project leader will submit the documentation to the Forensic DNA Analysis Management Team for consideration (Refer the section 4.4), with a due date for feedback.

4.4 Forensic DNA Analysis Management Team – Consideration of Project Proposal

The Forensic DNA Analysis Management team will consider the change management project proposal documents as outlined in section 4.3. It is not necessary for all Management Team members to read and approve every proposal; however a quorum of the Management team must approve the proposal. The quorum <u>must include</u> the Managing Scientist, Team Leaders, Quality and Projects Senior Scientist, Senior Scientist that has Line Management of the staff/project and Senior Scientist/s of areas significantly affected by the project. For major projects and validations a technical reviewer suggestion should also be provided to the management team for consideration (Refer to section 4).

Consideration of the proposal should include:

- A determination of the impact of the proposed change on all stakeholders
- 2. Cost/Benefit Analysis of the project
- 3. Risk Assessment (Workplace Health & Safety and Business Risks)
- 4. A communication plan for all project participants and stakeholders

The Forensic DNA Analysis Management Team will then make a recommendation as follows:

- Implement proposal. If the proposal is approved, the project documentation will be e-signed by the Management Team. The project leader/appointed staff can initiate the project.
 - Project work must be conducted by a technically experienced and competent person (Refer QIS <u>10662</u>)
 - For projects that are >3 months, the Senior Scientist Quality and Projects will meet with each project team ~ every 2 weeks to ensure project progression, and to provide advice and resources as required.
- Implement proposal after change. If the Management Team requires additions/edits to the project proposal, the Management team will return the document to the project leader/appointed staff with feedback. The project

documents will need to be edited and resubmitted (as per section 4.3.) before further consideration by the Management Team.

Abandon process. Refer to Section 4.7 for details.

After the due date for feedback project leader/line manager should:

- Make edits (if required).
- Create a PDF of the project proposal and project risk assessment documents
- Store/save the document to relevant project folder in <u>I:\Change Management</u>

Management team must:

- Provide feedback on the proposal
- Complete the risk assessment

4.5 Implementation and Final Report (Step 3)

On completion of the change management project - a final report is required, this is usually written by the project leader (Refer to QIS 23402 for report preparation details). A Technical Review - if it is required (Appendix 2), and an Implementation Plan (Refer to Appendix 3) must also be prepared. The implementation plan will be a list of the steps required to be completed either before the change is implemented, or shortly after implementation. Although a proposal may not be implemented on completion, a basic implementation plan that can be refined closer to implementation should still be completed and submitted. On completion of the report, technical review and implementation plan, they are to be forwarded by email to your Line Manager. The Line Manager/project leader will submit the final report, technical review and implementation plan to the Forensic DNA Analysis Management Team for consideration/acceptance.

If the final report is accepted by the Forensic DNA Analysis Management Team it will be esigned and the project/change management process closed. If the Management Team requires additions/edits to the final report, it will be returned to the project leader/appointed staff with feedback. The final report will need to be edited and resubmitted for consideration by the Management Team.

After the due date for feedback project leader/line manager should:

- Make edits (if required).
- Create a PDF of the project proposal and project risk assessment documents
- Ask the management team to e-sign the document.
- Store/save the document to relevant project folder in <u>I:\Change Management</u>

Management team must:

- Provide feedback on the final report
- E-sign the documents as/when requested by the project leader/line manager.

After acceptance of the final report the Forensic DNA Analysis Management team will recommend that the:

- Change is implemented into routine use (Refer to Appendix 3 for implementation plan for project leaders).
- Change is accepted but will be implemented at a later date (Refer to Appendix 3 for implementation plan for project leaders).
- Change is abandoned (Refer to Section 4.7 for details).

After completion of the project, all stakeholders must receive communications about the findings and outcomes of the project. This may include presentations at meetings, or the provision of final reports to stakeholders. For significant projects, a summary of the project is to be presented at team meetings.

On completion of the final e-signature by the Managing Scientist a communication is to be sent to the Quality team so that they can ensure all documents have been finalised. Quality team can then lock and store data files by loading them to the Forensic Register.

*Please note: in the event the work is to be published, please consider if the publication needs to reviewed by the FSS Ethics committee. Refer to QIS 32177 FSS Publication checklist.

4.6 Responsibilities in Signing Documentation

When a project proposal or report is submitted for review, it is the responsibility of the reviewer to ensure that all feedback is provided by the due date. Any feedback provided after the due date may not be considered (based on the merit of the feedback).

It is acceptable for a reviewer from the Forensic DNA Analysis Management Team to seek advice from other members of staff where it is deemed appropriate (e.g. where another person may have more experience in the subject of the report). In this instance, it is the responsibility of the person seeking the advice to provide the feedback to the project officer and to do so by the due date.

4.7 Abandoned/Cancellation

Should a change proposal not be approved, or if at any time the change is no longer required, the change management process may be abandoned/cancelled. This shall be recorded on the change management documents (to be forwarded to the Quality Team). If the project is abandon mid-way through a process an electronic file note can be created to detail the date and reason for project cessation.

It is possible to re-start abandon change management processes at a later date, and there are relevant sections in the change management forms to record a restarted process.

4.8 Recording Feedback

Project feedback, including feedback on project proposal and reports, is to be tabulated and stored in the relevant change management folder (under the appropriate project number folder).

All email communications regarding the project are also to be stored in the relevant change management folder.

5 Records

- All change management documentation (plans, reports, data etc.) are to be stored electronically in a network drive (e.g. I:Drive)
- On completion of projects all records (plans, reports, excel files etc) are to be stored in Forensic Register. To store records in FR:
 - Create new FR case Job Type=Research
 - Subject/Complainant=Project number and short title
 - Offence Class=Miscellaneous
 - Location=Forensic DNA Analysis Quality
 - Project documents loaded as an examination summary

6 Associated Documentation

QIS: 10662	FSS Guidelines for Method Validation
QIS: 22872	Project Risk Assessment for Change Management in Forensic DNA Analysis
QIS: <u>23401</u>	Forensic DNA Analysis Validation and Verification Guidelines
QIS: 23402	Writing Guidelines for Validation and Change Management Reports
QIS: 29100	Health & Safety Risk Assessment Form
QIS: 29106	Risk Management Guideline - conducting and evaluating Health and Safety
	risk assessments
QIS: 31052	Forensic DNA Analysis - Change Management Budget
QIS: 31543	Initial Request Form for Change Management in Forensic DNA Analysis
QIS: 31548	Minor Process Change Form for Change Management in Forensic DNA
	Analysis
QIS: 32177	Human Ethics Review Checklist - FSS Publications
QIS: 33017	FSS Research and Development short form
QIS: 33268	Human Ethics Review Checklist - Police Services
QIS: 33333	Participant Information and Consent Form (PICF) - Common Biological
	Samples
QIS: <u>33334</u>	Participant Information and Consent Form (PICF) - Semen Samples
QIS: 33335	Participant Information and Consent Form (PICF) - Vaginal Samples

7 Amendment History

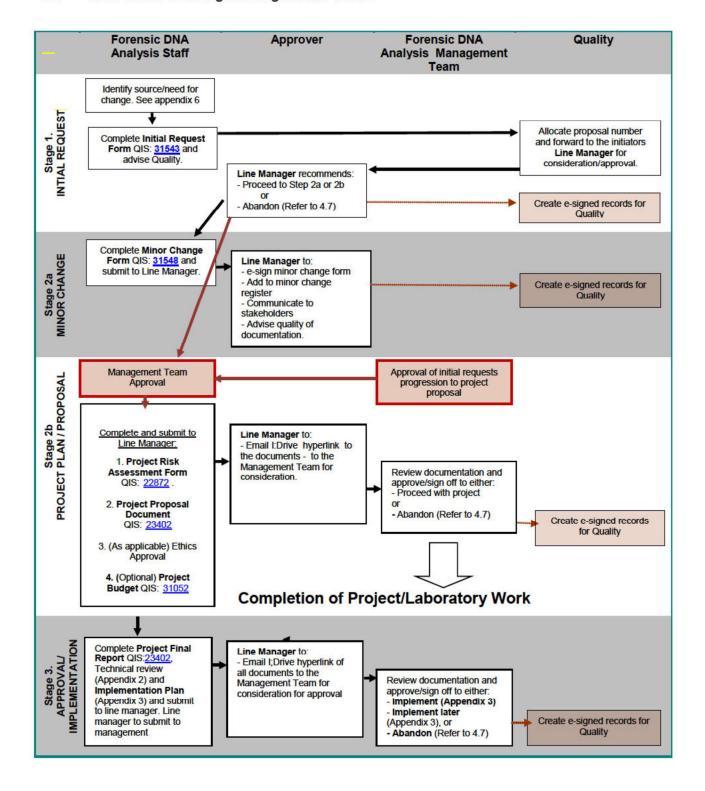
Version	Date	Author/s	Amendments
1	25 Aug 2005	Mary Gardam	First Issue
2	27 Feb 2007	J Olsson, M Gardam V Ientile	Format Changed to include Project Management.
2	April 2008	QIS2 Migration Project	Headers and Footers changed to new CaSS format. Amended Business references
3	25 Sept 2008	Robyn Smith Crystal Revera	Formatting, Changes made to reflect new Laboratory name & Contacts
4	14 May 2012	Shannon Thompson Kirsten Scott	Major revision/re-write as the change management process changed.
5	21 Jan 2013	Kirsten Scott	Update QIS numbers and headers. Add records, associated documents and minor edits.
6	26 Mar 2013	Kirsten Scott	Clarify point 3 in section 4.4. Update hyperlinks
7	6 June 2014	Kirsten Scott	Remove Assessment Phase. Change in actions required by line managers for approving initial plan and minor change documents.
8	19 June 2015	Kerry-Anne Lancaster	New template. Added milestone register and implementation plan. changed AUSLAB to LIMS, defined project proposal and responsibilities of the reviewer. Add QIS 33017
9	21 Oct 2015	Kirsten Scott	Inclusion of consent forms in associated documents. Option for mandatory projects to proceed directly to project plan. Inclusion of RDAC processes & Quality Checklist
10	25 Nov 2015	Kirsten Scott	Inclusion of a technical review for major projects and validations, and minor text update in other section as a result of technical review
11	20 Sept 2016	Kirsten Scott	Specify implementation plan as mandatory, Section 4.5 and 5 add a note on locking of data by quality. Section 3 clarify RDAC requirements

12	1 June 2018	Kirsten Scott	Remove milestone register (section 4.3, 4.8). Add comms and project numbering to section 4. Addition of FR instructions section 5. Add technical review template as appendix 2.
13	19 Nov 2019	Kirsten Scott	Add Human ethics checklist section 6. Additions to section 4.4: meetings with Quality Sen/Sci., and staff competency requirements. Header added to appendix 8.4
14	2 Oct 2020	Kirsten Scott	Edit document to reflect change from hardcopy records to electronic sign-off processes. Additions to appendix 4
15	14 July 2021	Abbie Ryan	Addition of Appendix 5 – e-sign procedure. 4.2 Addition of extra signature step to minor change procedure for Quality Senior Scientist. 4.3.1 Changed title of document 22872 to Project Risk assessment.
16	10 Dec 2021	Kirsten Scott	New header, remove optional Gantt chart for projects, add ethics QIS links and requirements and emerging/novel practices (Appendix 6)
17	30 Mar 2022	Abbie Ryan	Updated Appendix 3 – implementation plan tasks.

8 Appendices

- APPENDIX 1: Change Management Process
- APPENDIX 2: Technical Review Template
- APPENDIX 3: Implementation Plan for project leaders
- APPENDIX 4: Checklist of documents required for a Change Management Project
- APPENDIX 5: Procedure for e-signing documents
- APPENDIX 6: New and emerging novel practices checklist

8.1 APPENDIX 1: Change Management Process



8.2 APPENDIX 2: Technical Review Template

Technical review of Proposal #Project number Project title

General project observations:

Experiment 1:
Program settings checked: Yes / No / Not Applicable. Comments:
Formulas checked: Yes / No / Not Applicable Comments:
Data transformations checked: Yes / No / Not Applicable Comments:
Calculations checked: Yes / No / Not Applicable Comments:
Experimental observations (design/results etc):
Experiment 2: (add additional experiments as required)
Program settings checked: Yes / No / Not Applicable. Comments:
Formulas checked: Yes / No / Not Applicable Comments:
Data transformations checked: Yes / No / Not Applicable Comments:
Calculations checked: Yes / No / Not Applicable Comments:
Experimental observations (design/results etc):

Technical Reviewer

Name	Position	Signature	Date

Project Manager

Name	Position	Signature	Date

8.3 APPENDIX 3: Implementation Plan for project leaders

Successful project implementation may require numerous tasks to be completed either prior to implementation, or shortly after the implementation date. Some of the considerations/tasks that may be required are listed below; however, this is not intended to be a comprehensive list of tasks as each project will have different implementation requirements. Project leaders must devise and submit a comprehensive implementation plan for management review prior to the final report being signed off. Ideally, this implementation plan should be provided at the same time as the draft final report.

Once complete, the checklist should be submitted to the quality team for filing with the signed project documents.

Project Title:	Project Number:	roject Number:	
Task	Details	Responsible Line Manager/ Allocated to:	Date Completed
e.g. Create new procedur	es New SOPs and training modules to be		

		Allocated to:
e.g. Create new procedures	New SOPs and training modules to be written and approved	
e.g. Update procedure/s	Existing SOPs and training modules to be revised and approved	
e.g. Staff training	Project members and relevant to staff to be issued with CTT statements as required	
	CTT staff to train relevant staff	
e.g. Software setup	Final version of software to be setup and reviewed on instrument	
	Check if Macro updates are required	
e.g. Equipment tasks	Add equipment to QIS	
	Add equipment to LIMS	
e.g. Consumable tasks	Add consumables to LIMS.	
	Addition of products to FAMMIS	
	Order new consumables	
e.g. Forensic Register development/requirements		
e.g. DNA interpretation/reporting	May include: Model Maker requirements and assessment, Statement of Witness appendix update	
e.g. Impacts/risks assessed	Any risks identified in risk assessment are addressed.	
e.g. Add to minor change register	Ensure that implementation has been added to the minor changes register	
e.g. Communication	Communicate to staff and other stakeholders – by meetings and emails.	

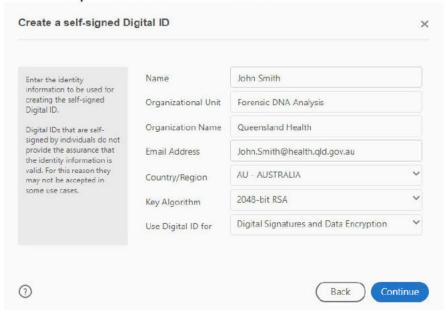
8.4	.4 APPENDIX 4: Checklist of documents required for a Change Management Project		
	Project Number:		
Minor	Change:		
☐ Init	tial Request Form (31543) (May not be required for mandatory projects)		
☐ Mir	nor Change Form (<u>31548</u>)		
☐ Ad	ded to Minor Change Register and emerging or novel practices register		
□ Im	plementation (Comments added to SOPs (if required) and communication to staff)		
Major	Project:		
☐ Init	tial Request Form (31543) (May not be required for mandatory projects)		
☐ Pro	oject Risk Assessment Form (<u>22872</u>)		
☐ Pro	oject Proposal Document		
□ (O	ptional) Project Budget (<u>31052</u>)		
☐ Eth	nics checklist and/or approval - if applicable (33268)		
☐ Ris	sk Assessment (As applicable for new equipment and laboratory procedures 29100)		
□ Pro	oject Final Report		
□ Те	chnical Review (for validations and major projects only)		
☐ Im	plementation Plan		
** Cor	nsent forms for staff collections should have been previously provided to quality if applicable.		
RDAC	project:		
	OAC Application Form (Copy only, original stored with Research Office)		
	OAC Final Report – if the project is funded (Copy only)		
□ Ex	cel Sheet – with Names of Management Team for acknowledgment of project.		
□ Qi	uality team have loaded all key project documents to FR for storage		
Check	dist completed by: Date:		

8.5 APPENDIX 5: Procedure for e-signing documents in Adobe

First time process to set up digital signature:

- Scan an image of your personal signature and save to your desktop.
- 2. Open up a PDF document in Adobe
- 3. Click tools and Open Certificate
- 4. Choose "Digitally Sign"
- Drag the box to point in PDF document where you want to apply your digital signature.
- 6. Select Configure Digital ID
- 7. Select Create a new Digital ID then continue
- 8. Select "Save to File" then continue
- 9. Ensure that you place all your credentials in the name section. (Do not use symbols)

Sample of how and areas to fill out:



Note: you can change the place where you save your credentials, the default saving file location is generally where the adobe program files are kept.

- Enter a password of your choice. You will use this password every time that you apply it
- 11. Last step in the process is to attach a copy of your 'signature'. Click continue
- 12. Click on the create button
- 13. Select image then select "Browse" to import in your signature from the file location
- 14. Click save.
- 15. To now digitally sign the PDF document, enter password and click sign.

Note: – if you are the final approver, e.g. expenditure delegate, line manager approving the document, you must check the 'Lock document after signing' checkbox. This will lock the entire document down and cannot be edited once this has been done.

- You will be asked to save the PDF file.
- 17. If the PDF document requires further electronic approvals, it can be forwarded to the next approval for their Digital ID. If the check box is checked 'lock document after signing', then the document can no longer be edited or signed.

For all future PDF documents, when you click Digitally sign, you will be asked to select the area to sign and then can select the Digital ID, enter your password and sign the document.

Approver/s: Cathie ALLEN

8.6 APPENDIX 6: New and emerging novel practices checklist

This checklist is provided as a template/processes by which staff can consider what to do - when they have seen something new, wish to do something new or are unsure how to proceed with a decide or idea. The emphasis is on the documentation and communication of decisions and thought processes - in line with best quality practices.

Step 1: Gather the facts and define the issue/problem.

Step 2: Make an assessment of your idea or what you have seen: taking into account:

- the case implications
- possible expenditure of resources (time and money)
- impact on clients
- health and safety etc. (refer to Section 4 above).

This will allow you to determine who is accountable for the decision, and how big the required and appropriate process will need to be.

Step 3: Action and documentation: For any issue that have a cost implication (resources or significant staff time), or implications for clients - the full change management process would apply (refer to this document above). For new observations and/or emerging novel practices that are smaller in nature - it maybe more appropriate to use following document to detail the issue, your thinking and the decision:

I:\Change Management\Change Register - Minor Changes and emerging or novel practices.xls

Step 4: Communicate to appropriate audience

Example: Raised by: John Smith Date started: 20/01/2022

Define your issue:

Apparent artefact at D18S51. Artefact shifts between labelling as a 17.1 or 17.2 variant allele. No stutter is observed for this artefact. Only observed in samples from peri-anal, rectal or penile areas.

Has it been seen before? Yes

Where? Case XXXXXXXXXX

Who can make the decision? Myself

Assessment

Adds contributor to otherwise single source assumed known contributor, height of artefact not consistent with another contribution dropping out. No expenditure of money, time or resources required.

Actions

- Removed artefact from FR GeneMapper table.
- Annotated epgs and re-loaded to Forensic Register
- Notations added to case in Forensic Register.
- Added to Change Register Minor Changes and emerging or novel practices document

Communication

Who	When	How
All reporters via Microsoft Teams	02/02/2021	Posted
Line manager	01/02/2021	Email

Queensland Health

Forensic and Scientific Services



Forensic DNA Analysis Validation and Verification Guidelines

1 Purpose and scope

Validation is the developmental process used to acquire the necessary information: to assess the ability of a procedure to obtain a reliable result, to determine the conditions under which such results can be obtained, and to determine the limitations of the procedure (National Association of Testing Authorities, 2020). Method validation and verification provides objective evidence that a method is fit for purpose, meaning that the particular requirement for a specific intended use are fulfilled (National Association of Testing Authorities, 2020). Verification studies are typically smaller than those that are required for validation. For full details refer to National Association of Testing Authorities, 2020 specific documents.

The Forensic DNA Analysis laboratory is certified by the National Association of Testing Authorities (NATA) and is obliged to meet these specifications. ENFSI (2010) states that for DNA based tests, validations/verifications must demonstrate that the profile/s obtained under the new regime will be of the same or better quality than those obtained under the previous regime.

The purpose of this procedure is to describe validation and verification guidelines for use within Forensic DNA Analysis. Test methods, equipment, computer/software systems and information management systems must be shown to be fit for purpose before they are used by the laboratory to generate results. Validations will be required in Forensic DNA Analyses for:

- all new methods developed "in-house";
- methods laboratory/commercial that have been modified;
- methods without validation data adopted from other laboratories or from literature;

Verifications will be required in Forensic DNA Analyses for:

- use of a previously published and validated method
- use of commercial kits

This procedure shall apply to all validation/verification projects conducted within Forensic DNA Analysis. The final decision regarding the extent and scope of the study shall be made by the Managing Scientist.

2 Definitions

Accuracy and Precision

<u>Accuracy (trueness):</u> is the closeness of agreement between the test result and the "true" or accepted value.

<u>Precision:</u> is a measure of closeness (degree of scatter) between independent test results under stipulated conditions (National Association of Testing Authorities, 2020). High precision does not necessarily imply high accuracy.

An example of accuracy and precision measures would be, a determination of the proportion of correct genotypic assignment of samples, and a review of the number of alleles correctly assigning to the expected 0.5bp window/bin.

Repeatability is a measure of the maximum acceptable difference between two test results obtained at the same time by the same analyst under identical conditions on the same material. ENFSI (2010) recommends repeatability studies contain a minimum of five replicates, while NATA (2020) specifies at least six degrees of freedom (e.g. 4 times in a series with 2 samples or 3 times in a series with 3 samples). A repeatability test might be: 12 samples on a plate 7 times with standards and/or controls in an amplification plate and processed by a single operator (suggest that the DNA extract of a defined concentration is prepared in a large volume, and aliquot out to PCR plate or CE plate etc. This will ensure pipetting error is minimised in the preparation of multiple samples to an equivalent concentration).

Reproducibility

- Within laboratory (in-house) reproducibility A measure of the maximum acceptable difference between two test results obtained on the same material by different analysts at different times.
- Between-laboratory reproducibility A measure of the maximum acceptable difference between two test results obtained on the same material by different laboratories. It is most conveniently determined in collaborative trials.

Reproducibility in Forensic DNA Analysis could be assessed by: several DNA samples being prepared on an amplification plate by one operator, and the same DNA samples prepared on an amplification plate by a second (different) operator.

Sensitivity is the rate of change of the measured response with change in the concentration of analyte National Association of Testing Authorities, 2020). For PCR-based assays, validation studies must consider the stochastic effects of PCR; particularly as it relates to DNA concentration. ENFSI (2010) recommends sensitivity tests have a minimum of 5 dilutions tested.

3 Principle

Validation provides objective evidence that the particular requirements for a specific intended use are met. There is no one method of validation that is universally agreed upon, however the validation guidelines below are consistent with NATA criteria (National Association of Testing Authorities, 2020), and are consistent with Scientific Working Group on DNA Analysis Methods (SWGDAM 2020) recommendations for the minimum criteria for the validation of DNA profiling processes (ENFSI, 2010).

4 Actions

The planning and implementation of a validation/verification project in Forensic DNA Analysis should occur as follows:

- a. Determine if it is a verification or a validation that is required. For example if a standard published method, with full validation data, and a commercially available kits is to be implemented within the laboratory - a verification not validation would be required (prior to its introduction). If a new methodology is developed - a validation would be necessary.
- b. Using the 'Procedure for Change Management in DNA Analysis' standard operating procedure QIS <u>22871</u>, a project proposal must be prepared. In the planning the work consider the following:

- Validation studies require an assessment of reproducibility, repeatability, sensitivity, accuracy and precision (ENFSI, 2010). Refer to definitions section 2 for details.
- Qualifying Test For validation studies the use of known samples and where
 possible authentic case samples should be used. This may be accomplished
 through the use of proficiency test samples, or samples that the laboratory
 routinely analyses (e.g. controls). Where previous typing results are available
 concordance of genotypes should be assessed.
- Mixture Studies Forensic casework laboratories must define and mimic the range of detectable mixture ratios. Studies should be conducted using samples that mimic those typically encountered in casework (e.g. postcoital vaginal swabs)
- The laboratory must ensure that the procedure/s minimise contamination that
 would compromise the integrity of the results (QIS <u>22857</u>). The laboratory
 should employ appropriate controls and implement quality practices to
 assess contamination and demonstrate that its procedure minimises
 contamination.
- Manufacturer's information and previous published validation studies should be used to inform the laboratories validation process.
- Refer to all NATA and ENFSI documentation listed in the reference list section 6 for specific and detailed validation study requirements
- Refer to QIS 10662 for additional resources.
- The project proposal must then be submitted to the Forensic DNA Analysis Management Team for approval prior to the initiation of experiment work.
- On completion of the experimental component of the validation, a final report will need to be written using the final report template QIS <u>23402</u>. The final report is to be submitted to the Forensic DNA Analysis Management Team for consideration

5 Records

Minimum records required for a validation are:

Project Risk Assessment for Change Management in Forensic DNA Analysis 22872. Project Proposal document. (see Writing Guidelines for Validation and Change Management Reports QIS 22871 & 23402).

Implementation Plan (Refer QIS 22871)

Final Report (Refer QIS 22871 & 23402).

Additional requirements (as applicable):

Ethics approval (Refer QIS 32177)

Technical review (Refer QIS 22871)

Forensic DNA Analysis - Change Management Budget (Refer QIS 31052).

6 References

National Association of Testing Authorities. (2020). NATA – National Association of Testing Authorities, Australia. Available at: https://nata.com.au/nata/ [Accessed 27 Aug. 2020].

ENFSI (2010) Recommended Minimum Criteria for the Validation of Various Aspects of the DNA Profiling Process. ENFSI DNA Working QA/QC subgroup. Issue No 1.

ENFSI (2020) European Network of Forensic Science Institutes. Available at: http://enfsi.eu/ [Accessed 27 Aug. 2020].

Scientific Working Group on DNA Analysis Methods (SWGDAM). (2020). Available at https://www.swgdam.org/ [Accessed 27 Aug. 2020].

7 Associated documents

QIS: <u>10662</u> - FSS – Guidelines for Method Validation

QIS: 22871 - Procedure for Change Management in Forensic DNA Analysis

QIS: <u>22872</u> - Project Risk Assessment for Change Management in Forensic DNA Analysis

QIS: 23402 - Writing Guidelines for Validation and Change Management Reports

QIS: 31052 - Forensic DNA Analysis - Change Management Budget

8 Amendment history

Version	Date	Author/s	Amendments
0	06 Sep 2005	Mary Gardam	First Issue
1	April 2008	QIS2 Migration Project	Headers and Footers changed to new CaSS format. Amended Business references from QHSS to FSS, QHPSS to CaSS and QHPS to Pathology Queensland
2	25 July 2008	C Revera	New Title, Changed Forensic Biology to DNA Analysis, authorised by C Allen, Chief scientist to Managing scientist. Purpose and scope combined, hyperlinks updated, definition of verification included.
3	4 Dec 2012	K Scott	New header. Complete rewrite to fit with new change management procedures in DNA Analysis
4	18 June 2014	K Scott	Update organisational name, document names and hyperlinks
5	20 Nov 2015	K Scott	Update header/template, references and minor text updates
6	8 Aug 2017	K Scott	Update references
7	27 Aug 2020	K Scott	Minor updates all areas
8	15 March 2022	K Scott	Template update, document names and references updated

9 Appendices

1 Appendix A Additional terms used in validation studies

Appendix A

Additional terms used in validation studies

Functional Specification: Defines how it is expected to function - these functions are typically outlined by the manufacturer of equipment/software.

Installation Qualification: Verifies design specification, the physical components of the system have been designed/constructed/supplied/installed in compliance with the design specifications. This is usually completed by the company performing the installation.

Lower limit of detection (LOD) - The lowest concentration or amount of analyte that can be reliably distinguished from zero, but not necessarily quantified, by the test method.

Limit of reporting/quantitation (LOR) - The lowest concentration of analyte that can be determined with acceptable repeatability and accuracy by the test method.

Operational Qualification: Verifies the functional specification, that the system functions as intended throughout anticipated operating ranges.

Performance Qualification: Verifies that the system will consistently produce results meeting user requirement specifications and quality attributes under both normal and worst-case conditions.

Uncertainty - The spread of values within which the true value would be expected to lie, with the stated degree of confidence (usually 95%).

User Requirement Specification: Defines how the system is expected to perform - this is usually set out in the tender document requirements.

JH-6



ASSESSMENT INFORMATION DOCUMENT

Your facility is due for a reassessment.

This Assessment Information Document seeks specific background information from facilities on the current scope of NATA accreditation, any changes required, and the specific resources available to meet the requested changes.

Some sections may not apply to your facility. Please cross-reference relevant sections from your management system documentation where appropriate.

Please upload a completed copy of this Assessment Information Document and required documents/records as detailed in covering letter to the NATA Portal or email to:

Ms Madelen Chikhani at By: 09 April 2022

Delays or failure to provide the requested information may result in delays to the accreditation process.

The personal information collected in this document and other management system documentation supplied for the assessment briefing is used for conducting the assessment, reporting on the assessment and the process of continuing accreditation. It may be disclosed to NATA staff members, all of whom have signed confidentiality agreements. Aggregated data gathered from the assessment process may also be provided to third parties in a de-identified format. It may also be disclosed to agencies to which NATA has a legal obligation or with which NATA has formal agreement.

Personal information collected such as name, business telephone and mobile phone numbers and email address of the Authorised Representative or the Site Contact may be made available to enquiries requiring the service of NATA accredited facilities. The Site Contact details may be included in the NATA website directory.

NATA's Privacy Policy contains information on access and correction to the personal information held by NATA and the compliant process associated with breaches of the Australian Privacy Principles. NATA's Privacy Policy is available from the NATA website, www.nata.com.au.

Accreditation No: 41 Site No: 14171 Job No: 82214

FACILITY DETAILS

In preparation for the accreditation activity, please review the information below to confirm (or change) the details of your facility and the site to be assessed. Use the shaded boxes to provide corrected or changed details.

FACILITY (the name in which accreditation is held)		
Accreditation No: 41		
Facility Name: Queensland Health Forensic and Scientific Services		
Facility Trading Name (see note 1):		
ABN or ACN: 66-329-169-412		
Mailing Address:		
Street address (if different from above):		
Facility web address (optional): www.health.qld.gov.au		
Phone:		
INVOICING DETAILS (for all sites under your facility)		
Mailing address:		
Phone:		
Email:		

The following details are specific to your Facility's Authorised Representative.

(The rights and responsibilities of the Authorised Representative are outlined in the General Accreditation Criteria: Responsibilities of authorised representatives, available on the NATA website.)

Accreditation No: 41 Site No: 14171 Job No: 82214

Authorised Representative: Ms Helen Gregg	To change the appointed Authorised Representative please complete the <u>Facility</u> <u>Details Update (FDU) form</u> available from the NATA website.		
Position: Quality Manager			
Direct Phone:			
Email:			
SITE DETAILS			
Site No: 14171			
Site Name: Forensic and Scientific Services			
Site Trading Name (see note 1):			
Availability of services: Services conditionally available to external clients	 □ Services available to external clients □ Services conditionally available to external clients □ Services not available to external clients 		
Street address (physical location): Liaison Unit			
Site Contact (full name including title):			
Phone:			
Mobile:			
Indicate the Site Contact's primary contact num	ber: ☐ Phone ☐ Mobile		
Email:			
Do you wish to publish the Site contact informa ☐ Yes ☐ No			
(The name of the contact person and preferred phone number and email address will be listed in our records as the person to contact with enquiries about the Site's activities (i.e. from potential clients) and may be listed on the NATA website.)			

Site No: 14171 Job No: 82214 Accreditation No: 41

Note 1: Trading name(s) (optional)

Providing this information indicates the applicant is seeking approval to issue reports in its trading name(s), in addition to the name of the Facility. Trading names may be provided for a Facility and/or for individual Sites.

In order to able to issue reports in a trading name the following criteria need to be met.

- There must be a clear and reasonable link between the name of the Facility and the trading name(s) supplied, such as an ownership link or a link by virtue of a registered trading name;
- Activities reported in a trading name(s) will have been performed by the staff of the accredited Facility/accredited Site to which the trading name(s) applies, using the same techniques and procedures s those covered by the Scope(s) of Accreditation of the applicable accredited Facility/accredited Site;
- The scope of reporting applicable to the trading name(s) is the same as or a subset of the Scope of Accreditation of the applicable accredited Facility/accredited Site.

Should trading name(s) be provided you will be contacted to further explore this option.

Accreditation No: 41 Site No: 14171 Job No: 82214

NATA SCOPE OF ACCREDITATION

A copy of your current scope of accreditation is attached.

Annotate this scope to indicate the approximate frequency of all laboratory activities.

		2020		20	2021		2022			
	Quarter	Jul-Sep	Oct-Dec	Jan-Mar	Apr-Jun	Jul-Sep	Oct-Dec	Jan-Mar (in part)	Total	Weekly average
	InTube samples	4938	4727	5461	5210	5457	5423	4756	35972	404.18
- 0	Items examined (not SAIK)	1447	1281	1445	1415	1544	1461	1340	9933	111.61
ogy of ateri	SAIKs	96	81	121	112	128	117	104	759	8.53
c Biolo ation cal ma	Phadebas Supernatant	199	104	141	95	177	143	99	958	10.76
U 10 11	Microscopic	1142	684	742	734	961	733	687	5683	63.85
Forensi Examin biologid	Presumptive TMB/AP/p30/Phadebas paper	462	356	566	531	697	574	482	3668	41.21
ic .	Extraction (Reference Maxwell)	1074	714	839	825	545	595	382	4974	55.89
	Extraction (Diff Lysis, incl Supernatant)	673	457	689	663	876	706	637	4701	52.82
	Extraction (Casework Maxwell)	1250	917	956	1022	690	876	536	6247	70.19
	Extraction (QIAsymphony)	1826	1689	1927	2140	1903	1917	2536	13938	156.61
	Extraction (PCIA - Bones)	5	10	11	10				36	0.69
. <u>12</u>	Integrated (QIA extraction and quantification)	4248	4032	4536	4320	4464	4752	3168	29520	331.69
enetic Analysis	Post Extraction processing (Nucleospin, Microcon)	398	403	362	242	310	386	245	2346	26.36
Ā	Quantifications	10374	9275	10258	10004	9820	10554	7821	68106	765.24
etic	Amplifications (PCR)	9983	8590	8485	9003	7627	8143	6161	57992	651.60
Gen	Capillary Electrophoresis (3130, 3500)	15316	13210	14219	15491	12566	13371	9180	93353	1048.91

Animal health facilities only: please also complete the attached Supplement document.

Changes to the scope of accreditation

Surveillance visit: Additions will not normally be considered during a surveillance visit as such visits will not include a technical assessor. Where requested a decision will be made as to how best to meet the request without compromising the aim and focus of the surveillance visit. Accordingly, a variation visit may be arranged concurrently or as a separate visit once all information concerning the request has been considered. Charges will be incurred to accommodate the variation visit in accordance with NATA's Fee Schedule current at the time. Please be aware that any extensions to scope of accreditation may also result in an increase to your annual membership fees.

Reassessment: Any requests for additional activities to be added to the scope of accreditation as part of a scheduled reassessment will only be accommodated where such requests do not compromise the purpose of the reassessment (to review the existing scope of accreditation to determine ongoing compliance with the accreditation criteria). Where additional resources and time are required to accommodate the request, a concurrent variation visit may be arranged, and charges will be levied in accordance with the current Fee Schedule available from the NATA website. Please be aware that any extensions to scope of accreditation may also result in an increase to your annual membership fees.

Do you wish to request additions or deletions or editorial amendments to the scope of accreditation?

Accreditation No: 41 Site No: 14171 Job No: 82214

Amendments as follows:

As advised by K.Scott in email 25 May 2018 and 31 May 2018 to Lyndon Thomas (NATA):

- QIAsymphony (DNA extraction) technique to be added to DNA profiling for criminal casework and DNA profiling for relationship testing
- 3500xl capillary electrophoresis to be added to DNA profiling for criminal casework and DNA profiling for relationship testing
- Microcon to be added to DNA profiling for criminal casework and DNA profiling for relationship testing
- Nucleospin to be added to DNA profiling for criminal casework and DNA profiling for relationship testing

As advised by K.Scott in email 25 July 2018 and 7 August 2018 to Lyndon Thomas (NATA):

• Maxwell 16 is a DNA extraction process and not capillary electrophoresis technique.

Instrument and software changes since last NATA visit in December 2020:

- February 2021 3130xl CE instruments removed from use. 3500xL use ongoing.
- May 2021 Implementation of STRmix v2.8 (project #231)
- June 2021 Verification of DCS v4 on 3500xL (project #223)
- January 2022 Forensic DNA Analysis replaced the AB 9700 PCR machines with AB Proflex PCR machines (project #199)

Other changes:

- Feb 2021 Verification of commercial H&E stain (project #220)
- March 2021 Removal of "Hair examination" from NATA scope of accreditation

New projects currently in progress:

- Y-Filer (project #206)
- Verifiler (project #213)
- Ion Chef and S5 MPS (Project #216)

Note: Changes to calibration and measurement capability (CMC) may be considered as additions to the scope of accreditation.

☐ Yes. Please complete the	<u>Application t</u>	<u>for changes</u>	to the	scope of	accreditation t	<u>form</u> available
from the NATA website.						

☑ No

ASSESSMENT INFORMATIO	N DOCUMENT	
Accreditation No: 41	Site No: 14171	Job No: 82214
Regulatory requirements ap	plicable to laboratory a	ctivities
	atory requirements? For	cope of accreditation subject to, or used by example, do you test products covered by rement, food regulation, etc.?
□ Yes		
⊠ No		
	vant regulation (including	copy of your current scope of accreditation gregulatory body and/or regulatory ruling), For example:
		in accordance with AS/NZS 1249:2003 sumer Protection Notice No. 10:2008
Testing of human specimens	<u>s</u>	
Are any of your laboratory acti	vities covered by your so	cope of accreditation on human samples?
⊠ Yes		
		ese samples are used in legal investigations ot come under the framework for In Vitro
□ No		
subject to the Therapeutic God Framework and assessed aga	ods Administration (TGA) inst the National Patholo	ccreditation. Note that such testing may be In-Vitro Diagnostic (IVD) medical device gy Accreditation Advisory Council (NPAAC) te In Vitro Diagnostic Medical Devices.
Sampling		
Since your previous assessment NATA scope of accreditation?	ent, are there changes to	any sampling conducted covered by your
☐ Yes. Please complete the Afrom the NATA website.	Application for changes to	o the scope of accreditation form available
⊠ No		

 $\hfill\square$ Not applicable

Accreditation No: 41 Site No: 14171 Job No: 82214

Off-Site Laboratory Activities

Since your previous assessment, has your facility commenced performing laboratory activities offsite, for example, field testing or at clients' premises, and do you require this to be covered by your scope of accreditation?

Note: Refer to relevant documents in the NATA Accreditation Criteria (NAC) package applicable to your scope of accreditation, available from the NATA website, for any specific requirements for such laboratory activities.

	Yes. Please complete the <u>Application for changes to the scope of accreditation form</u> available from the NATA website.
\boxtimes	No

Accreditation No: 41 Site No: 14171 Job No: 82214

STAFF

In the following spaces provided (or on a separate sheet if is insufficient room), list the current facility staff. Please also indicate whether any staff work on a shift or part-time basis.

Staff responsible for quality management Part-time staff (as at 07/03/2022) Extended leave (as at 07/03/2022)

Name and qualifications	Position (Please also specifically identify staff responsible for technical and quality management)	Date started in the facility
ACEDO, Pierre [BSc]	Scientist	2006
ADAMSON, Angela [BSc (Hons)]	Reporting Scientist	2003
AGUILERA, Maria [BSc]	Scientist	2006
ALLEN, Catherine [BSc, MSc (For Sc)]	Managing Scientist	1999
ANDERSEN, Belinda [B Biomed Sc, GDFor]	Senior Scientist	2005
ANGUS, Chantal [BA (Hons)]	Laboratory Assistant	2017
AVDIC, Kevin [HNC Chem]	Forensic Technician	2014
BRADY, Susan [BAppSc. (Biotech); Grad.Dip. (For Inv)]	Scientist - Leave	2004
BRISOTTO, Paula [BSc, MSc (For Sc)]	Team Leader	2001
BROOKS, Julie	Laboratory Assistant	2016
CALDWELL, Valerie [B.AppSc. (Med Sc)]	Scientist	2006
CAUNT, Emma [BSc (Hons)]	Reporting Scientist	2007
CHANG, Cindy [BSc; PGDip Clin Biochem]	Scientist	2001
CHENG, Amy [BSc]	Scientist	2006
CIPOLLONE , Melissa [B.AppSc.]	Scientist	2006
CONNOLLY, Yvonne [BA, DipBus; Cert II &Cert III B.Admin]	Administration	2014
DARMANIN, Alanna [BA, BSc (Hons); MSc For Arch & Crime Scene Invest, Cert For Stat]	Scientist	2010
DWYER, Tegan [BForSc]	Reporting Scientist	2010
EBA, Ryu	Laboratory Assistant	2011
ENTWISTLE, Josie [BSc BA]	Reporting Scientist	2005
ESTREICH, Kim	Laboratory Assistant	2019
FARRELLY, Lisa [BAppSc]	Scientist	2013
FINCH, Patricia [BSc]	Reporting Scientist - Leave	2002
FLANAGAN Cecilia [Cert IV Lab Tech; Cert II Gov]	Administration	2021
FRENCH, Naomi [Cert IV Lab Tech]	Laboratory Assistant	2019
GALLAGHER, Claire [B.Tech. PG.Cert]	Reporting Scientist	2006
GOODRICH, Michael	Laboratory Assistant Supervisor	2010
GULLIVER, Maddison	Laboratory Assistant	2021
HARMER, Wendy [Cert II BA, DipMgt]	Administration	2005
HART, Michael [City and Guilds Level 3 (UK); Cert IV Lab Tech] DipLabTech	Forensic Technician	2014
HUNT, Matthew [BSc (Hons)]	Reporting Scientist	2009
HOWES, Justin [BSc, BA, MSc (For Sc), DipMgt]	Team Leader	2005
JAMES, Cassandra [BSc MSc (For Sc)]	Reporting Scientist	2016

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Accreditation No: 41 Site No: 14171 Job No: 82214

Name and qualifications	Position (Please also specifically identify staff responsible for technical and quality management)	Date started in the facility
JOHNSTONE, Sharon [BSc; MSc (For Sc), DipMgt]	Forensic Scientist Advanced	1999
KAITY, Adam [BSc (Hons I); PhD]	Scientist	2008
KELLER, Angelina [BAgSci (Hons), MSc (ForSc)]	Reporting Scientist	2004
LANCASTER, Kerry-Anne [B.AppSc, GDip For Inv]	Reporting Scientist	2005
LE, Lai-Wan [BSc (Med Lab), MSc (For Sc)]	Scientist	2005
LLOYD, Allison [B.AppSc; MSc (For Sc)]	Forensic Scientist Advanced	2006
LUNDIE, Generosa [BSc (Biomed Sc)]	Scientist	2006
MARGETTS, Michelle [BSc For Sc, Cert IV Lab Tech, DipLabTech]	Scientist	2011
MATHIESON, Megan [B.HSc., B.BioMedSc, GDFor]	Reporting Scientist	2005
McINDOE, Phillip [BTecONC]	Laboratory Assistant	2019
McKEAN, Sandra	Laboratory Assistant	2008
McNEVIN, Allan [B. AppSc. (Med Lab Sc)]	Reporting Scientist	2004
MICIC, Biljana [BSc]	Scientist	2005
MOELLER, Ingrid [BSc (Hons), PhD]	Reporting Scientist	2004
MORGAN, Amy [B.AppSc]	Scientist	2014
MORTON, Kristina [BSc For Sc.]	Scientist	2020
NICOLETTI, Deborah [BSc (MLS)]	Reporting Scientist	2005
NURTHEN, Thomas [BSc (Hons)]	Reporting Scientist	2004
NYDAM, Sharelle [BSc (Hons)]	Scientist	2014
PARRY, Rhys [BSc (Hons)]	Reporting Scientist	2006
PENDLEBURY-JONES, Victoria	Administration	2015
PIPPIA, Adriano [B. AppSc.]	Reporting Scientist	2000
PROWSE, Tara [B. AppSc.]	Scientist	2009
QUARTERMAIN, Alicia [BHSc, MSc (For Sc)]	Reporting Scientist	2005
RIKA, Kylie [BSc, PGDipFor, DipMgt]	Forensic Scientist Advanced	2005
ROSELT, Nicole [B.For Sc. & BCCJ]	Scientist	2016
RYAN, Abigail [BSc (Hons) For Sc]	Scientist	2008
RYAN, Luke [BSc, MSc (For Sc), Dip Gov(Sec), DipMgt]	Forensic Scientist Advanced	2013
SANDERSON, Suzanne	Laboratory Assistant	2006
SAVAGE, Chelsea [B.For Sc. & BCCJ]	Scientist (Quality)	2015
SCOTT, Kirsten [BSc (Hons). PhD. GCEd, GDEd., DipMgt]	Forensic Scientist Advanced – Quality and Projects	2007
SEYMOUR-MURRAY, Janine [B. AppSc.]	Scientist	2006
TAYLOR, Penelope [BSc (Hons)]	Reporting Scientist	2001
WAIARIKI, Stephanie [BSc For Sc, DipLabTech]	Laboratory Assistant	2022
WILLIAMS, Helen [B. AppSc. (Med Lab Sc), PGDip (Biotech)]	Scientist	2003
WILSON, Jacqueline [B.AppSc. MSc]	Reporting Scientist	2006

Note: NATA will list individuals in the Report on Assessment where there is a regulatory framework or is covered in a Deed of Agreement, Memorandum of Understanding or other binding agreement with a third party. If this is applicable to any of your laboratory activities, indicate in the table any nominated individuals or changes to nominated individuals who are authorised to release results under such an arrangement, including the arrangement in place. Please provide resumes for any new individuals not previously listed.

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Accreditation No: 41 Site No: 14171 Job No: 82214

ENSURING THE VALIDITY OF RESULTS

Has your facility participated in any proficiency tests, measurement audits or inter-laboratory comparison programs since your last assessment?

Refer to the General Accreditation Criteria: Proficiency testing document available from the NATA website for the policy on participation in such programs.

\boxtimes	Yes
	No

If yes, please provide details in the table below. Records of participation in these programs must be available for review during the NATA assessment, together with details of action taken in response to unsatisfactorily performance.

Name of provider, program and activities undertaken	Frequency of program	Last date of participation
CTS Collaborative Testing Services	38 tests in total	March 2022
FB5701 Forensic Biology	1	Jan 2021
FB5801 DNA-Mixture	1	Jan 2021
FB5840 DNA Database - Saliva	3	Feb 2021
FB5870 DNA Parentage	3	Feb 2021
FB5702 Forensic Biology	1	March 2021
FB5802 DNA-Semen	1	March 2021
FB5781 Body Fluid Identification	1	March 2021
FB5703 Forensic Biology	1	April 2021
FB5803 DNA-Blood	1	April 2021
FB5871 DNA Parentage	3	May 2021
FB5704 Forensic Biology	1	July 2021
FB5804 DNA-Semen	1	July 2021
FB5843 DNA Database - Saliva	3	July 2021
FB5872 DNA Parentage	2	Aug 2021
FB5705 Forensic Biology	1	Sept 2021
FB5805 DNA-Blood	1	Sept 2021

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FB5782 Body Fluid Identification	1	Sept 2021
FB5706 Forensic Biology	1	Oct 2021
FB5806 DNA-Mixture	1	Oct 2021
FB5701 – Forensic Biology	1	Jan 2022
FB5801 – DNA-Mixture	1	Jan 2022
FB5840 DNA Database - Saliva	3	Feb 2022
FB5870 DNA Parentage	2	Feb 2022
FB5702 Forensic Biology	1	March 2022
FB5802 DNA-Semen	1	March 2022
FB5781 Body Fluid Identification	1	March 2022

[☑] If yes to the above, please provide a summary of your facility's performance in proficiency testing programs or inter-laboratory comparisons. This should include matrices/analytes covered and any outliers recorded (including actions taken).

All tests (mixture, semen, database, standard, parentage and body fluid) were consistent with manufacturers information with the exception of test FB5781 Body Fluid Identification test (March 2021). For this non-conformance OQI#55008 refers – see below for details and actions (Refer attached).

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Report for QIS OQI as of 11/03/2022 1:37:20 PM

Report for QIS OQI -

55008 CTS 21-5781 non-conformance

OQI Details

Status Closed Approved

Subject CTS results submitted by Forensic DNA Analysis for items 3 and 6 were

not consistent with manufacturers information as provided by CTS.

Source of OQI EQAP/Collaborative/Proficiency Test

Date Identified 22/06/2021

OQI Creator Contact Details

Creator Abigail RYAN

Organisational Unit/s Quality and Projects

Service/s Forensic and Scientific Service

Site Location/s Coopers Plains

Investigator/Actioner Contact Details

Actioner Allan MCNEVIN

Organisational Unit/s Evidence Recovery

Service/s Forensic and Scientific Service

Site Location/s Coopers Plains

Investigation Details

Investigation Completed **Investigation Details**

03/08/2021

Root Cause Type None - No ProblemItem 3

the scenario provided, the examiner and peer reviewer had not considered testing the Item for the possible presence of saliva. The swab was stained with what appeared to be blood, which tested positive for the presumptive presence of blood. On review, the scenario does not provide any clear indication that item should have been tested for saliva. The testing carried out in FSS Forensic DNA Analysis is consistent with that reported by other laboratories, with 136 of 244 laboratories that responded to the CTS reporting saliva as "Not Tested".

The item was re-examined for the presence of saliva and tested positive.

Item 6

The item tested was black and pink in colour. The area of the item that tested positive for Phadebas was part of the fabric that was entirely black. The material did not appear faded, and there was no visual indication of the presence of blood. The scenario provided did not strongly indicate which body fluids the Item should be tested for, so it was tested for the presence of semen and saliva, and visually inspected for the presence of blood. Of the 244 laboratories that responded to the CTS there were 15 laboratories that reported the

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> item as negative for blood, 1 as inconclusive for blood and 15 that did not test the item for blood.

The item was re-examined for the presence of blood. It was noted that there was no visual indication of the presence of blood. The examiner did not recall any discolouration of the Phadebas test paper at the time of initial examination that may have provided some indication that blood was present. A TMB test was conducted on the area previously to be determined as Phadebas positive, and a positive result was obtained. The area which in which the DNA was located was an area of fabric with an intense black colour (i.e. not visibly faded in any way). At present, the laboratory does not have any validated protocols for observing for biological fluids under alternative light sources. This kind of testing is performed by QPS, and subsamples of possible blood staining are usually submitted as in-tube items. However, items requested for Saliva testing are usually submitted as whole items.

Preformed By

Allan MCNEVIN

Action Details

Action Complete 03/08/2021 Title

Action Fix Type No Action PossibleOutcomes

Action Description The testing of CTS tests does not perfectly correlate with routine processes.

This is not unexpected as the CTS test has to cater to a larger number of laboratories with varying workflow practices. With respect to Item 3, for our laboratory, the testing of swabs is usually performed when swabs are submitted as "in-tube" samples, where the swab head is submitted inside a tube ready for DNA extraction. Testing for blood on these items is performed by QPS prior to submission to FSS. It is incumbent on the officer submitting the item for testing to request any additional testing (semen, saliva). Generally, case history information is either not provided, or only provided in relation to the specific items submitted for testing, and the laboratory staff are not called on to decide whether additional testing is required. As the sample type and location of Item 3, and the scenario provided would not flag any immediate thoughts of the need for saliva testing, it is not considered necessary that further actions are required to be carried out with respect to the results obtained from this specific test.

For Item 6, it is unclear what could have been done different, as on reexamination, there was no visible blood staining, most likely due to the intense colouration of the fabric being tested. At present, the laboratory does not have any validated protocols for observing for biological fluids under alternative light sources. This kind of testing is performed by OPS, and subsamples of possible blood staining are usually submitted as in-tube items. However, items requested for Saliva testing are usually submitted as whole items. At this stage, no actions as a result of the findings of this OQI have been determined.

The outcomes from this OQI to be discussed with the team next team meeting.

Task Details

No Tasks found

Follow-up And Approval

Accreditation No: 41 Site No: 14171 Job No: 82214

> Follow-up Status Comment

> > Accept investigation and actions as written

Approver Approval/Rejection Date Approval/Rejection Comment

Paula BRISOTTO 04/08/2021

4/08/2021 1:23:26 PM Paula BRISOTTO:

Approved of investigation and actions

Associations

No Associations found

Records

No Records found

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Job No: 82214

MONTH			CTS P	Proficiency T	roficiency Tests 57x require sample screening	5					
	CTS Proficien	icy Tests 58x a	are sample spe proficie	cific and pre-	CTS Proficiency Tests 58x are sample specific and pre-screened. No screening techniques are required and therefore this set of proficiencies should <u>NOT</u> be used for sampling scientists.	es are required and thereforsts.	e this set of				
	CTS type	Participant Number (Web Code)	Lab Number(s)	FR Number	Staff	Role	Received Date	Due Date	Date Submitted	Follow-up completed	Feedback presented
	1000000				Kerry-Anne Lancaster	Reporter				3	
	FB5701		SOUCCESCE		Matthew Hunt	Reviewer	ACADA PORTA	-cocicoio	*concora*	10000101	A COCCOUNTY
	Forensic Biology		34470000		Kristina Morton	Sampler	1202112021	8103/2021	15/02/20/21	1104/2021	1702/50/11
					Mike Hart	Reference sampler					
January					Kylie Rika	Reporter					
	FB5801		200000000		Anne Finch	Reviewer					
	DNA-Mixture		1344700/3		Valerie Caldwell	Sampler	1202/11/0/51	DIGISTRAL	LZOZICDIC	1704/2021	1707/50/11
					Mike Hart	Reference sampler					
	2000	100			Sharelle Nydam	Reporter					
	FB5840				Helen Williams	Reviewer	100000000	-			
	UNA Database -		/34420128		Valerie Caldwell	Sampler	11/02/2021	8/03/2021	4/03/2021	29/04/2021	11/05/2021
					Phillip Mcindoe	Reference sampler					
		W		ž.	Lisa Farrelly	Reporter					
	FB5840		724400427		Biljana Micic	Reviewer	14,000,000	PE0672018	B102/20094	200040005	11/05/20124
	Saliva		10107+101		Valerie Caldwell	Sampler	110212021	707 500	ologico de	23/04/2021	1100/2021
					Phillip Mcindoe	Reference sampler					
					Megan Mathieson	Reporter					
	FB5840		SALOCARCE		Tara Prowse	Reviewer	4440000044	-coctcore	*0001001*	PODOLOGO	11/05/2024
	Saliva		041074461		Valerie Caldwell	Sampler	110515051	1 707/50/0	TO TO TO	12071502	1702/2071
Cohemany					Phillip Mcindoe	Reference sampler					
Dinaiy		/		E	Angelina Keller	Reporter					
	FB5870		TOAKODANOT		Rhys Parry	Reviewer	DE INCOMPA	19/04/2021	4Cinetions	141050004	141051000
	DNA Parentage		18107+60		Helen Williams	Sampler	170717007	03/05/2021)		1703/50/1	1702/20/11
					Mike Hart	Reference sampler					
				2	Penelope Taylor	Reporter					
	FB5870		DOCUCARCT		Ingrid Moeller	Reviewer	25,000,0004	19/04/2021	4500470034	11/05/0024	11/05/2021
	DNA Parentage		00707+60		Helen Williams	Sampler	1707170077	03/05/2021)		1703/5071	1702/2021
				7	Michelle Margetts	Reference sampler					
					Jacqui Wilson	Reporter					
	FB5870		7344200745		Adrian Pippia	Reviewer	1500000130	19/04/2021	42/04/2024	11/05/0024	11/05/2021
	DNA Parentage		0		Cindy Chang	Sampler	1707170	03/05/2021)		7070011	7070000
				֡							

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CTS Style Participant Laboral Maria La	HINCH			CTS	Proficiency Te	ests 57x require sample screening						
Foreign Vivel Code Number(s) FR Number Staff Role Role Role Staff Role Role Staff Role Staff Role Staff Role Staff Role Staff Role Staff		CTS Proficien	ncy Tests 58x a	re sam <mark>ple spe</mark> proficier	cific and pre-s	screened. No screening techniques a <u>VOT</u> be used for sampling scientists.	are required and therefore	e this set of				
Forest Mark		CTS type	Participant Number (Web Code)	Lab Number(s)	FR Number	Staff	Role	Received Date	Due Date	Date Submitted	Follow-up completed	Feedback presented
Foreste Biology LACETRY Adde Right Reference sampler Ref						Thomas Nurthen	Reporter					
Forestic Biology (ARTERY)		FB5702	U2483J			Sharon Johnstone	Reviewer	PCMC/2018	*COC/SOFE	PCACTABLY?	SEMERODA	+cucrous
Michele Margets Reference sampler Reponsition Michele Margets Reference sampler Reponsition Michele Margets Reference sampler Reponsition Michele Margets Reference sampler Reference		Forensic Biology				Abbie Ryan	Sampler	CONTENT	Signification	7717177	201001202	202/00/0
Page						Michelle Margetts	Reference sampler	XII.				
FBS002 U4483C						Ingrid Moeller	Reporter					
PMA - Semen F42XX Valeria Caldwell Sampler Sa		FB5802	U2483C			Deborah Nicoletti	Reviewer					
Michelle Margetis Reference sampler Reference sampler Reference sampler Reference sampler Reference sampler Reference sampler Reviewer 1,106,2021 1,006,20		DNA - Semen	(F432XC)			Valerie Caldwell	Sampler	6/03/2021	3/05/2021	29/04/2021	25/05/2021	6/08/2021
Station Bytes Rejenter Allian McLlevin Rejenter Allian McLlevin Sampler team 3 Sampler team 3 Sampler team 4 Abbie Ryan Sampler team 5 Sa						Michelle Margetts	Reference sampler					
FBS/03	March					Sharon Byrne	Reporter					
FBS781 U44834 Kristina Morton Sampler Lem 5 Source						Allan McNevin	Reviewer					
FBS 70						Abbie Ryan	Sampler Item 1					
February Name February Fe		FB5781	07483.1			Kristina Mortan	Sampler Item 2					
Felcin Williams Sampler team 4 Felcin Williams Sampler team 5 Sampler team 6 Sa		Body Fluid	(NZWC37)			Valerie Caldwell	Sampler Item 3	8/04/2021	24/05/2021	11/05/2021	4/08/2021	6/08/2021
FBE703 UZ483C FBE704 Cindy Chang Sampler Rem 6 Sam						Helen Williams	Sampler Item 4	أاريا				
Janine Seymout-Murray Sampler team 6						Cindy Chang	Sampler Item 5					
FBS103 U2483C Aligela Adamson Reviewer Trov.vv Aligela Adamson Aligela Ad						larine Savmour Murray	Sampler from 6					
FBS703 Uz483C						Samue Seymouranay	Sample remo					
Fabina						Allison Lloyd	Керопег	7				
Febso		FB5703				Angela Adamson	Reviewer	28/04/2024	24/06/2024	15/05/2021	13/07/2021	5.08/2024
FBS03		Forensic Biology				Janine Seymour-Murray	Sampler					
FBS803 U2483A DIASTIN Howes Reviewer TTCVJV) Kristina Morton Sampler TTCVJV) TTCVJVV TTCVJV TTCVJ	Annil		- 00			Mike Hart	Reference sampler	200				
FB6803 U24834 Liverage Control of the contr						Justin Howes	Reporter					
NA Parentage (195222) Control of the control		FB5803	U2483A			Josie Entwistle	Reviewer	14 MEYONDA	P4 10E/2024	4E/06/2024	12/07/2024	ACOC18012
PB6811 (a) Uv483F Emma Caunt Howes Reporter Tuma Caunt Howes Reporter Tuma Caunt Howes Reporter Tuma Caunt Reviewer Tuma Caunt Tuma Caunt Reviewer Tuma Caunt Reviewer Tuma Caunt Tuma Caunt Tuma Caunt Reviewer Tuma Caunt Tuma Caunt Tuma Caunt Reviewer Tuma Caunt Tuma Ca		DNA - Blood	(TZGVJV)			Kristina Morton	Sampler	7970001	1300000	1700000	7071000	100000
PB6871 (a) U2483F Emma Caunt Reviewer T1/06/2021 2008/2021 12/07/2021 24/08/2021						Kevin Avdic	Reference sampler					
FB6871 (a) U2483F Convertage 1 UA483F Convertage 1 UA483F Convertage 1 UA483F Convertage						Justin Howes	Reporter					
Michelle Margets Abbie Ryan Sampler Michelle Margets Abbie Ryan Abbie Ryan Abbie Ryan Sampler Abbie Ryan Sampler Abbie Ryan		FB5871 (a)	U2483F			Emma Caunt	Reviewer	reactaurs	PEDC15UIC	PCOC/ZOICP	PCUCIONIAC.	500,000,00
Michelle Margetts Reference sampler Reporter		DNA Parentage	(LWRCQC)			Abbie Ryan	Sampler	11000000	- CONTROL	1500115051	20200112	20100100
PB6871 (b) U2483C Charles Ch						Michelle Margetts	Reference sampler					
FB6871 (b) U24835 Sharon Johnstone Reviewer 11/06/2021 19/07/2021 19/						Josie Entwistle	Reporter					
ONA Parentiage (NH7JEA) Abbie Ryan Sampler 1100c.402 1300c.402 1300		FB5874 (b)	U2483G			Sharon Johnstone	Reviewer	A A LINE COLUMN	- COLLEGE	Provizoros.	-COCOCUPAC	Cociocios
Mike Hart Reference sampler Thomas Nurthen Reporter V3483H Kylio Rika Reviewer (U65223) Abbie Rvan Sampler	imay	DNA Parentage	(NH7JEA)			Abbie Ryan	Sampler	1110012021	EIONI EUCH	1207 I DIGI	24/10/2021	201031202
U2483H Kylio Rika Reviewer 11/06/2021 2/09/2021 24/08/2021 (U65223) Abbie Rvan Sampler 11/06/2021 2/09/2021 24/08/2021						Mike Hart	Reference sampler					
UZ483H (U85223) Kylio Rika Reviewer Sampler 11/06/2021 2/00/2021 24/08/2021						Thomas Nurthen	Reporter					
(U85223) Abbie Rvan Sampler 11/06/2021 24/08/2021		FB5871 (c)	U2483H			Kylio Rika	Reviewer				-	0000000
		DNA Parentage	(U8G223)			Abbie Ryan	Sampler	11/06/2021	2/08/2021	16/0//2021	24/08/2021	30/09/2021

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				CTS Proficie	CTS Proficiency Testing Schedule 2021						
MONTH			CTS	Proficiency Te	CTS Proficiency Tests 57x require sample screening						
	CTS Proficien	ıcy Tests 58x a	ire sample spe proficier	cific and pre-s	CTS Proficiency Tests 58x are sample specific and pre-screened. No screening techniques are required and therefore this set of proficiencies should <u>NOT</u> be used for sampling scientists.	es are required and thereforests.	e this set of				
	CTS type	Participant Number (Web Code)	Lab Number(s)	FR Number	Staff	Role	Received Date	Due Date	Date Submitted	Follow-up completed	Feedback presented
				A.S.	Emma Caunt	Reporter					
	FB5704 Forensic				Angelina Keller	Reviewer	-	-			-
	Biology		134422100		Kristina Morton	Sampler	15/0/1/2021	1703/50/1	74/08/2021	200/180/05	SOURCE STATE
					Michelle Margetts	Reference sampler					
					Adrian Pippia	Reporter					
	FB5804		- PECCASON		Alicia Quartermain	Reviewer		***************************************	PEOCYGOIC	***************************************	*0000000
	DNA-Semen		1177461		Valerie Caldwell	Sampler	100112021	1303/2021	1703/507	1707160106	20103150105
					Michelle Margetts	Reference sampler					
					Generosa Lundie	Reporter					
July	FB5843 (a)		TOLLOCALOT		Sharelle Nydam	Reviewer	+cucizuloc	37,000,0034	1506/80/36	10M010034	PENCICKE
Sans	Saliva		134477133		Valerie Caldwell	Sampler	2310112021	2110312021	1707190197	130700161	1707710
					Julie Brooks	Reference sampler					
					Pierre Acedo	Reporter					
	FB5843 (b)		TRANSPORT		Megan Mathieson	Reviewer	1CUCIZUIOC	27/00/2021	34/08/3024	19/40/2024	PEDESCHIE
	Saliva		117766		Michelle Margetts	Sampler	2310112021	170315051	201001115	1307001361	1707717
					Julie Brooks	Reference sampler					
	Contraction of the				Melissa Cipollone	Reporter					
	FB5843 (c)		TOLLOGE		Lisa Farrelly	Reviewer	1 COUNTRIOC	*COCIONTC	PC06/80/46	*COCIONO*	ACOUNTE ALC
	Saliva		66 177461		Michelle Margetts	Sampler	2310112021	1703/5021	1707100147	120700161	707717
					Julie Brooks	Reference sampler					
					Claire Gallagher	Reporter					
	FB5872 (a)		234477700		Jacqui Wilson	Reviewer	100000001	PENCHONSE	274013034	ATM4 PODDA	ACOCICAIC
	DNA Parentage		00/7745/		Helen Williams	Sampler	1707/20/01	707/01/67	17070111	170711111	1707010
					Louise Benincasa	Reference sampler					
August					Ingrid Moeller	Reporter					
	FB5872 (b)		DOTCCLACT		Kylie Rika	Reviewer	10000001	SEMOCOURSE	10000000	ATM4 (2024	PEDENCENC
	DNA Parentage		66 1774401		Helen Williams	Sampler	1707/20/01	1707/01/67	1207/01/01	170711111	1707717
					Louise Benincasa	Reference sampler					

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				CTS Proficie	CTS Proficiency Testing Schedule 2021						
MONTH			CTS	Proficiency T.	CTS Proficiency Tests 57x require sample screening						
	CTS Proficier	ncy Tests 58x a	are sample spe proficie	cific and pre-	CTS Proficiency Tests 58x are sample specific and pre-screened. No screening techniques are required and therefore this set of proficiencies should <u>NOT</u> be used for sampling scientists.	s are required and therefore s.	this set of				
	CTS type	Participant Number (Web Code)	Lab Number(s)	FR Number	Staff	Role	Received Date	Due Date	Date Submitted	Follow-up completed	Feedback presented
					Penny Taylor	Reporter					
	FB5705 Forensic		208004407		Allan McNevin	Reviewer	1CUCOUIUC	PC0C14418	10000010181	PC0024241C	PROPERTY
	Biology		20027545		Helen Williams	Sampler	20/03/2021	1707/11/0	1202/01/01	707777	707010
					Louise Benincasa	Reference sampler					
					Angela Adamson	Reporter					
	FB5805		TAGGETTE		Cassie James	Reviewer	recount	ACOC! ANIO	SCOCIONISC.	PEOCIE PIC	ACOULCASE.
	DNA-Blood		110771101		Cindy Chang	Sampler	177103177	2071110	1707101117	70707	TO TO TO TO
Contombos					Louise Benincasa	Reference sampler					
eptellinei					Valerie Caldwell	Reporter					
					Allison Lloyd	Reviewer					
					Janine Seymour-Murray	Sampler Item 1					
	FB5782		SCOCCAACT		Cindy Chang	Sampler Item 2	111000031	2214412024	10001110	4,047,3033	44,0472033
	Identification		-		Kristina Morton	Sampler Item 3	170701		-	-	110111011
					Michelle Margetts	Sampler Item 4					
					Kristina Morton	Sampler Item 5					
					Helen Williams	Sampler Item 6					
					Matthew Hunt	Reporter	3-1				
	FB5706		NOSCENARY		Rhys Parry	Reviewer	244,0024	*C0C1C110C	HOUGHON	4430473033	4410412022
	Forensic Biology		100771101		Michelle Margetts	Sampler	17071117	17077107	3151516	7707110111	7707110111
Ortobor					Louise Benincasa	Reference sampler					
october					Claire Gallagher	Reporter					
	FB5806		SUSCENEE		Jacqui Wilson	Reviewer	244,0024	*COCIC FIOC	PCOC/CH/8	44104/2022	44 104 12022
	DNA-Mixture		one your		Janine Seymour-Murray	Sampler	17071117	170777107	1202/2110	77071011	7707110111
					Louise Benincasa	Reference sampler					

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			CTS	Proficiency	Testing Schedule 2022					
MONTH			CTS Profi	ciency Tests	57x require sample screening			¥		
	CTS Proficiency To				ened. No screening technique be used for sampling scient		erefore this set			
	CTS type	Participant Number (Web Code)	Lab Number(s)	FR Number	Staff	Role	Received Date	Due Date	Date Submitte	
					Luke Ryan	Reporter				
	FB5701				Allison Lloyd	Reviewer			L. LEGGER	
	Forensic Biology		723188457		Valerie Caldwell	Sampler	- 18/01/2022	7/03/2022	10/02/202	
V.					Kevin Avdic	Reference sampler	-24			
January					Kerry-Anne Lancaster	Reporter	_ 7			
	FB6801		tation in the		Deborah Nicoletti (Rogers)	Reviewer	Salumetra es	120000000	100000000000000000000000000000000000000	
	DNA-Mixture		723188468		Helen Williams	Sampler	- 18/01/2022	7/03/2022	1/03/2022	
					Mike Hart	Reference sampler				
					Helen Williams	Reporter				
	FB5840 (A)				Pierre Acedo	Reviewer	5			
	DNA Database - Saliva		734422951		Janine Seymour-Murray	Sampler	- 10/02/2022	4/04/2022		
	Sailva				Kim Estreich	Reference sampler	-X			
					Biljana Micic	Reporter				
	FB5840 (B)				Kirsten Scott	Reviewer				
February	DNA Database - Saliva		734422960		Kristina Morton	Sampler	- 10/02/2022	4/04/2022	9/03/202	
	Saliva				Madison Gulliver	Reference sampler				
					Tara Prowse	Reporter				
	FB5840 (C)				Abbie Ryan	Reviewer				
	DNA Database - Saliva		734422974		Amy Morgan	Sampler	- 10/02/2022	4/04/2022	6/03/202	
	Sallva				Madison Gulliver	Reference sampler				
					Alicia Quartermain	Reporter				
	FB5870 (A)				Angelina Keller	Reviewer	100000000000			
	DNA Parentage		734423057		Cindy Chang	Sampler	- 24/02/2022	18/04/2022		
					Mike Hart	Reference sampler				
					Adrian Pippia	Reporter				
	F85870 (B)				Rhys Parry	Reviewer	·			
	FB5870 (B) DNA Parentage		734423068		Michelle Margetts	Sampler	- 24/02/2022	18/04/2022		
						Mike Hart	Reference sampler	->-		
			1		Justin Howes	Reference sampler Reporter				
	ERE703				Josie Entwistle	Reviewer	-			
		FB5702 Forensic Biology		734423181		Valerie Caldwell	Sampler	- 10/03/2022	2/05/2022	
					Valetie Caldwell	Reference sampler	- %			
		+			Sharon Johnstone	Reporter				
	FB5802				Thomas Nurthen	Reviewer	¥7			
	DNA - Semen		734423190		Helen Williams	Sampler	- 10/03/2022	2/05/2022		
					neien vvillams	Reference sampler	-			
March		+					-			
						Reporter				
						Reviewer	-			
	FB5781					Sampler Item 1	4			
	Body Fluid					Sampler Item 2	- (p.,			
	Identification					Sampler Item 3	-8			
						Sampler Item 4	No.			
						Sampler Item 5	100			
	(C)			0		Sampler Item 6				

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EQUIPMENT

Equipment includes, but is not limited to, measuring instruments, reference standards and analytical systems.

Note: Refer to the General Accreditation Criteria: Equipment assurance, in-house calibration and equipment verification, available from the NATA website, for further information.

Please complete the table below (or attach a separate sheet) indicating whether the equipment is calibrated in-house or externally.

Calibration of equipment is necessary when:

- the measurement accuracy of measurement uncertainty affects the validity of reported results;
- the equipment is required to establish the metrological traceability of reported results.

Where calibration of equipment is deemed not necessary, it is still required that the facility ensure equipment has been verified that it conforms with specified requirements (e.g. method requirements; manufacturer's requirements).

		Calibrated	
		In-house	Externally
Equipment description	Yes	Procedure (as per Methods Manual, national or international standard, etc)	Yes
Genetic Analysers : Forensic DNA Analysis currently has two 3500xl instrumen 3130xl instrument that was taken out of use on 15/02/2021. To be suitable fo service requirements and continue to pass internal spectral checks			
200418261 ; 3130 (B) - Analyser , 3130xl			Yes
200440262 : 2500 (A) Analyses 2500vl			Yes
200418262 ; 3500 (A) - Analyser , 3500xL			
200418262 ; 3500 (A) - Analyser , 3500xL 200418263 ; 3500 (B) - Analyser , 3500xL			Yes
	n external p	rovider. The instruments	
200418263; 3500 (B) - Analyser, 3500xL QuantStudio: has 6 monthly maintenance, and 2 yearly calibration check by a	n external p	provider. The instruments	

and pipetting robotics, the PCS can do single channel POVAs to 5000uL. Both the MVS and PCS instruments are calibrated prior to use, using either a plate or calibration solutions (refer QIS#31956 and 26628). All reagents, consumables and calibration plate/solutions of the MVS and PCS systems are traceable back to the NIST Standard. The MVS calibrations plates are sent out to external providers

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200418246 ; ARTEL MVS Calibration Plate	Yes
200418247 ; ARTEL MVS Calibration Plate	Yes
Balance: Receives 1 year service and 3 year NATA calibration by an external provider. Mor completed internally. The balance is deemed suitable for use if it meets all NATA calibratio continues to pass internal 1 & 6 monthly checks.	
200418260 ; Balance , Electronic XS105DU	Yes
BSD FTA punching instrument receives annual servicing by an external provider. No calibra suitable for use if servicing finds no issues with instrument function	ation is required. Instrument is
200422004 ; BSD 600 Ascent A2	Yes
Centrifuges: Within Forensic DNA Analysis we have both critical centrifuges and non-critical used for DNA extraction, microcon, nucleospin processing, semen testing or phadebas sup Critical centrifuges are calibrated externally. They must pass external calibration to be suite	ernatant testing are deemed critical
200418244 ; Centrifuge , Eppendorf 5424	Yes
200418251 ; Centrifuge , Eppendorf 5424	Yes
200421429 ; Centrifuge , Eppendorf 5425	Yes
200422136 ; Centrifuge , Eppendorf 5425	Yes
200422137 ; Centrifuge , Eppendorf 5425	Yes
200422138 ; Centrifuge , Eppendorf 5425	Yes
200422139 ; Centrifuge , Eppendorf 5430	Yes
200421645 ; Centrifuge , Eppendorf 5804	Yes
200418255 ; Centrifuge , 333506	Yes
200419296 ; Centrifuge , 333506	Yes
200418254 ; Centrifuge , Sigma 41640	Yes
ı ,	

and below 5% for ≥11µL, plus 6 monthly preventative maintenance by an external provider. The instrument is suitable for use if it meets both internal verifications and external servicing.

200418618 ; Liquid Handler , Hamilton STARlet (B)	Yes	QIS#26628	Service
200418619 ; Liquid Handler , Hamilton STARlet (A)	Yes	QIS#26628	Service
200418620 ; Liquid Handler , Hamilton STARlet (C)	Yes	QIS#26628	Service

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Microscopes : receive annual servicing by an external provider. No calibration is r servicing finds no issues with function	required. Instrument is suitable for use if
200418265 Microscope , Olympus BX41	Service
200418266 Microscope , Olympus BX41	Service
200418267 Microscope , Olympus BX41	Service
200418268 Microscope , Olympus BX41	Service
200420451 Microscope , Nikon Eclipse Ci-L	Service
200421945 Microscope , Nikon Eclipse Ci-L	Service

POVAs have been assessed as non-critical pieces of equipment. The checks that are in place to ensure pipettes are within range and suitable for use include: positive and negative controls on batches, initial NATA calibration certificates and internal 3 monthly checks with traceable ARTEL equipment and reagents. To be suitable for use POVAs must - pass initial NATA calibration and 3 monthly internal verifications using ARTEL to criteria given in QIS#26628 (MVS) and QIS#31956 (PCS) which is relative inaccuracy and co-efficient of variation below 10% for \leq 10 μ L and below 5% for \geq 11 μ L

POVA 10-100u ,	I10767K	Yes		Initial
POVA 20-200uL,	G42714K	Vaa		Initial
Eppendorf Reasearch Plus		res		Initial
POVA 20-200uL ,	G42713K	Yes		Initial
	15404714			
,	J54217K	Yes		Initial
POVA 0.5-10uL ,	J54848K	Vec		Initial
Eppendorf Research Plus		103		IIIIIIai
•	G37613K	Yes		Initial
	00704516			
•	G37615K	Yes		Initial
	V22491			1 '4' 1
Finnpipette		Yes	QIS#3156	Initial
POVA 1-10ul , Thermo	FK26794	Yes		Initial
Finnpipette		103		IIIIIai
	FK26795	Yes	QIS#26628	Initial
• •	1,00,10,1			
POVA 1-10ul , Thermo Finnpipette	V22491	Yes		Initial
POVA 1-10ul Cliptip,	NH47298	Yes		Initial
• • •	KH30542	Yes		Initial
	11 102020			
	JH92826	Yes		Initial
*	I10323K		-	
Eppendorf Research Plus	11002010	Yes		Initial
	Eppendorf Research Plus POVA 20-200uL , Eppendorf Reasearch Plus POVA 20-200uL , Eppendorf Reasearch Plus POVA 0.5-10uL , Eppendorf Research Plus POVA 0.5-10ul , Thermo Finnpipette POVA 1-10ul Cliptip , Thermo POVA 1-10ul Cliptip , Thermo POVA 1-10ul Multi Channel , Thermo POVA 10-10ul ,	Eppendorf Research Plus POVA 20-200uL , Eppendorf Reasearch Plus POVA 20-200uL , Eppendorf Reasearch Plus POVA 0.5-10uL , Eppendorf Research Plus POVA 0.5-10ul , Thermo Finnpipette POVA 1-10ul Cliptip , Thermo POVA 1-10ul Cliptip , Thermo POVA 1-10ul Multi Channel , Thermo POVA 10-100uL , I10323K	Eppendorf Research Plus POVA 20-200uL , Eppendorf Reasearch Plus POVA 20-200uL , Eppendorf Reasearch Plus POVA 0.5-200uL , Eppendorf Research Plus POVA 0.5-10uL , Thermo Finnpipette POVA 1-10ul , Thermo Finnpipette POVA 1-10ul , Thermo Finnpipette POVA 1-10ul Cliptip , Thermo POVA 1-10ul Cliptip , Thermo POVA 1-10ul Multi Channel , Thermo POVA 1-10ul Multi Channel , Thermo POVA 1-10ul Nulti Channel , Thermo POVA 1-10ul Multi Channel , Thermo POVA 1-10ul Cliptip , Thermo POVA 1-10ul Multi Channel , Thermo POVA 1-10ul Multi	Eppendorf Research Plus POVA 20-200uL , Eppendorf Reasearch Plus POVA 20-200uL , Eppendorf Reasearch Plus POVA 20-200uL , Eppendorf Reasearch Plus POVA 0.5-10uL , Eppendorf Research Plus POVA 0.5-10ul , Thermo Finnpipette POVA 1-10ul Cliptip , Thermo POVA 1-10ul Cliptip , Thermo POVA 1-10ul Multi Channel , Thermo POVA 10-10ouL , I10323K Yes

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200422604	DOVA 10 100ul	LIAFORON	<u> </u>		
200422691	POVA 10-100uL,	H15260K	Yes		Initial
	Eppendorf Research Plus			_	
200422692	POVA 10-100uL , Eppendorf Research Plus	I10319K	Yes		Initial
	POVA 10-100uL,			-	
200422693		I10766K	Yes		Initial
	Eppendorf Research Plus POVA 10-100uL ,			-	
200422694		H15277K	Yes		Initial
	Eppendorf Research Plus POVA 100-1000uL ,			-	
200422633	Eppendorf Research Plus	R12542J	Yes		Initial
	POVA 100-1000uL,				
200422634	Eppendorf Research Plus	R12384J	Yes		Initial
	POVA 100-1000uL,				
200422635	Eppendorf Research Plus	R11936J	Yes		Initial
	POVA 100-1000uL,			-	
200422636	Eppendorf Research Plus	R12624J	Yes		Initial
	POVA 100-1000uL,				
200422637	Eppendorf Research Plus	R12137J	Yes		Initial
	POVA 100-1000uL ,		.,		
200422638	Eppendorf Research Plus	R12253J	Yes		Initial
	POVA 100-1000ul ,	2112224	.,	-	
200418335	Thermo Finnpipette	CH32624	Yes		Initial
00040000	POVA 100-1000uL ,	3102245			
200422602	Socorex Acura 825	4	Yes	QIS#3156	Initial
000400000	POVA 100-1000uL,	3102142	Yes	or	Initial
200422603	Socorex Acura 826	0	res	QIS#26628	Initial
200440969	POVA 100-1000ul,	0063000	Yes	Q10#20020	Initial
200419868	Socorex Calibra 822	9063000	165		IIIIIIai
200420149	POVA 100-1000ul Clip	JH91415	Yes		Initial
200420149	Tip , Thermo	3091413	163		IIIIIai
200421992	POVA 100-1000ul Clip	RH15216	Yes		Initial
200421992	Tip , Thermo	KH13210	163		IIIIIai
200418298	POVA 100-1000ul Cliptip,	JH91419	Yes		Initial
200410290	Thermo	30191419	163		IIIIIai
200418300	POVA 100-1000ul Cliptip,	JH91424	Yes		Initial
200410300	Thermo	01191424	103		IIIIIai
200418334	POVA 2-20ul , Thermo	CH45188	Yes		Initial
200410334	Finnpipette		100		miliai
200418342	POVA 2-20ul , Socorex	1405115	Yes		Initial
200410042	Acura 825	0	100		miliai
200422676	POVA 2-20uL , Eppendorf	Q48353J	Yes		Initial
200122010	Research Plus	3 100000	. 55		
200422677	POVA 2-20uL , Eppendorf	Q48385J	Yes		Initial
200122011	Research Plus	3 100000	. 55		
200422678	POVA 2-20uL , Eppendorf	Q48435J	Yes		Initial
	Research plus	<u> </u>			
	POVA 2-20uL, Eppendorf	Q48386J	Yes		Initial
200422679	Research Plus	(340.)00.			

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	DOV/A 0 00 1 E 1 f	1	1		
200422680	POVA 2-20uL , Eppendorf Research Plus	J35823B	Yes		Initial
200422681	POVA 2-20uL , Eppendorf Research Plus	J35801B	Yes		Initial
200422682	POVA 2-20uL , Eppendorf	J35800B	Yes	-	Initial
200422727	Research Plus POVA 20-200uL ,	G42318K	Yes		Initial
	Eppendorf Reasearch Plus		165	-	IIIIIIai
200422601	POVA 20-200uL , Socorex Acura 825	3101230 5	Yes		Initial
200422729	POVA 20-200uL , Eppendorf Reasearch Plus	G42676K	Yes		Initial
200422730	POVA 20-200uL,	G42675K	Voc	-	Initial
	Eppendorf Reasearch Plus		Yes	_	Initial
200422879	POVA 20-200uL , Socorex Calibra 822	9062270	Yes		Initial
200422880	POVA 20-200uL , Socorex Calibra 822	1001206 1	Yes		Initial
200422881	POVA 20-200uL , Socorex Calibra 822	9062274	Yes	-	Initial
200422885	POVA 20-200uL ,	G42369K	Yes	QIS#3156	Initial
	Eppendorf Research Plus		163	or	IIIIIai
200422732	POVA 20-200uL , Eppendorf Reasearch Plus	G42656K	Yes	QIS#26628	Initial
200418343	POVA 20-200ul , Socorex Acura 825	1407351 3	Yes		Initial
200422604	POVA 20-200uL , Socorex Acura 826	3011107 7	Yes		Initial
200418331	POVA 20-200ul , Eppendorf Research	2022626	Yes		Initial
200418320	POVA 20-200ul , Thermo Finnpipette	JH10553	Yes		Initial
200418310	POVA 20-200ul Cliptip , Thermo	JH74655	Yes	_	Initial
200418313	POVA 20-200ul Cliptip , Thermo	KH09750	Yes	_	Initial
200419811	POVA 20-200uL Cliptip , Thermo	KH09754	Yes		Initial
200421986	POVA 5-50uL , Thermo Finnpipette	GH71377	Yes		Initial
200418321	POVA 5-50ul , Thermo Finnpipette	GH71376	Yes		Initial
200418328	POVA 5-50ul , Thermo	GH35698	Yes		Initial
200418329	Finnpipette POVA 5-50ul , Thermo Finnpipette	GH27001	Yes		Initial
200418339	POVA 5-50ul , Sealpette	EL16316	Yes	1	Initial
L	1		1	<u> </u>	

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200418305	POVA 5-50ul Cliptip , Thermo	JH75442	Yes	QIS#3156	Initial
200418307	POVA 5-50ul Cliptip , Thermo	JH75445	Yes	or QIS#26628	Initial
200420053	POVA 5-50uL Cliptip , Thermo	JH75443	Yes		Initial
200419981	POVA 50-300uL Multi Channel , Labsystems Finnpipette	E43203	Yes	QIS#26628	Initial
200421847	POVA Multipette E3 , Eppendorf	K18155I	Yes	QIS#26628	Initial
200422610	POVA Multipette Repeater , Eppendorf Multipette M4	M48200J	Yes	QIS#26628	Initial
200422611	POVA Multipette Repeater , Eppendorf Multipette M4	M46990J	Yes	QIS#26628	Initial

QIAsymphony instrument has two parts SP and AS modules. Both modules are serviced annually by an external provider. The AS module will also have 3 monthly verifications for dispensing volumes using the ARTEL MVS instrument. The QIAsymphony will be suitable for use if servicing finds no issues with instrument function and if 3-monthly checks in the ARTEL pass criteria given in QIS#26628 (MVS).

200418249 ; QIAsymphony AS A , QIAsymphony AS	Yes	QIS#26628	Service
200420328 ; QIAsymphony AS B , QIAsymphony AS	Yes	QIS#26628	Service
200418248 ; QIAsymphony SP A , QIAsymphony SP	N/A	N/A	Service
200420192 ; QIAsymphony SP B , QIAsymphony SP	N/A	N/A	Service

Thermal cyclers: Forensic DNA Analysis previously used six 9700 thermal cyclers, these were taken out of use on 10/01/2022. They have been replaced with six Proflex thermal cyclers that were implemented on 10/01/2022. Annual checks by an external provider, and internal weekly cycle and rate checks. The instruments are suitable for use if they pass external annual checks and weekly internal checks.

200418274 ; Thermal Cycler (B) , ABI 9700	Service
200418275 ; Thermal Cycler (C) , ABI 9700	Service
200418276 ; Thermal Cycler (D) , ABI 9700	Service
200418277 ; Thermal Cycler (E) , ABI 9700	Service
200418278 ; Thermal Cycler (F) , ABI 9700	Service
200418279 ; Thermal Cycler (G) , ABI 9700	Service
200420445 ; Thermalcycler Proflex 1 Base, Thermo 200422684 ; Thermalcycler Proflex 1 Samp Block, Thermo	Service

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000400440 TI	
200420446 ; Thermalcycler Proflex 2 Base, Thermo	Service
200422685 ; Thermalcycler Proflex 2 Samp Block,	Service
Thermo	
200420447; Thermalcycler Proflex 3 Base, Thermo	Service
200422686 ; Thermalcycler Proflex 3 Samp Block,	
Thermo	
200420448; Thermalcycler Proflex 4 Base, Thermo	Service
200422687; Thermalcycler Proflex 4 Samp Block,	Service
Thermo	
200420449; Thermalcycler Proflex 5 Base, Thermo	Camiaa
200422688; Thermalcycler Proflex 5 Samp Block,	Service
Thermo	
200420576 ; Thermalcycler Proflex 6 Base, Thermo	Service
200422689; Thermalcycler Proflex 6 Samp Block,	
Thermo	
THOMAS	

Thermometers: Within Forensic DNA Analysis we have both critical and non-critical thermometers. Fridges and freezers within Forensic DNA Analysis are monitored by a BMS system (with alarms), however in addition to the BMS many fridges and freezers have non-critical thermometers in them for easy of user observation only. Non-critical thermometers are not included below. Thermometers that are deemed critical are those used for DNA extraction water-baths, nucleospin clean-ups and the CE denaturation blocks. Critical thermometers are checked internally every six months (single point) and a full check completed every 5 years. Thermometers are deemed suitable for use if they pass all internal checks (as per QIS#10670)

200419950 ; Thermometer - Alcohol 24 , Alcohol/Glass 76mm Part/Immer	Yes	QIS#10670	
200419951 ; Thermometer - Alcohol 25 , Alcohol/Glass 76mm Part/Immer	Yes	QIS#10670	
200418670 ; Thermometer - Alcohol 32 , Alcohol/Glass 76mm Part/Immer	Yes	QIS#10670	
200418668 ; Thermometer - Alcohol 9 , Alcohol/Glass 76mm Part/Immer	Yes	QIS#10670	

Timers: Within Forensic DNA Analysis we have both critical and non-critical timers. Timers that are used to remind staff to return to samples post-denaturation, or during extraction are non-critical (non-critical timers are not listed below). Timers that are used for making a "result" reading on presumptive tests (AP and PSA) are deemed critical. Timers that are deemed critical are checked internally every 6 months against the National Measurement Institute (NMI) WebTimer, they must pass this internal check to be suitable for use (as per QIS#10672)

200420325 ; Timer 2 , Electronic	Yes	QIS#10672	
200418259 ; Timer 34 , Electronic	Yes	QIS#10672	
200420501 ; Timer 4 , Electronic	Yes	QIS#10672	
200418257 ; Timer 7 , Electronic	Yes	QIS#10672	
200421923 ; Timer 41, Lab Co	Yes	QIS#10672	
200422531 ; Timer 42, Lab Co	Yes	QIS#10672	

[★] For facilities performing in-house calibrations: please provide a copy of the test method and statement of capability of each in-house calibration identified above.

NATIONAL ASSOCIATION OF TESTING AUTHORITIES, AUSTRALIA (ABN 59 004 379 748)

Accreditation No: 41 Site No: 14171 Job No: 82214 SUBCONTRACTING, AGENCY OR FRANCHISING ARRANGEMENTS Since your previous assessment, does your facility now operate under formal subcontracting, agency or franchising agreement with another organisation which you have not advised NATA of? ☐ Yes ⊠ No Note: While we do not sub-contract out any work from Forensic DNA Analysis to an external group, we do complete small scale commercial work (validations and environmental sample monitoring) for external organisations such as ARUMA.

Note: As per clause 5.3 the laboratory cannot claim conformity with ISO/IEC 17025 for externally provided laboratory activities on an ongoing basis.

TEST REPORTS, SAMPLING REPORTS AND CALIBRATION CERTIFICATES

If yes, please provide details of the arrangement and the principal organisation.

Please provide an example copy of a recently completed test report and/or sampling report and/or calibration certificate.

Note: Refer to the General Accreditation Criteria: use of the NATA emblem, NATA endorsement and references to accreditation, available from the NATA website, for criteria relating to endorsing reports.

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Forensic and Scientific Services

STATEMENT OF WITNESS

Peer Reviewed..... Case Analyst. Peer Analyst... Date Issued 23/07/2021

Client Reference Report Number

QP2100607105

QUEENSLAND) TO WIT)

I, Allan Russell MCNEVIN, of Brisbane in the State of Queensland, do solemnly and sincerely declare that:-

- 1. I am employed by Queensland Health Forensic and Scientific Services (QHFSS) at Coopers Plains, Brisbane.
- 2. I hold the position of scientist in the Forensic DNA Analysis laboratory of QHFSS.
- 3. I was awarded a Bachelor of Applied Science from Queensland University of Technology.
- 4. I am a member of the Australian and New Zealand Forensic Science Society.
- 5. This is my statement in relation to the alleged offence that Occurrence Number QP2100607105 refers. The defendant in this matter is The complainant in this matter is

te solely to the item(s) and/or sample(s) as received.

Allan Russell MCNEVIN

...... Date 23 July 2021

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STATEMENT OF WITNESS Client Reference QP2100607105 6. Laboratory records show that on 8 April 2021, S/CONST BRETT ANTHONY WINNETT delivered the following 17 1094408050, 1094408065, 1094408078, 1094408084, 1094408099, 1094408131, 1094408154, 1094408177, 1096024457, 1096024462, 1096024474, 1096024488, 1096024493, 1096024509, 1096024510, 1096024534, 1096024548 7. Laboratory records show that on 13 April 2021, STELLA CONDOLEON delivered the following reference sample: 1095780816 8. Laboratory records show that on 15 April 2021, S/SGT STEPHAN PAUL FOXOVER delivered the following reference 1095780802 9. The results of the scientific examinations conducted in the laboratory are as follows: The DNA profile of has been determined from the reference sample. 1095780802 The DNA profile of has been determined from the reference sample. 1094408050 - EXH A- DRIED RED STAIN 1CM X 1CM [SWBL] from the vehicle keys in the loungeroom The mixed DNA profile obtained from this sample indicates the presence of DNA from two contributors. As such, an assumption of two contributors has been made for statistical analysis. Each of the reference DNA profiles associated to this case has been compared with this mixed DNA profile separately, in order to assess whether or not any of them may have contributed DNA. Based on statistical analysis, the results are as follows: It is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if he had not. It is estimated that the mixed DNA profile obtained is approximately 14 times more likely to have occurred if had not contributed DNA rather than if she had. 1094408065 - EXH B- DRIED RED STAIN 3CM X 3CM [SWBL] from the checked shirt in the main bedroom The mixed DNA profile obtained from this sample indicates the presence of DNA from three contributors. Since this sample is said to have been collected from an item of clothing belonging to unexpected to find DNA which could have come from him. In order to interpret this mixed DNA profile an assumption of DNA from three contributors, one of whom is has been made. The DNA profile of has been compared to this mixed DNA profile, to assess whether or not she may have contributed DNA along with Based on statistical analysis, it is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if she had not. The results relate solely to the Item(s) and/or sample(s) as received. Allan Russell MCNEVIN Date 23 July 2021 Page 2 of 12

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STATEMENT OF WITNESS QP2100607105 Client Reference 1094408078 - EXH C- DRIED RED STAIN 3CM X 3CM [SWBL] from the checked shirt in the main bedroom The mixed DNA profile obtained from this sample indicates the presence of DNA from two contributors. Since this sample is said to have been collected from an item of clothing belonging to it would not be unexpected to find DNA which could have come from him. In order to interpret this mixed DNA profile an assumption of DNA from two contributors, one of whom is has been made. The DNA profile of has been compared to this mixed DNA profile, to assess whether or not she may have contributed DNA along with Based on statistical analysis, it is estimated that the mixed DNA profile obtained is greater than 100 billion times more had contributed DNA rather than if she had not. likely to have occurred if 1094408154 - EXH K- EXCISED DRIED RED STAIN 2CMX2CM [FABRIC] from the checked shirt in the main bedroom The mixed DNA profile obtained from this sample indicates the presence of DNA from three contributors. Since this sample is said to have been collected from an item of clothing belonging to unexpected to find DNA which could have come from him. In order to interpret this mixed DNA profile an assumption of DNA from three contributors, one of whom is has been made. The DNA profile of has been compared to this mixed DNA profile, to assess whether or not she may have contributed DNA along with Based on statistical analysis, it is estimated that the mixed DNA profile obtained is greater than 100 billion times more had contributed DNA rather than if she had not. likely to have occurred if 1094408084 - EXH D- INVISIBLE STAIN [SWBL] from the steering wheel The mixed DNA profile obtained from this sample indicates the presence of DNA from two contributors. As such, an assumption of two contributors has been made for statistical analysis. Each of the reference DNA profiles associated to this case has been compared with this mixed DNA profile separately, in order to assess whether or not any of them may have contributed DNA. Based on statistical analysis, the results are as follows: It is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if he had not. It is estimated that the mixed DNA profile obtained is approximately 16000 times more likely to have occurred if had contributed DNA rather than if she had not. 1094408099 - EXH E- DRIED RED STAIN 1CM X 1CM [SWBL] from the handbrake The mixed DNA profile obtained from this sample indicates the presence of DNA from two contributors. As such, an assumption of two contributors has been made for statistical analysis. Each of the reference DNA profiles associated to this case has been compared with this mixed DNA profile separately, in order to assess whether or not any of them may have contributed DNA. Based on statistical analysis, the results are as follows: It is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if he had not. The results relate solely to the item(s) and/or sample(s) as received. Allan Russell MCNEVIN Date 23 July 2021

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can be excluded as having contributed DNA to this mixed DNA profile.

1094408131 - EXH I- STAIN INVISIBLE [SWBL] From the Iphone case in the loungeroom

The mixed DNA profile obtained from this sample indicates the presence of DNA from two contributors. As such, an assumption of two contributors has been made for statistical analysis.

Each of the reference DNA profiles associated to this case has been compared with this mixed DNA profile separately, in order to assess whether or not any of them may have contributed DNA.

Based on statistical analysis, the results are as follows:

It is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if he had not.

It is estimated that the mixed DNA profile obtained is approximately 3 times more likely to have occurred if had not contributed DNA rather than if she had.

1094408177 - EXH L-EXCISED FABRIC WITH DRIED RED STAIN 1CM X1CM [FABRIC] from the cloth in the drivers side door

The DNA profile obtained from this sample matches the DNA profile of Based on statistical analysis, it is estimated that the DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if he had not.

1096024457 - JB1 - BLOOD SWAB 20CMX2CM DRY RED STAIN [SWBL] on front stairs railing

The mixed DNA profile obtained from this sample indicates the presence of DNA from two contributors. As such, an assumption of two contributors has been made for statistical analysis.

Each of the reference DNA profiles associated to this case has been compared with this mixed DNA profile separately, in order to assess whether or not any of them may have contributed DNA.

Based on statistical analysis, the results are as follows:

It is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if she had not.

It is estimated that the mixed DNA profile obtained is approximately 1.2 billion times more likely to have occurred if I had contributed DNA rather than if he had not.

1096024462 - JB2 - BLOOD SWAB 8CMX4CM DRY RED STAIN [SWBL] on lamp on bedside table in front

The mixed DNA profile obtained from this sample indicates the presence of DNA from two contributors. As such, an assumption of two contributors has been made for statistical analysis.

Each of the reference DNA profiles associated to this case has been compared with this mixed DNA profile separately, in order to assess whether or not any of them may have contributed DNA.

Based on statistical analysis, the results are as follows:

It is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if she had not.

The results relate solely to the Item(s) and/or sample(s) as received.

Alian Russell MCNEVIN ...

Date 23 July 2021



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STATEMENT OF WITNESS Client Reference QP2100607105 It is estimated that the mixed DNA profile obtained is approximately 30 times more likely to have occurred if had contributed DNA rather than if he had not. 1096024474 - JB3 - BLOOD SWAB 20CMX13CM DRY RED STAIN [SWBL] on right side of doons on bed in front bedroom The DNA profile obtained from this sample matches the DNA profile of Based on statistical analysis, it is estimated that the DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if she had not, 1096024488 - JB4 - BLOOD SWAB 2.5CMX2CM DRY RED STAIN [SWBL] on right side of doona on bed in front bedroom The DNA profile obtained from this sample matches the DNA profile of Based on statistical analysis, it is estimated that the DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if he had not. 1096024493 - JB5 - BLOOD SWAB 1.5CMX2CM DRY RED STAIN [SWBL] on pillow on floor on right side of bed The mixed DNA profile obtained from this sample indicates the presence of DNA from two contributors. As such, an assumption of two contributors has been made for statistical analysis. Each of the reference DNA profiles associated to this case has been compared with this mixed DNA profile separately, in order to assess whether or not any of them may have contributed DNA. Based on statistical analysis, the results are as follows: It is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if he had not. It is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if she had not. 1096024509 - JB6 - BLOOD SWAB 2CMX2CM WET RED STAIN [SWBL] on bathroom tile floor The DNA profile obtained from this sample matches the DNA profile of Based on statistical analysis, it is estimated that the DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if she had not. 1096024510 - JB7 - BLOOD SWAB 20CMX4CM DRY RED STAIN [SWBL] on kitchen fridge door The mixed DNA profile obtained from this sample indicates the presence of DNA from two contributors. As such, an assumption of two contributors has been made for statistical analysis. Each of the reference DNA profiles associated to this case has been compared with this mixed DNA profile separately, in order to assess whether or not any of them may have contributed DNA. Based on statistical analysis, the results are as follows: It is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if she had not. It is estimated that the mixed DNA profile obtained is approximately 2 times more likely to have occurred if had not contributed DNA rather than if he had. Allan Russell MCNEVIN The results relate solely to the item(s) and/or sample(s) as received. Date 23 July 2021 Page 5 of 12

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1096024534 - JB9 - BLOOD SWAB 5CM X 3CM DRY RED STAIN [SWBL] on handle of knife

The mixed DNA profile obtained from this sample indicates the presence of DNA from two contributors. As such, an assumption of two contributors has been made for statistical analysis.

Each of the reference DNA profiles associated to this case has been compared with this mixed DNA profile separately, in order to assess whether or not any of them may have contributed DNA.

Based on statistical analysis, the results are as follows:

It is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if she had not.

It is estimated that the mixed DNA profile obtained is approximately 1100 times more likely to have occurred if had not contributed DNA rather than if he had.

1096024548 - JB10 - BLOOD SWAB 4CM X 2CM DRY RED STAIN [SWBL] on blade of knife

The mixed DNA profile obtained from this sample indicates the presence of DNA from two contributors. As such, an assumption of two contributors has been made for statistical analysis.

Each of the reference DNA profiles associated to this case has been compared with this mixed DNA profile separately, in order to assess whether or not any of them may have contributed DNA.

Based on statistical analysis, the results are as follows:

It is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if had contributed DNA rather than if she had not.

It is estimated that the mixed DNA profile obtained is approximately 580 million times more likely to have occurred if had not contributed DNA rather than if he had.

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APPENDIX

Procedural and technical overview of DNA profiling at Forensic DNA Analysis, Forensic and Scientific Services

Forensic Biologist

It is a forensic biologist's role to:

- 1. Report on the examination of items submitted in relation to a case for the presence of possible biological material. If identified, a sample of the biological material is analysed in an attempt to obtain a DNA profile.
- 2. Report on the DNA profiles obtained from samples submitted by the Queensland Police Service (QPS) in relation to a case.

Any DNA profiles that are obtained from these samples are compared with the DNA profile obtained from an individual's reference sample to assess whether or not the individual may be a contributor of DNA. Where multiple reference DNA profiles are analysed for a case, they are all genetically different to each other unless otherwise specified. That is, they all possess differing allelic designations - see the DNA Profiling section below.

Examinations

Unless otherwise stated, the examinations of items for biological material were conducted by staff within the QPS. Sub-samples from these items were forwarded to Forensic and Scientific Services (FSS), for the purposes of conducting DNA analysis.

The descriptions of items and/or samples submitted to Forensic DNA Analysis by the QPS, and listed within this Statement, are derived from the descriptions entered by the QPS into the Forensic Register.

Receipt details are listed for items reported in this statement, including reference samples where relevant. The period of testing on these items is from the date of the first submission, to the date of this Statement. Details of specific dates of testing are retained within the Laboratory Information Management System (LIMS). Where relevant, any items that are still in progress at the time of issuing this Statement are identified and final results will be reported in an addendum Statement.

Forensic DNA Analysis operates under the agreement that the QPS are responsible for item prioritisation, sample selection, selection of screening/sampling methods, anti-contamination procedures and the application of Standard Operating Procedures (SOPs) on work undertaken on the items/samples prior to submission to the Forensic DNA Analysis laboratory. As such, forensic biologists may not be able to provide information or opinion on possible biological origin of DNA profiles that may be obtained from these samples.

At the discretion of the QPS, some items may be submitted to this laboratory for the purposes of both examination and DNA profiling. These examinations are performed in accordance with the SOPs of this laboratory. For these items, contemporaneous notes are made during the examination by the examining scientist and these notes form part of the casefile.

Forensic DNA Analysis casefiles and any samples remaining are available for independent examination and / or testing upon request.

As a representative of the laboratory, I am only able to comment on the processes performed within Forensic DNA Analysis.

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Chain of Custody

All Forensic DNA Analysis case files and exhibits are electronically tracked, monitored, and securely stored to ensure that appropriate chain of custody and continuity measures are maintained. The QPS case number and sample submission information is provided from the QPS via an electronic interface to FSS, and this information is cross-checked against labelling on the exhibit packaging prior to processing.

Entry into Forensic DNA Analysis is restricted to authorised persons only, via electronic proximity access cards. Forensic DNA Analysis forms part of a Queensland Health campus site wherein access is controlled and monitored by a security team. Records of visitors to Forensic DNA Analysis are retained.

Accreditation

Forensic DNA Analysis first achieved accreditation by the National Association of Testing Authorities (NATA) to conduct forensic DNA analyses in 1998, and has continuously maintained NATA accreditation since this date. NATA ensures continued compliance with the accreditation requirements through routine reassessment (every 3 years) and an intermediate surveillance visit (at approximately 18 months between reassessment surveys).

NATA accredited facilities are assessed against best international practices based on the ISO/IEC 17025 standard. Laboratories are deemed able to competently perform testing and analysis activities if the laboratory demonstrates that it meets compliance with the standard within the scope of their accreditation.

The parameters assessed during accreditation include:

- · Organisation and management
- Quality management system
- Personnel
- · Evidence management
- · Methods and procedures
- . Quality control and Proficiency Testing
- Equipment
- · Reporting of results
- · Procurement of services and supplies
- · Accommodation and safety
- · Security and access

For details of the current ISO/IEC 17025 Standard, refer to Standards Australia.

http://www.nata.com.au

DNA Profiling

Deoxyribonucleic acid (DNA) is a complex chemical found in almost all cells of the human body. It carries genetic information that determines the physical and chemical characteristics of a person. Forensic DNA Analysis uses two main systems for the generation of DNA profiling results. In this case, the PowerPlex® 21 System was used, which examines 21 regions (loci) of DNA, 20 of which contain highly variable short tandem repeats (STRs). The 21st region gives an indication as to the genotypic sex of the donor (for details see Table 1). The generation of a DNA profile involves a method known as the polymerase chain reaction (PCR), which is used to produce numerous copies of these specific regions of the DNA. In this way, minimal amounts of DNA isolated from small or degraded samples can be increased to a level where the DNA can be detected, profiled, and compared with DNA profiles from other samples.

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The individual components (alleles) of a DNA profile are represented by a series of peaks on a graph that are measured and given a designation, using standard sizing ladders. A person will have two alleles (represented graphically as peaks) for each locus, one inherited from the biological mother and one inherited from the biological father. However, if the same allele is inherited from both parents, only one peak will be graphically represented at that locus.

A DNA profile obtained from biological material such as blood, semen, saliva, hair or cellular material (eg. touch DNA) can be compared with the DNA profile obtained from the reference sample from any person.

Table 1: PowerPlex® 21 system, list of loci

Abbreviated Name	Scientific Name	Chromosomal Name	
Amel	AMELOGENIN	Sex (X and Y)	
D3	D3S1358	3	
D1	D1S1656	1	
D6	D6S1043	6	
D13	D13S317	13	
Penta E	Penta E	15	
D16	D16S539	16	
D18	D18S51	18	
D2	D2S1338	2	
CSF	CSF1PO	5	
Penta D	Penta D	21	
TH01	TH01	11	
vWA	HUMVWAFA31/A	12	
D21	D21\$11	21	
D7	D7S820	7	
D5	D5S818	5	
TPOX	TPOX	2	
D8	D8S1179	8	
D12	D12S391	12	
D19	D19S433	19	
FGA	HUMFIBRA	4	

Statistical Analysis of DNA Profiles

STRmix[™] is an expert statistical DNA profile analysis system developed and validated in Australia and New Zealand. Forensic DNA Analysis uses the STRmix** software to assist in the interpretation of DNA profiles and the calculation of likelihood ratios for DNA profiles generated using the PowerPlex® 21 system.

DNA profiles are initially assessed to determine the number of contributors. This value will be the minimum number of people that are required to reasonably explain the observed profile, however, it is noted that there is always the possibility that the profile is a result of a different number of contributors.

As such, if there is no indication of a contribution by more than one person, then a DNA profile is described as being from a "single contributor". If less than 40 alleles are present in a DNA profile, this is referred to as a "partial" or "incomplete" DNA profile. If there are indications of two or more contributors, then a DNA profile is described as being a "mixed" DNA profile.

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DNA Profiles Assumed To Originate From One Person (Single Source)

A person can be excluded as a possible source of the biological material if the alleles of the crime-scene DNA profile are different from the corresponding alleles of the person's reference DNA profile. If the corresponding alleles of the crime-scene DNA profile contain the same information, then that person, along with any other person who has the same corresponding alleles as the crime-scene DNA profile, can be considered as a potential contributor of the DNA.

The evidential significance of such a finding is assessed by considering two competing propositions:

Proposition 1: The crime-scene DNA originated from the person of interest.

Proposition 2: The crime-scene DNA originated from someone other than, and unrelated to, the person of interest.

The resultant figure (termed the 'Likelihood Ratio') compares the two opposing propositions. The likelihood ratio describes how likely the DNA profile obtained from the biological material is to have occurred if Proposition 1 were true (the DNA originated from the person of interest) rather than if Proposition 2 were true (the DNA originated from someone other than, and unrelated to, the person of interest).

The likelihood ratio is calculated by taking into account the characteristics of the DNA profile and the frequency of occurrence of the individual alleles that make up the DNA profile.

If less than the 20 STR regions of DNA are observed in a DNA profile, the likelihood ratio will be smaller than the likelihood ratio calculated for a full DNA profile obtained from the same individual. In other words, the more incomplete a DNA profile is, the greater the likelihood that the obtained DNA profile could have come from someone other than, and unrelated to the person of interest.

DNA Profiles Assumed To Originate From More Than One Person (Mixed DNA Profiles)

In order to assess whether a person may or may not have contributed to a mixed DNA profile, a set of competing propositions (similar to the single source DNA profile example) are considered. For example, for a two-person mixture:

Proposition 1: The crime-scene DNA originated from the person of interest and an unknown person, unrelated to the person of interest.

Proposition 2: The crime-scene DNA originated from two unknown people, both unrelated to the person of interest.

The likelihood ratio provides a statistical assessment of the possibility that the DNA profile obtained was the result of a DNA contribution by the person of interest.

The likelihood ratio will not always favour Proposition 1 (the DNA originated from the person of interest and an unknown person unrelated to the person of interest). The likelihood ratio could favour Proposition 2 (the DNA originated from two unknown people unrelated to the person of interest).

In certain circumstances, if the ownership of an item is established or if the sample was collected from an identified person's body, then it is possible to make the reasonable assumption that the identified person has contributed DNA to the resultant mixed DNA profile. In these cases, a mixed DNA profile can be 'conditioned' on the DNA profile of the assumed contributor, such that the presence of the alleles corresponding with the assumed contributor's reference DNA profile can be factored into the statistical interpretation. This may facilitate a more meaningful statistical analysis of potential second and/or third contributors to the DNA profile. In this situation, the likelihood ratio is based on the following propositions:

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Proposition 1: the DNA has originated from the assumed contributor and the person of interest.

Proposition 2: the DNA has originated from the assumed contributor and an unknown individual, unrelated to the person of interest.

DNA profiles with an uncertain number of contributors

Forensic DNA Analysis is currently validated for the interpretation of DNA profiles with 1-4 contributors. Once the assessment of the number of contributors has been made, this information is used in the STRmix"

When a DNA profile is deemed unsuitable for interpretation it may be described as 'complex'. This can occur under the following circumstances:

- When a DNA profile could have a large number of contributors and therefore it is difficult to exclude individuals and/or determine whether a person could be a potential contributor to the DNA profile
- When there is very limited information available and/or the quality of the profile is poor.

It is important to note that should further information be received that renders the assumptions used in an analysis invalid, the DNA profile will require additional statistical interpretation.

Datasets Used in Statistical Analyses

Three validated datasets consisting of DNA profiles obtained from individuals of the Australian Caucasian, Aboriginal, and South-East Asian populations are used to calculate the likelihood ratio. A population correction factor. 0 (theta), is applied to all likelihood ratio calculations in order to correct for the common genetic ancestry of people within a particular population (sharing of DNA components inherited from a common ancestor). In Forensic DNA Analysis, a likelihood ratio that represents a stratified population statistic is reported irrespective of whether the DNA profile of interest is single source or a mixed DNA profile.

The likelihood ratio is calculated using all three datasets and the relative proportions of each ethnic grouping in the overall Australian population based on data sourced from the Australian Bureau of Statistics. This calculation, which is known as stratification, allows for the possibility that a random unknown person, unrelated to the person of interest, from anywhere in Australia could have contributed to the DNA profile obtained. Therefore, the likelihood ratio is not dependent on the ethnicity of the person of interest, but it is representative of the relative proportion of the person of interest's ethnic population within the larger Australian population.

Theta cannot account for close blood relatives. Closely related people, such as siblings, will have a greater chance of sharing similar components within their DNA profiles. However, due to the nature of parental DNA recombination during conception, the probability that two siblings would share the same 40 alleles would be very small. As this relationship becomes more distant, the probability of two relatives having the same DNA profile becomes smaller still. If it were thought that a close blood relative might have been a contributor of DNA, the most meaningful approach to interpretation would be to submit the reference sample from the relative in question for DNA analysis and subsequent direct comparison to the crime-scene DNA profile.

Often the calculated likelihood ratio produces numbers of thousands (1000s) or even millions (1000,000s) of billions. To avoid the use of potentially confusing mathematical terminology, a "ceiling figure" for the likelihood ratio of 100 billion is used (this is called "truncation"). For example, a calculated likelihood ratio of "150 000 billion times more likely", would be reported as "greater than 100 billion times more likely". The actual calculated figure can be provided upon request.

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Touch DNA / Transfer of DNA

When a person touches a surface, it is possible for their DNA to be transferred onto that surface. This transferred DNA can often be recovered (sampled) by a swab, tape lift, or excision depending upon the nature of the surface in question, and the sample can then be subjected to DNA profiling.

This transferred DNA can originate from cells or cellular material within sweat and/or oils on the skin. The generation of a DNA profile will depend on many factors. These include the amount of DNA transferred, the nature of the surface being touched, and the amount of genetic material available for transfer. The persistence of any transferred genetic material to a surface will depend largely upon the nature of the surface and the conditions the surface has been subjected to in the time between deposition and recovery of the DNA. For example, DNA could be lost from the surface by washing or mechanical action such as abrasion.

It therefore follows that the inability to obtain a DNA profile from a touched surface does not necessarily mean that a person has not had contact with the surface. It is possible for a person to contact a surface without a detectable amount of their DNA being transferred or subsequently recovered for analysis.

JUSTICES ACT 1886

I acknowledge by virtue of Section 110A (6C) (c) of the Justices Act 1886 that:

- This written statement by me dated 23 July 2021 and contained in the pages numbered 1 to 12 is true to the best of my knowledge and belief; and
- (ii) I make it knowing that, I would be liable to prosecution for stating anything that I know is false.

....

Allan Russell MCNEVIN

Signed at BRISBANE on 23 July 2021

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PROCEDURES

Please provide a list and copy of all non-standard test or calibration or inspection procedures (including in-house procedures) covered by the scope of accreditation.

Major Equipment Documents:

#10670 Procedure for Thermometer Checks

#10672 The Verification of Timing Devices

#26628 Verifications using the ARTEL MVS

#31956 Verifications using the ARTEL PCS Pipette Calibration System

#33315 Procedure for Verification and Maintenance of Equipment

Major Forensic DNA Analysis procedures:

#17091 Organisation and Management in Forensic DNA Analysis

#17117 Procedure for Case management

#17146 Internal Security and Access to Forensic DNA Analysis

#17154 Procedure for Quality Practice in Forensic DNA Analysis

#22871 Procedure for Change Management in Forensic DNA Analysis

#28801 DNA Analysis Unit Management Review template

#30800 Investigating Adverse Events in Forensic DNA Analysis

#33773 Procedure for Profile Data Analysis using the Forensic Register

#33800 Examination of Items

#34006 Procedure for the release of results using the Forensic Register

#34035 Forensic Register FTA Processing

#34229 Explanations of Exhibit Results for FR

#34245 Reference Sample Result Management

#34247 Creating and Reviewing Links - FR

#34281 Procedure for the Use and Maintenance of the Forensic DNA Analysis Elimination **Databases**

Additional minor documents can be provided on request

Please provide an example of an estimation of measurement uncertainty (MU) and a list of the procedures for which MU estimates have been made.

Documents:

#10670 Procedure for Thermometer Checks

#10672 The Verification of Timing Devices

Changes to least uncertainties of measurement:

oxtimes Not applicable. There are no changes to least unc	certainties of measurement
---	----------------------------

☐ If there are changes to least uncertainties of measurement, provide uncertainty calculations and supporting data for their derivation.

Note: Changes to calibration and measurement capability (CMC) may be considered as additions to the scope of accreditation. Please complete the Application for changes to the scope of accreditation form available from the NATA website.

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WORKPLACE HEALTH & SAFETY

Assessments are conducted by a team comprising of NATA staff and voluntary technical assessors. This team will need to attend your premises to have discussions with your staff and to observe activities covered by your scope of accreditation being performed.

To prepare for the assessment and to ensure the health and safety of the assessment team while on-site (including any field work), please respond to the following:

General issues

Issue	Yes	No
Have relevant WHS requirements been implemented, including provision of appropriate amenities for the NATA assessment team (e.g. washrooms, potable water supply)?		
Does your facility, or a site to be visited, have a company alcohol and testing policy which the NATA assessment team would be subject to? If yes, please attach a copy of the policy.		×
In response to the COVID-19 pandemic		
Does your facility comply with government guidelines pertaining to social distancing in addition to other provisions such as hand sanitation facilities and visitor register (to allow for contact tracing)?	×	
Can your facility provide COVID-19 PPE, as required?		
Will the assessment team be subject to additional measures to those of relevant health directives relating to COVID? If yes, please provide detail in the space below or on a separate sheet.		×
Additional measures:		

Specific hazards

Location	Hazard	Precaution	
e.g. Abattoir e.g. Radiography laboratory	Q Fever Radiation	Vaccination required Film badge	
All laboratory areas	Contaminations of exhibits	PPE required	
All laboratory areas	Standard chemical use	PPE required	
All laboratory areas	Biological hazard	PPE required	

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MANAGEMENT SYSTEM

☑ Please provide a copy of your facility's current management system documentation and any associated management system procedures.

Refer to document #19259 - FSS Quality Management System Guide Additional documents can be provided on request.

ISO/IEC 17025:2017 requires the facility to implement a management system in accordance with either Option A or Option B.

Option A requires clauses 8.2 to 8.9 of the Standard to be addressed.

Option B requires that a management system to be implemented in accordance with ISO 9001.

Your facility has established a management system in accordance with which option of the standard?

 □ Option A ☐ Option B

If the management system established is in accordance with Option A, it will be assessed against clauses 8.2 to 8.9 of the Standard.

If the management system established is in accordance with Option B, the records to be reviewed on-site by the NATA Lead Assessor may be reduced subject to the following:

- 1) the management system is certified by a certification body (CB) accredited by JAS-ANZ, or by another signatory to the International Accreditation Forum (IAF) Multilateral Recognition Agreement (MLA);
- 2) the CB's accreditation covers ISO/IEC 17021 Parts 1 and 3. If Part 3 is not specifically listed in the CB's scope of accreditation, then it must be clear that its accreditation covers the certification of Quality Management Systems (QMS) to ISO 9001 (which may be included in the scope of accreditation or other documentation provided by the accreditation body signatory to the IAF MLA);
- 3) copies of the most recent certification audit report(s) issued by the CB covering your facility's management system in full is (are) provided to NATA;
- 4) confirmation from the CB of the close out of any non-conformities raised during certification audits is provided to NATA;
- 5) the certification of the management system covers the laboratory activities proposed to be covered by your NATA scope of accreditation.

Evidence in support of 1) to 5) is requested to be submitted with a copy of your facility's management system documentation. The latter is required to allow the assessment team to familiarise itself with your system. The records to be reviewed on-site will be dependent on the extent of the evidence provided and the extent of the audits performed by the CB.

> Should evidence

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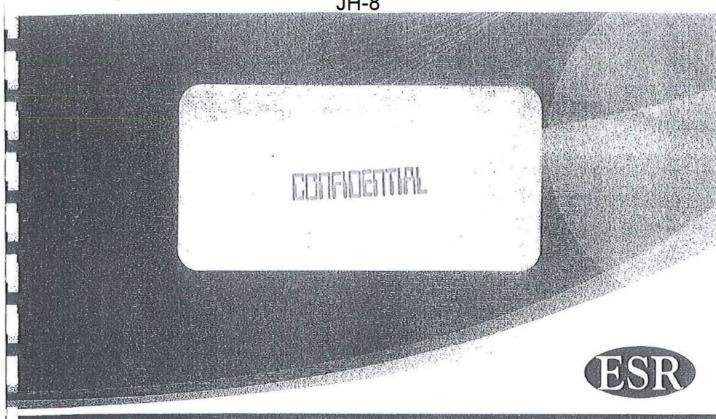
> supporting points 1) to 5) not be provided, NATA will assess your management system in accordance with Option A (i.e. clauses 8.2 to 8.9 of the Standard).

JH-7

Snip of Project Experimental Design and Reports provided to NATA in 2022

- #199 Proflex Final report
- #199 Verification of ProFlex thermalcyclers experimental design
- #220 Experimental design commercial H&E stain
- #220 Verification of commercial H & E stains Final Report for esign
- #223 Verification of DCS v4.0 on 3500xL experimental design
- #223 Verification of DCS v4.0 on 3500xL_Final report
- #231 Experimental design_STRmix v2.8
- #231 Verification of STRmix v2.8_Report_Final





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REVIEW OF THE USE OF QUANTIFILER™ HUMAN QUANTITATION SYSTEM AT QUEENSLAND HEALTH SCIENCE SERVICES

Susan Petricevic 8 April 2005 REVIEW OF THE USE OF QUANTIFILER IM HUMAN QUANTITATION SYSTEM AT QUEENSLAND HEALTH SCIENCE SERVICES Suc Petricevic, 8 April 2005



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REVIEW OF THE USE OF QUANTIFILER™ HUMAN QUANTITATION SYSTEM AT QUEENSLAND HEALTH SCIENCE SERVICES
Sue Petricevic, 8 April 2005



Preface

An independent review of Queensland Health Scientific Services (QHSS) Forensic Sciences was commissioned in March 2005. This review was a response to staff and public concern over aspects of services provided by QHSS Forensic Sciences. Although initial media allegations related particularly to DNA processes at the John Tonge Centre, the terms of reference for the review of QHSS forensic science services were wide-ranging and commissioned the review team to:

- 1. Conduct a review of organisational structure, staff levels, staff qualifications and competencies and working arrangements in forensic sciences.
- 2. Review management processes and information systems including staff and client communications and relationships.
- 3. Assess quality and validation processes of scientific analyses and equipment.
- 4. Benchmark the Forensic Sciences service in terms of turnaround time and productivity against other forensic service providers in Australia and overseas.

The outcomes expected from the review included the provision of a report to be presented to the Director General Queensland Health, outlining findings and recommendations.

The independent review is being undertaken by a team of scientists with significant forensic experience, coordinated by the Institute of Environmental Science and Research Limited (ESR), a Crown Research Institute in New Zealand. ESR is the leading provider of forensic science services in New Zealand and is accredited by the American Society of Crime Laboratory Directors – Laboratory Accreditation Board (ASCLD-LAB). The review team also includes a representative of the Australian National Institute of Forensic Science (NIFS).

This Report is the **first** of a series of reports that record the outcomes of the independent review. These reports will, in sequence:

- Examine issues relating to the validation and use of the QuantifilerTM Human DNA Quantification system at QHSS;
- Record the outcomes from a detailed technical audit of the forensic DNA work undertaken at the John Tonge Centre; and
- Provide an overview of the findings of the review team in accordance with the Terms of Reference, in relation to the operations of QHSS Forensic Sciences, with recommendations for improvements.

This Report has primarily been authored by one member of the review team but has been peer-checked and is endorsed by the team as a whole.

Wayne Chisnall Keith Bedford Susan Petricevic Veronica Borrett 8 April 2005 REVIEW OF THE USE OF QUANTIFILER HUMAN QUANTITATION SYSTEM AT QUEENSLAND HEALTH SCIENCE SERVICES

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Executive Summary

- There is no evidence that any DNA profiles obtained subsequent to Quantifiler introduction at QHSS have resulted in incorrect DNA profiling results.
- The developmental validation of the QuantifilerTM Human DNA Quantification Kit system, used in conjunction with an ABI PRISM® 7000 Sequence Detection System (7000 SDS) and prepared by Applied Biosystems complies with the validation requirements of NATA and the DAB Guidelines.
- The internal validation of the same system, completed at QHSS before implementation into casework complies with the requirements of NATA Guidelines. Documentation for the validation is held in the Biology group of QHSS and available on the Intranet.
- It is commendable that Ms Belzer and others raised the issue in OQI 11401 for review and examination. It must be emphasised that this type of questioning and re-evaluation is vital to ensure that the highest forensic analytical standards are being met. Under no circumstances should the expression of staff concern be discouraged or ignored.
- However, it is the opinion of this review that comments expressed in OQI 11401, specifically in relation to the use of Quantifiler and the term "accuracy", are scientifically incorrect. It appears that Ms Belzer was not familiar with the scientific principles and purpose of quantification nor with the use of the word "accuracy". This indicates an issue with staff training rather than with the Quantifiler technique.
- The DAB Guidelines require a Qualifying Test to be carried out by each analyst and documented prior to undertaking casework. Implementation of this would be a quality improvement and is recommended to QHSS.
- Validation documents, detailing an anomaly with the commercially manufactured DNA standards could be submitted to an internationally refereed scientific journal for publication
- On-going development and extensive additional validation of the quantitation system is continuing in QHSS. This is a normal expectation of a high-quality and professional DNA testing facility. The boundaries of the internal PCR control should be further investigated.

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- A high rework rate for casework samples can be due to several factors including in-house interpretation standards requiring full DNA profiles for each sample, difficulties in DNA quality and quantity assessment due to degradation and inhibition and the desire to obtain full profiles in a sensitive system where previously none would be obtained. Extraction methods should be reviewed to ensure that high quality initial DNA template is achieved.
- Re-engineering of the process for designating reworks should be undertaken, together with additional training and on-going method re-evaluation.

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1. Scope of this Review

As a result of staff and public concern over the use of new systems to quantitate DNA samples at the QHSS, a review of the system validation carried out has been undertaken.

Staff Concerns

The staff concerns expressed were summarised in "Opportunity for Quality Improvement" (OQI) 11401, a report made by former staff member Deanna Belzer, under the Queensland laboratory's Quality System (see Appendix I for the full report). The main concerns are summarised below:

- Staff concerns that there is no evidence of adequate validation being conducted with the introduction of the QuantifilerTM Human DNA Quantification Kit system and Real-Time PCR for use with forensic casework samples.
- 2. Staff repeated concerns regarding quantitation inaccuracy dating back to Quantifiler's implementation in April 2004 and a study was presented in a staff meeting on 5 January 2005. If 'proven' that Quantifiler was inaccurate and a critical instrument in DNA analysis, this would be in breach of NATA regulations.
- Staff concerns that method changes may have led to an increase in sample rework rates, costs and reporting inefficiency.

As part of this review, existing staff in the Biology section of QHSS have been interviewed and laboratory protocols, practices and documentation examined. An additional Technical Review has been undertaken in association with this report.

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2. Background - DNA Analysis Using PCR

DNA analysis at the Queensland Health Scientific Services laboratory in Cooper's Plains, Brisbane is generally carried out using the Polymerase Chain Reaction (PCR) in conjunction with the Applied Biosystems TM Profiler Plus Kit. This kit enables the amplification and analysis of nine sites on the DNA molecule. DNA fragments are detected using an Applied Biosystems 3100 genetic analyser. This is a state-of-the-art DNA analysis system, as used in forensic laboratories all over the world.

The PCR acts as a type of "molecular photocopier", amplifying certain sites on DNA within samples to produce sufficient for detection. This amplification process is largely responsible for the increased sensitivity achieved in modern DNA profiling applications.

Generally, the more DNA is present in a sample, the less sample template is required for amplification. Introduction of excess DNA template can result in an overamplified product that is outside interpretation guidelines.

Conversely, if insufficient template DNA is present, there may be insufficient suitable DNA present to copy. This situation may be unavoidable when there is simply insufficient DNA in a sample.

In intermediate cases, the addition of insufficient DNA template in a PCR may give a partial DNA profile. This means that DNA results are obtained from some, but not all, of the nine DNA sites examined. This situation does not mean that an incorrect DNA profile or incorrect result would be obtained, but means that only part of the profile would be obtained.

Therefore, it is advisable to attempt to estimate the amount of template DNA present in each sample prior to amplification. The aim of this estimation, or quantitation, is to achieve the highest quality DNA profile possible from a particular sample. It may not be possible to estimate the very low amount of DNA present in some samples; however it is adequate to indicate that a low level exists and the maximum should be amplified.

Further complicating factors exist at the DNA amplification stage. Although an estimate of DNA quantity is made, additional factors such as inhibition and degradation can effect production of an optimal profile. PCR inhibitors are defined as compounds present in the sample matrix that interfere with the ability of the PCR to amplify DNA. Common inhibitors such as wood and leather processing chemicals, certain metals, dyes and haem may affect the ability to assess the quantity of DNA required for an optimal amplification. Sample degradation from age and environmental exposure can reduce the presence of DNA regions suitable for amplification and effect DNA assessment.

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Samples, such as reference blood or buccal samples from individuals may not be routinely quantitated prior to amplification. This is because the amount of DNA present is less variable, and already estimated.

Therefore, although DNA quantitation may be described as a "critical" step in the analytical process, what is required is an estimate of the amount of DNA present, not an exact value of the DNA concentration.

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3. DNA Quantitation Methods in Use at QHSS

DNA quantitation, or quantification, is undergoing a period of transition. This transition is largely due to advances in DNA profiling technology and the desire to develop automated systems.

3.1 Quantiblot Human DNA Quantitation System

QHSS, like many forensic laboratories around the world, was using the QuantiBlot® Human DNA Quantitation system supplied by Applied Biosystems until June 2004 when they changed to Quantifiler. The Quantiblot system is promoted by the manufacturer as specific for human DNA. Samples and standards appear as a series of signals of differing intensities on films. The largest and darkest spots indicate the highest quantity of DNA. Sample results are read by direct visual comparison to the standards.

Quantiblot does not provide an exact determination of the amount of DNA present in a sample or standard due to the limitations of this system. What is required, and estimated, is an assessment of template DNA within ranges that will produce a suitable DNA profile in the DNA amplification system used. The Quantiblot system was relatively insensitive; therefore samples with trace quantities of DNA below the Quantiblot detection threshold often provided good quality DNA profiles. Laboratory practice at QHSS, and internationally, recommended amplification of extracts giving negative assessments to ensure that all possible profiling results were obtained.

3.2 QuantifilerTM Human DNA Quantification Kit systems

The latest technologies for routine DNA quantitation in forensic laboratories are Real-Time PCR based systems that can be readily automated. By assessing the amount of PCR product manufactured against standard starting template concentrations, it is possible to assess the amount of template DNA present in a sample. The QuantifilerTM Human DNA Quantification Kit system is an example of this state-of-the art technology, often used for example in conjunction with an ABI PRISM® 7000 Sequence Detection System (7000 SDS).

By definition, RT-PCR systems use PCR technology so have the same complications with sample degradation and inhibitors. The primers used to amplify DNA in a this RT-PCR system differ to those used in a STR amplification kit such as Profiler Plus. Therefore, it is possible that different PCR amplification success could occur in each system. This means that a positive quantitation estimation may not necessarily lead to an optimal DNA profile, and a negative quantitation may not necessarily predict any DNA profile being obtained. As in Quantiblot, a safeguard is to amplify all samples with negative quantitation values. However, a PCR based system, such as Quantifiler,

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is considered to be a better indicator of amplifiable DNA than an enzyme based system such as Quantiblot.

The Quantifiler kit uses an Internal PCR Control (IPC) as an additional indicator of PCR inhibition. This is designed to indicate whether a negative quantitation result is due to a lack of suitable DNA or whether it is due to an inhibition event. This quality improvement measure allows analysts to better predict the optimal template input for individual samples.

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4. Accreditation Requirements for Method Validation

The QHSS laboratory is accredited under NATA, the National Association of Testing Authorities, Australia. The relevant NATA clauses 5.4.5.2 and 5.4.5.3 governing method validation are detailed in Appendix II. A further ISO/IEC 17025 Application Document, 2002 Version 1 prescribes Supplementary Requirements for Accreditation in the Field of Forensic Science.

Although a corporate Validation Document exists in QIS (the Queensland Health Service Quality System, QHSS Validation of Test Methods Document, 10663R2) no specific Validation Document for the Biology team is available.

Internationally accepted Quality Assurance Standards for Forensic DNA Testing Laboratories, known as the DAB Guidelines, regulate Forensic DNA Laboratories globally. These Standards, developed by SWGDAM (the Scientific Working Group for DNA Analysis Methodologies) and issued by the FBI are prerequisites for accreditation with international programmes such as ASCLD/LAB (The American Society of Crime Laboratory Directors/ Laboratory Accreditation Board) and will have eventual amalgamation with ISO17025.

It must be stressed that compliance with the more stringent DAB Guidelines is not a prerequisite for NATA accreditation. However, due to their international acceptance, the DAB requirements (detailed in Appendix II) are also used in this review as a comparison point for the validation undertaken at QHSS.

4.1 Developmental Validation

It is useful to consider the DAB Guidelines (DAB Standard 8), where validation is classified as either developmental or internal validation:

DAB Standard 8.1.

To successfully satisfy Standard 8.1, compliance must be demonstrated with all of the subcategories of Standard 8. Validation is the process used by the scientific community to acquire the necessary information for accessing a procedure's reliability to obtain a specific, desired result. The validation process also serves to identify critical aspects of a procedure that must be controlled and monitored, while defining the limitations of the procedure.

Developmental validation must precede the introduction of a novel methodology for forensic DNA analysis. A novel methodology may include an existing technology or testing procedure that has been developed for a specific technology (e.g., medical testing, genetic analysis) that is not currently applied to forensic DNA analysis. Citations in peer-reviewed scientific journals that provide the underlying scientific basis for a novel methodology should be available.

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Developmental validation may be carried out by manufacturers of novel products, prior to their use by the forensic community (DAB and NATA ISO/IEC 17025 cl 5.4.5.1). The QuantifilerTM Human DNA Quantification Kit system is used in conjunction with an ABI PRISM® 7000 Sequence Detection System. Extensive developmental validation studies undertaken by Applied Biosystems are outlined in the QuantifilerTM Human DNA Quantification Kit Users Manual. Each DAB clause is noted and the complying validation follows,

DAB standard 8.1.2 accuracy, precision, and reproducibility of the procedure is defined, refer Users Manual sections 6-3 to 6-7,

DAB standard 8.1.2.1 documentation is available that defines and characterizes each locus, refer Users Manual section 1-2 and Profiler Plus documentation

DAB standard 8.1.2.2 species' specificity, sensitivity, stability, and mixture studies are completed, refer Users Manual sections 6-10 to 6-22. Degradation studies and comparisons to other methods are also detailed in sections 6-23 to 6-28.

NATA validation requirements refer to standard and non-standard methods, defined as follows,

NATA Clause 5.4.5.2 Method Validation,

"(the) laboratory must validate non-standard methods, laboratory designed/developed methods, standard methods used outside their intended scope and amplifications and modifications of standard methods. Records for method validation must include results obtained, procedure used, and a statement as to whether the method is fit for the intended use."

The Quantifiler system is a standard method used within its intended scope without amplifications and modifications. This standard requires documented results, procedure and suitability statement (see developmental validation above).

The manufacturer's studies fulfill the developmental validation requirements of DAB and NATA for the Quantifiler™ System and ABI Prism 7000 SDS and will not be reviewed in further detail.

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4.2 Internal Validation

In addition, internal validation studies are required by laboratories introducing the new system (DAB standard 8.1.3);

DAB Standard 8.1

To successfully satisfy Standards 8.1.2 and 8.1.3, compliance must be demonstrated with all of the subcategories of these standards. Prior to implementing a new DNA analysis procedure or an existing DNA procedure developmentally validated by another laboratory, the forensic or database laboratory must first demonstrate the reliability of the procedure internally. The internal validation studies conducted by the forensic laboratory should be sufficient to document the reliability of the technology as practiced by that laboratory. Summaries must be written for all internal validation studies and approved by the technical manager/leader.

Each new instrument or performance-based software change (including upgrades) requires a performance check. A performance check is an evaluation of a validated procedure existing in the laboratory system to ensure that it conforms to specifications and may include such studies as reproducibility and sensitivity.

FBI DNA Quality Assurance Audit Document Issue Date 07/04 (Rev. #6)

Under NATA, the ISO/IEC 17025 Application Document for Forensic Science, NATA 2002 Version 1, regulates internal validation,

ISO/IEC 17025 Clause 5.4 includes requirements that methods be generally accepted, clarifies and extends the earlier requirements by adding "procedures used must be demonstrably capable of producing valid results" (cl 5.4.2 (a)).

ISO/IEC 17025 Clause 5.4.2 (a), (b) when introducing laboratory-new methods, a "..laboratory must first demonstrate reliability of the procedure in-house against any documented performance characteristics of that procedure. As a minimum, the method must be tested using known samples. It is recommended using non-probative samples... Records of performance verification must be maintained."

ISO/IEC 17025 Clause 5.4.2 (c) "where a test can be performed by more than one method, there must be documented criteria for method selection...where appropriate the degree of correlation between the methods must be established and documented."

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ISO/IEC 17025 Clause 5.4.5.1 defines that "the validation process identifies critical aspects of a procedure which must be carefully controlled and monitored". The remainder of this clause approximates wording in DAB standard 8, discussed above, although is less prescriptive with "may" rather than "must" used to indicate testing requirements such as specificity.

Application of the accreditation requirements to the internal validation carried out at QHSS for the introduction of QuantifilerTM follows.

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5. Internal Validation of Quantifiler use at QHSS

Validation records of assessments completed prior to Quantifiler introduction, and ongoing development were reviewed by discussions with staff, review of original documents and subsequent reports. These documents were then assessed with regard to the NATA and DAB requirements detailed above and with respect to the actual timeline of introduction.

Timeline (QHPSS DNA Processing Project, 16/3/05)

- March 2004- a second capillary electrophoresis 3100 instrument was implemented for casework and initial validation and thresholds estimated.
- 2. April 2004- Quantifiler and ABI Prism Sequence Detector purchased and installed. User training provided.
- 3. April-May 2004- Quantifiler initial validation conducted.
- 4. June 2004- Quantifiler quantitation system introduced for forensic casework.
- November 2004- ABI called in to retest the 7000 Prism SDS and 3100B.
- 6. December 2004- evaluation of TE buffer quality.
- January 2005-review of Quantifiler performance and extended validation checks.
- March 2005- review of Amplification repeat rates and Contributing Factors; review of progress to date. Quantifiler Research Plan- DNA Processing Improvement Project to further investigate standards variability, pipetting and operator variability.

5.1 March 2004 - Implementation of a New 3100 Instrument

A second capillary electrophoresis instrument was implemented for casework, the 3100B. The reviewers have a draft document entitled "Validation of Casework Samples on the ABI Genetic Analyser". Although the document is not dated, we were advised it was written in approximately May 2004, detailing previous validation carried out. Staff were concerned that there was considerable management pressure to implement this system quickly; however standards were not compromised.

A brief review of the implementation is included in this review for completeness as although the focus is on Quantifiler validation, 3100 performance affects the profiles obtained.

The study ran samples amplified at 2ng on the 3100, as for the previous 377 instrument. This resulted in split peaks, pull-up and overloading. This is not an unexpected finding as 3100 instruments are thought to have higher sensitivity than 377s. Ten samples were quantified using Quantiblot and run at 1ng and the results deemed acceptable. Approximately 250 casework samples were rerun on the 3100 and

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corresponding DNA profiles obtained. It is uncertain if these were probative or nonprobative samples.

Minimum thresholds for heterozygote peaks were determined by analysis of 306 samples with baselines being recorded. The mean baseline peak height was set at +3 standard deviations. From these data, the threshold was set at 150 relative fluorescence units (rfu) for heterozygotes. Peak heights between 75 and 150 were deemed non-reportable but would be recorded. Peak heights below 75 rfu were not recorded.

Homozygote threshold levels were similarly determined as reportable above 300 rfu, recordable between 150 and 300 rfu and non-recordable below 150 rfu.

- These threshold levels are in-line with other forensic laboratories. The exact values will vary between laboratories and instruments.
- The reviewers are impressed by the expertise and experience of the 3100 validation team. It remains for QHSS to verify that a final report would have been prepared and will be available in the laboratory.

5.2 April 2004

The ABI PRISM® 7000 Sequence Detection System was installed in mid April 2004 and installation calibration process carried out. The initial calibration record was unavailable for review. Training of approximately eight analysts was carried out by Applied Biosystems approximately April 27-28.

Known human DNA samples were prepared from each of three reference samples at 1, 1/10 and 1/100 along with one mixed sample. The 10 samples were quantitated in parallel using Quantiblot and Quantifiler at one nanogram of template DNA, calculated according to the Standard DNA Series used in each quantitation system. The results are shown in Appendix IV, Table 1, Comparison of Quantifiler and Quantiblot- amplification of known samples.

Six of the samples gave a higher quantitation result with Quantifiler, one with Quantiblot and three gave no result in either system. This is likely to reflect the difference in quantitation mechanism and DNA standard series used.

The aim of quantitation is to estimate the amount of template DNA required to provide the best quality DNA profile from a given sample. Although the actual profiles were not sighted, the "full profile" results achieved indicate that the Quantifiler assessment was at least as successful as Quantiblot in providing an estimate of DNA required. These results conform to expectations. The assessment criteria include passing the QHSS rule on allelic imbalance and peak height threshold rules.

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It should be noted that in this small study, Quantifiler sensitivity appears to be higher than that of Quantiblot; however there are indications of different concentrations being returned with the different DNA standards used. This was not noted at the time.

5.3 April- May 2004

Records indicate that further testing was carried out on 56 samples (non-probative) previously quantitated using Quantiblot and either the 377 Genescanner or 3100 Genetic Analyser. These are described as "a range of substrates that would be encountered in routine casework and also a range of concentrations" and included blood, semen, hair and cell samples. The results are shown in Table 2, Comparison of Quantiblot and Quantifiler Results, repeated and non-probative samples.

It is interesting to note that in 38 samples, the Quantifiler estimate was higher (indicated in red) than the Quantiblot estimate for the same sample. No quantitation results were obtained from either system for 8 samples. However, as expected, some DNA profiling results were obtained when these were amplified. The annotation in blue indicates the best profile returned; this is slightly in favour of the Quantifiler prediction. It is also interesting to note that sample 64693 (semen) returned additional alleles with the Quantifiler estimate amplification. It is unknown if this indicates increased sensitivity using Quantifiler and the 3100 or a contamination event.

The results of sample amplifications at 0.5 and 1 ng have also been viewed. From the results of this testing, the analysts concluded that Quantifiler estimation at 1ng of DNA provided good quality DNA profiling results when compared to the historic Quantiblot system and that the correct, corresponding DNA profiles were obtained from the samples tested.

The QHSS SOP Amplification of Extracted DNA using the AmpFlSTR COfilerTM Kit 17200R4 recommends amplification of 1.0 – 2.5ng of DNA in a volume of 20μL.

The reviewers agree that the internal validation results shown above and reviewed in original format indicate that Ing of DNA, as estimated by the Quantifiler system, was suitable for amplification and production of DNA profiles comparable to those previously obtained using the Quantiblot system.

The review now considers the application of the NATA accreditation requirements tabled above, at the time of Quantifiler and ABI Prism 7000 SDS introduction into forensic casework:

ISO/IEC 17025 Application Document for Forensic Science, NATA 2002 Version 1, Clause 5.4

The reliability of the system (to estimate DNA template suitable for production of a DNA profile) was tested by examining 66 known and non-probative samples and obtaining correct DNA profiling results (cl 5.4.2 (a), (b) and records of performance verification were maintained. The system was demonstrably capable of producing valid results (cl 5.4.2 (a)). Although peak heights in the 377 to 3100 systems, are not directly

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comparable, the attainment of full profiles is considered a valid comparison. The degree of correlation between the methods (cl. 5.4.2 (c)) was established by comparison (see Tables 1 and 2, Appendix IV) and documented. QHSS proposed to change to Quantifiler as it can be automated, Quantiblot cannot.

Comment on standards ISO/IEC 17025 (cl. 5.4.2 (c)) and DAB 8.1

These refer to a written summary of the internal validation studies that must be approved by the technical manager/leader.

It appears this initial validation was retained in laboratory records; however it was not correlated as a formal document prior to introduction of the system. There is also a requirement that a "fitness for client purpose" be documented. Since the supporting validation material appears to have been available in folders within the laboratory and reviewed by the technical manager/leader (Vanessa Ientile), this requirement is met. It is recommended that a formal report be prepared, reviewed and available before a new system is introduced; however this is not considered to be a non-compliance.

Comment on ISO/IEC 17025 Clause 5.4.5.1

This requires that "the validation process identifies critical aspects of a procedure which must be carefully controlled and monitored".

DNA quantitation is an estimate. Although it may be regarded as a critical aspect of DNA analysis, the exact numerical value returned is not critical. The critical requirement is that an indication of the amount of DNA required to produce a reliable DNA profiling result be returned. This is shown in the validation carried out. Two separate quantities of DNA were amplified, 0.5 and 1.0 ng. The higher amount was assessed as more suitable from the results obtained.

It is noted that later experiments will indicate that the standard DNA control material used to make this assessment may be of uncertain concentration. However, at the time the initial validation was completed the critical aspect, DNA amount, was identified by the validators and tested under controlled and monitored conditions.

FROM EXAMINATION OF THE DOCUMENTS AND DATA PROVIDED, IT IS THE OPINION OF THIS REVIEW THAT THE QUANTIFILER SYSTEM WAS VALIDATED BY QHSS, AS REQUIRED UNDER ITS NATA ACCREDITATION, PRIOR TO ITS IMPLEMENTATION IN FORENSIC CASEWORK.

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5.3.1 Additional Comments, DAB Guidelines

As a tool for on-going process improvement, the validation is also assessed under the higher standards required by the DAB Guidelines:

DAB Standard 8: Validation

8.1 Does the laboratory use methods and procedures for forensic DNA analysis that have been validated prior to casework implementation?

DAB 8.1.3.1(b) Has the reproducibility and precision of the procedure been monitored and documented using human DNA control(s)?

The reproducibility of the system was measured by RT-PCR replicate readings and comparison of different amplifications for the same samples. However, it would have been desirable to analyse replicate samples and assess the precision. It may be possible to calculate the precision of the sample estimates retrospectively from the duplicates shown in Table 2. It should be noted that the reproducibility and precision of the 3100 genetic analyzer, where the final profile is recovered, have been monitored and documented as part of the implementation threshold evaluations.

Investigation of the effectiveness of the internal PCR control (IPC) was not conducted; however a small trial has been carried out subsequently. Although covered in the developmental validation performed by the manufacturer, it is recommended that additional experiments be conducted at QHSS to define the in-house boundaries of this IPC system.

DAB 8.1.3.3 Has the analyst or examination team successfully completed a qualifying test using the DNA analysis procedure prior to its incorporation into casework applications?

The reviewers did not see records of the qualifying tests performed by analysts so are unable to confirm if these occurred. A qualifying test is usually carried out by an analyst as part of an initial competency assessment when acquiring a new skill. Subsequent proficiency tests, conducted on a regular basis check that the skill is maintained. It is recommended that qualifying tests and regular proficiency tests be undertaken for analysts using this system. This can be achieved by incorporating the "new" test system into regular QA trials.

The reviewers were informed that the Method (219633 Real Time PCR Quantification of Human DNA using Quantifiler Human DNA Quantification Kit (Applied Biosystems), Appendix V) and Training Module (22622R0– Real Time PCR Quantification of Human DNA using Quantifiler Human DNA Quantification Kit, Appendix V) were available prior to commencement.

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On inspection, the Method amendment history records 26 May 2004 AF, LH, GM and 25 Oct 2004 TG/CI as implementation and amendment dates. This indicates the Method was in place at the time of Quantifiler introduction.



However, the amendment history of the Training Module is recorded as Revision 0 30 Nov 2004, K Kulman, First Issue. This needs clarification as it indicates that the Training Module was not written at the time of Quantifiler introduction in June 2004.

5.4 June 2004

The system was introduced for routine casework at the start of June 2004.

5.5 September -October 2004

The initial decision on introduction was to amplify Ing of template DNA assessed by Quantifiler. It appeared anecdotally that sample rework rates had increased markedly by September 2004. This has cost and quality implications. The Biology section conducted a study in September and December 2004 that indicated a rework rate of approximately 37-40%. Previous rework rate was estimated at 15%. The results of this and further studies are discussed below (March 2005 review).

5.6 November 2004

Documentation within the Biology team now indicated that the rework rate had increased, ABI were called in to recalibrate the 3100B (and 7000SDS) systems.

A DNA Processing Improvements project, investigating possible causes for the increase in reworks was commenced. A detailed project plan is available to all staff on QIS with links to reports as completed.

The aims of this project included the identification of possible causes for the increased repeat amplification rate observed for crime samples (a separate report: Review of Amplification Repeat Rates and Contributing Factors, was prepared) and a review of the Quantifiler system.

Laboratory trials indicated that Quantifiler DNA standards were stable over a minimum two week period (results not sighted) and were prepared two weekly as recommended.

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5.6.1 Review of TE Buffer for Amplification (dates between approximately October 2004-February 2005)

A study was conducted to determine if the type of TE buffer used was affecting the results obtained from Quantifiler. Several different batches and products were compared by amplification of products and analysis on the 3100 instrument. The study concluded that USP Grade TE⁻⁴ generally gave better results and a recommendation was made to use this for Profiler amplifications. It is unclear whether this implementation has yet assisted in reduction of the allelic imbalance seen in casework.

The TE has been identified as a critical reagent and a record of the date of receipt retained and the expiry date logged and adhered to. Each batch is to be tested prior to routine use. This is a definite quality improvement instigated by QHSS. DAB Guidelines require traceability of all reagents used in DNA profiling applications.

DAB 9.2.3(a) Has the laboratory identified and evaluated the reagents critical to the analysis process prior to use in casework?

Reagents must be labeled with the identity of the reagent and a tracking mechanism identifying preparation or expiration date and component sources. Records must be maintained that identify the preparer of the reagent and the quality control measures (if any) used to check the reliability of the reagent. The laboratory must identify the reagents critical to the analytical processes used and evaluate each, prior to their use on evidence and convicted offender samples.

THIS REVIEW COMMENDS THE INITIATIVE IN THE BIOLOGY TEAM AND RECOMMENDS THAT THE TEAM ASSESS AND RECORD OTHER CRITICAL REAGENTS IN THE DNA PROFILING PROCESS, AS REQUIRED UNDER DAB GUIDELINES.

5.6.2 DNA Quantitation Standards

The introduction of a positive control for use with the Quantifiler system was initiated as a quality improvement by QHSS; it had not been a requirement of Applied Biosystems. The control used was the Applied Biosystems Human DNA Control 9947A, provided with the Profiler Plus kit, at a concentration listed in the User's Manual as 0.1 ng/µL. The concentration of this standard is not guaranteed by Applied Biosystems as it is produced by another company.

After repeated testing of the positive control using Quantifiler, it became clear that the Quantifiler assessment of the DNA concentration was 0.2 $ng/\mu L$; double that of the labelling. This difference was not apparent when quantitating using the A260 absorbance method (as used by the manufacturer).

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A Promega guaranteed Human Male gDNA Standard (a mixture of genomic DNA sources) was used for comparisions to different Quantifiler standard batches. The results indicate that Quantifiler overestimates the concentration by a factor of 1.5 to 2. Repeated testing at different times and against various standards also indicated there were inconsistencies.

This information is contained in a comprehensive report: "A Report of the Investigation into DNA Quantitation Using the Quantifiler System (Applied Biosystems)", V Hlinka and I Muharam, 5 January 2005. It is hypothesised that "findings of inaccurate DNA concentrations in both the ABI AmpFISTR Human DNA Control 9947A and the ABI Quantifiler Human DNA Standard, with assayed concentrations that are half their expected values".

Several sets of Quantifiler standards from different batches were compared, as were several commercial control DNA products. This information is detailed in the report: Results of Quant Test Amp Standards 11.1.05 and A260 Comparison, Hlinka V, January 2005. The report concluded that "differences in quantitation values occur depending on the standard used for analysis".

As a result of this testing, the Biology section decided to increase the amount of Quantifiler assessed DNA template added to each Profiler Plus reaction from 1ng to 2 ng. This is because the Quantifiler estimate of 1ng would approximate 0.5ng of the Quantiblot assessed result. This was still within the optimal amount of template DNA for the Profiler Plus system of 1-2.5ng of template DNA (reference: QIS 17200R4 Amplification of Extracted DNA using the AmpFISTR COfiler Kit). As noted above, a low input of template DNA can result in partial DNA profiles.

Initial indications are that this template increase produced fewer partial DNA profiles. (Review of Quantifiler Implementation and Performance document, March 2005, original results sighted by reviewers).

5.6.3 Scope of the Problem

The problem of obtaining different DNA quantitation results from different DNA control standard curves has been observed in other forensic laboratories in Australia and France (personal communication, S Petricevic and QHSS staff).

A comparative review and quantitation trial has recently been reported by NIST, the National Institute of Standards and Technology. The study collated 287 data sets from 80 participants using 19 different DNA quantitation methodologies. These included Quantiblot and Quantifiler systems. The pertinent comments from this report follow;

Kline MC, Duewer DL, Redman JW and Butler JM, Results from the NIST 2004 DNA Quantitation Study. J Forensic Sci, May 2005;50 (available online at www.astm.org) p7:

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"The quality of the standards used to calibrate the various assays is a potential source of among-laboratory...and/or among method.. variability.." "An evaluation in our laboratory of four different commercially-available DNA standard materials, two of which were nominally identical but from different production batches and had very different storage histories, confirms this magnitude of difference among current DNA quantitation standards. On this evidence, we expect that calibration of working standards to a homogeneous, stable, and properly characterised DNA quantitation reference material will help reduce among-laboratory variability."

It seems clear that the lack of a suitable primary DNA standard for PCR quantitation is a recognised problem internationally. NIST have developing a human DNA quantitation Standard Reference Material, SRM2372 in response to this need. This affirms the validation work from QHSS.

Interestingly, this phenomenon is indirectly addressed in the Quantifiler Kits User's Manual and paraphrased below.

Quantifiler Kits User's Manual section 6-7, Reproducibility

"Six different human DNA samples were tested for reproducibility of the quantification results. Using the concentrations provided by the supplier, the DNA samples were diluted to 2.0 ng/ μ L (A), 0.5 ng/ μ L (B), and 0.1 ng/ μ L (C).

All samples and dilutions were tested in successive runs using the Quantifiler Human kit in triplicate. The mean DNA quantity and standard deviations were calculated for each sample at 95% confidence interval values (mean +/- two standard deviation units) and expressed as a percentage of the mean quantification result".

An extract from the results of the 2.0ng dilutions, Table 6-3 follows.

Reproducibility using the Quantifiler Human kit

Sample	DNA(ng/µL				Std Devn	95% Confidence (+/-; percent)
** ******	Run 1	Run 2	Run 3	Mean		
	2.580	2.830	2.900	2.770	0.168	12.15
	2.300	2,240	2,210	2.250	0.046	4.07
	1.360	1.350	1.360	1.357	0.006	0.85
	1.810	1.790	2.240	1.947	0.254	26.12
	1.920	1.800	1.770	1.830	0.079	8.67
	1.720	1,860	1.700	1.760	0.087	9.91

It can be seen that different DNA concentrations are returned, depending on the starting DNA control material used. A mean range of 2.770 to 1.357 ng/μL of DNA (an approximate factor of two) is reported for the controls tested. Therefore, this phenomenon should not be surprising to users of the Quantifiler system.

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It is important, and necessary, that validation of this RT-PCR system focuses on the assessment of the appropriate amount of template DNA to be amplified when assessed compared to the DNA controls of choice.

5.6.4 The RT-PCR Technique - Accuracy of the Results

Staff have expressed concerns that the Quantifiler method is inaccurate. It is important to remember that RT-PCR based technology such as the Quantifiler technique is based on PCR amplification. By definition, the PCR doubles the amount of DNA product produced every PCR cycle. Therefore, given that RT-PCR assesses DNA template based on threshold values (Ct) a difference in Ct values of 1 equates to a two-fold difference in initial template amount. To return to the NIST report,

Kline MC, Duewer DL, Redman JW and Butler JM, Results from the NIST 2004 DNA Quantitation Study. J Forensic Sci, May 2005;50 (available online at www.astm.org) p7:

"A factor of two uncertainty in the amount of template DNA is equivalent to a +/- a single PCR amplification cycle"

A RT-PCR assessment of DNA quantity is an estimate of the amount of DNA present, not an exact value. This is due to the biological limits of the assay. Variation in statistical confidence is also listed at between 0.85 and 26.12 percent for the Quantifiler Developmental Validation cited above.

Samples must be estimated in, as a minimum, duplicate reactions and the results averaged. A numerical value is provided for the DNA quantity present in each sample. The presence of several decimal points in this value is a reflection of the instrumental measurement of this estimate, not the accuracy of the technique. The result remains a best estimate of DNA quantity within the bounds of a PCR assessment, as was the Quantiblot method. It is designed to assist in obtaining a high quality DNA profile in a subsequent STR amplification system.

The QHSS Validation of Test Methods Document, 10663R2 defines accuracy and precision as follows,

- "3.1 Accuracy The closeness of agreement between the test result and the "true" or accepted value. Accuracy is determined by replicate analysis of a reference material of known composition, or by replicate analyses of a spiked matrix.
- 3.2 <u>Precision</u> The closeness of agreement between independent replicate test results. There are two measures of precision, repeatability and reproducibility. High precision does not necessarily imply high accuracy."

Given the inherent ranges of a RT-PCR technique, it is appropriate that estimates of DNA template indicate ranges of DNA present in samples. It is accepted that it is difficult to define the "true" value, given the variability of available standard DNA

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control material and the concerns of staff are acknowledged. However, in effect the "true" value is not required as the role of quantitation is to assess appropriate amount of template DNA to use in a validated system analysis to obtain true values, or results. These true results are the resultant accurate DNA profiles.

Turning to the NATA requirement,

NATA Clause 5.4.5.3 Range and Accuracy

"ensure the range and accuracy of the values obtainable from validated methods are relevant to the clients' needs."

This clause is interpreted by the review as requiring that the overall DNA profiling method is "fit for purpose". The validation work carried out shows correspondence between samples tested under Quantiblot and Quantifiler assessed amplifications. There is no suggestion, nor any evidence that incorrect DNA profiles have been obtained by a change in the quantitation system. The system has been found appropriate to client's needs- the production of correct DNA profiling results.

5.7 March 2005 - Review of Reports and Progress

5.7.1 Sample Rework Rates

The following information is taken from the report: "Review of the Amplification Repeat Rates and Contributing Factors, March 2005."

The initial decision on introduction was to amplify 1ng of template DNA assessed by Quantifiler. It appeared anecdotally that sample rework rates had increased markedly by September 2004. This had cost and quality implications. The Biology section conducted a study in September and December 2004 that indicated a rework rate of approximately 37-40%. Previous rework rate was also estimated at 15%. An earlier set of data (although compiled March 2005) indicates the non-purified samples rework rate for Quantifiler to be 67% (April to May 2004) with Quantiblot at 38% (May-June 2004), presumably prior to improvements and implementation.

With increase of template DNA from 1ng to 2ng, the rework rate in January to February was calculated as approximately 30%. Although this result shows an improvement, it is still high for routine sample amplification.

It is interesting that approximately 61% of the last study samples returned quant values between 0.0 and 0.5 ng. It is unknown what proportion of these were under 200ng, a value where stochastic effects require special attention.

Further evaluation of this data is required by QHSS to assess whether the input of 2ng has significantly improved the rework rate; this is not clear from the data provided.

It is noted that samples returning quants at low levels would already have been amplified at maximum (20uL) so increasing the template would have no effect. If this

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is shown to be correct, it is recommended that the procedure for deciding rework of samples be re-engineered. Factors such as gel reader experience and training and recent increases in allele calling thresholds will also have effects.

5.7.2 Additional Comments: Factors that can influence sample rework

This review has been asked to comment on the reduction of sample amplification reworks. Partial DNA profiles can be caused by many factors, additional to those discussed above. These are commented on below.

- The quality of DNA template recovered after extraction can effect amplification. It is noted that QHSS use the Chelex method with Nucleospin clean-up when required. Chelex may not provide the high quality of DNA template returned by organic, column-based or modern magnetic bead based protocols. With the move to automated DNA analysis anticipated, it is recommended that an overhaul of current extraction protocols be undertaken.
- DNA profile interpretation guidelines may need review. There is the possibility that samples are being reworked routinely, without a check on whether or not the rework is required. There may be instances where a full DNA profile obtained for one sample in a case negates the need for full profiles from all other corresponding samples in the same case. It may be that partial profiles could be acceptable if they correspond at the sites for which results are compared. However, this will vary on a case to case basis. A system for examining the profiles obtained before an automatic rework is carried out may reduce the analyses required.
- New DNA technology (3100 and Profiler Plus) appears to be returning DNA profiling results with higher sensitivity than the previous system used at QHSS. Therefore, now results may be obtained from samples where previously none could be obtained. Some of these results will be partial profiles due to the low amounts of template DNA present. These may never yield full profiles, even if concentrated before amplification. Extraction processes for trace samples should be optimized so that the maximum is amplified each time.

Samples returning negative quantitation results, or those at very low levels, should be amplified at maximum volume ($20\mu L$). Since this has been the practice at QHSS, ambiguity in quantitation standard results outlined above will have limited effect on the rework of these low level standards. Since the maximum was amplified, the best possible profile under the systems used should already have been obtained.

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5.8.3 Additional Comments - Detection Sensitivity

Concern has been expressed by staff that amplification of insufficient DNA may result is non-reporting/detection of alleles from a minor DNA contributor in a mixed DNA sample. This concern reflects the responsibility and integrity of the QHSS biology staff in striving to deliver the best results possible. Quantitation should aim to prevent this type of "profile drop out", due to below threshold signals or to insufficient template, wherever possible.

It must be accepted that it is never possible to lower interpretation thresholds to completely eliminate this effect. The presence of stochastic effects in the PCR means that safety limits at the profile analysis stage must be imposed for threshold allele detection.

In addition, a more sensitive DNA technique, such as use of the 3100 over the 377, may develop and allow the detection of lower amounts of DNA. The converse is also true, results may be obtained where none were previously available. It is agreed that an under- estimate of DNA template can result in a partial DNA profile. Validation of these methods must ensure that optimal DNA amounts are amplified to minimize loss of data.

Partial DNA profiles are a consequence of being able to amplify very small amounts of DNA; therefore the increased signal sensitivity observed with 3100 analysis is extremely important. This should be seen as a quality improvement introduced by the Biology team at the QHSS laboratory.

5.7.4 Review of Operator and pipetting variability; continued review of DNA standards

Quantitation results and amplification success can be affected by the care taken in standard preparation, pipette calibration, solution homogeneity and storage.

A further, well- presented and meticulously developed draft plan was prepared on behalf of the QHSS Biology team. Additional investigations of the variability and precision of the Quantifiler assessments are presented. These reiterate previous DNA standard studies and hypothesis reasons for variations observed. Manual and automatic pipettors are compared, as are operators and sample position homogeneity. This is an interesting and thorough survey.

Parts of this QHSS report, along with data in previous reports by the same authors (Hinka, Muharam *et al.*) could be considered as suitable for publication in an internationally reviewed scientific journal. Comments by this review on DNA standards are presented earlier in this review document and have support in the scientific community.

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- DATA CONCERNING DNA QUANTITATION, STANDARDS AND RT-PCR ACCURACY SUPPORT THE FINDINGS AT QHSS. THESE PROBLEMS ARE ALSO BEING PURSUED BY FORENSIC LABORATORIES GLOBALLY.
- THE REWORK PROBLEM HAS BEEN INVESTIGATED
 APPROPRIATELY AND RECOMMENDATIONS FOR REDUCING
 REWORKS ARE BEING INTRODUCED AT QHSS. PROCEDURES
 AND STAFF TRAINING SHOULD ALSO BE REVIEWED BEFORE
 THE REWORK RATE IS REASSESSED.
- IT IS RECOMMENDED THAT THE VALIDATION REPORTS AND REVIEWS PREPARED WITHIN THE QHSS BIOLOGY SECTION BE SUBMITTED TO AN INTERNATIONALLY RECOGNISED PEER-REVIEWED SCIENTIFIC JOURNAL FOR PUBLICATION. THIS MEASURE WILL VALIDATE THE QHSS FINDINGS WHILE RAISING THE SCIENTIFIC PROFILE OF THE TEAM.

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Summary of Review Recommendations

- Ensure that future validation studies are available to all staff prior to use of new techniques.
- Prepare a scientific paper for submission to an internationally refereed scientific journal outlining the validation work conducted for the Quantifiler system at QHSS and the problems encountered with DNA standards.
- Develop a comprehensive Validation SOP for the Biology group. This
 document must address the requirements of NATA and DAB for
 developmental and/or internal validation as appropriate
- Continue with staff training in DNA analysis and profile interpretation and continue to develop SOPs and training modules.
- Ensure that qualifying tests (in addition to regular Proficiency testing) are completed by every analyst prior to starting casework.
- Reassess the extraction protocols in use. Automated analysis will require cleaner DNA extracts and revised extraction protocols.
- Reassess guidelines for the acceptance of DNA profiling results, including specifically defining when a partial profile will suffice.
- Continue to encourage staff to use the OQI system to express concerns and suggest quality improvements. Ensure that these are reviewed, acted upon as appropriate and the outcome discussed with staff.

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Appendix I: Opportunity For Quality Improvement (OQI) 11401

OQI No.:

11401

What Stage?:

ACTION

Created By:

BELZER, Deanna

On:

12-FEB-2005

Event History:

28-FEB-2005 08:05 PM: Investigated IENTILE, Vanessa

28-FEB-2005 08:14 PM: Actioned IENTILE, Vanessa

02-MAR-2005 01:22 PM: Action Redirected NILSEN, Sharyn

Corporate

Scientific Services (QHSS)

Unit*:

Site/Location*: Coopers Plains

Centre/Group: Forensic Sciences (FS)

Department*: Description:

Forensic Biology (SS) I am concerned that we (Forensic Biology) are in breach of both NATA regulations

and section 95A of the Evidence Act describing the conditions of the DNA

evidentiary certificate.

According to this legislation we are unable to write DNA certificates given that it has been 'proven' that Quantifiler is not accurate. Section 95A part 8 states "Any equipment used in testing the thing at the laboratory is to be taken to have given accurate results in the absence of evidence to the contrary." The repeated suspicions and concerns of staff directed to supervising scientists, dating back to Quantifilers implementation in April, regarding the accuracy of this quantitation process have finally been verified by the recent study presented in the staff meeting on 5 January 2005.

Secondly, NATA requires that all equipment is validated for forensic sampling. There is no evidence that a validation was ever conducted for this equipment (or a cost-benefit assessment that the equipment would actually be suitable for our requirements). The Appendix of our Statutory Declaration states statements are prepared in accordance with the requirements of NATA. Clearly NATA guidelines in this instance have been breached when a critical instrument in the DNA analysis procedure has not been validated and further proven to be inaccurate. I question if we are breaching the Justices Act when we sign statements which declare all information in the forgoing pages are accurate to the best of the scientists knowledge.

Furthermore, the consequences of this change in process without a pre-assessment and validation have lead to the dramatic increase in reworking samples and hence an overwhelmingly high financial cost, and an inefficiency in reporting results within reasonable turn around times.

I suggest an independent auditor review DNA master, the DNA workbook, and other OQI's relating to inconsistent DNA profiles, all indicative of an ineffective system / equipment that significantly increases costs because of inadequacies in validations and scientific knowledge in managing the processes appropriately.

Source of OOI: Internal Problems (OHPSS)

What Needs

Report/Result

Fixing?:

Action By

IENTILE, Vanessa

Whom?: In Corporate

Scientific Services (QHSS)

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Unit:

In

Coopers Plains

Site/Location:

In

Forensic Sciences (FS)

Centre/Group:

In Department: Forensic Biology (SS)

Investigation:

Quantitation is used to estimate the amount of DNA in a sample. This is used to calculate an appropriate volume to obtain a DNA profile from Profiler Plus. Quantifiler was introduced to replace the Quantiblot system which was labour intensive, time consuming and did not identify degraded DNA or potential inhibitors. Quantifiler is a Real Time PCR system that allows identification of potential DNA inhibitors but is still an estimation of the amount of DNA. Quantifiler was installed in April 2004 and initial validation studies were performed. Updates on the installation, validation and implementation process were discussed at staff meetings and minuted between Feb & May 2004.

Quantifiler was chosen over other RT PCR systems because of its internal positive

control that allows identification of potential inhibitors.

After implementation it was expected that the number of reworked samples would decrease. In March, April 2004, the 3100 was implemented for crime scene samples. This instrument is more sensitive than the previously used 377 and validation was completed to establish new reporting thresholds. A review of results indicated that the repeat rate had not decreased but it was uncertain whether this was as a result of the Quantifiler, the sensitivity of the 3100, the new reporting thresholds or other

optimisation issues relating to TE buffer.

A project was started to review these procedures and this process is ongoing. A project page was created detailing the testing and results for these projects as well as the changes implemented as a result.

Part of the ongoing process reviews is to continue to monitor the repeat rates as an indication of the system function.

Date

28-FEB-2005

Investigated:

Root Cause:

Other

Action:

The ongoing process reviews and changes can be accessed via

http://qhpss.health.qld.gov.au/qhss/forensic-

sciences/FSEP/forensic_biology_projects/dna-process/default.asp

An audit of the validation and ongoing improvement processes will be scheduled to monitor these improvements.

Date Actioned

28-FEB-2005

or Due:

Introduce Controls

How Fixed?: Accepted?;

Pending

Reason for Acceptance:

The follow-up has been performed by Sharyn Nilsen - Principal Quality Advisor as Ms Belzer is no longer an employee of QHPSS. The above investigation does not directly address whether NATA requirements or the Evidence Act have been breached. It does also not address the issue of inaccuracy of the results. All have

been discussed at length and assurances have been given that none of these are an issue. However, I would appreciate more detail in the OQI record itself. SN 02-03-05

Line Manager: KELLY, Robyn

Approval Type: Pending

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Appendix II: NATA and DAB Method Validation Standards

NATA Clause 5.4.5.2 Method Validation

laboratory must validate:

- non-standard methods
- · laboratory designed/developed methods
- · standard methods used outside their intended scope
- amplifications and modifications of standard methods
- records for method validation must include results obtained, procedure used, a statement as to
 whether the method is fit for the intended use

NATA Clause 5.4.5.3 Range and Accuracy

 ensure the range and accuracy of the values obtainable from validated methods are relevant to the clients' needs.

DAB Standard 8; Validation

8.1 Does the laboratory use methods and procedures for forensic DNA analysis that have been validated prior to casework implementation?

Discussion

To successfully satisfy Standard 8.1, compliance must be demonstrated with all of the subcategories of Standard 8. Validation is the process used by the scientific community to acquire the necessary information for accessing a procedure's reliability to obtain a specific, desired result. The validation process also serves to identify critical aspects of a procedure that must be controlled and monitored, while defining the limitations of the procedure.

8.1.1 Have developmental validation studies been conducted and appropriately documented?

Discussion

Developmental validation must precede the introduction of a novel methodology for forensic DNA analysis. A novel methodology may include an existing technology or testing procedure that has been developed for a specific technology (e.g., medical testing, genetic analysis) that is not currently applied to forensic DNA analysis. Citations in peer-reviewed scientific journals that provide the underlying scientific basis for a novel methodology should be available.

- 8.1.2 Have novel forensic or database DNA methodologies used by the laboratory undergone developmental validation to ensure the accuracy, precision, and reproducibility of the procedure?
- 8.1.2.1 Is there documentation and is it available that defines and characterizes each locus?
- 8.1.2.2(FO) Have species' specificity, sensitivity, stability, and mixture studies been conducted?
- 8.1.3 Has the laboratory completed and documented internal validation studies?

Discussion

To successfully satisfy Standards 8.1.2 and 8.1.3, compliance must be demonstrated with all of the subcategories of these standards. Prior to implementing a new DNA analysis procedure or an existing DNA procedure developmentally validated by another laboratory, the forensic or database laboratory must first demonstrate the reliability of the procedure internally. The internal validation studies conducted by the forensic laboratory should be sufficient to document the reliability of the technology as practiced by that laboratory. Summaries must be written for all internal validation studies and approved by the technical manager/leader.

- 8.1.3.1(a) Has the procedure been tested using known and non-probative evidence samples?
- 8.1.3.1(b) Has the reproducibility and precision of the procedure been monitored and documented using human DNA control(s)?

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- 8.1.3.3 Has the analyst or examination team successfully completed a qualifying test using the DNA analysis procedure prior to its incorporation into casework or database applications? (CO8.1.3.2)
- 8.1.3.4 Have material modifications to analytical procedures been documented and subjected to validation testing?
- **8.1.4(FO)** If methods are not specified, does the laboratory, wherever possible, select methods that have been published by reputable technical organizations or in relevant scientific texts or journals or that have been appropriately evaluated for a specific or unique application?

Discussion

For laboratory systems that consist of more than one laboratory, each of the laboratories must complete and maintain performance-based validations (e.g., sensitivity and precision), while basic validation studies may be shared among all locations in a laboratory system. The internal validation materials must be documented, summarized, and approved by the technical manager/leader. Summaries of a system's internal validation studies must be available at all sites. Each new instrument or performance-based software change (including upgrades) requires a performance check. A performance check is an evaluation of a validated procedure existing in the laboratory system to ensure that it conforms to specifications and may include such studies as reproducibility and sensitivity.

FBI DNA Quality Assurance Audit Document Issue Date 07/04 (Rev. #6).

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Appendix III: QHSS Validation of Test Methods Document, 10663R2

QHSS - Validation of Test Methods

1 PURPOSE

This document provides guidelines for the approach to validation of test methods within QHSS.

It is intended that a consistent approach be adopted throughout the organisation towards method validation and the documentation of validation results.

Note: QIS 13025 is a suitable form for summarizing validation information for a method.

2 SCOPE

Validation is intended to demonstrate:

- that a method is technically sound and appropriate for the purpose to which it is to be applied;
- that any variation to a standard method is technically justified and supported by documented evidence;
- what limitations are associated with a method.

3 DEFINITIONS

- 3.1 Accuracy
 The closeness of agreement between the test result and the "true" or accepted value. Accuracy is determined by replicate analysis of a reference material of known composition, or by replicate analyses of a spiked matrix.

 (Note: Analysis of reference materials is preferable to using spikes, because in the latter the analyte may be in a different form or may not be bound to the matrix as closely as in a naturally contaminated substrate.)
- 3.2 <u>Precision</u>The closeness of agreement between independent replicate test results. There are two measures of precision, repeatability and reproducibility. High precision does not necessarily imply high accuracy.
- 3.3 Repeatability A measure of the maximum acceptable difference between two test results obtained at the same time by the same analyst under identical conditions on the same material. Normally, repeatability is calculated at the 95% confidence level (any two correctly obtained results will not differ from one another by more than the repeatability value in more than 1 in 20 cases).
- 3.4 Within laboratory (in-house) reproducibility A measure of the maximum acceptable difference between two test results obtained on the same material by different analysts at different times. Normally, reproducibility is calculated at the 95% confidence level. This value of reproducibility is the one generally used to estimate the limits of uncertainty of a result. (Note: by its nature, it is usually not possible to determine in-house reproducibility for microbiological tests.)

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- 3.5 Between-laboratory reproducibility A measure of the maximum acceptable difference between two test results obtained on the same material by different laboratories. Normally, between laboratory reproducibility is calculated at the 95% confidence level. It is most conveniently determined in collaborative trials.
- 3.6 <u>Linearity (linear response range)</u> The range of concentrations between which the method produces a linear calibration line.
- 3.7 Lower limit of detection The lowest concentration of analyte that can be detected, but not necessarily quantified, by the test method. This is most commonly taken to be the concentration at which there is only a 5% chance that the result obtained will be within the range normally obtained for zero concentration. There are different approaches to estimating this, but the most common is to take the concentration corresponding to 3 times the standard deviation of the result obtained from the analysis of blanks.
- 3.8 <u>Limit of reporting</u> The lowest concentration of analyte that can be determined with acceptable repeatability and accuracy by the test method. Depending on the level of certainty required (e.g. whether or not the analysis is for legal purposes), this is usually taken as 6 or 10 times the limit of detection.
- 3.9 Sensitivity The slope of the calibration function (graph).
- 3.10 <u>Selectivity</u> The ability of the test method to measure an analyte accurately in the presence of potentially interfering substances.
- 3.11 <u>Test/testing</u> In this document, refers to any process of analysis or examination intended to ascertain the composition or properties of a sample.

4 GUIDELINES

- 4.1 Each discipline within QHSS (chemistry, microbiology, pathology etc.) shall adopt detailed documented procedures for method validation. These may be either:
- 10662, QHSS Guidelines for Method Validation: Physical Sciences.
- An Australian Standard, or failing that an international standard, for method validation in the
 discipline (if one exists). This could include validation methods used by well recognized authorities
 such as ASTM.
- 4.2 The following categories of test methods must undergo some form of validation before use in the provision of services:
- new methods developed "in-house";
- existing laboratory methods that have been modified in any manner which could affect performance;
- methods without validation data adopted from other laboratories or from literature;
- standard methods modified in any manner which could affect performance.

In addition, older standard methods often do not include data such as repeatability and reproducibility. Such methods shall be treated as in-house methods for the purpose of assessing the extent of validation required.

NOTE: Standard methods published with full validation data, and methods using commercially available kits, must still be trialled in the laboratory prior to being used in service. As a minimum, this would normally take the form of analysing a standard material to assess accuracy. Some types of tests or kits may require further validation to assess laboratory performance, typically to gauge factors such as suitability of instrumentation and laboratory environment, and the skill of the operator.

- 4.3 Where applicable the following parameters should be established by the validation process:
- accuracy
- precision (repeatability and in-house reproducibility);
- limits of detection and reporting, and sensitivity;

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- linear response range;
- selectivity (interferences).

Note – Where a method determines a range of related analytes, eg chemically related pesticides, it may not be necessary to establish the full range of validation parameters for every analyte in cases where they would be expected to be similar. It is up to the professional judgement of the users of the method to decide which parameters can justifiably be extrapolated from one analyte to another.

- 4.4 Procedures employed to determine these parameters may include:
- testing of reference materials and/or comparison of results with those from an accepted method of known performance (accuracy);
- · replicate testing of a sample (repeatability);
- · testing of blanks (specificity/interferences);
- testing of spiked samples (accuracy via recovery, specificity, repeatability, and limits of detection and reporting);
- testing of standards (accuracy, repeatability, sensitivity, linearity and limits of detection and reporting);
- · comparison of results obtained by different operators (in-house reproducibility);
- testing of interlaboratory proficiency samples or reference materials from recognised sources (accuracy and between-laboratory reproducibility);
- assessment of the results for clinical validity using suitable patient groups (sensitivity and specificity).
- 4.5 Validation should take place under normal working conditions.

The number of parameters to be determined during the validation process should be decided by the scientist performing the work in conjunction with the team leader. Factors to be taken into account include the future demand foreseen for the method (a method likely to be used only occasionally will probably not justify exhaustive validation), concentration range of the analyte and the number of substantially different matrices likely to be encountered. The team leader must be able to give reasons why particular aspects of validation were omitted.

Normally, the minimum level of validation that is acceptable is determination of accuracy using a reference material of known properties, or spiking of a matrix of a similar kind to the test material. If there is any doubt as to the extent of validation required, the client shall be consulted prior to use of a method in a particular case.

All possible validation need not necessarily be done before a method is initially put to use, subject to client requirements as in 4.8 below. It can be added to as usage expands and more validation becomes justified or required.

- 4.6 The validation process must be carried out by a technically experienced and competent person. The Team Leader (or delegate) must then evaluate the performance of the assay by reviewing all results obtained during the validation procedures. This review process may involve staff with relevant technical and scientific expertise, and clients.
- 4.7 Quality control acceptance criteria for the test method should be established as part of the validation process. This will provide data for ongoing assessment of the method.
- 4.8 Results obtained by a test method must not be released to a client unless the method has been validated at least to the extent required to meet the requirements of clients or certification authorities.

5 RECORDS

5.1 Each team should develop a reliable means of recording and storing validation data. These records should provide evidence that all necessary parameters have been determined and that the performance characteristics meet expectations.

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- 5.2 If the validation process detects limitations of a particular method, these must be clearly documented. Should the method be adopted for use by the laboratory, clients must be made aware of these limitations if they could affect the validity of the work requested by them.
- 5.3 The validation data for methods in current use must be readily available for inspection when required. Validation data for superseded methods must be archived for the period required for retention of test records.

6 ASSOCIATED DOCUMENTS

10662 QHSS - Guidelines for Method Validation: Physical Sciences 13025 Electronic Method Validation Information

7 REFERENCES

- AS/ISO 9001:2000 Quality management systems requirements. 7.5.2 Validation of processes for production and service provision. 8.2.3 Monitoring and measurement of processes.
- 2 AS/ISO 17025 1999 General requirements for the competence of testing and calibration laboratories
- 3 NATA ISO 17025 Application Documents for each field of testing. 5.4 Test and calibration methods and method validation.
- 4 NATA Technical Note 17 April 1994 Requirements for the Format and Content of Test Methods and Recommended Procedures for the Validation of Chemical Test Methods; 3. Method Validation

8 AMENDMENT HISTORY

pdating of document

REVIEW OF THE USE OF QUANTIFILER IM HUMAN QUANTIFATION SYSTEM AT QUBENSLAND HEALTH SCIENCE SERVICES
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Appendix IV: <u>Internal Validation Data</u>, <u>Prior to Quantifiler</u> <u>Introduction at QHSS</u>

Table 1 Comparison of Quantifiler and Quantiblot- amplification of known samples

Sample	Quantiblot Result Ng/uL	Quantifiler Result ng/uL	Volume amped (1ng), Quantiblot	Volume amped (1ng), Quantifiler	Result from Quantiblot	Result from Quantifiler
	2	3.38	0.5	0.3	Full profile	Full profile
	0.06	0.075	15	13	Full profile	Full profile
	NVP	Undetermin ed	20	20	Partial profile	Full profile
	0.25	0.465	5	2	Full profile	Full profile
	0.03	0.006	20	20	Full profile	Full profile
	NVP	Undetermin ed	20	20	NSD	NSD
	0.5	0.896	2	1	Partial profile	Full profile
	NVP	0.007	20	20	NSD	Partial profile
	NVP	Undetermin ed	20	20	NSD	NSD
	0.25	0.549	5	2	Full profile	Full profile

Where there is a preferred result from a sample, this is indicated in blue.

Table 2 Comparison of Quantiblot and Quantifiler Results, repeated and non-probative samples

Sample	Quantiblot	Quantifiler	Quantifiler	Quantifiler		Profile Result	
	Ng/uL	Result 1	Result 2	Mean		Quantiblot	Quantifiler
	T-"-					Mixed full	mixed full
	1	1.4	1.15		1.28	profile	profile
	2	4.36	4.46		4.41	full profile	full profile
	>2	3	2.95		2.97	full profile	full profile
	1	2.52	2.19	1	2.35	full profile	full profile
	0.125					full profile	full profile
	nvp					5 alleles	full profile
	•					xs not	
	0.25	3.57	3,42		3,49	reportable	full profile
	nvp	0.302	0.267	0.285		Nsd	full profile
	пур					Nsd	full profile
	0.25	0.83	0.767	0.799		full profile	full profile
	0.25	0.668	0.636	0.652	-	full profile	full profile
	2	7,55	7.44		7.49	full profile	more alleles- mix
•	2	4.97	5.47		5.22	full profile	full profile
	0.13	undetected	undetected			full profile	overloaded
	>>2	25.05	26.37	2.5	5.71	full profile	full profile
	0.25	0.895	0.828	0.861		Mixed full profile	mixed full profile
	0.5						
	>>>2	1764.53	1666.37	1715	5.45	xs not reportable	tull profile
	Į	2.78	2.59		2.69	full profile	Xs not reportable

REVIEW OF THE USE OF QUANTIFILER IM HUMAN QUANTITATION SYSTEM AT QUEENSLAND HEALTH SCIENCE SERVICES Sue Petricevic, 8 April 2005



Table 2 ctd, Comparison of Quantiblot and Quantifiler Results, repeated and non-probative samples

nvp					Nsd	5 alleles
nvp					partial profile	partial profile
2	3.56	3.22		3.39	full profile	full profile
1	4.84	4.81		4.83	full profile	full profile
nvp	undetected	undetected			Nsd	Nsd
>>2	8.76	8.35		8.55	full profile	full profile
0.5	0.488	0.551	0.519		partial profile	partial profile
>2	18.1	17.41		17.75	full profile	full profile
2	28.84	28.44		28.64	full profile	full profile
nvp	0.155	0.177	0.166		partial profile	partial profile
0.5	0.707	0.664	0.685		partial profile	full profile
2	1.32	1.28		1.3	full profile	full profile
2	5,94	5.47		5.71	full profile	full profile
>2	9.76	10.62		10.19	full profile	full profile
1	0.328	0.311	0.32		full profile	partial profil
nvp	0.222	0.159	0.19		full profile	full profile
nvp	undetected	undetected			Nsd	Nsd
0.5	2	1.69		1.84	full profile	full profile
>>>2	231.94	235.18		233.56	full profile	full profile
						less
1	0.112	0.096	0.104		partial profile	information
nvp	undetected	undetected			Nsd	Nsd
						more
nvp	34.17	33.37		33.77	partial profile	information
2	8.63	9.01		8.82	full profile	full profile
n∨p	undetected	undetected			Nsd	Nsd
0.13	0.408	0.497	0.492		Nsd	full profile
0.5	3.2	3.06		3.13	full profile	full profile
2	12.54	12.36		12.45	full profile	full profile
0.5	undetected	undetected			Nsd	Nsd
0.5	16.91	16.57		16.74	full profile	full profile
0.03	0.737	0.768	0.753		full profile	full profile
>2	17.13	16.79		16.96	full profile	full profile
				•••••	1	Xs not
1	3.96	3.99		3.97	full profile	reportable
2	0.032	0.036	0.034		Nsd	partial profil
0.125	0.676	0.753	0.714		full profile	full profile
nvp	undetected	undetected			Nsd	Nsd
2	4.53	4.15		4.34	full profile	full profile
1	2.35	2.23		2,29	full profile	full profile
2					full profile	full profile

Where there is a preferred result for a sample, this is indicated in blue.

REVIEW OF THE USE OF QUANTIFILER $^{\rm to}$ HUMAN QUANTITATION SYSTEM AT QUEENSLAND HEALTH SCIENCE SERVICES Sue Petricevic, 8 April 2005



Appendix V: Quantifiler Training Module and Standard Operating Procedures (SOP)

Training Module - Real Time PCR Quantification of Human DNA using Quantifiler Human DNA Quantification Kit

PURPOSE

To outline the training and competency requirements for new staff in the quantification of Human DNA using Quantifiler Human DNA Quantification kit from Applied Biosystems

ASSOCIATED METHODS

21963

Real time PCR Quantification of Human DNA using Quantifiler Human DNA

Quantification kit.

21962

Quantifiler plate map

17175

Operational Practices in the DNA Dedicated Laboratories

ASSOCIATED READING

- 1 Quantifiler Kits, Quantifiler Human DNA Quantification Kit and Quantifiler Y Human Male DNA Quantification Kit User's Manual
- 2 Real-Time Compendium 7000 SDS ver. 3.0 User's Manual
- 3 Sequence Detection Systems, ABI PRISM 7900HT, 7000, 7700; GeneAmp 5700 Chemistry Guide
- 4 ABI PRISM 7000 Sequence Detection System User Guide

DEFINITIONS (Training & Experience Levels)

Inexperienced officer

staff member being trained in new method or procedure.

Introduced

staff member has read method and associated reading and

observed method being performed.

Competent officer

staff member can perform method without supervision and has

completed competency tests. Supervisor has issued authority

to perform test.

Experienced officer

staff member has performed method routinely for at least six

months and is experienced enough to train new staff.

Assessed during performance appraisals.

REVIEW OF THE USE OF QUANTIFILERTM HUMAN QUANTITATION SYSTEM AT QUEENSLAND HEALTH SCIENCE SERVICES

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TRAINING PROTOCOL

- Read method and associated reference material.
- Observe competent officers perform (at minimum) the quantitation of Human DNA at least three times
- Discuss relevant safety precautions for sample and reagent preparations including minimising contamination.
- Perform a minimum of six quantitations under supervision by an experienced officer.
- Prepare or observe preparation of associated reagents as listed in 21963.
- Prepare quantitation worksheets and perform sample id checks under supervision.

amendment history

Revision	Date	Author/s	Amendments
0	30 Nov 2004	K Kulman	First Issue

appendices

Competency Test and Authority to Perform Analysis

COMPETENCY TEST AND AUTHORITY TO PERFORM ANALYSIS

Training Checklist:

- Preparation of sample worksheets
- · Reagent preparation
- · Competently perform quantitations
- Awareness of safety and sample handling issues
- Sample storage

Training Officer:	 Date complete:	
Trainee:		

COMPETENCY TEST

- Describe the principle of Real-time PCR.
- 2 What is the IPC and its function?
- 3 Describe the steps involved in setting up the standards.
- 4 Why is accurate pipetting so important?
- 5 What volume of DNA extract is added to PCR mix in each well?
- 6 What value of 'r2' will determine if the standard curve will provide accurate results?
- Briefly outline some techniques that are used to reduce the chances of cross contamination during the quantification procedure.

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8	How do you determine if cross contamination or operator	contamination has occurred?
9	How do you determine if a sample has inhibition?	
	ining Objectives Completed: YES/NO	
Con	ments:	
	ining and competency test completedorm this method.	is now authorised to
Sup	erylsor:	Date:
Offi	cer:	Date:

REVIEW OF THE USE OF QUANTIFILER M HUMAN QUANTITATION SYSTEM AT QUEENSLAND HEALTH SCIENCE SERVICES

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SOP: Real Time PCR Quantification of Human DNA using Quantifiler Human DNA Quantification Kit (Applied Biosystems)

SCOPE

- This method is used as the method of choice for quantification of extracted DNA samples.
- This system is designed to quantify the total amount of amplifiable human DNA in a sample using a real-time PCR assay.
- 3. This quantification assay uses the ABI PRISM 7000 Sequence Detection System (7000 SDS).

REFERENCES

- Quantifiler Kits, Quantifiler Human DNA Quantification Kit and Quantifiler Y Human Male DNA Quantification Kit User's Manual
- 6 Real-Time Compendium 7000 SDS ver 3.0 User's Manual
- 7 Sequence Detection Systems, ABI PRISM 7900HT, 7000, 7700; GeneAmp 5700 Chemistry Guide
- 8 ABI PRISM 7000 Sequence Detection System User Guide

PRINCIPLE

Real-time PCR assay measures the amount of a nucleic acid (DNA) target during each amplification cycle of the PCR.

This DNA quantification assay combines a target-specific (human DNA) assay with an internal PCR control (IPC) assay.

The Internal Positive Control (IPC) system is used to distinguish between a true negative sample result and reactions affected by the presence of inhibitors, assay setup or a chemistry or instrument failure. The IPC is present within the Master Mix components and is added to each and every sample, and thus undergoes the same PCR cycles as the samples in question.

A set of prepared standards and samples are added in duplicate to a 96-well reaction plate. Each well on the reaction plate is flooded with light from a tungsten-halogen lamp, which excites the fluorescent dyes in each well of the plate. A CCD camera collects the differing wavelengths of light emitted. Data analysis algorithms are then applied to the raw data collected.

REAGENTS

- 1 For kit reagents, see Quantifiler Kits User's Manual.
- 2 For reagents made in the laboratory, see 17199.

ASSOCIATED DOCUMENTS

1 21962 Quantifiler Plate Map

STORAGE OF KIT REAGENTS

All reagents supplied with the kit are to be stored at 2 to 8°C. Glycogen is to be stored below 20°C.

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QUALITY CONTROL PROCEDURES

- 1 Crime scene samples and person/reference samples are not processed together in the same 96-well reaction plate at the same time. These samples will be run separately from each other to avoid contamination issues arising.
- 2 All samples, including standards are in duplicate on each 96-well plate. This is to ensure a more accurate result is obtained for each sample.
- 3 An Internal Positive Control (IPC) is contained within the Master Mix. The IPC system is used to distinguish between a true negative sample result and reactions affected by the presence of inhibitors, assay setup or a chemistry or instrument failure.

STORAGE OF DOCUMENTS

Sample sheets and attached methods are filed together by date in the Analytical area (Room 6116).

amendment history

- 26 May 2004 AF, LH, GM
- 2 25 Oct 2004 TG/CI

REVIEW OF THE USE OF QUANTIFILERTM HUMAN QUANTITATION SYSTEM AT QUEENSLAND HEALTH SCIENCE SERVICES

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Casework/Reference (please circle)	
Run ID:	Date:
Worksheet Attached:	Operator:
Quant Date Entered into Workbook:	
DNA Standards Lot #:	
DNA Standards prepared by & date:	
DNA Master Updated:	Worksheets Created:
Quantifiler Log Updated for Amp Date etc. : []	Workbook Updated with [], amp date etc.:
Preparation of Standard Dilutions	
	from a serial dilution of Quantifiler Human DNA Standards are stable for up to 2 weeks at 2-8 °C. It is

Steps 1-7 are to be carried out in Room 6122 (Quant Room)

NOTE 2: The recipe for T₁₀E_{0,1} buffer, refer to S.O.P. #17199.

- 1. Label eight 0.5mL sterile tubes 1 through 8.
- 2. Vortex Quantifiler Human DNA Standard (located in freezer in Quant room) to mix thoroughly.
- 3. Prepare TE-Glycogen buffer in a 5ml tube using:
 - 5ml T₁₀E_{0.1} Buffer
 - 5μl Glycogen (20mg/ml) located in freezer in PCR Prep Room

NOTE: T₁₀E_{0.1} Buffer with glycogen to be made fresh each time.

- Pipette 300μL of TE-Glycogen buffer (10mM Tris-HCl (pH 8.0), 0.1mM Na₂EDTA, 20μg/mL glycogen) into the first sterile tube labeled "1".
- 5. Aliquot 200μL of TE-Glycogen buffer into the remaining seven tubes.
- Add 100μL of Quantifiler Human DNA Standard to the tube labelled 1. Vortex thoroughly and transfer 100μL from tube 1 into tube 2. Vortex to mix thoroughly.
- Continue serial dilution through to tube 8 using 100µL aliquots each time.

Quantifiler Human DNA Std Concentrations				
DNA Standard	ng/μL			
1	50.00			
2	16.70			
3	5,560			
4	1.850			
5	0.620			
6	0.210			
7	0.068			
8	0.023			

REVIEW OF THE USE OF QUANTIFILER M HUMAN QUANTITATION SYSTEM AT QUEENSLAND HEALTH SCIENCE SERVICES

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Creation of new Plate Document

Steps 1-12 are to be carried out in Room 6123 (PCR Room)

- 1, Turn on the laptop computer.
- Turn on the ABI PRISM 7000 Sequence Detecting System (7000 SDS) instrument.
- 3. Launch 7000 SDS System software (icon appears as an R with multi colours).
- 4. From the MAIN MENU select FILE -> NEW or click the PLATE DOCUMENT Icon.
- Make sure that the assay is ABSOLUTE QUANTIFICATION and the container is 96-WELL CLEAR.
- Select the template to be used for the new document from the TEMPLATE LIST use the TEMPLATE.sdt template from the drop-down menu.
- Click OK.
- Open the Weil Inspector: VIEW -> WELL INSPECTOR
- Select the first pair of wells. Select the SAMPLE NAME box in the WELL INSPECTOR and type in the DNA number, then press ENTER. Continue this process to name and locate the remainder of the samples.
- 10. When you are finished setting up the plate document, close the WELL INSPECTOR.
- From the MAIN MENU window, select FILE -> SAVE AS to open the SAVE AS dialogue box.
- Name the file with the Run ID and Date (e.g. QF#666 30-02-04 and CW or Ref). Click SAVE.
- 13. Find someone to sequence check your sample sheet.

Preparation of samples

Steps 1-2 are to be carried out in Room 6122 (Quant Room)

- Thaw extracted DNA samples, vortex briefly and pulse spin.
- 2. Set out samples in rack according to the worksheet.

Steps 3-7 are to be carried out in Room 6120 (Reagent Prep Room)

 Prepare PCR Mix using ART tips. Calculate appropriate volumes required and record Lot Numbers.

Master Mix (prepared in sterile 5mL tube)							
11.5µL of Quantifiler Human Primer Mix							
14.0µL of Quantifiler PCR Reaction Mix							

REVIEW OF THE USE OF QUANTIFILER M HUMAN QUANTITATION SYSTEM AT QUEENSLAND HEALTH SCIENCE SERVICES

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- Gently vortex PCR mix, 3-5 seconds.
- Place new 96-well reaction plate in a black splash free support base.
- Label 96-well reaction plate with the date and Run ID.
- Dispense 23µL of PCR mix into each reaction well.

Steps 8-13 are to be carried out in Room 6122 (Quant Room)

- Vortex the eight DNA standards.
- Using aerosol resistant tips add 2μL of each of the standards to the appropriate well according to the sample sheet and mix thoroughly. (Stds used: 50, 16.7, 5.56, 1.85, 0.62, 0.21, 0.068, 0.023ng/μL)
- Using acrosol resistant tips add 2μL of extracted DNA samples to the appropriate wells in duplicate and mix thoroughly.
- 11. Seal 96-well reaction plate with the optical adhesive cover.
- Centrifuge the plate at 3000rpm for approx. 20 seconds in a table top centrifuge with plate holders to remove any bubbles.
- Transfer reaction plate to PCR room (Room 6123)

Steps 14-16 are to be carried out in Room 6123 (PCR Room)

14. Place compression pad over the optical adhesive cover with the grey side down and the brown side up and the holes positioned directly over the reaction wells. Put plate in machine with barcode facing out.

NOTE: Before starting the run, ensure that all other windows are closed except the PLATE DOCUMENT currently in use.

- 15. In the main window select the INSTRUMENT tab.
- Click START. Check on the INSTRUMENT tab that the machine actually starts the run.

Results Analysis and Workbook Entry

- After the run has completed (Ihr:45min) take the Quantifiler Disk into the PCR Room (Room 6123) and insert into Quantifiler computer.
- In the main Quantifiler window click on the green "Analyze" arrow.
- 3. Click on the "Results" tab then click on the "Standard Curve" tab. Ensure that the r² value is greater than 98% (0.98). If it is less than 98% determine which standards have given inaccurate results. Return to the plate map and using the well inspector click "Omit Well" for any standards which are inaccurate. After omitting wells, once again click "Analyze". [N.B. on the standard curve the standard at top left is Std#8.]
- 4. When the standard curve is acceptable choose File->Export->Results and export the results to the Quantifiler disk - select A: drive and save the results table using a similar name to the quantifiler (e.g. CW#66 23-08-04).

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Steps 5-11 are to be carried out at the DNA Master computer

- Insert the Q'filer disk into the DNA Master computer and run the (green star) macro to send Q'filer results to DNA Master. [Macros located in G;\DNAMaster\dnaUPLOAD,xls]
- 6. After checking that there is A4 paper in the Epson printer, run the (pink cylinder) macro to print the amp sheet and create the 3100 sample sheets. Check the log and assign the next 3100 plate name, and record this in the log. This is the name of the samplesheet (e.g. CW#66).
- Assign an amp date in the log and update the DNA workbook with the quant results, amp date and GS number for each sample.
- If any samples were inhibited a sheet will print up a list of any samples which were fully or partially inhibited.
- For any partially inhibited samples enter that sample ID in AUSLAB and record a SPECIMEN NOTES communication. Remember to press F4 to save each note.
- 10. For any completely inhibited samples (which do not appear on the amp sheet) resubmit for Nucleospin cleanup and once again notify CS through specimen notes. Samples should be resubmitted as a red number and the results field in DNA master for the original sample should be updated to read "QF Inhibition – See red # near..."
- 11. At the end of the macro the sample sheet will automatically be saved in the "G:\forbio\plate_import_template_files" folder.

JH-9

From: Cathie Allen

Allan McNevin; Justin Howes; Kirsten Scott; Kylie Rika; Matthew Hunt; Paula Brisotto; Sharon Johnstone; To:

Megan Mathieson; Saan Orion Luke Ryan; Wendy Harmer

FW: Proposal #184 Subject:

Monday, 31 July 2017 11:00:43 AM Date:

Attachments: Project Proposal Evaluation of the efficacy of Microcons July2017.doc

Project Plan Evaluation of the efficacy of Microcons July2017.doc image003.jpg

Hi Everyone

Cc:

Please find attached a Project Proposal, Project Plan and Budget for Project #184 - Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

These documents are held in I:\Change Management\Proposal#184 - Evaluation of the efficacy of Microcons

ACTION: Please consider the documents, undertake a risk assessment for your team and add this to the Project Plan and provide feedback to Justin Howes by Thursday, 17th of August 2017.

Cheers Cathie



HSQ's vision | Delivering the best health support services and solutions for a safer and healthier Queensland.

Queensland Health acknowledges the Traditional Owners of the land, and pays respect to Elders past, present and future.



Project Proposal #184

Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon[®] Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

July 2017
Justin Howes and Cathie Allen



Project Proposal #184 Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon® Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

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For more information contact:

Forensic DNA Analysis, Forensic and Scientific Services, Department of Health, GPO Box 48, Brisbane QLD 4001.

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Project Proposal #184 – Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon® Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

Document Details

Contact for enquiries and proposed changes

If you have any questions regarding this document or if you have a suggestion for improvements, please contact:

Contact officer: Justin Howes

Title: <u>Team Leader – Forensic Reporting and Intelligence Team</u>

Phone: Email:

Version history

Version	Date	Changed by	Description
1.0	25/07/2017	Justin Howes	Document Created.

Document sign off

This document has been approved by:

Name	Position	Signature	Date
Cathie Allen	Managing Scientist		

The following officers have endorsed this document

Name	Position	Signature	Date
Justin Howes	Team Leader FRIT		

Name	Position	Signature	Date
Paula Brisotto	Team Leader ER & Q		

Name	Position	Signature	Date
Megan Mathieson	A/Senior Scientist Analytical		

Name	Position	Signature	Date
Allan McNevin	Senior Scientist ER		

Name	Position	Signature	Date
Kirsten Scott	Senior Scientist Q & P		

Name	Position	Signature	Date
Sharon Johnstone	Senior Scientist Intel		

Name	Position	Signature	Date
Matthew Hunt	A/Senior Scientist Reporting 1		

Name	Position	Signature	Date
Kylie Rika	Senior Scientist Reporting 2		

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1. Purpose and Scope

1.1. Background

Microcon[®] Centrifugal Filter Devices are centrifugal filter devices that desalt and concentrate macromolecular solutions such as DNA-containing solutions. They employ Amicon's low binding, anisotropic, hydrophilic regenerated cellulose membrane [1].

The use of Microcon® filters to concentrate extract has been a standard post-extraction process within Forensic DNA Analysis to reduce the volume of extract from approximately 100uL to \leq 20µL for AmpF ℓ STR® Profiler Plus® and \leq 35µL for PowerPlex® 21 (PP21) -requested samples.

Since the implementation of PP21 amplification kit within Forensic DNA Analysis for casework samples in December 2012, extracts with low Quantification values were recommended to be concentrated. Templates of <0.132ng were found to exhibit marked stochastic effects after amplification ^[2]. Consequently, a workflow that directed extracts automatically to a concentration step based on Quantification value was implemented ('auto-microcon' process).

Anecdotally, the suitability to provide the Queensland Police Service (QPS) with DNA profile intelligence from extracts that have been concentrated has been noted to be limited. Furthermore, extracts that are of low quant value that have been automatically concentrated have been observed to rarely yield DNA information for QPS.

Project #163 – Assessment of results obtained from 'automatic-microcon' samples [3] was conducted to evaluate the results of samples that were processed with the 'auto-microcon' process. A recommendation of this project was to re-evaluate after the introduction of the Forensic Register in conjunction with the use of Quantifiler® Trio DNA Quantification Kit.

1.2. Purpose

The purpose of this project is to evaluate the suitability for interpretation of DNA profiles that may be obtained after the post-extraction concentration step using the Microcon® centrifugal filter devices. This evaluation will include an assessment of those samples that underwent the 'auto-microcon' process.

1.3. Scope

This evaluation will be based on a data mine of extracts in the year 2016 that were concentrated with Microcon® centrifugal filter devices, and will assess the 'suitability' of PP21 profile outcomes as a function of quant values obtained from using the Quantifiler® Trio DNA Quantification Kit.

This evaluation will look at two data sets as a function of the quantification value:

- PP21 DNA profile outcomes from extracts that were processed through the 'auto-microcon' process;
- 2. PP21 DNA profile outcomes from all extracts that were concentrated with the Microcon[®] filter devices.

NB. The recommendation from Project #163 was to reassess after Forensic Register (FR) implementation. This recommendation was based on the perceived ease of retrieving data from the FR as opposed to AUSLAB, and with the thought that the FR would soon be implemented. For the purposes of this project, it is not considered essential to have the FR implemented if the data can be retrieved from AUSLAB. However, it is considered important that the data be spanning a sufficient period of processing, and be based on the same Quantification system namely the Quantifiler® Trio DNA Quantification Kit.

1.4. Definitions

Auto-microcon: Samples with extracts quantified in the range 0.001ng/ μ L to 0.0088ng/ μ L that were automatically processed for a concentrat on step using Microcon® centrifugal filter devices.

NCIDD: National Criminal Investigation DNA Database

2. Governance

Project Personnel

 Project Manager: Justin Howes – Team Leader, Forensic Reporting and Intelligence Team.

Decision Making Group

 The Management Team (including the Project Manager), are the decision making group for this project and may use the defined acceptance criteria in this project to cease part or all of the experimentation at any stage. The Decision Making Group may also make modifications to this Experimental Design as required, however this must be documented and retained with the original approved Experimental Design.

Reporting

The Project Manager will provide a fortnightly project status update to the Decision Making Group at the Management Team meetings and by exception as required.

3. Resources

The following resources are required for this validation/project:

Forensic DNA Analysis staff and computer time to retrieve data from AUSLAB and to use Microsoft Excel.

4. Methods

4.1. Data retrieval from AUSLAB (LIMS)

The data date range is to encompass all samples quantified with Quantifiler® Trio DNA Quantification Kit in 2016, with the following criteria:

- Quantification value above the Limit of Detection (Quantification) for the Quantifiler[®] Trio DNA Quantification Kit (ie. 0.001ng/µL);
- Extracts to be from samples with DNA Priority 2 (High Priority PP21 amplification kit);
- Extracts to have undergone a concentration step using Microcon[®] centrifugal filter devices;
- Exhibit report outcome (interpretation).

Data will be exported to Microsoft Excel for interrogation.

4.2. Data interrogation

The data will be interrogated by assessing the DNA profile outcome results reported as Exhibit Report lines (from AUSLAB) as a function of the quantification value.

The data will exclude samples that have not returned a DNA profile result, are Quality samples (including environmental monitoring samples), have no quant value in the data export, or have quality issues noted.

The DNA profile outcome will be assessed as either 'fail' or 'success' with the following definitions:

- 'Fail': DNA profile interpretation outcomes of 'Complex unsuitable for interpretation', 'No DNA profile', 'Partial unsuitable for interpretation';
- 2. 'Success': All other DNA profile outcomes.

5. Experimental Design

5.1. Experiment 1: Assessment of 'auto-microcon' results

Intent

Evaluate the 'success' or 'fail' outcomes for PP21 samples that were processed in 2016 through the 'auto-microcon' workflow.

Data Analysis

The samples applicable to this experiment will have quantification values in the range 0.001ng/ μ L to 0.008ng/ μ L.

DNA profile interpretation outcomes will be grouped into either 'success' or 'fail' as a function of the quantification value.

A percentage of samples that fall into these categories will be determined.

Of the DNA profile interpretation outcomes of 'success', the type of outcome will be broken down further to determine:

The percentage of these samples that were reworked; and,

The percentage of samples that led to an upload of DNA information to NCIDD.

Assessment Criteria

The Decision Making Group will determine if the percentages obtained are significant enough to inform a new workflow strategy in consultation with QPS.

Factors to consider include, but not limited to:

- Effect on turnaround time for samples in this range considering the success/fail results;
- Effect on intelligence provision that is provided to QPS from DNA profiles uploaded to NCIDD for samples in this range;
- Cost of processing samples (including reworks where appropriate) including staff and consumables considering the success/fail results;
- Opportunity cost of not being able to process other samples that could lead to meaningful information for QPS, including processing with further techniques (eg. Y-STR profiling).

5.2. Experiment 2: Assessment of all DNA profile results from extracts that have had a concentration step.

Intent

Evaluate the 'success' or 'fail' outcomes for PP21 samples that were processed in 2016 and underwent a post-extraction concentration step using Microcon® centrifugal filter devices.

Data Analysis

The samples applicable to this experiment will have quantification values in the above $0.001 ng/\mu L$.

DNA profile interpretation outcomes will be grouped into either 'success' or 'fail' as a function of the quantification value.

A percentage of samples that fall into these categories will be determined.

Of the DNA profile interpretation outcomes of 'success', the type of outcome will be broken down further to determine:

- 1. The percentage of these samples that were reworked; and,
- The percentage of samples that led to an upload of DNA information to NCIDD

Assessment Criteria

The Decision Making Group will determine if the percentages obtained are significant enough to inform a new workflow strategy in consultation with QPS.

Factors to consider include, but not limited to:

- Effect on turnaround time for samples in this range considering the success/fail results;
- Effect on intelligence provision that is provided to QPS from DNA profiles uploaded to NCIDD for samples in this range;
- Cost of processing samples (including reworks where appropriate) including staff and consumables considering the success/fail results;
- Opportunity cost of not being able to process other samples that could lead to meaningful information for QPS, including processing with further techniques (eg. Y-STR profiling).

6. Results and Data Compilation

The assessment criteria for each experiment will be used to make an overall assessment as to whether there is sufficient information to inform a new workflow strategy for low quant samples.

The decision points will be based on two data groups:

- 1. Samples in the 'auto-microcon' range;
- Samples in an extended quant range (and what that extended range may be).

The Decision Making Group is responsible for assessing the need for further work to assist in making a decision, and will inform the Project Manager.

A final report will be produced which will compile all analyses, conclusion and recommendations.

7. References

- QIS 19544v11 Concentration of DNA Extracts Using Microcon Centrifugal Filter Devices
- [2] PowerPlex® 21– Amplification of Extracted DNA Validation. Megan Mathieson, Thomas Nurthen, Cathie Allen. December 2012. Forensic DNA Analysis.
- [3] Project #163 Assessment of results obtained from 'automatic-microcon' samples. Josie Entwistle, Allison Lloyd, Kylie Rika, Thomas Nurthen, Cathie Allen. August 2015. Forensic DNA Analysis.

Fundin	g Source (if applicable)		
Funding Source	Amount Requested	Appro	ved
FSS Internal Fund		NO	•
Partner Contribution		NO	•
External Fund		NO	•
Total		0	

		For duration of project -
Expenditure Type	Monthly	Months:1
<u>Labour</u>		
DNA Analysis Personnel (Salaries and on-costs - for this email Business Management Information (BMI) Team for a salary forecast).	5085	
Other:		
<u>Subtotal Labour:</u>	5085	0
Non- Labour		
Equipment	0	
Consumables	0	
Travel	0	
Publication Costs	0	
Other:	0	
Subtotal Non-labour:	0	0
Total:	5085	0

Justification of Funding (Non-Labour Costs)

Justify in terms of	need and co	st, each budget item i	n previous table.	Please attach	quotes where necessar	y
---------------------	-------------	------------------------	-------------------	---------------	-----------------------	---

Total of 2 weeks work time for the Project Manager (HP6), including data interrogation and administration.

Justification of Funding (Labour Costs)

If project includes an appointment to assist or backfill, please provide details in the table below and submit to the BMI teamfor a detailed quote.

Status	✓ Permanent
	Part-Time
	Temporary (<12mths)
	Casual
Classification	Administrative
	Operational Officer
	✓ Health Practitioner
Time Period	2 weeks
Position Title	Team Leader
Qualification Required	Nil
Budget Required - Salary:	5085
On-Costs:	
Total:	5085
Cost Centre	787164

Any other information that you think maybe relevant please explain below:	

Authorised by C.Allen QIS31052V1



HealthSupport Queensland Forensic and Scientific Services

Project Plan

Stage 2			
and a		Project #:	184
Name/s of Project	Justin Howes	Start Date:	25/07/2017
Staff:	Justill Howes	Due Date:	09/08/2017
Name Project Team Leader :	Justin Howes	Contact Phone Number:	
Technical Reviewer/s	Rhys Parry		
Project Title:	An Evaluation of the Efficacy of Using the Microcon® Centrifug Intelligence.		Lack To the Live and the second secon
Project type	Administration IT/LIMS Data mining/analysis External		
Project Background (r	nay include a literature review):		
Forensic DNA Analysis AmpF&STR® Profiler Pl Since the implementa in December 2012, ex Templates of <0.132n Consequently, a work Quantification value w Anecdotally, the suita from extracts that have	filters to concentrate extract has be to reduce the volume of extract finds and ≤35½L for PowerPlex® 21 (tion of PP21 amplification kit withing tracts with low Quantification value gowere found to exhibit marked staffow that directed extracts automatically to provide the Queensland Provide the Q	rom approximately PP21) -requested so in Forensic DNA Arries were recomme ochastic effects afterically to a concentrocess). Police Service (QPS) ted to be limited.	y 100uL to ≤20⊡L for samples. nalysis for casework samples nded to be concentrated. Her amplification. Itration step based on) with DNA profile intelligence Furthermore, extracts that are
evaluate the results of recommendation of the conjunction with the u The purpose of this pr	ment of results obtained from 'auto f samples that were processed with his project was to re-evaluate after use of Quantifiler® Trio DNA Quanti oject is to evaluate the suitability	h the 'auto-microc · the introduction of ification Kit. for interpretation of	on' process. A of the Forensic Register in of DNA profiles that may be
A TOTAL PROPERTY OF THE PROPER	st-extraction concentration step us an assessment of those samples t	Not the second of the second o	and the state of t

This evaluation will be based on a data mine of extracts in the year 2016 that were concentrated with Microcon° centrifugal filter devices, and will assess the 'suitability' of PP21 profile outcomes as a function of quant values obtained from using the Quantifiler° Trio DNA Quantification Kit.

This evaluation will look at two data sets (from 2016) as a function of the quantification value:

- PP21 DNA profile outcomes from extracts that were processed through the 'auto-microcon' process;
- PP21 DNA profile outcomes from all extracts that were concentrated with the Microcon® filter devices.

Potentially, a new workflow could be designed based on the success/fail rates observed in the data. This could create time and cost savings for the laboratory, and increase the ability to process other higher DNA-yielding samples more quickly.

Proposed Methodology:

The evaluation will look at two data groups:

- Evaluate the 'success' or 'fail' outcomes for PP21 samples that were processed in 2016 through the 'auto-microcon' workflow. The samples applicable to this experiment will have quantification values in the range 0.001 ng/uL to 0.0088 ng/uL.
- Evaluate the 'success' or 'fail' outcomes for PP21 samples that were processed in 2016 and underwent a post-extraction concentration step using Microcon[®] centrifugal filter devices. The samples applicable to this experiment will have quantification values in the above 0.001ng/uL.

DNA profile interpretation outcomes will be grouped into either 'success' or 'fail' as a function of the quantification value.

- A percentage of samples that fall into these categories will be determined.
- Of the DNA profile interpretation outcomes of 'success', the type of outcome will be broken down further to determine:
 - 1. The percentage of these samples that were reworked; and,
 - 2. The percentage of samples that led to an upload of DNA information to NCIDD.

Expected Outcome:

It is expected that the data, especially the data generated for 'auto-microcon' samples will match the anecdotal information from case managers which has been gathered from years of experience. It is expected that the vast majority of DNA profile outcomes would be in the 'fail' category ie. mostly reported as 'complex unsuitable for interpretation'.

It is expected that there will be some 'success' and that this would include DNA profiles that would have been loaded to NCIDD and possibly obtained linking information for the QPS.

It is an expectation that any recommendations are communicated with QPS in order to agree on possible new workflow strategies. This could include not automatically processing low quant samples

77.7	t to hold and communicate 'low DNA' d on case assessment by QPS.	\ quant' to QPS. San	ples could	l be pro	ocessed
It is an expectation	that Critical Priority (P1) samples be	processed with the	'auto-mici	rocon' p	process.
	ct Milestones: (Ensure that the Char ment\Change Management Milesto		ilestone R	egister	is filled out
Description of Outpu	its/Milestones:		Expected date:		Completed date:
1.Data generation	and compilation		02/08/2	017	
2. Report writing a	nd submission to Mgt Team		04/09/2	017	
3. Workflow strate	gy communication and decisions		03/10/2	017	
4.Implementation	of any agreed decisions		06/11/2	017	
5.					
If expected due da	te/s not met - explanation of reaso	n required:			
Project Budget:		Total Project Budg	et		
Prepare using QIS 3105	2 (and attach to Project Plan)	\$5085			
Gantt Chart (for la	rge projects): If required, refer to Quality	team for help preparing	(and attach t	to Project	t Plan)
RISK ASSESSMENT:			. A. Jan		
If a risk is identified identification and i	d: Refer to QIS document <u>29100</u> and management.	29106 for further i	ntormatio	n on ris	sk
Team:	Details of Risk/s Identified			Туре	of Risk/s:
				Bus OH	siness Risk &S
Evidence				Signatu Line Ma	12000
Recovery:					
				Bus OH	siness Risk &S
Analytical :				Signatu Line Ma	



		Business Risk OH&S
Intel:		Signature Line Manager
		Business Risk OH&S
Reporting 1:		Signature Line Manager
		Business Risk OH&S
Reporting 2 :		Signature Line Manager
		Business Risk OH&S
Quality and Projects		Signature Line Manager
(includes OO):		
		Business Risk OH&S
Admin :		Signature Line Manager
		Business Risk OH&S
Team Leader ER &Quality :		Signature Team Leader
Team Leader	Potential risks of samples not going to NCIDD – expected to be a low percentage of samples. Samples could always	Business Risk OH&S
FRIT:	be microconned if the case circumstances warrant eg. P1	Signature Team Leader

	case. Collaboration with QPS and to occur.	d communication o	f risks	JAH
Project Proposal ap	pproved by:			
Signature Team Leader and Quality:	ER	Date:		
Signature Team Leader FRIT:		Date:		
Signature Managing Scientist:		Date:		
	·			

Comments:

Please send to Quality Team after completion

JH-10

Erin Shearer

From: Justin Howes

Sent: Wednesday, 30 August 2017 10:50 AM

To: Kylie Rika

Subject: RE: Microcon project

Thanks JAH



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Forensic & Scientific Services, Health Support Queensland, **Department of Health**

HSQ's vision | Delivering the best health support services and solutions for a safer and healthier Queensland.

Queensland Health acknowledges the Traditional Owners of the land, and pays respect to Elders past, present and future.

From: Kylie Rika

Sent: Wednesday, 30 August 2017 10:18 AM

To: Justin Howes

Subject: RE: Microcon project

No feedback

thanks



Kylie Rika Dip Mgt BSc PGrad Dip (Forensic)

Senior Reporting Scientist - Forensic Reporting and Intelligence Team

Forensic DNA Analysis | Forensic & Scientific Services, Health Support Queensland, Department of Health

HSQ's vision | Delivering the best health support services and solutions for a safer and healthier Queensland.

Queensland Health acknowledges the Traditional Owners of the land, and pays respect to Elders past, present and future.

From: Justin Howes

Sent: Wednesday, 30 August 2017 9:25 AM

To: Kylie Rika

Subject: Microcon project

Hey Kylie,

I am only waiting on your feedback for the proposal #184.

Please fill in risks to the Project Plan in I:\Change Management\Proposal#184 - Evaluation of the efficacy of Microcons

I want to print the Proposal for Mgt Team in next day or so.

Thanks JAH



Justin Howes

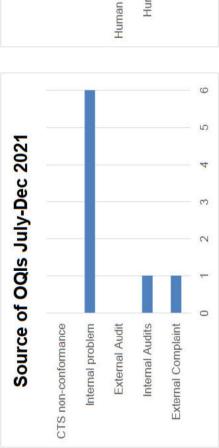
Team Leader - Forensic Reporting and Intelligence Team

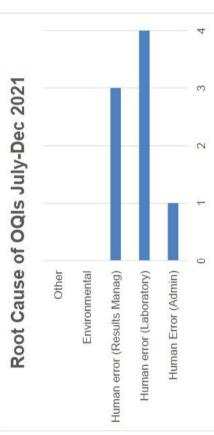
Forensic DNA Analysis, Forensic & Scientific Services, Health Support Queensland, **Department of Health**

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	lan- lin 2019	Int-Day 2019	Jan- Jun 2020	Int-Dar 2020	lan- lim 2021		Tarriet
		00000000	0411 541 E0 E0	04 DOC 5050	Jan 1941 2021	04 DCC 202 I	laiget
OQIs raised	8	7	8	10	13	8	000
OQls open >1 year	0	0	0	0	0	0	0
OQIs completed <90days	75%	100%	100%	%08	84%	%29	>15%
Average days OQIs open	59.6	36	24	47	64	86	<60





Queensland 1591th

OQI's

OQ! Number	Title	Creator	Date Identified Days Open	Days Open	Root Cause Type	Actioner	Status
	Reporting of a Result Matching to the						
52076	55076 Incorrect Reference Sample	Allison LLOYD	15/07/2021	41	Other	Justin HOWES	Closed Approved
					Unintended Human		
55172	55172 Mismatching Profile	Cassandra JAMES	12/08/2021	4	Error	Abigail RYAN	Closed Approved
						Vicki	
		Vicki PENDLEBURY-			Unintended Human	PENDLEBURY-	
55253	55253 Timesheet/Published Roster discrepancies	JONES	10/08/2021	37	Error	JONES	Closed Approved
	Error in the registration of a DNA Extract for				Unintended Human		
55357	55357 storage transition	Kirsten SCOTT	15/09/2021	33	Error	Adam KAITY	Closed Approved
	Incorrect process applied to				Unintended Human		
55369	55369 reference/suspect check comparisons	Adrian PIPPIA	26/08/2021	166	Error	Allison LLOYD	Closed Approved
	CDNAEXT20211101-11 - Failed QIAsymphony		j) jes		Unintended Human		
55612	55612 Extraction	Luke RYAN	8/11/2021	44	Error	Luke RYAN	Closed Approved
	Positive control Microscopy Slide Negative				Unintended Human		
55782	55782 for Sperm	Allison LLOYD	11/11/2021	50	Error	Luke RYAN	Action
55409	55409 Sample Not Loaded to NCIDD	Penelope TAYLOR	15/09/2021	107		Allison LLOYD	Investigation

*Need to improve TAT, action speed on OQIs

Queensland Realth

Stakeholder Issues/Complaints

55076 Reporting of a Result Matching to the Incorrect Reference Sample

OQI Details

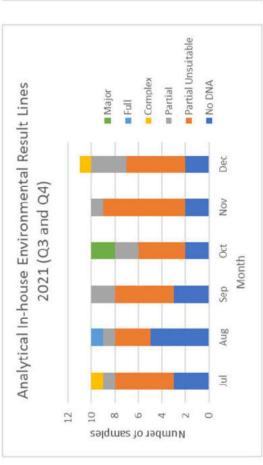
Status Closed Approved

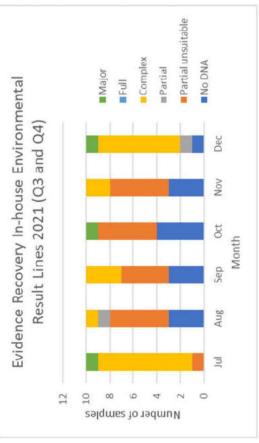
Subject A DNA result line for Sample was updated after comparison to reference DNA profiles. The result line was validated after being updated against the incorrect reference sample. This was discovered at Statement writing stage when it was corrected.

Source of OQI External Complaint

Date Identified 15/07/2021

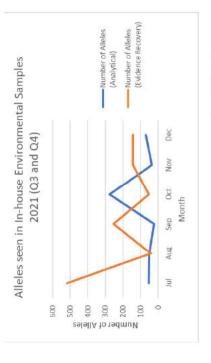
Environmental Sampling







Environmental Sampling



Evidence Recovery submitted 10 samples

November and 11 samples in December.

Analytical submitted 10 samples in July-

each month from July to December.

Evidence Recovery

5 x staff matches:

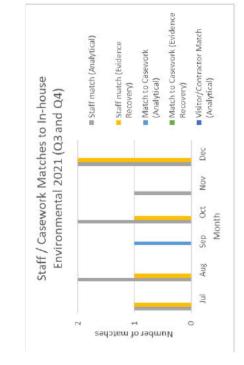
5 x ER staff members

Analytical

8 x staff matches:

- 5 x Analytical staff members
- 3 x Laboratory Assistants





Audits

	Jan-Jun 2019	Jul-Dec 2019	Jan-Jun 2020	Jul-Dec 2020	Jan-Jun 2021	Jul-Dec 2021	Target
Audits Raised	9	3	4	10	8	2	≥3
Risk Assessment/WHS Audits	-	က	5	9	2	က	≥1
Audits Overdue	0	0	0	1	0	0	0

Nil NATA assessments in this period. Next visit scheduled mid-2022

- 2 x Standard audits completed as scheduled
 - JIC kits
- Equipment & Calibrations
 Nil OQIs raised from these audits

3 x Risk assessments

- Removal of BSD Ascent 600 punch guard
- MPS project: Ion Chef system and Ion Genestudio S5 Plus reagents
 - First Aiders

Proficiency Testing

12 x proficiency tests completed - all by due date

6 x Standard

3 x Saliva / FTA

1 x Body Fluid

2 x Parentage

12 tests have follow-up completed

All results consistent with manufacturers data

Document Control

	Jan-Jun 2019	Jul-Dec 2019	Jan-Jun 2020	Jul-Dec 2020	Jan-Jun 2021	Jul-Dec 2021	Target
Total Documents	219	214	212	209	203	205	Į.
Number documents overdue for review	12	œ	7	7	-	0	<5
Average days past review	43	19	25	118	က	51	<45
Overdue notifications	548	214	258	204	450	480	<500

New Documents:

#36045 Multi-kit stutter calculator #36061 Procedure for resolving DNA profile interpretation #36067 Forensic DNA Analysis Newsletter #36070 Delivery of the gradual exposure checklist

#36071 Gradual exposure training checklist #36129 Forensic DNA Analysis values and behaviours in action

Management Review 2021 Q3 & Q4 - Quality & Projects

Next period expecting some Verifier specific documents will be generated



Finance

their need. This area can be impacted by COVID-19, as QPS officers don't wish items listed on the SOA due to the COVID-19 pandemic. Computer costs have and Computers. Revenue was lower, which is beyond the work unit's control, as the Queensland Police Service submit person samples for testing based on was overbudget in 3 main areas: Health Practitioner Labour, Clinical Supplies (with Police Services Stream overspent by \$181,605). Forensic DNA Analysis to be in close proximity to others. Clinical supplies can fluctuate due to the Australian Dollar (some supplies being purchased from overseas) and some At the end of Dec 2022, Forensic DNA Analysis was overspent by \$125,500 trended upward over the financial year.

Miscellaneous Topics

- Training and Professional Development:
- Created Gradual Exposure training program
- SHO OHR:
- First Aid Risk Assessment completed 2 staff per block
- Procurement/Supplier performance: Nil issues
- Calibrations:
- Some delays in accessing BTS services and external services
 - Nil internal issues

Projects

CM#	Project Name	Started	Started Completed	Document Status	Project Status
199	Proflex	8/08/2018	11/01/2022	Complete	Complete
200	Statement format and wording revision	31/08/2018		Minor change in progress	On hold pending FR enhancement
206	Y Filer plus	5/04/2019		Project proposal signed	Lab work in progress
211	Streamlining of DNA profile result reporting workflow	25/06/2019		Initial request signed	On-hold pending Verifiler validation
213	Verifiler	10/09/2019		Report writing in progress	Report writing in progress
216	Validation of Ion chef and S5	21/06/2021		Ethics approved	Lab work started
221	Impact of Magnetic Fingerprint Powders on Bead-Based Trace DNA Extraction	1/07/2020		Project proposal in draft	Query continuation of project
222	Profiling of Spermatozoa from Microscopy Slides	10/02/2021		Ethics approval submitted	Query continuation of project
226	Collection of Sperm from Pubic Hair	TBA		Nil started	Query continuation of project
227	Baseline method trial	7/12/2020		Final report in draft	Final report in draft
229	Paternity Index Distributions by Locus in PP21	27/10/2020		Final report in draft	JAH to evaluate/review implementation
232	Paternity calculations for mixed DNA samples	13/04/2021		Ceased	Ceased
233	Bone sampling and demineralisation protocol	30/04/2021		Project proposal in draft	Nil progress
234	Process mapping of interpretation and reporting	11/06/2021		On hold	On hold
235	2021 FR version upgrade	10/02/2021		N/A	UAT in progress
237	Reduction in physical case file creation	30/07/2021 15/09/2021	15/09/2021	Complete	Complete



Change Management

Implementation date	Details	Project Leader	Area Affected
14/07/2021	Action from OQI 54954 - Kinship program no longer has the dataset STRBASE ALL (1036) visible for selection. The only datasets visible for selection are three PP21 datasets, and three STRBASE datasets for paternity CTS cases. At the same time as making this minor change, the default datasets in Kinship are now marked to be the three PP21 datasets, here QLD datasets no longer visible for selection. If these datasets are required account at the Administration tab.	ЈАН	Reporting
19/07/2021	SS with high stutter guidelines implemented today. Comment added to SOP 17117. Email sent to all Case managers. See also G:\ForBiol\AAA Forensic Reporting & Intel\AAA Reporting guidelines\Proposed SS guidelines.	KDR	Case Managers
28/07/2021	HP2 Forensic Technicians to sample blood cloth samples in line with exisiting competency for the sampling of FTA cards and Guthrie cards	ARM	Evidence Recovery
20/09/2021	Admin team to no longer create casefiles for Category 1 and 2 cases with statement requests unless they are Sexual Assaults. Comments added to SOPs 17117 and 34248. Email to be sent to all Reporting Scientists and to Admin.	AKL	Reporting Scientists, Admin
19/10/2021	Minor Process Change: Promega change of PP21 QC detection from 3130 series to 3500 series instruments.	LBR	Analytical
12/11/2021	Minor change to process - Any manual reference samples that are received e.g. hair and other non-FTA samples should be assessed by Evidence Recovery and the category type checked. If the category type is anything other than "reference" it should be referred to the Quality team to have the category changed to "reference". A notation will be added to the sample to state the reason for changing the category type, for example "Category manually changed from "Hair" to "Reference" by Quality team to ensure correct reference processing".	Quality	Evidence Recovery



Administration Team

Summary of Happenings

July, August, September

OHS (Riskman) - Nil incidents

- Training attended
 - Recruitment
- Flexible Work Sessions x 2
- Accidental Counsellor Training Workshop (Lifeline)
- o Forensic DNA Analysis Newsletter back into production.
- Worked with SSDU regarding courses for Admin staff
- o OQI 55253 "Timesheet / Published Roster Discrepancies" raised as a result of HR Audits/ discrepancies in HR recordings.
- October, November, December
- Training attended –Job Seeking Skills
- We had some fun / laughter with Fun Activities / Dress Up
- o Two (2) admin staff involved in Values & Behaviours Presentation
- Project #237 commenced creating case files as per Category
- Change to system used within court room for video evidence in some courts / areas.



Administration Team

Highlights / achievements

- myHR Audits undertaken working towards a concurrent timeframe in the future
- Values & Behaviours Admin Team Members assistance provided towards videos, documentation & posters
- FR configuration of storage begannered case files Forensic Chemistry (this was a long journey, but success at last)

Most difficult problem/issue for your team has encountered in the last 6 months.

change. (Payroll were training new staff in "new e-forms" and then the new processing staff were applying that knowledge to current myHR processes, even though Change in how Payroll required leave forms for part days to be recorded, and that there was no communication about such to the myHR Superusers about this the e-forms had not yet been rolled out to our department).

emails) with a Payroll Leader, and a two (2) page document being prepared to assist staff in completing forms. Forms are still, on occasions, not being completed fully/correctly and admin staff are making amendments to the comments fields on forms after checking against timesheets and leave record entries, which is very The change required all the hours of work/breaks/leave to be broken down in the comments fields and resulted in many queries from staff, meetings (and many time consuming.

If there was one thing that could be changed or improved to assist you, what is it.

myHR users. We still await news of the rollout of the new "e-forms" and other updates (Pilot has been running since the start of October 2021). There is a definite Admin staff worked through this issue and came up with all the solutions to the problems being faced due to changes by Payroll which had not been conveyed to frustration at the lack of information being cascaded down to admin teams who need to ensure forms are being correctly lodged and prepare for the new processes. We feel that there is not much we can do within our unit to address this – other than making written enquiries with Payroll Services Leaders.

continued ...



Administration Team

Most difficult problem/issue for your team has encountered in the last 6 months.

e leave directions handed down by Qld Health were open to interpretation and have led to much confusion and differing advice being provided to staff. This has caused some frustration to admin team members, who have worked hard to provide a good service to the team but have COVID related leave – without hard and fast rules admin have struggled to know what leave staff should be applying for. The information / flow charts were just been unsure themselves that the information they are providing is not going to change / be interpreted differently along the workflow. updated towards end of Januar

If there was one thing that could be changed or improved to assist you, what is it.

changing so quickly. Our admin team, with the support of our Team Leaders, have done all we could do to support staff through all the changes, and we can't think We are finally through the toughest part of this issue, at least we hope so, and have finally received better instructions as to how forms should be completed and of anything more we could now do to improve matters. The collective Police Services Admin Team, worked together to ensure a consistent approach for leave what leave is relevant to each situation (COVID related). Our leave codes/options and instructions are now dated, which has been a big help as things were requests for this pandemic across Police Services Stream.



Administration Team

• Statements (including Evidence Certificates, Re-issued)

0	o July	tatements (of which 5 had "Peer Reviewer initials added to Statement as per FR Record)
0	August	78 Statements (of which 7 had "Peer Reviewer initials added to Statement as per FR Record)
0	September	54 Statements (of which 1 had "Peer Reviewer initials added to Statement as per FR Record)
0	October	45 Statements (of which 4 had "Peer Reviewer initials added to Statement as per FR Record)
0	November	71 Statements (of which 5 had "Peer Reviewer initials added to Statement as per FR Record)
0	December	51 Statements (of which 6 had "Peer Reviewer initials added to Statement as per FR Record)

393 Statements

9 more statements for this period than in Q1 & Q2 of this year.

	\rightarrow	←	\leftarrow
356	348	384	393
m	ന	ന	ന
22	8	32	75
Q1 & Q2	Q3 & Q4	21 & Q2	33 & Q4
δ	g	g	Ö
	_		
2020	2020	2021	2021
L V	· · ·		· ·



Administration Team

· Page Numbering of Case Files (adding case identifier where required) followed by review of each file.

6566 pages (3564 pages of which were numbered by Laboratory Assistants) 5737 pages (2242 pages of which were numbered by Laboratory Assistants) 3853 pages (2755 pages of which were numbered by Laboratory Assistants) 5486 pages (1527 pages of which were numbered by Laboratory Assistants) 2871 pages (443 pages of which were numbered by Laboratory Assistants) 1916 pages (778 pages of which were numbered by Laboratory Assistants September November December October August o July

Total 26429 pages

Down 8172 pages from that done in Q1 & Q2.

2020 Q3 & Q4 33610 2021 Q1 & Q2 34601↑ 2021 Q3 & Q4 26429



Administration Team

Creating Case Files

- July 70 case files
- August 54 case files
- September 28 case files
- October 19 case files
- November 19 case files
- December 16 case files

Total 206 case files

(121 less than Q1 & Q2 due to Project #237)

2021	20 Q	→ /0×
2020	Q1 & Q2	307
2021	Q1 & Q2	327 ↑
2021	Q3 & Q4	206 ↓
2021 2021 2021	Q3 & Q4 Q1 & Q2 Q3 & Q4	267 \\ 327 \\ 206 \\

Request for Copy of Case Files

Each file is scanned (copied) and a watermark added, then reviewed prior to distribution to the requestor.

July - 4 case files

August - 2 case files

September - 1 case file

October - 5 case files

November - 1 case file

December - 3 case files

Total 16 files

(5 less than Q1 & Q2)

2020	Q1 & Q2	9
2020	Q3 & Q4	27 ↑
2021	Q1 & Q2	21 \
2021	Q3 & Q4	16 →

Queensland Health

Quarter 3 - 2021

Administration Team

• HR forms processed (Stats from myHR Dashboard)

			655 forms			66 - 10.07%	470 - 71.75%	119 forms	
	Totals		Submitted		Received	Late	On Time	Early	
	Early	21	2	20	12	12	16	21	15
	On Time	58	20	62	69	22	09	47	29
Received	Late	13	80	Ŋ	5	80	80	80	11
Number of Forms	- ubmitted	92	09	28	86	2.2	84	92	83
Pay Period		01/2122	02/2122	03/2122	04/2122	05/2122	06/2122	07/2122	08/2122

myHR Forms Returned from Admin Peer Review to staff members for this period –

14 of 205 forms (August) 6.82% 14 of 173 forms (July) 8.09%

17 of 199 forms (September) 8.54%



Quarter 4 – 2021

Administration Team

• HR forms processed - (Stats from myHR Dashboard)

			528 forms			69 - 13.06%	324 - 61.36%	135 forms			
	Totals		Submitted		Received	Late	On Time	Early		%	%
	Early	∞	41	29	29	33	22		18 of 180 forms (October) 10%	18 of 247 forms (November) 7.28%	15 of 177 forms (December) 8.47%
	On Time	56	42	72	45	55	54		18 of 180 forn	18 of 247 forn	15 of 177 forn
Received	Late	7	6	15	12	9	20		ers for this period –		
Number of Forms	Submitted	71	65	116	98	94	96		Peer Review to staff membe		1192
		122		122					myHR Forms Returned from Admin Peer Review to staff members for this period –		2020 Q1 & Q2 1192
Pay Period		09/2122	10/2122	11/2122	12/2122	13/2122	14/2122		myHR Forms R€		

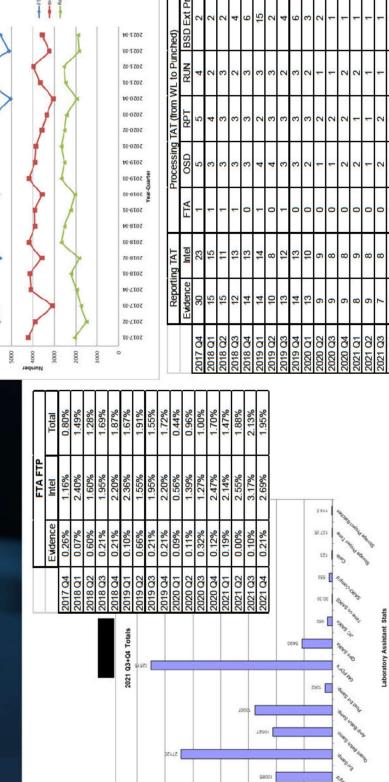
Queensland Health

2021 Q1 & Q2 874 ↓ 2020 Q3 & Q4 1275 ↑

2021 Q3 & Q4 1183

FTA Quarterly Totals

Quality & Projects Team Laboratory Assistant Stats



Queensland Health

Management Review 2021 Q3 & Q4 - Quality & Projects

Quality & Projects Team Highlights

- Possibility of a replacement for STORstar Tube tracker (Microelectronics)
- runs on currement operating system
- improved OHS, nil pinch risk

BSD

- · easy clean, smaller instrument
- potential to scan and store 3D-barcodes from NUNC tubes into plate map or LIMS
- 2. Commercial work with ARUMA
- External work that has become routine.
- Established processed for processing and reporting
- Excellent communication and scientific communication history
- 3. KDS added to HEC committee opportunity to learn new skills for the laboratory



Quality & Projects Team

Difficult issue/problem:

function/s, actual errors, result duplications. All these functional issues result in · Slow IT, particularly as it impacts on FR use: causes issue with incorrect batch records that are difficult and time consuming to correct (if found)

Things that could change or improve:

For all staff involved in project to

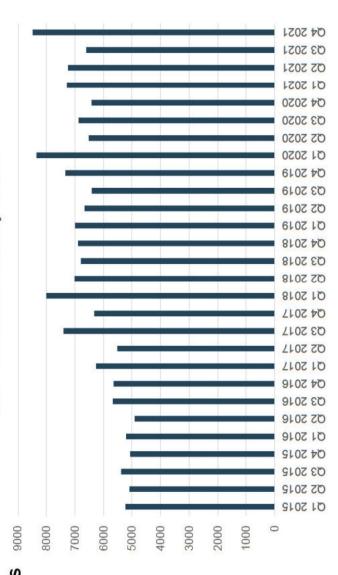
- Set realistic goals in terms of due dates
- Be committed to meeting the target dates they set
- Keep excellent records as they progress through projects
- Finish projects: with all paperwork steps completed without prompting by quality

Evidence Recovery - KPI data overall picture

Total Exhibits Received by Quarter Averaging over 7000 exhibits 9000

The general trend is still increasing.

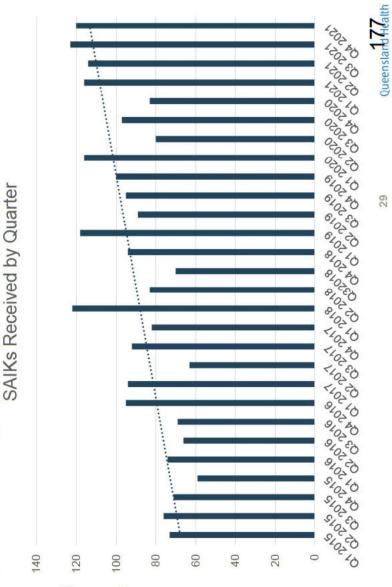
each quarter.



Evidence Recovery - KPI data SAIKs

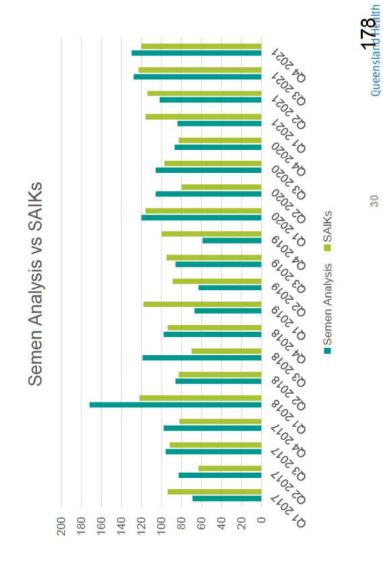






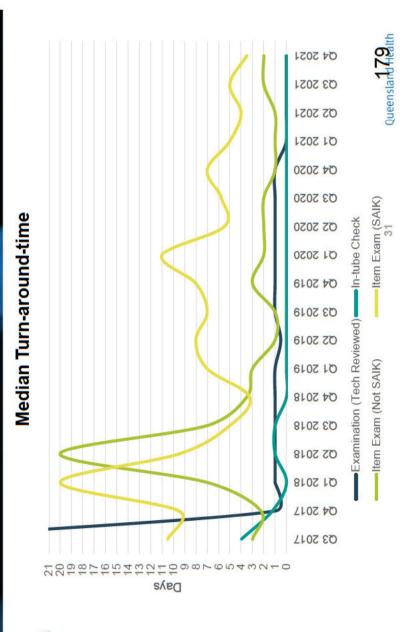
Evidence Recovery - KPI data Semen examinations

semen testing were higher than the number or AIKs Whole items received for received in Q3 and Q4.

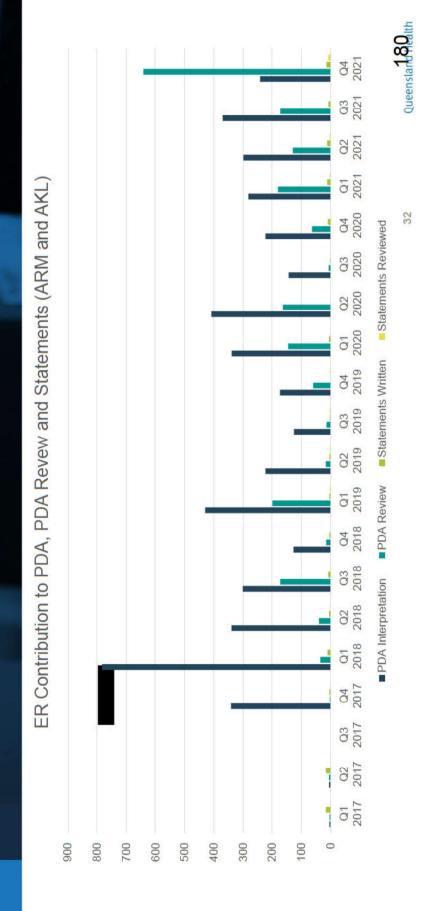


Evidence Recovery - Turnaround times

Overall, turn-around times are still low. The have increased somewhat for Non-SAIK whole items however not significantly.



Evidence Recovery - KPI data PDA and PDA review



Queensland Realth

Evidence Recovery - Other

- Supply
- No impacts on supply noticed in the reporting period
- Workplace Health and Safety
- o No major events affecting the team
- Client engagement
- Nothing specific to note

Evidence Recovery – Other continued

Projects

- provisional approval pending ethics review submission on hold Proposal#222 (Profiling of Spermatozoa from microscopy slides) - FSS research submission g
- Proposal#233 (Bone sampling and demineralisation protocol) Planning commenced, involved quite a bit of research into various protocols and equipment, awaiting AKL review

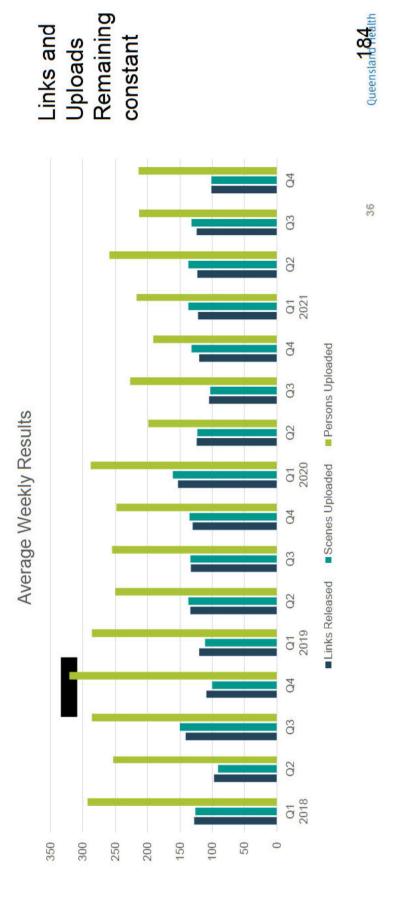
Other

some upheaval to the team. AKL still continuing to learn ER tasks, complete outstanding Significant change to supervising staff of ER team near end of Q4 2021 which caused General feeling is that AKL time is being spread too thin between both ER and Intel work including updating many documents and build relationships within the team. duties and is not ideal until more competence is developed on the ER side.

Intelligence - KPIs

	2019 Q3-4	2019 Q3-4 2020 Q1-2 2020 Q3-4	2020 Q3-4	2021 Q1-2	2021 Q3-4
Links Released	3447	3729	2809	3184	3343
Crime Scenes Uploaded	3537	3798	2926	3571	3398
Person Samples Uploaded	6595	6528	5248	6191	6312
PDA	2176	269	121	421	218
PDA Reviewed	5481	5443	4235	4657	2320 Note: tallies stopped in Nov

Intelligence - Weekly Averages



Queensland Plealth

Intelligence - Statements and Court Attendance

Statements Written Q3: 57

Statements Written Q4: 1

Statements Reviewed Q3: 61

Statements Reviewed Q4: 1

Tallies for statements written and reviewed stopped in November

• Court: 3

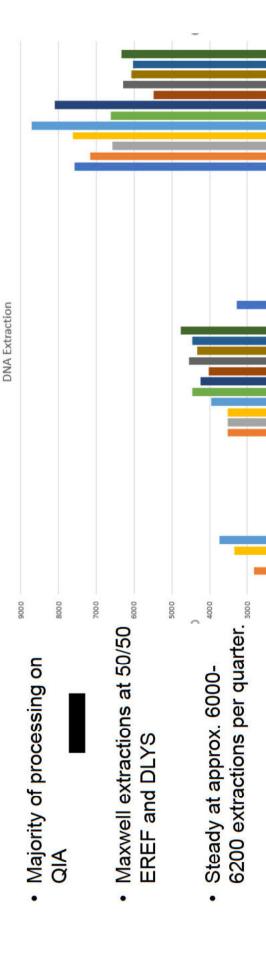
Intelligence - Other

- Audits NIL
- WH&S issues MIL
- OQIS NIL
- Training NIL
- undertaken. Approx. 150 sample determined to have not been uploaded from around o Mini project – internal audit of samples from old cases not uploaded to NCIDD 2000-2003. Not actioned as of yet.

Intelligence - Additional Impacts

- Cold cases Op Pearl AKL
- 6 weeks admignin HP6 role AKL
- instead of Intel. Changing teams had a big impact on staff however output remained Intel in November. All PDA and statement tallies were then added to the ER results - Changes to team, AKL started in the ER senior scientist role whilst still maintaining fairly consistent.

DNA Extraction - Sample input



#Q12019 #Q22019 #Q32019 #Q42019 #Q42019 #Q42019 #Q42020 #Q42020 #Q42020 #Q42020 #Q42020 #Q42021 #Q42021 #Q42021

2000

24734 extractions in 2021

1000

Highlight 1 - Priority 1s

Large number of P1s during this period.

Processing times very fast (measured typically in hours).

Anecdotally P1 processed seems to be getting faster.

Is a pressure on team but also brings team together

Highlight 2 - VFP

- Significant progress made on VFP project.
- Analytical CW Final Report approved.
- African Microvariant report approved.
- EREF and Direct nearing completion.

Queensland Health

Highlight 3 – Keep calm and carry on

- Despite COVID and disruptions to:
- Staff availability and schools
- Supply of reagents and consumables
- Availability of engineers

Work has continued without significant disruption to business continuity.

Staff have been flexible and adaptable.

Most Difficult Issue - Staffing and BCsC

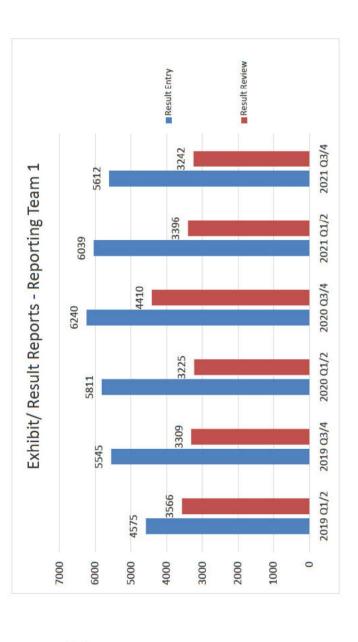
Rotating HP4 creates constant state of flux and lack of stability and continuity.

 Lack of certainty regarding Intel role and training not provided to all staff creates disharmony. • Slow progress on Business Cases for Change (BCsC) gives perception of "stereotypical government" - slow, ineffective and demotivating.

One Change

• Finalise BCsC to provide certainty and stability at all levels within FDNA.

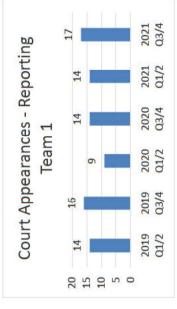
PDA / Rev

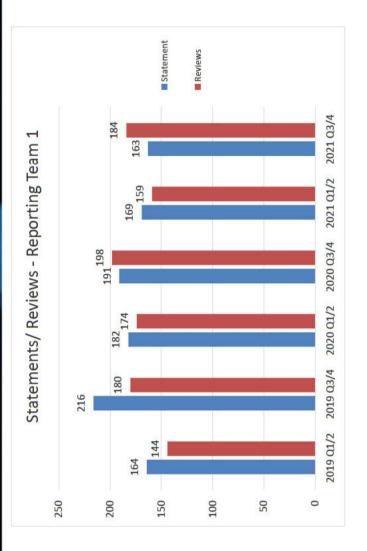


Relatively steady

DNA Extraction - Sample input

- Many more required in 2nd part of the year
 - Reviews higher than written as project people find doing a quick review a good way to contribute
- Court highest half for 3 years





Most Difficult Issue - Media attention & incoming work

- Media attention and the requirement to focus on a specific case.
- Disruption to meny ataff hands on having to divert attention to this case for auditing of samples and fulfilling external requests
- Disruption to focus on current work
- Negative impacts to confidence especially with court representation
- More work than we could handle was coming in the door
- Statement requests prior to Christmas
- (63 more statements written in reporting in total this year than last year)
- Outstanding PDA climbed adding extra pressure

Highlight 1 – Team unity

- particular Thys in the face of a constant media barrage. The team were very supportive of each other and in
- Many staff fronted to court and represented us well during that time
- Information was shared to benefit all

Queensla 200 Ith

Highlight 2 – Priority 1s

- Large number of P1s during this period.
- Each scientist accepted responsibility for the task when asked

Highlight 3 – Maintaining service to QPS

- Lock downs continued through this period and staff adapted termsrk from home with short notice
- Staff have been flexible and adaptable.
- PDA and statement writing and reporting of coronial cases Training continued with new staff sometimes remotely for

One Change

Asked for help sooner / shed non-core work.

When PDA samples were rising we asked for help from other teams e.g plate reading roster

Project work could have been more limited to assist in maintaining PDA / review numbers

KPI Data

	Trend	<u>\</u>	\$	\$	<u>*</u>	\$
	Q3 & Q4 2021	132	126	4463	3784	9
Totals	Q1 & Q2 2021	128	136	4031	2769	2
	Q3 & Q4 2020	107	108	4319	3002	2
	Q1 & Q2 2020	122	149	4453	3647	7
		Stateme nts	Stateme nt reviews	PDA entry	PDA review	Court

Management Review 2021 Q3 & Q4 - Reporting Team 2

<u>Other</u>

- · 10 × P1
- 4 x Significant Cold Casework
- 1 x 95a

Highlights/achievements

YFP progress

Court testimony from home via Pexip

Attained more paternity competency within the team

Resilience in face of media events

Team Challenges

Increase in workload

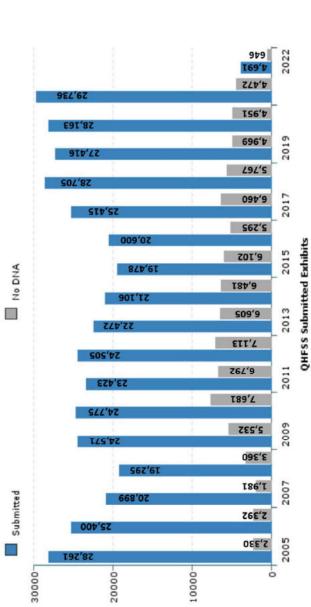
Significant defence challenge for 98 case

URL rejected issue

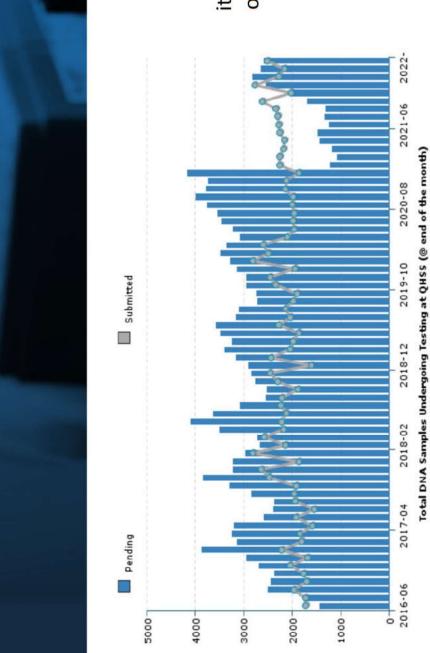
29

Improvement Idea

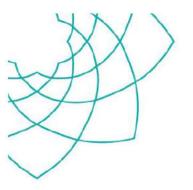
Ability to take from a pool of competent Case Managers when FRIT workload gets high



Peer reviewed results amended: 52 – cause: unintended human error (previous 6 months: 46)



items to be reported on at the end of Dec 2021: 2842 **Total outstanding**



JH-12



The Examination for and of Spermatozoa

1 Purpose

The presence of spermatozoa is a confirmatory test for the presence of semen. This document describes the method by which a scientist performs microscopic examination for the presence of spermatozoa which includes the preparation of microscopic slides from exhibits, staining of slides and interpretation of the microscopic smears/slides for spermatozoa and other cellular material.

2 Scope

This Standard Operating Procedure (SOP) applies to all scientists performing the examination of items for the presence of semen.

3 Definitions

In this document, where reference is made to spermatozoa, it refers to human spermatozoa unless otherwise specified.

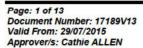
4 Background

The investigation of sexual assault cases may require the testing of exhibits collected as part of a forensic medical examination or scene examination for the presence of semen. Within the laboratory the detection of spermatozoa confirms the presence of semen. A reliable and accurate staining method is essential to aid the examining scientist the ability to differentiate between cellular types; most significantly spermatozoa from epithelial, yeast and white blood cells.

Currently the Haematoxylin and Eosin (H&E) stain is adopted for this process. The H&E staining method has been used for this purpose within the laboratory for many years. The haematoxylin (basic stain) stains the deoxyribonucleic-acid (DNA)/histone rich base of the spermatozoa head deep purplish-blue. The eosin (acidic stain) stains the acrosomal cap pink and the tail pink if the spermatozoa are intact (N.B. because Forensic DNA Analysis uses a water based eosin stain, the acrosomal cap often appears very light pink or clear). The use of counterstaining differentiates spermatozoa from most cellular debris.

Confusion with yeasts, especially monilia, can occur and extreme care must be taken when monilial infections such as thrush are suspected. With experience, spermatozoa and yeasts can be distinguished by size and/or the presence of cell walls.

Haematoxylin is a natural dye. Its active colouring agent is haematin, which is formed by the oxidation of haematoxylin. This oxidation process or "ripening" occurs when haematoxylin solutions are allowed to stand for several days. However, the process can be accelerated with the introduction of an oxidising agent such as sodium iodate. During





oxidation the haematoxylin loses two atoms of hydrogen, and its formula changes from $C_{16}H_{14}O_6$ to $C_{16}H_{12}O_6$. Sufficient haematoxylin should be left unoxidized in the solution so that natural oxidation can continue thus prolonging the shelf life and useability of the stain. Completely oxidized haematoxylin becomes colourless. As the oxidation process occurs when haematoxylin is exposed to light and continues over the life of the solution, haematoxylin should be stored in dark bottles until ready for use. Haematoxylin is an excellent nuclear stain. Haematin, via the aluminium ion mordant, binds to the anionic sites in the nuclei (a mordant is a substance that causes certain staining reactions to take place by forming a link between the tissue and the stain). At this stage the nuclei stain red, which is then converted to the blue-black colour when the pH is raised (by a weak alkali wash such as Scott's tap water substitute in some H&E staining methods) known as "blueing".

To avoid stain precipitation on the slide, the haematoxylin solution must be filtered. It should be changed immediately if staining quality deteriorates.

Eosin is an acid dye which combines electrostatically with the acidophilic tissue components such as cytoplasm. (an anionic dye that stains the cationic tissue components). Alcoholic and acidified solutions of eosin tend to stain much more vividly than do the aqueous solutions. With water soluble eosin, rinse in water very quickly or else eosin will wash out.

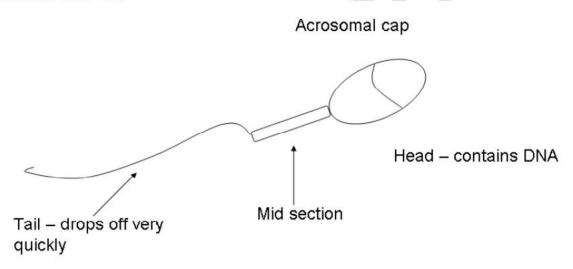


Figure 1 Spermatozoa

5 Actions- Staining procedure

5.1 Slide Preparation

Use new slides and clean with ethanol. Label with the sample ID, date, case number and sampler's initials using a pencil only. Use clean, flamed instruments.

Create a suspension from the exhibit by one of the following methods,

Scrape the stained area into a 1.5/2ml tube. Add between100-300µl nanopure
water with a POVA pipette to the tube until the scraping is moist. Vortex
thoroughly.

- Excise the stained area and cut into small pieces. Place pieces into a 1.5/2ml tube and add between100-300µl nanopure water with a POVA pipette to the tube until the pieces are moist. Vortex thoroughly.
- 3. If slide is being prepared from a swab, excise the material from the swab and cut the material into small pieces. Place the pieces of material into a 1.5/2ml tube and add between100-300µl nanopure water with a POVA pipette to the tube until the pieces are covered (approx 200µl). Vortex thoroughly.

Add a drop of the recently vortexed suspension to the labelled slide.

Dry the slide on a heat block. If a heat block is not available, heat-fix the slide by passing it over a flame with the material to be stained uppermost once the suspension has dried.

5.2 Slide Staining

Microscopic slides are stained using Haematoxylin and Eosin (H&E). The method for preparing the staining reagents, and performing manual staining is as per procedure detailed in Appendix 1.

Note: registration of the reagent and association to individual exhibits is detailed in QIS 24469 (Batch Functionality in AUSLAB)

5.3 Microscopic Examination

Examine slide using the x40 or x100 objective using the oil immersion, or the x40 objective using the dry microscope. Score the number of spermatozoa observed (use the standard microscopy form, QIS <a href="https://doi.org/10.2101/j.nc.2016/j.nc.

0 (0)None seen <+ (<1+)Very hard to find (Use England Finder Graticule) (1+)Hard to find + Easy to find (2+)(3+)Very easy to find +++ (4+)Abundant

Note whether spermatozoa are intact (heads and tails) or non-intact (heads only). Look for epithelial cells and whether there are bacteria or yeast present. Human spermatozoa are distinguished from non-human mammalian sources by their morphology and by their behaviour toward H&E, resulting in a purplish/blue head and light pink/clear cap (see Section 5.4).

If less than ten spermatozoa are located, for at least one spermatozoa, note the location on the slide with the use of the England Finder Graticule (see Appendix 3).

5.4 Animal Semen

Animal spermatozoa are morphologically different to human spermatozoa and react differently to staining. Where suspected spermatozoa are located which are morphologically different to human spermatozoa, the examining scientist should consider the possible presence of animal spermatozoa. N.B. Forensic DNA Analysis does not identify or characterise animal spermatozoa.

5.5 Spermatozoa Interpretation

If slides are stained properly spermatozoa should be easily distinguished from epithelial cells, cellular debris, fibres etc. Spermatozoa heads can look similar in shape and colour to yeasts. If in any doubt consult an experienced examiner.

The recovery of semen is dependent on a number of factors but not limited to

- The amount of spermatozoa in the ejaculate
- The amount of ejaculate
- · The environment the ejaculate is deposited on
- Washing
- Douching
- Menstruation
- Efficiency of the sampling process
- Time between ejaculation and sampling
- Storage of the samples
- Natural drainage or degradation of spermatozoa in certain environments

With respect to the above influences, the time since ejaculation has occurred can only be estimated. A number of studies have been conducted regarding the persistence of spermatozoa in the vagina. References to these studies can be found in Appendix 2.

6 Records

Nil

7 Quality assurance/acceptance criteria

Controls are used to test the quality and validity of the staining reagents prior to use. A positive control slide should be tested prior to the staining of slides for microscopy, (once daily), each time a new batch of Haematoxylin and Eosin solution is prepared and when positive control slides are prepared.

A positive control slide is a known sample of human semen, which has been diluted.

7.1 Creation of H&E control slides

Collect human semen in a sterile green-capped "Falcon" tube. The tube is to be labelled with the following information:

- Sperm donor number
- Date and time of collection

The semen is to be stored within a freezer until required to create H&E positive control slides using the following process:

- 1. Clean heating block using bleach and 70% ethanol solution.
- Clean frosted microscope slides with ethanol and label with white label (H&E Pos Ctrl: Sperm donor number; Lot No.).
- Spread slides out on heating block to heat before use.
- 4. Clean automatic pipette with bleach and 70% ethanol solution.
- 5. Using a new filtered pipette tip, add 20µL of the neat semen to 10.0ml nanopure water using a clean 10ml tube. Vortex.
- Add 20 µL of the diluted semen solution to each slide.

- 7. Heat fix the slides on a heating block at 50°C for approximately 30 minutes.
- Store the slides in labelled plastic slide box "Unstained H&E Positive Control Slide Storage", and store the box in Rm 6124.

7.2 Testing and interpretation of control slides

The following process is used to test, interpret and record control slide results:

- Remove a H&E control slide from slide box, label with date, initials, lot number and batch, and stain with H&E using the method in Appendix 1.
- 2. Dry slide on heating block at 50°C.
- 3. Coverslip slide using mounting medium.
- Examine slide microscopically in Rm 6119 as per section 5.3.
- Completed control slides get transferred to a plastic box labelled H&E Control Slide Storage box #
- 6. Once a slide box is full of completed positive control slides, transfer the box to Rm 6106B, 'Exhibit Room' for long term storage.
- 7. Add audit entry to document that the control slide has passed control and what box it has been stored in (i.e. 2)

Acceptance of the reagents is based on the interpretation of the Positive control slide. The following criteria must be met before passing the reagent for use:

- Spermatozoa head stains a deep purplish-blue.
- Acrosomal cap stains light pink/clear.
- 3. Tail stains pink.

In the event the control slide fails the following process is to be completed:

- Repeat the staining procedure with a new control slide and assess as above.
- 2. If the control slide fails again then discard the in-use staining batch and stain a new slide using a fresh batch from the stock solutions and assess slide as above.
- If the control slide fails then a new batch of stock solutions must be prepared and the old solutions discarded, and assess the validity of the reagents as per this section.



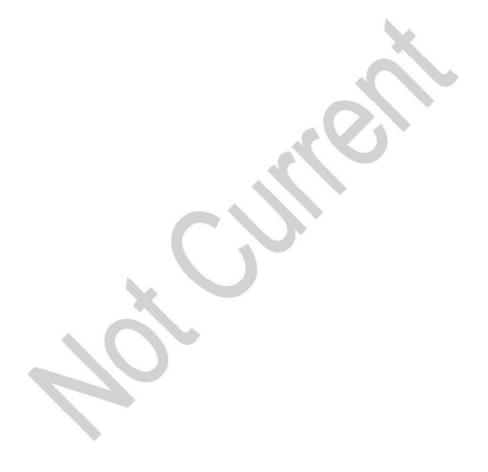
8 Associated Documentation

- QIS: <u>17037</u> Microscopy of Smears Form
- QIS: 17142 Examination of Items
- QIS: <u>17185</u> Detection of Azoospermic Semen in Casework Samples
- QIS: 17186 The acid Phosphatase Screening Test for Seminal Stains
- QIS: 20080 Digital Imaging in Forensic DNA Analysis
- QIS: <u>23849</u> Common Forensic DNA Analysis Terms and Acronyms
- QIS: 30800 Investigating Adverse Events in Forensic DNA Analysis Unit
- QIS: 24469 Batch Functionality in AUSLAB
- QIS: 32106 Examination of Sexual Cases

9 References

- 1 Biology Methods Manual, Metropolitan Police Forensic Science Laboratory, Great Britain, 1978.
- 2 Allard, J.E (1997). "The collection of data from findings in cases of sexual assault and the significance of spermatozoa on vaginal, anal and oral swabs." Science and Justice V37(2): April; 99-108.
- 3 Allery, J.P., Telmon, N., Mieuset, R., Blanc, A., Rouĝe, D. (2001). "Cytological Detection of Spermatozoa: Comparison of Three Staining Methods." *Journal of Forensic Sciences* V46(2): 349-351.
- 4 Brown, G. (1978) "An Introduction To Histo Technology"
- 5 Chiasson, D.A., Vigorito, R., Lee, Y.S., Smialek, J.E. (1994). "Interpretation of postmortem vaginal acid phosphatase determinations." *American Journal of Forensic Medicine and Pathology* 15(3): 242-246.
- 6 Collins, K.A., Bennett, A.T. (2001). "Persistence of Spermatozoa and Prostatic Acid Phosphatase in Specimens from Deceased Individuals During Varied Postmortem Intervals." *American Journal of Forensic Medicine and Pathology* 22(3): 228-232.
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10 Amendment History

Revision	Date	Author/s	Amendments
0	Unknown	Unknown	Unknown
1	Unknown	Unknown	Unknown
2	Unknown	Unknown	Unknown
3	27 Nov 2002	V lentile	Format updated, manual staining to appendix.
	10000		Removed notes on examination of swabs,
			removed unpublished paper, as work wasn't
			completed.
4	19 Nov 2003	L Freney	Updated references
5	12 Jul 2006	J Howes/A	"Reference" put after "Actions".
		Williamson	
6	05 Aug 2006	J Howes	Added in Sexual Assault Investigation
	100		Flowcharts, examination of SAIK Swabs,
			Photograph or Witness required for ++ (1+)
21	(M)		sperm and PSA test.
7	23 Oct 2006	J Howes	Reporting results Eg. ++ or 2+
8	25 Jun 2007	J Howes	Unified grading scale comments. Added
			Crimelite flowchart.
Version	Date	Updated By	Amendments
9	13 Mar 2008	QIS2 Migration	Headers and Footers changed to new CaSS
		Project	format. Amended Business references from
			QHSS to FSS, QHPSS to CaSS and QHPS to
			Pathology Queensland
10	16 July 2010	A Lloyd	Removal of Crimelite in scope and the
			Crimelite flowchart. Changed section 2.2 to
			include use of suspensions. Removal of
			section 2.8 – Vaginal Secretions. Changes to
	h.,		section 2.10 to remove AP testing on smears
			positive to spermatozoa. Photograph or
			locations required for smear with 1 or 2 sperm
			seen. Clarification of flowchart regarding
			previously screened items by QPS. Changes
			to SAIK flowchart. Removal of animal sperm
			diagrams and insertion of photographs of
44	00 5-1- 0044	Allered	animal sperm.
11	03 Feb 2011	A Lloyd	Amended use of vernier for slides to use of the
12	24 0-4 2042	Allend	England Finder Graticule.
12	31 Oct 2013	A Lloyd	Removed animal sperm photos. Amended
		J Seymour-	workflow charts, changed headings from CASS
		Murray	to HSSA. Change H&E solutions and staining,
			add England Finder information. Updated
13	03 July 2015	I Covmour	some hyperlinks. New template, update hyperlinks, some
13	03 July 2015	J Seymour-	
		Murray	formatting updates and minor wording
S		A	changes.

11 Appendices

- 1 Appendix 1: Preparation of H & E Stain and Manual Staining Procedure
- 2 Appendix 2: Persistence of Spermatozoa in the Vagina
- 3 Appendix 3: England Finder Package Insert

11.1 Appendix 1: Preparation of H & E Stain and Manual Staining Procedure

11.1.1 Chemical Hazards

Eosin (yellowish)

WARNING: Eosin (yellowish) can cause serious damage to the eyes. Avoid contact, wear PPE and eye protection.

Haematoxylin

WARNING: Haematoxylin: the toxicological properties have not been investigated. Prevent contact with skin and eyes. Do not inhale or ingest. Wear PPE and eye protection.

Sodium iodate (NalO3)

WARNING: Sodium iodate causes burns and is harmful if inhaled or swallowed. Protect eyes and skin.

Wear PPE and eye protection.

Glacial acetic acid

WARNING: Acetic acid is extremely corrosive and is harmful if inhaled or swallowed. Protect eyes and skin. Wear PPE and eye protection.

Pertex Mounting medium

WARNING: Pertex is irritating to eyes, respiratory system and skin.

Xvlene

WARNING: Flammable. Harmful by inhalation and in contact with skin. Irritating to skin.

NOTE: All reagents prepared in the laboratory shall bear a label:

```
(enter details eg 10% NaOH)....
Prepd from Lot/batch:.....

Date: ../../. Initials: ......

Expires:../../. Store at:....°C

WARNING: Contains ......
```

Or an individual label printed by the Bar-One Lite system (B1Lite on short cut).

11.1.2 Preparation of Eosin

Eosin –Water soluble (CI 45380)

- Eosin 5.0g
- Nanopure water 500mL

Procedure

- Weigh 5.0g Eosin and add 500mL nanopure water in a flask.
- 2. Mix on the magnetic stirrer until completely dissolved.
- Label the reagent bottle according to laboratory standards.

11.1.3 Preparation of Haematoxylin

Haematoxylin (CI 75290)

Haematoxylin 2.5g



- Sodium iodate (NalO3) 0.2g (Must be accurate)
- Potassium aluminium sulphate dodecahydrate (KAl(SO4)2.12H2O (potassium alum) 25.0q
- Nanopure water 350mL
- Glycerol 150mL
- Glacial Acetic acid 10mL

Procedure

- Weigh 2.5g Haematoxylin and dissolve in 350mL nanopure water in a flask. Mix on the magnetic stirrer. (Haematoxylin must be fully dissolved before adding the other reagents)
- Weigh 25.0g potassium aluminium sulphate dodecahydrate and add to haematoxylin solution, continue stirring.
- Weigh 0.2g sodium iodate and add to Haematoxylin solution, continue stirring until reagents are dissolved.
- Measure 10mL glacial acetic acid and add to haematoxylin solution, continue stirring.
- Measure 150mL glycerol and add to haematoxylin solution, continue stirring for 5 minutes.
- 6. Filter into a clean dark reagent bottle. (Haematoxylin oxidises)
- Label the reagent bottle according to laboratory standards.

11.1.4 Register lot details of reagent

 Register lot details using QIS 24469 Batch Functionality in AUSLAB and QIS 17165 Receipt, Storage and Preparation of Chemicals, Reagents & Kits (Appendix 3).

11.1.5 Manual Staining Procedure

Staining is performed in the staining fumehood in Rm 6124, main laboratory, Forensic DNA Analysis.

Procedure:

- Place slide on staining rack over sink, stain with haematoxylin for five minutes (add one volume and let rest)
- 2. Wash with nanopure water.
- 3. Stain with eosin for one minute.
- 4. Wash with nanopure water (quick wash).
- Allow to dry on hot plate.
- 6. Mount coverslip in Pertex

11.1.6 Staining Quality Controls

The following quality steps should be implemented:

- Haematoxylin should be filtered before use as the crystals in solution can result in stain deposit on the slides which affects the reading of the slide. (Once a week)
- Haematoxylin differentiates better if it is matured for 3 to 4 days before use.
- Eosin should be filtered once a week.

When stored in dark bottles, haematoxylin may keep for up to 12 months and eosin for up to 12 months (dependent on control slide result).

11.2 Appendix 2: Persistence of Spermatozoa in the Vagina

Literature provides a range of time periods for the persistence of non-motile spermatozoa in the vagina:



- Up to 24 Hours¹
- Up to 3-4 days²
- Up to 9 days or 12 days in the cervix, sometimes after menstruation³
- Up to 3 to 4 days, but may be longer⁴

Literature provides a range of time periods for the persistence of motile spermatozoa in the vagina:

- The number of motile spermatozoa discernible in the vagina may be normal after one hour and markedly decreased after 2 hours; after 3 hours normally no spermatozoa are found. Menstruation often prolongs motility in the vagina to as long as 4 hours compared with the normal period of 30 to 45 minutes.⁵
- Spermatozoa remain motile in the vagina for 2 to 3 hours and in the cervix for 48 to 110 hours⁶
- Normally 10% of the spermatozoa are alive in the vagina at the end of 2 hours post coitum.
 Variations in number and motility depend upon the pH of the vagina and semen, quantity of semen deposited, bacteria and flora of the vagina and the time examined post-coitally. The author has seen motile spermatozoa in the vaginal pool after 8 hours.⁷
- In several cases in which repeated examinations were possible before conception occurred, all motility ceased within one hour after intercourse. A fall of motility to 10% within 30 minutes is compatible with fecundity. On the other hand, spermatozoa may continue to move for 3 hours in a normal untreated vagina.⁸
- The motility of the spermatozoa in the specimen may give a clue to their length of stay as they remain motile from 30 to 60 minutes after deposition in the vagina.⁹



¹ O.J. Pollack. 1963 Arch. Pathology 35 p140-184

² Gordon, Turner and Price 1965 *Medical Jurisprudence*

³ Morrison 1972 Brit. J. Vener. Dis 48 p141

⁴Gordon, Turner and Price 1965 Medical Jurisprudence

O.J. Pollack. 1963 Arch. Pathology 35 p140-184

⁶ Weisman 1941 Spermatozoa and Sterility

⁷ Wm.Heinmann Medical Books Ltd 1945 Fertility in Women

⁸ Hamish Hamilton Medical Books 1948 Sterility and Impaired Fertility

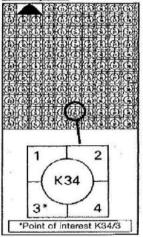
⁹ Gonzales, Vance, Helpern and Umberger 1954 Legal Medicine

11.3 Appendix 3: England Finder Package Insert

The England Finder

The England Finder is a glass slide marked over the top surface in such a way that a reference position can be deduced by direct reading, the relationship between the reference pattern and the locating edges being the same in all finders. The object of the Finder is to give the microscopist an easy method of recording the position of a particular field of interest in a specimen mounted on a slide, so that the same position can be re-located using any other England Finder on any microscope.

Description



The England Finder, a section of which is illustrated, consists of a glass slide 3" x 1" marked with a square grid at 1mm intervals. Each square contains a centre ring bearing reference letter and number, the remainder of the square being subdivided into four segments numbered 1 to 4. Reference numbers run horizontally 1 to 75, and letters vertically A-Z (omitting I). The main locating edge is the bottom of the slide which is used in conjunction with either the left or right vertical edge of the slide, according to the fixed stops of the stage of the microscope, all three locating edges being marked with arrow heads. The label on the finder should always appear visually at the bottom left corner when through most microscopes the reference image will appear correct.

In the illustration (part shown), the point of interest is marked with a cross, and will be seen to lie in the third segment of the square of reference K34, hence the England Reference is K34/3.

Method of Use

- 1. Mark the specimen slide with a label on the left indicating with arrows which sides are to be used for location. Place the slide on the stage of the microscope bringing the bottom long edge in contact with the base stops of the stage and then sliding either left or right into contact with the vertical fixed stops as appropriate. It is important always to obtain the main location of the slide and finder on the base stops first.
- Having examined the specimen in the normal way and found a point of interest, bring this to the centre of the field of view (a crosswire in the eyepiece is useful in this respect).
- 3. Taking care not to alter the position of the fixed stops of the stage, remove the slide and replace with the England Finder, again bringing the bottom edge in contact first and sliding to the appropriate vertical stop, the label of the Finder being at the bottom left corner.
 - 4. The reference pattern of the Finder will now be seen through the microscope (adjusting the focus if necessary). The reference number of the main square is recorded followed by an oblique stroke and the number of the segment in which the centre of the field of view lies (1 to 4 or 0 if in the centre circle). The boundary lines of the main squares are easily distinguishable as these are the only continuous straight lines of the pattern.
 - 5. The reverse procedure is adopted to re-locate the point of interest, The England Finder is placed on the stage as outlined above and the stage is adjusted until the appropriate reference square and segment appear in the centre of the field of view. Remove the finder and replace with the specimen slide with label to left and appropriate vertical slide in contact with the fixed stop, when the point of interest will appear in the centre of the field of view.

11.3.1 England Finder Graticule Use

Before use ensure stage slide holder is in correct position (slide holder should be in the correct position as it is not removed on cleaning but if not-hold with one hand push holder back to full extent against the screws, tighten screws while holding and check for correct positioning).

- Place graticule on stage with labelled corner at LHS front and clear edge against back of slide holder
- 2. Using the 10x objective (and Kohler illumination) locate co-ordinates
- Proceed to 40x objective and adjust focus as required (using oil if applicable). Locate co-ordinates and revert back to the 10x objective.
- Taking care not to alter the position of the fixed stops of the stage, remove the graticule and replace with the slide of interest.
- Proceed stepwise to 40x objective (oil or dry as applicable)
- 6. Adjust focus and locate sperm

NB: If the stage has moved repeat from step 1.



Document Management: 17189 - V9.9H-Fyamination For & Of Spermatozoa

Supposed and Spinos	als Notifications	Comments	Controlled Copies	Version History Associations	Associations
Records Workflow					
Comments By	Comment Date	Response By		Response Date	Comment Noted
Melissa CIPOLLONE 25/0	25/09/2009	Allison LLOYD	29/09/2009		Noted
Kate PIPPIA 03/0	03/09/2008	Allison LLOYD	03/09/2008		Noted
✓ Abigail RYAN 14/:	14/12/2009	Allison LLOYD	15/12/2009		Noted
Comments	14/12/2009 1 Slide prepari suspensions.	14/12/2009 12:01:06 PM Abigail HOULDING: Slide preparation needs amending in the next revision to include making slides from suspensions.	I HOULDING: Iding in the next r	evision to include	making slides fron
Response	15/12/200	15/12/2009 8:29:06 AM Allison LLOVD:	Trovb:		



JH-14

Pathology & Scientific Services Biomedical Technology Services

CHANGE TEMPLATE

PART A: REQUEST FOR CHANGE

Proposal # 31

TITLE: PSA, Sg, salivary amylase and DNA profiling from the one tube

PROPOSED BY: Kate Lee and Adrian Pippia

DATE: 0/01/08

URGENT

Predicted Scope (Mark boxes):

Internal Change

Project

THE PROBLEM: (Issues driving the project / Justification for change: (approx 100 words)

The ability to perform all confirmatory tests and DNA analysis on the one sample tube will ensure minimal sample loss and therefore maximise the potential for obtaining a DNA profile. It is proposed that samples or subsamples can be placed in a 1.5mL tube with extraction buffer and extracted as per the directions for RSID/PSA testing. The enzymes used for these tests remain in the supernatant of the sample while the DNA containing cells are pelleted at the bottom of the tube. The supernatant can therefore be used on the test strips and the remaining sample in the tube be submitted to the analytical section.

This proposal is in conjunction with the change proposals for RSID semen and saliva and PSA testing (#24,#27,#28 and #30). Testing will be performed as part of these proposals.

Forensic Biology	V	Forensic Pathology	Skills Development Unit
- Major Crime	_	- Histopathology	QPS
- Volume Crime	V	- Mortuary	DNA Unit
- Analytical	1	- Coronials	Public Health
- Administrative		FSLU	QHPS
- Operational		Property Point	Canteen
Forensic Toxicology		CSR	
Forensic Chemistry		AUSLAB	

SUBMIT to Quality Representative
For review by Forensic Biology Management

		r.S
RECOMME	ENDATIONS from Forensic Biology Management Team	17/01/08 (Date)
STATUS:	Proposal directed to Minor Change Register Proposal approved to continue Part B planning process Proposal approval to continue Part C planning process (Proposal not approved at this time	
REASON:_	To find a suitable presumptive/ confimatory testing protol amenable to automotion.	Proposed Review Date:

DOCUMENTATION REQUIRED by Forensic Biology	Management Team	(Please mark box):
Internal Change process (Complete Part B, D, E	E) Due:	(date)
Project (Complete Part C, D, E)	△ Due:	(date)
FB Management Representative: Sam CAVE (Print Name)	(Sign)	17 · 01 · 08 (Date)

JH-15



Pathology & Scientific Services Biomedical Technology Services

Proposal #.....24.....

CHANGE TEMPLATE

PART A: REQUEST FOR CHANGE

al second record contract cont	A STATE OF THE STA
TITLE: Change to supplier of PSA test cards	
PROPOSED BY: Allison Lloyd (previously by Kate Lee)	
DATE: 31st March, 2011	URGENT

Predicted Scope (Tick boxes): Internal Change Project

THE PROBLEM: (Issues driving the project / Justification for change: (approx 100 words)

The Princeton BioSign PSA Wb Rapid Test is used within the laboratory to detect PSA in azoospermic semen. These tests cost approximately \$900 for a box of 35. Current usage of these tests is approximately 3-4 boxes per month (105-140 tests) at a cost of approximately \$2700-3600. The ABAcard p30 Test is an alternatively branded PSA test that works on the same principle as the BioSign Test. The ABAcards cost approximately \$205 for a box of 25 (equivalent usage \$861-1148). Changing to the cheaper brand of cards will save the laboratory an estimated \$22068-29424 per year. (KL, RJP 23/04/07)

Updated 19/11/07 by KL and RJP: Early tests show that ABAcard may be more sensitive than the Biosign cards at high dilutions. Want to investigate further with a series of trials with a variety of different testing methods. Data will be used for a comparative report with BioSign. ABAcard and RSID semenogelin (see change proposals for RSID project #28 and BioSign project #27).

Updated 31/03/11 by AKL: The BioSign PSA Wb Rapid Test is no longer available for purchase. Testing performed by KL and RJP have verified that the ABAcard p30 is fit for purpose within the laboratory. Further testing is wanted to optimize parameters for current sampling and analytical procedures. This testing will investigate the best extraction solution, appropriate sample size, extraction/incubation time, a standard procedure to deal with potential high dose hook effect, and continue specificity testing of non-human semen.

STAKEHOLDERS (Tick boxes)				
Forensic Biology	1	Forensic Pathology	Skills Development Unit	
- Evidence Recovery	✓	- Histopathology	QPS	
- FRIT	1	- Mortuary	DNA Unit	
- Analytical		- Coronials	Public Health	
- Administrative		FSLU	QHPS	
- Operational		Property Point	Canteen	
Forensic Toxicology		CSR		
Forensic Chemistry		AUSLAB		

SUBMIT to Quality Representative For review by Forensic Biology Management

Change Prop	osal Te	mplate
-------------	---------	--------

RECOMMENDATIONS from Forensic Biology Manageme	ent Team	(Date://)
STATUS: Proposal directed to Minor Change Regi Proposal approved to continue Part B pl Proposal approval to continue Part C pla Proposal not approved at this time	anning process	
REASON:		Proposed Review Date:
DOCUMENTATION REQUIRED by Forensic Biology	/lanagement Te	eam (Please tick):
☐ Internal Change process (Complete Part B, D, E) OR	Due:	(date)
☐ Project (Complete Part C, D, E)	Due:	(date)
FB Management Representative:		
FB Management Representative:(Print Name)	(Sign)	(Date)
FBMT request completion of Part B (further information	CALL DE LA CALLES	YES NO
PART B: INTERNAL CHANGE PLANNING	Propo	sal #24
Start Date: Wednesday 28th November		
D		
Proposed Implementation Date: 2008		
What is changing? (approximately ½ page) The Princeton Biosign PSA test is routinely used to a ABAcard PSA tests work using the same principle, howe the Biosign kits. This will save the lab a significant amoun	ever, they are a	A STATE OF THE PARTY OF THE PAR
Attachment:		
<u>'</u>		
Who is involved? Change Leader, Change Team and Kate Lee	d Sponsor	
Rhys Parry		
Justin Howes		
Kylie O'Neil Jayde Keating		
VALIDATION PLAN:	Required:	YES NO
Attachment: G:\ForBiol\Change Management\Proposal#2	24\Validation Pla	an ABAcard

DOCUMENTATION AFFECTED:

Document #	Document Title		
17185	Detection of Azospermic Semen in Casework Samples		
17189	Examination For & Of Spermatozoa		
23608	Training Module - Detection of Azospermic Semen using the Biosign PSA Wb Rapid Test for Prostate Specific Antigen		
16003	Training Module Examination of Microscopy Slides for the Detection of Spermatozoa		
24443	Examination of Microscopy Slides for the Detection of Spermatozoa (Level 2 - Interpretation of Findings) Training Module		
16002	Training Module - Acid Phosphatase Screening (Level 1)		
17186	The acid Phosphatase Screening Test For Seminal Stains		
24275	Training Module - Acid Phosphatase Screening Test for Seminal Fluid (Level 2)		

IMPLEMENTATION PLAN:				
Activities Required	Person/Area Responsible	Estimated Completion Date	Quality Officer Confirmation of Completion	
prepare samples for testing	K Lee/R Parry	21/01/08		
Test samples for sensitivity and specificity	K Lee/R Parry	29/02/08	1	
	2			
		8		
	•		and the later hand	

Continue to Parts D and E and then notify Quality Officer or submit electronically Change Leader: (Print Name) (Sign & Date) FBMT request completion of Part C: YES NO PART C: PROJECT PLANNING Proposal #.....24..... 1. Download Project Plan Template (Project Plan – Forensics, QHSS Project Group site on the QHSS intranet.) 2. Project planning / advice can be organized via the Principal Project Manager 3. Completed Project Plans and / or Gantt charts can be submitted to: FB-Projects@health.gld.gov.au 4. The Principal Project Manager will distribute project plans to the Project Control Group for review prior to PCG meetings 5. Depending on the outcomes from the PCG - the PART C proposal may be: Approved. (Proceed to PART D) Not Approved. Undertake further planning and Re-submit Part C by......(date)

(Sign)

Follow-Up Approval(date) (Proceed to PART D)

(Print Name)

PCG Representative:

(Date)

6. Summary of PCG outcomes is shown below:

OUTCOME OF PCG DISCUSSIONS (To be completed by Chair of the PCG)			
DATE	ACTIVITY PCG Meeting	ACTION / RESOURCES ALLOCATED	

Proposal # ...24... PART D: IMPACT & RISK ASSESSMENTS

identifying any impacts to their work-areas.

Major Crime: Comments:	Presented by: Assessed by:	Date: Date:	
Analytical:	Presented by:	Date:	
Comments:	Assessed by:	Date:	
X Volume Crime	Presented by:	Date:	
Comments	Assessed by:	Date:	
Operational:	Presented by:	Date:	
Comments	Assessed by:	Date:	
0	December	Data	
Quality: Comments	Presented by: Assessed by:	Date: Date:	
SDU:	Presented by:	Date:	
Comments	Assessed by:	Date:	
X O H & S:	Presented by:	Date:	
Comments	Assessed by:	Date:	

Change Proposal Template

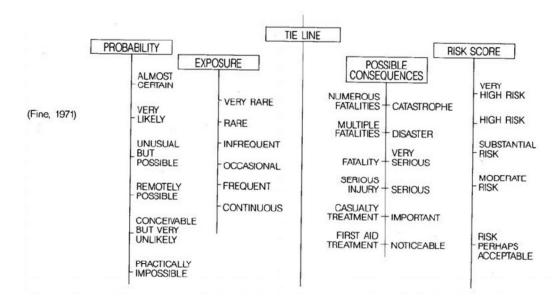
Requisitions: Comments	Presented by: Assessed by:	Date: Date:	
AUSLAB: Comments	Presented by: Assessed by:	Date: Date:	
QPS: Comments	Presented by: Assessed by:	Date: Date:	
FSLU: Comments	Presented by: Assessed by:	Date: Date:	
Property Point: Comments	Presented by: Assessed by:	Date: Date:	
Comments	Presented by: Assessed by:	Date: Date:	
X	Presented by: Assessed by:	Date: Date:	

RISK ASSESSMENT: (To be used if risks of impacts are identified by Stakeholders)

The following form can be used to determine Safety Risks / Risks to a Process, Risks to a Result etc.

Once the risk is identified:

- Determine the probability that the event will occur, mark the probability line at that point
- Determine how often the event is likely to occur, mark the exposure line at that point
- Determine the consequences of this event occurring refer to the comments on the right hand side (the left hand side is generally for safety related risks); mark the possible consequences line at that point.
- Draw a line from the position marked on the probability line to the position marked on the exposure line. Continue the line through to the Tie Line.
- Draw a line from the Tie Line through the position marked on the Possible Consequences line and continue the line through to the Risk Score Line.
- The point at which the line crosses the Risk Score is the Risk. f.



Risks Identified in Ir	mpact Assessments	☐ No ☐ Not Required	
Risk Level:			
Risk Assessment/s	Attached: Yes / N/A		
Risk Assessment u	ndertaken by	Date	
Risk Comments/Justification:			
-			
Chief Scientist Appr	oval:Sign/Date	(Required if Risk =>Mod	erate)
	Signibate		
	To SUBMIT TO FBMT (Internal Change) CLICK HERE	To SUBMIT TO PCG (Projects) CLICK HERE	

PART E: APPROVAL		Propos	al #24
FORENSIC BIOLOGY N	MANAGEMEN	IT APPROVAL (Interr	nal Change)
PCG APPROVAL (Proje	ect Planning)		
Implementation Date:			
		Approved (Circle Applicable)	
Major Crime:			
	(Sign & Date)	YES / NA	
Volume Crime:			
	(Sign & Date)	YES / NA	
Analytical:			
	(Sign & Date)	YES / NA	
Quality Representative:			
	(Sign & Date)		
Principal Project Manager:			
	(Sign & Date)	YES / NA	
Chief Scientist:			
omer colonidat.	(Sign & Date)		
Managar Faranaisas	(Sign & Date)		
Manager Forensics:			
	(Sign & Date)	YES / NA	
PART F: REVIEW:	Pro	posal # 24	
	. 10	P	
Register Updated Docui	ments Activated	☐ Internal Change Filed	Project Filed
Completed by Quality Representative:	(Print Name)	(Sign & Dat	e)





Pathology & Scientific Services **Biomedical Technology Services**

CHANGE TEMPLATE

PART A: REQUEST	FO	R CHANGE		Proposal #28
TITLE: Validation of RSID	sem	enogelin test (alleged confirm	natory	y test for semen)
PROPOSED BY: Kate Lee	•			
DATE: 19th November 200	7			URGENT
Predicted Scope (Tick boxe	s):	Internal Change		✓ Project
THE PROBLEM: (Issues of	Iriving	g the project / Justification for c	hange	e: (approx 100 words)
		elin will be investigated due to ositives) and sensitivity comp		reported greater specificity (thus to PSA detection methods.
While RSID is likely to be a superior system, literature suggests that semenogelin is less stable than PSA and thus breaks down sooner. This will be investigated. If found to be the case, it is foreseen that PSA kits will not be replaced entirely as a detection method but rather they will become a secondary system to be used if RSID fails to detect semen. Hence the need to further validate PSA and the importance of determining the most suitable PSA kit to use as a secondary detection mechanism (see change proposals for BioSign #27 and ABAcard #24)				
STAKEHOLDERS (Tick be	oxes)		
Forensic Biology	V	Forensic Pathology	L	Skills Development Unit
- Major Crime	1	- Histopathology		QPS
- Volume Crime	1	- Mortuary		DNA Unit
- Analytical		- Coronials		Public Health
- Administrative		FSLU		QHPS
- Operational		Property Point		Canteen
Forensic Toxicology		CSR		- Cantoon
Forensic Chemistry		AUSLAB	1	
SUBMIT to Quality Representative For review by Forensic Biology Management RECOMMENDATIONS from Forensic Biology Management Team (Date://) STATUS: Proposal directed to Minor Change Register Proposal approved to continue Part B planning process (Internal Change) Proposal approval to continue Part C planning process (Project Planning) Proposal not approved at this time REASON: Proposed Review Date:				
DOCUMENTATION REQU	IREI	D by Forensic Biology Man	ager	ment Team (Please tick):
☐ Internal Change pro OR	cess	(Complete Part B, D, E)	[Due:(date)
☐ Project (Complete P	art (C, D, E)	[Due:(date)
Authorized by V lentile		Dogo 1 of 0		2207204 444

Proposal	

Change Proposal Template
FB Management Representative:(Print Name) (Sign) (Date)
(Philic Name) (Sign) (Date)
FBMT request completion of Part B (further information required): YES NO PART B: INTERNAL CHANGE PLANNING Proposal #28
Start Date: Wednesday 28 th November
Proposed Implementation Date: 2008
Community of the Commun
What is changing? (approximately ½ page) Currently azoospermic semen is detected by using the Princeton BioSign PSA kits. These kits tests for the presence of prostate specific antigen (PSA). PSA has been found to be present in some other body fluids and therefore there is a risk of false positive results. The RSID tests in this change proposal detect Semenogelin, a protein secreted by the seminal vesicles and epididymis. Together with fibronectin, Semenogelin gives rise to the gel like coagulum of newly ejaculated semen. The RSID test uses two monoclonal antibodies which are specific for human semenogelin. This greater specificity means we are able to more confidently say that, in the absence of spermatozoa, an acid phosphatase positive stain is in fact human semen. The sensitivity and specificity of the RSID semen tests will be examined to determine whether it is appropriate for use within the laboratory.
Attachment:
Who is involved? Kate Lee Rhys Parry Justin Howes Kylie O'Neil Jayde Keating
VALIDATION PLAN: Required: YES NO
Attachment: G:\ForBiol\Change Management\Proposal#28\Validation Plan RSID
DOCUMENTATION AFFECTED: (If change made from PSA to RSID)
Document # Document Title

Document #	Document Title
17185	Detection of Azospermic Semen in Casework Samples
17189	Examination For & Of Spermatozoa
23608	Training Module - Detection of Azospermic Semen using the Biosign PSA Wb Rapid Test for Prostate Specific Antigen
16003	Training Module Examination of Microscopy Slides for the Detection of Spermatozoa
24443	Examination of Microscopy Slides for the Detection of Spermatozoa (Level 2 - Interpretation of Findings) Training Module
16002	Training Module - Acid Phosphatase Screening (Level 1)
17186	The acid Phosphatase Screening Test For Seminal Stains
24275	Training Module - Acid Phosphatase Screening Test for Seminal Fluid (Level 2)

IMPLEMENTATION PLAN:					
Person/Area Estimated Quality Officer Activities Required Responsible Completion Confirmation of					
n.	——————————————————————————————————————		Date	Completion	
	pare samples for testing	K. Lee	07/01/08		
Test samp	oles for sensitivity and specificty	K Lee	18/01/08		
				¥	
		a .	h d		
Contin	ue to Parts D and E and then r Change Leader: Kate Lee)		electronically	
FBMT requ	est completion of Part C:	YES	NO		
PART C:	PROJECT PLANNING		Proposal #	#	
1. Download	Project Plan Template (Project	Plan – Forensics, (QHSS Project Group site or	the QHSS intranet.)	
2. Project pla	nning / advice can be organized	d via the Princip	pal Project Manage	r	
	d Project Plans and / or Gantt ch	arts can be su	bmitted to:		
	pal Project Manager will distribu or to PCG meetings	te project plans	s to the Project Cor	trol Group for	
5. Depending	on the outcomes from the PCG	- the PART C	proposal may be:		
Appr	oved. (Proceed to PART D)				
□ Not A	Approved. Undertake further pla	nning and Re-	submit Part C by	(date)	
Follo	w-Up Approval(date) (Proceed to	PART D)		
PCG Rep	PCG Representative: (Print Name) (Sign) (Date)				
6. Summary	of PCG outcomes is shown below	w:			
0	UTCOME OF PCG DISCUSSIO	NS (To be con	npleted by Chair of	the PCG)	
DATE	ACTIVITY		ACTION / RESOURCE	S ALLOCATED	
	PCG Meeting				

	Change Proposal Template

PART D: IMPACT &	RISK ASSESSMENTS	Proposal #
IMPACT ASSESSMENTS: stakeholders for comment. identifying any impacts to the	Boxes with checks X MUS	der must present the proposal to it be completed by the stakeholders,
Major Crime: Comments:	Presented by: KL Assessed by:	Date:14/1/08 Date:
Analytical: Comments:	Presented by: Assessed by:	Date: Date:
X Volume Crime Comments	Presented by: KL Assessed by:	Date:16/1/08 Date:
Comments	Presented by: Assessed by:	Date: Date:
Quality: Comments	Presented by: Assessed by:	Date: Date:
SDU: Comments	Presented by: Assessed by:	Date: Date:
O H & S: Comments	Presented by: Assessed by:	Date: Date:

Change Proposal Template

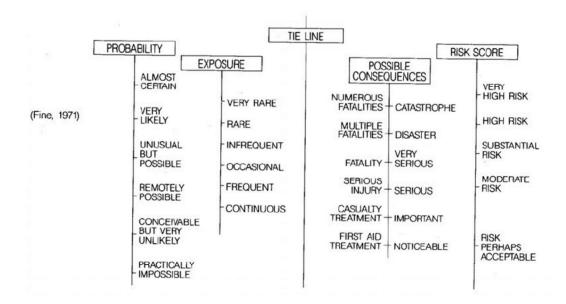
Requisitions: Comments	Presented by: Assessed by:	Date: Date:	
X AUSLAB: Comments	Presented by: Assessed by:	Date: Date:	
QPS: Comments	Presented by: Assessed by:	Date: Date:	
FSLU: Comments	Presented by: Assessed by:	Date: Date:	
Property Point: Comments	Presented by: Assessed by:	Date: Date:	
X	Presented by: Assessed by:	Date: Date:	
X: Comments	Presented by: Assessed by:	Date: Date:	

RISK ASSESSMENT: (To be used if risks of impacts are identified by Stakeholders)

The following form can be used to determine Safety Risks / Risks to a Process, Risks to a Result etc.

Once the risk is identified:

- Determine the probability that the event will occur, mark the probability line at that point
- Determine how often the event is likely to occur, mark the exposure line at that point
- Determine the consequences of this event occurring refer to the comments on the right hand side (the left hand side is generally for safety related risks); mark the possible consequences line at that point.
- Draw a line from the position marked on the probability line to the position marked on the exposure line. Continue the line through to the Tie Line.
- Draw a line from the Tie Line through the position marked on the Possible Consequences line and continue the line through to the Risk Score Line.
- The point at which the line crosses the Risk Score is the Risk. f.



Risks Identified in Impact Assessments □Yes □ No □ Not Required	
Risk Level:	
Risk Assessment/s Attached: Yes / N/A	
Risk Assessment undertaken by	
Risk Comments/Justification:	
Chief Scientist Approval: (Required if Risk =>Modera	te)

To SUBMIT TO FBMT (Internal Change) **CLICK HERE** FB-Change@Health.qld.gov.au

To SUBMIT TO PCG (Projects) **CLICK HERE** FB-Projects@health.qld.gov.au)

PART E: APPROVAL		Propos	al #	
FORENSIC BIOLOGY MANAGEMENT APPROVAL (Internal Change)				
PCG APPROVAL (Proje	ect Planning)			
Implementation Date:				
		Approved (Circle Applicable)		
Major Crime:				
	(Sign & Date)	YES / NA		
Volume Crime:				
	(Sign & Date)	YES / NA		
Analytical:				
	(Sign & Date)	YES / NA		
Quality Representative:				
	(Sign & Date)			
Principal Project Manager:	, ,			
- Timo par Troject manageri	(Sign & Date)			
	(Sign & Date)	TEO / IVA		
Chief Scientist:				
	(Sign & Date)			
Manager Forensics:				
	(Sign & Date)	YES / NA		
PART F: REVIEW: Proposal #				
☐ Register Updated ☐ Docu	ments Activated	☐ Internal Change Filed	Project Filed	
Operated to Operate State Control of the Control of				
Completed by Quality Representative:	(Print Name)	(Sign & Date	e)	

JH-17



Pathology & Scientific Services Biomedical Technology Services

CHANGE TEMPLAT	ſΕ			
PART A: REQUEST	FOR CHANGE	Proposal #27		
TITLE: Further validation	of BioSign PSA cards and refi	nement of standard operating procedure		
PROPOSED BY: Rhys Pa	arry and Kate Lee			
		UD OF UZ		
DATE: 19 th November 20		URGENT _		
Predicted Scope (Tick box	res): ✓ Internal C	hange Project		
THE PROBLEM: (Issues	driving the project / Justification	for change: (approx 100 words)		
absence of sperm is being relying on the literature for cross-reactivity and washing our case load means that near future due to the factorial relationship.	ing challenged/questioned more theoretical knowledge of PS ing of exhibits). How much be transfer based PSA determinent that most cases are not resources reactivity, determination	e mechanism of detection of semen in the ore vigorously. At present we are largely A in a wide variety of situations (especially etter to be able to quote in-house studies? nations will be relevant for the foreseeable olved through to trial for at least two years. of optimal elution volumes, and direct vs		
Additionally, the SOP states that samples are to be soaked in approximately 50-100uL water and then 70uL are to be removed for testing (obviously this doesn't work when only 50 was added in the first instance) - but there is no justification for these amounts. Other labs use higher volumes which increase convenience at the expense of over dilution of the PSA to sub-detection concentrations. Concentration of elution fluid minimises false negatives but increases the risk of DNA loss. We need to determine a good medium. Also with implementation of DNAIQ, large elution volumes will no longer be possible thus we will need to modify our sampling technique to maximize DNA recovery while still maintaining a method of reliably detecting PSA/Seminal presence. The sooner this is undertaken, the sooner we can develop a suitable work flow for the SOP which for all intents and purposes will become defunct with the modification of our techniques. Data will be used for a comparative report with BioSign, ABAcard and RSID semenogelin.				
STAKEHOLDERS (Tick I	boxes)			
Forensic Biology	1 1	Skills Development Unit		
- Major Crime	✓ Forensic Pathology ✓ - Histopathology	QPS		
- Volume Crime	✓ - Mortuary	DNA Unit		
- Analytical	✓ - Coronials	Public Health		
- Administrative	FSLU	QHPS		
- Operational	Property Point	Canteen		
Forensic Toxicology	CSR			

SUBMIT to Quality Representative
For review by Forensic Biology Management

AUSLAB

RECOMMENDATIONS from Forensic Biology Management Team	(Date: 23/11/07)
STATUS: Proposal directed to Minor Change Register	

Forensic Chemistry

			Change Pro	posal Template
	Proposal approved to continue Part B pla Proposal approval to continue Part C plan Proposal not approved at this time			
REASON:	-house studies / validation	10	Proposed R	teview Date:
	literature.			
DOCUMENTA	TION REQUIRED by Forensic Biology M	anagement Te	eam (Please	tick):
Internal OR	Change process (Complete Part B, D, E)	Due:	6/12/07	(date)
	(Complete Part C, D, E)	Due:		(date)
FB Manage	ment Representative: V (entile (Print Name)	(Sign)	23.1	(Date)
	t completion of Part B (further information re	equired):	YES	NO
PART B: I	NTERNAL CHANGE PLANNING	Propo	sal #	
Start Date:				
Proposed Imp	lementation Date:			
торооси ппр	nomentation bate.			
What is chang	ying? (approximately ½ page)			
Attachment:				
Who is involve	change Londer Change Teem and	Choncor		
who is involve	ed? Change Leader, Change Team and	Sponsor		
VALIDATION F	PLAN:	Required:] YES	□ NO
Attachment:				
DOCUMENTA	TION AFFECTED:			
Document#	Docume	ent Title		

CaSS Forensic and Scientific Services

Project #78 Verification of ABA cards p30

Report - Verification of ABA cards p30 test.

Allison Lloyd, Adrian Pippia and Thomas Nurthen, Kirsten Scott DNA Analysis Unit, Forensic and Scientific Services

Abstract

An alternative PSA/p30 presumptive test is required for use in the DNA Analysis Laboratory as the current test is being discontinued by the manufacturer. The ABA card p30 test kit was trialled for use against the BioSign WB PSA test with concordant results. Further experimentation on specificity and sensitivity for the ABA card was performed to modify the manufacturer's recommended procedure to provide optimal results within the laboratory. It was determined that Nanopure water was a suitable and stable extraction medium for the test and that the kit showed sufficient sensitivity required for case work purposes. The high dose hook effect was apparent in the neat and some of the 1 in 4 dilutions. However, samples diluted to either 1 in 10 or 1 in 100 are shown to remove any high dose hook effects from affected samples. The use of the ABA card p30 test has been shown to be an acceptable alternative to the BioSign WB PSA test and is suitable for routine use in the DNA Analysis Laboratory.

Background

Prostate Specific Antigen or p30 is a protein produced in the cells of the human prostate gland and is a widely used biomarker for the detection of the possible presence of seminal fluid in forensic cases. P30 cannot conclusively identify the presence of semen as the protein has been detected in other tissues and bodily fluids such as urine and breast milk, however the concentration of p30 in human semen far exceeds those of other tissues and is a useful indicator of the presence of semen. As p30 is expressed independently of spermatozoa, p30 is a useful indicator of semen from vasectomised or azoospermic males.

The test kits used for p30 detection rely on an immunoassay sandwich reaction between p30 in a sample and p30 antibodies in the test strips. The ABA card device has two result areas within the device window; the control "C" area and the test "T" area.

On completion of the test, a pink line in the "C" area is a positive control pass, and indicates that the test is functional. On test completion a pink line in the "T" test result area - is a positive test result i.e. presumptive positive for semen. The absence of a pink line in the "T" test area - is a negative test result i.e. presumptive negative for semen. When there is a faint pink line in the "T" test area (less intense than the "C" line), the result might be described as having a 'weak positive' result. For valid use of the ABA card test, the kits positive control line must be apparent on completion of the test, and the test must not be used after the expiration date. If there is no pink line visible in the "C" control area of the test, or it is a partial line, it is inconclusive and the test should be repeated.

The high dose hook effect occurs when the measured levels of antigen displays a significantly lower absorbance than actual concentration present in a sample. When an assay is saturated by a very high concentration of the p30 antigen binding to all available sites on both the solid phase antibody as well as the detection antibody, the sandwich formation is prevented. The saturated detection antibodies are washed off giving a false negative or low positive result.



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Project #78 Verification of ABA cards p30

3. Purpose and Scope of testing

The current testing method for the detection of PSA/p30 of suspected semen samples relies on the BioSign Wb PSA product which is being discontinued by the manufacturer and a suitable replacement is required. The ABA card p30 test by Abacus Diagnostics is a similar test, which uses the same principles of function and has been tested previously alongside the BioSign kit with concordant results in 2007 as part of Project #24. This verification aims to determine the optimal conditions of the ABA card kit by testing against samples that mimic routine case work items, to test sensitivity, and the propensity of the kit to show high dose hook effect. If the kits are shown to be prone to the high dose hook effect, appropriate semen dilutions will be assessed to ensure this effect can be negated.

Equipment and Materials

- ABA card p30 test kits (Abacus Diagnostics, USA) batch #23200702, exp April 2012 and #23200921 exp August 2012
- 3 x normal neat human semen samples
- 2 x azoospermic neat human semen samples from vasectomised donors
- Extraction medium determined in Experiment 4.1 known hereafter as Extraction Medium X
- Nanopure Water
- PBS 10x
- TE buffer prepared as per Receipt, Storage & Preparation of Chemicals, Reagents & Kits QIS Document no. 17165v8.
- Swabs (plastic applicator, rayon tipped) (Copan)
- 1.5ml tubes (Simport)
- Pasteur pipettes sterile (Copan, Italy)
- 200µl pipette (eppendorf, Germany) and tips (ART 200G Molecular BioProducts)
- Centrifuge (Quantum Scientific)
- Vortex (Molecular Solutions)
- Digital timer
- Measuring cylinders
- 50mL flasks/jars
- Recently laundered fabric substrates denim, cotton, satin-like, towelling, carpet
- Sellotage sticky tape for tape lifting
- Cat semen sample
- Bull semen sample
- Pig semen sample
- Possum semen sample
- Kangaroo semen sample
- Koala semen sample
- Data compiled as part of Project #24 using ABA card p30 test kits (Abacus Diagnostics, USA) batch #23260211 exp. Oct 2007

5. Methods

5.1. Semen Collection

Semen was collected from normal healthy males. Donors were instructed to abstain from ejaculation for 3-5 days. Ejaculate was collected in sterile 50mL tubes and stored at 4°C prior to use.

5.2. PBS dilutions

1. Diluted a portion of 10xPBS solution by 1/10 with Nanopure water by adding 600µL 10xPBS to 5400µL Nanopure water to make 1xPBS.



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Project #78 Verification of ABA cards p30

- Prepared 1/10 dilution of normal neat semen in 1xPBS by adding 60μL semen to 540μL 1xPBS.
- 3. Deposited 100µL of diluted semen onto 1 x swab head, allowed to dry. Cut cotton off swab head and placed into 1.5mL tube.
- Added 250μL of 1xPBS solution. Vortexed for 5s whilst rotating tube. Allowed to stand for 10 min then centrifuged for 3 min at 14000 rpm.

5.3. Nanopure water dilutions

- Prepared 1/10 dilution of normal neat semen in Nanopure water by adding 60µL semen to 540µL water.
- 2. Deposited 100µL of diluted semen onto 1 x swab head, allow to dry. Cut cotton off swab head and placed into 1.5mL tube.
- 3. Added 250µL Nanopure water. Vortexed for 5s whilst rotating tube. Allowed to stand for 10 min then centrifuged for 3 min at 14000 rpm.

5.4. TE buffer dilutions

- Prepared 1/10 dilution of normal neat semen in TE buffer by adding 60µL semen to 540µL TE buffer.
- 2. Deposited 100µL of diluted semen onto 1 x swab head, allowed to dry. Cut cotton off swab head and placed into 1.5mL tube.
- 3. Added 250µL TE buffer. Vortexed for 5s whilst rotating tube. Allowed to stand for 10 min then centrifuged for 3 min at 14000 rpm.

5.5. Sample preparation for high dose hook effect

The extraction mediums to be tested were PBS, TE buffer and Nanopure water. The extraction medium (Nanopure water) and preparation times (10 minutes) used for this preparation was determined in Experiment 6.1.

- Deposited 100µL of neat semen from each donor onto 1 x swab, 1 x tape lift, 2 x
 pieces of fabric of each type (denim, cotton, satin-like, towelling, carpet). Allowed to
 dry.
- Prepared suspensions of swabs and tape lifts in 1.5mL tubes by adding 250µL of Nanopure water. Vortexed for 5 seconds whilst rotating tubes. Allowed to stand for 10 mins. Centrifuged for 3 min at 14000 rpm.
- For each semen donor, selected 1 of each fabric type and excised stained area, cut into pieces and prepared suspension by adding 250µL of Nanopure water. Vortexed for 5 seconds whilst rotating tube. Allowed to stand for 10 min. Centrifuged for 3 min at 14000 rpm.
- 4. For each semen donor, selected the remaining pieces of fabric and scraped the stained area. Created suspensions from the scrapings in 1.5mL tubes by adding 250µL of Nanopure water. Vortexed for 5s whilst rotating tubes. Allowed to stand for 10 mins. Centrifuged for 3 min at 14000 rpm.
- 5. Suspensions created in steps 2-4 are now considered neat semen suspensions.
- 6. Prepared dilutions from the neat semen suspensions with Nanopure water: 1/4, 1/10, 1/100, 1/1000, 1/10000, 1/50000.



Project #78 Verification of ABA cards p30

5.6. Animal semen sample preparation

The extraction medium (Nanopure water) used for this preparation was determined in Experiment 6.1.

- 1. 250µL of Nanopure water was added to a 100µL sample of neat semen from each animal. Vortexed for 5 s whilst rotating tube. Allowed to stand for 10 min then centrifuged for 3 min at 14000 rpm. This is now known as the neat semen suspension.
- Prepared 1/10 and 1/100 dilutions from the neat semen suspensions of each animal in Nanopure water.

6. Experimental Design

6.1. Investigation into Extraction Buffer and Extraction Time

This experiment was to determine the best extraction medium (Nanopure water, 1xPBS or TE buffer) and the time required for extraction of p30 into solution. The inclusion of the 18 hr test time ensures that the p30 test will give a positive result if the test is unable to be performed until the following day as can occur in current case work circumstances.

- 1. Added 150µL of suspension (Nanopure water, PBS or TE) to ABA card test well kit and read result after 10 min as per the manufacturer's instructions.
- Changed extraction time or standing time intervals in 5.1 5.3 to 2 min, 30 min, 1 hr, 2 hr, 4 hr and 18 hr.

6.2. Investigation into sensitivity and the high dose hook effect

The ABA card kit has a documented p30 sensitivity of 4ng/ml. Normal seminal fluid has a p30 concentration between 2.0 x 10⁵ and 5.5 x 10⁶ ng/mL. Due to the high level of p30 in semen compared with blood (for which the kit was designed), false negative results may occur due to the high dose hook effect. This happens when an excessive number of the p30 molecules inhibit the binding of the mobile p30-antibody complexes to the immobilised antibodies in the test area. In effect, an overloaded sample will give a weakened positive reaction or no reaction at all. This validation assessed the sensitivity of the ABA card p30 test by using dilutions of semen on different substrates to investigate the high dose hook effect, and determine a suitable dilution factor. This dilution is to be included in the standard operating procedure where this effect is suspected.

 Deposited 150µL of each diluted sample (neat, 1/10, 1/100, 1/1000, 1/10000, 1/50000) from each donor into ABA card test wells. Waited for 10 min and read result as per the manufacturer's instructions.

6.3. Specificity using Animal Semen

It is well documented that p30 is not just found in human seminal fluid but in other tissues such as urine and breast milk. As part of Project #24 in 2007, previous testing of bodily fluids and other reagents and solutions generally found in the laboratory have given negative results with the ABA card p30 test kits with exception of male urine.

Table 1. Specificity samples run on ABA card p30 kits. Data compiled by KSL 2007. (Unpublished)

Substrate	p30 result at 10 minutes
Male Urine 1	+
Male Urine 1 diluted 1/10	+
Male Urine 1 diluted 1/100	-
Male Urine 1 diluted 1/1000	-
Male Urine 1 diluted 1/10000	-



Project #78 Verification of ABA cards p30

Male Urine 2	-
Female Urine 1	
Female Urine 2	-
Female Urine 3	-
Saliva 1	-
Saliva 2	-
Saliva 3	-
Saliva 4	
Saliva 5	
Sweat 1	- A
Sweat 2	- 4
Sweat 3	• (2.4)
Breast Milk 1	• X-2
Breast Milk 2	- #:
Faeces (Female) 1	<u> </u>
Faeces (Female) 2	. 32.1
Faeces (Female) 3	<u>-</u>
Mediwipes (direct application to test kit)	
Mediwipes solution (neat)	<u> </u>
Water	•
Bleach	- 1000 Wind
Ethanol	- L

This verification will expand previous research by conducting species specificity testing on samples of animal origin: cat, bull, pig, possum, koala, and kangaroo.

 Deposited 150µL of each of the neat suspension, 1/10 dilution and 1/100 dilution for each animal into ABA card test kit wells and read results after 10 min as per the manufacturer's instructions.

Results and Discussion

7.1 Casework samples run on ABA Card p30 kits alongside Biosign PSA kits

As part of Project #24 in 2007, 50 casework samples were run concurrently on the Biosign PSA kits and the ABA card p30 kits. For each sample, the results from both tests were identical. Note: weak positive results refer to a positive line in the 'T' area of the kit that is of less intensity than the control line.

		PSA Positive Results				
QPS Number	Exhibit Barcode	Item Description	Location of Stain	AP Result	Microscopy	PSA (Blosign/ABA card)
d		SAIK	Low vaginal swab	positive @ 60s	nil sperm seen	pos/pos
(SAIK	Vulval swab	positive @ 60s	nil sperm seen	pos/pos
(underwear	inside crotch	positive @ 20s	nil sperm seen	pos/pos
q		negligee	inside crotch	positive @ 30s	nil sperm seen	pos/pos
		underwear	o/s front RHS leg seam	positive @ 60s	nii sperm seen	pos/pos
		underwear	o/s front mid crotch area	positive @ 50s	nil sperm seen	pos/pos
		underwear	o/s back mid region	positive @ 60s	nil sperm seen	pos/pos



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	underwear	o/s back crotch region	positive @ 35s	nii sperm seen	weak pos/weak pos
_	g-string	i/s front	positive @ 18s	nil sperm seen	pos/pos
	SAIK	Vulval swab	positive @ 30s	nil sperm seen	weak pos/weak pos
	underwear	i/s front above gusset	positive @ 10s	nil sperm seen	weak pos/weak pos
	underwear	i/s front above gusset left seam	positive @ 10s	nii sperm seen	pos/pos
	underwear	i/s front above gusset	positive @ 10s	nil sperm seen	pos/pos
	underwear	i/s front gusset	positive @ 10s	nil sperm seen	pos/pos
	towel	area 2	n/a	nil sperm seen	weak pos/weak pos
	underwear	i/s crotch	positive @ 15s	nil sperm seen	pos/pos
	PSA Negative Results				
	SAIK	vaginal swab	positive @ 55s	nil sperm seen	neg/neg
	SAIK	vulval swab	positive @ 60s	nil sperm seen	neg/neg
	underwear	o/s front	positive @ 60s	nil sperm seen	neg/neg
	underwear	i/s crotch	positive @ 40s	nil sperm seen	neg/neg
	underwear	i/s back	positive @ 40s	nil sperm seen	neg/neg
	g-string	i/s front	positive @ 18s	nil sperm seen	neg/neg
	denim skirt	o/s front	positive @ 34s	nil sperm seen	neg/neg
	denim skirt	o/s front	positive @ 34s	nil spem seen	neg/neg
	denim skirt	o/s back	positive @ 40s	nil sperm seen	neg/neg
	denim skirt	i/s front	positive @ 45s	nil sperm seen	neg/neg
	underwear	l/s crotch	positive @ 1m 30s	nil sperm seen	neg/neg
		l/s crotch along left			
	underwear	seam	positive @ 1m 30s	nil sperm seen	neg/neg
-	SAIK	Low vaginal swab	positive @ 1m 30s	nil sperm seen	neg/neg
	SAIK	Vulval swab	positive @ 1m 50s	nil sperm seen	neg/neg
	SAIK	Low vaginal swab	positive @ 1m 20s	nil sperm seen	neg/neg
	SAIK	high vaginal swab	positive @ 55s	nil sperm seen	neg/neg
-	SAIK	Vulval swab	positive @ 60s	nil sperm seen	neg/neg
	SAIK	high vaginal swab	weak positive	nil sperm seen	neg/neg
	\$AIK	Low vaginal swab	weak positive	nil sperm seen	neg/neg
	SAIK	Vulval swab	weak positive	nil sperm seen	neg/neg
	SAIK	Low vaginal swab	weak positive	nil sperm seen	neg/neg
	SAIK	Vulval swab	weak positive	nil sperm seen	neg/neg
	g-string	front o/s crotch	positive @ 1m 20s	nil sperm seen	neg/neg
	g-string	front i/s crotch	positive @ 1m 10s	nil sperm seen	neg/neg
-	towel	area 1	n/a	nil sperm seen	neg/neg
	towel	area 3	n/a	nil sperm seen	neg/neg
	SAIK	perianal swab	weak positive	nil sperm seen	neg/neg
	SAIK	high vaginal swab	weak positive	nil sperm seen	neg/neg
	SAIK	Low vaginal swab	weak positive	nil sperm seen	neg/neg
	SAIK	sanitary pad	positive	n/a_	neg/neg
	underwear	i/s front	positive @ 1m 20s	nil sperm seen	neg/neg
	fitted sheet	area 3	positive @ 30s	nil spem seen	neg/neg
	SAIK	perianal swab	positive @ 1m 30s	nil spem seen	neg/neg
	SAIK	rectal swab	positive @ 1m 35s	nil sperm seen	neg/neg



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Project #78 Verification of ABA cards p30

7.2 Determination of optimal volume for use in the ABA card p30 kits

As part of Project #24 in 2007, experiments were conducted to determine the optimal volume of sample to be deposited in the ABA card p30 test kit well to obtain a result whilst limiting the potential loss of DNA by dilution of the sample in forming a suspension. The ABA card manufacturer's instructions recommend using a larger sample volume (150µL) than is required for the Biosign PSA kits (30µL) due to the lack of 'running' buffer'.

7.2.1 Sample Preparation and Experimental Design

- Neat semen was diluted with Nanopure water to give dilutions of 1/10, 1/100, 1/1000, 1/10000 and 1/50000.
- Swabs and pieces of cotton and polyester/cotton were loaded with different sample volumes (Refer to Table 3) from neat semen and from each of the dilutions. All substrates were allowed to dry.
- 3. A range of volumes (Refer to Table 3) of Nanopure water was added to each sample to extract the p30 from the sample, vortexed and allowed to stand for 10 mins.
- 4. Each sample was centrifuged at 14000 rpm for 3 mins.
- The test volume was removed from the sample and deposited in the ABA card p30 test kit wells. Results were read after 10 mins.

7.2.2 Results

It was determined that the ABA card p30 test kit required a minimum of 150 µL of test suspension in the test well in order to enable the capillary action of the kit and for the solution to reach the testing areas on the membrane. This volume is much higher than the one or two drops required for the Biosign PSA kits. However the Biosign kits include a 'running' buffer to ensure the capillary action.

As a result of this experiment, a test volume of 150 µL was used in the 2011 experiments in Section 6.

Table 3. Extraction Volumes and (Sample Volumes) per dilution. Results compiled by KSL/RJP, 2007. Unpublished)
(+ is positive result, - is negative result, W+ is weak positive result all at 10 mins)

SWABS				**		
	Neat	1/10	1/100	1/1000	1/10000	1/50000
150 μL (100 μL sample)	-	+	+	W+	-	•
200 μL (100 μL sample)	•	+	+	W+	-	-
250 μL (100 μL sample)	-	+	+	W+	- 8	
300 μL (100 μL sample)	-	+	W+	-	- 38	
300 μL (200 μL sample)	-	+	+	W+	-	
350 µL (200 µL sample)	-	+	+	W+		
250 µL (100 µL sample)	W+	+	+	-	• 6	-
250 μL (200 μL sample)	•	+	+	-	- #	
300 μL (100 μL sample)	W+	+	+	-	-	-
300 μL (200 μL sample)	-	+	+	-	-	<u>.</u>
COTTON						
	Neat	1/10	1/100	1/1000	1/10000	1/50000
100 μL (50 μL sample)	W+	+	+	-	-	**:-
150 μL (50 μL sample)	W+	+	+	-	-	_
150 μL (100 μL sample)	W+	+	+	_		•
200 μL (100 μL sample)	W+	+	+	-	-	, i =
250 μL (100 μL sample)	W+	+	+	-	-	
250 μL (200 μL sample)	-	+	+	_		

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300 μL (100 μL sample)	W+	+	+	-	-	
300 μL (200 μL sample)	-	+	+	-	•	
SATIN						
	Neat	1/10	1/100	1/1000	1/10000	1/50000
100 μL (50 μL sample)	+	+	W+	-		
150 μL (50 μL sample)	+	+	_			
150 μL (100 μL sample)	+	+	W+	- 4		-
200 μL (100 μL sample)	+	+	W+	•		-
250 µL (100 µL sample)	+	+				
250 μL (200 μL sample)	+	+	W+		•	
300 μL (100 μL sample)	+	+	-	-	•	
300 μL (200 μL sample)	+	+	W+	- ·		_

7.3 Investigation into Extraction Buffer and Extraction Time

Samples were made by depositing 100 µL of 1:10 diluted semen onto swab heads. The diluted semen was made as per Section 5.2-5.4. There appeared to be no difference between results with the use of different extraction buffers or extraction time. Nanopure water was selected as the preferred extraction medium due to cost and availability and was used throughout the remainder of the experiments in Sections 5 and 6. An extraction time of 10 minutes was selected as the Extraction Time in Sections 5 and 6 due to convenience. The results of the different extraction buffers and time are in Table 4.

Table 4. Determination of Optimal Extraction Medium and Extraction Time. (+ is positive p30 result at 10 mins)

Extraction Medium	2 min	10 min	30 min	1 hr	2 hr	4 hr	18 hr (4pm - 9am)
Nanopure water	+	+	+	+	+	#	+
1xPBS	+	+	+	+	+	+	+
TE buffer	+	+	+	+	+	+	+

7.4 Investigation into Sensitivity and the 'High Dose Hook Effect'

The results in Table 5 shows that neat semen extracted from different substrates and tested by the ABA card p30 test has the potential to demonstrate the High Dose Hook Effect. Although only one false negative was recorded with a tape lift of neat semen, the large numbers of weak positive results indicate that the effect was still occurring in the most of the neat and 1:4 dilution samples. Results show that the 1:10 diluted samples provided the highest number of positive results with the lowest number of weak positives for all fabric and sampling types. However for swabs and tape lifts, a dilution factor of 1:10 gave positive results though there is evidence that the hook effect was still occurring. The 1:100 dilutions gave the best results for swabs and tape lifts of semen stains.

The first true negative results were seen in the 1:100 dilutions of scraped fabric stains indicating that scraping does not recover nearly as much p30 (and thereby possibly DNA) as excision of stains. It appears that a dilution of 1:10 is the best possible dilution (of the dilutions tested) to provide a positive result if the high dose hook effect is suspected. If a negative result occurs and the effect is still suspected, a second dilution of 1:100 is recommended.



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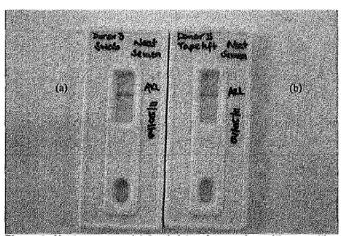


Figure 1. Neat semen on (a) Swab (showing weak positive result) and (b) Tape lift (showing a false negative result at 10 mins.



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Table 5. Sensitivity testing (+ is positive p30 result, - is negative p30 result, W+ is weak positive p30 result all at 10 mins)

										.93		165
Neat	Swab	Tape lift	Carpet Excised	Carpet Scraped	Cotton Excised	Cotton Scraped	Satin Excised	Satin Scraped	Denim Excised	Denim Scraped	Towel Excised	Towel Scraped
Donor 1	W+	W+	W+	W+	W+	+	W+	+	W+	*	W+	+
Donor 2	W+	W+	+	W+	W+	+	W+	+	W+	+	W+	+
Donor 3	W+	-	W+	W+	W+	W+	W+	W+	W+	+	W+	W+
Donor 4	+	+	+	+	+	+	+	+	W+	+	+	+
Donor 5	W+	W+	W+	W+	+	+	+	W+	W+	+	W+	+
1:4			Carpet	Carpet	Cotton	Cotton	Satin	Satin	Denim	Denim	Towel	Towel
dilution	Swab	Tape lift	Excised	Scraped	Excised	Scraped	Excised	Scraped	Excised	Scraped	Excised	Scraped
Donor 1	+	W+	W+	+	+	+	+	+	+	+	+	+
Donor 2	W+	W+	+	W+	+	+	+	+	W+	+	+	+
Donor 3	W+	W+	+	+	+	+	+	+ **	W+	+	W+	+
Donor 4	W+	W+	+	+	+	+	+	+	+	-2007 +	W+	+
Donor 5	W+	W+	W+	+	+ O-#	+	+	+	+	+	+	+
1:10 dilution	Swab	Tape lift	Carpet Excised	Carpet Scraped	Cotton Excised	Cotton Scraped	Satin Excised	Satin Scraped	Denim Excised	Denim Scraped	Towel Excised	Towel Scraped
Donor 1	+	+	+	+	+	W+	+	+	1	+	+	+
Donor 2	+	+	+	+	+	+	+	+	1	+	+	+
Donor 3	W+	+	+	+	+	+	+	+	•	+	. +	W+
Donor 4	+	+	+	+	+	+	+	+	•	+	+	W+
Donor 5	W+	W+	W+	+	+	+	+	+	+	+	+	+
1:100 dilution	Swab	Tape lift	Carpet Excised	Carpet Scraped	Cotton Excised	Cotton Scraped	Satin Excised	Satin Scraped	Denim Excised	Denim Scraped	Towel Excised	Towel Scraped
Donor 1	+	+	+	+	+	-	W+		+		**************************************	W+
Donor 2	+	+	+	+	+	+	+	W+	+	W+	+	+
Donor 3	+	+	W+	+	+	W+	W+		+	-	. +	-
Donor 4	+	+	+	+	+	+	+	+	W+	- &	+	W+
Donor 5	+	+	+	+	+	+	+	-	+		+	-
1:1000 dilution	Swab	Tape lift	Carpet Exclsed	Carpet Scraped	Cotton Excised	Cotton Scraped	Satin Excised	Satin Scraped	Denim Excised	Denim Scraped	Towel Excised	Towel Scraped
Donor 1	W+	W+	W+		_	-	-	-	-	- ****		_
Donor 2	W+	+	W+	+	_	-	W+	-	-		W+	
Donor 3	W+	+	-	-	-	_	-	-	-	- 9		
Donor 4	W+	W+		-	-	-	-	· .	-	- 🥳		6536500
Donor 5	-	W+	W+	-	_	-	-	-	_	- %	*	_
1:10000 dilution	Swab	Tape lift	Carpet Excised	Carpet Scraped	Cotton Excised	Cotton Scraped	Satin Excised	Satin Scraped	Denim Excised	Denim Scraped	Towel Excised	Towel Scraped
Donor 1	-			_			-				Market 1	-
Donor 2	-		_	-	_							<u> </u>
Donor 3		-	-	-			-	-		-		
Donor 4		-	-	_	_	-	-	_	_	-		A
Donor 5	-	-	-	-	_	_	-		-		-	
		-									7.7	A STATE
1:50000 dilution	Swab	Tape lift	Carpet Excised	Carpet Scraped	Cotton Excised	Cotton Scraped	Satin Excised	Satin Scraped	Denim Excised	Denim Scraped	Towel Excised	Towel Scraped
Donor 1	-	-	-		-	-	-	-		-	•	
Donor 2	-	•	-	-	-	-	-		-	-		- Aller -
Donor 3	-	-	-	-	-	_	-	-	-	_		
Donor 4		-	-	-	-	-	-	_	-	- :::	11.5° <u>-</u>	
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7.4 Investigation into specificity using animal semen

Semen samples of different animal species were tested using the ABA card p30 test kits to determine whether the kits were human specific. Results (Refer to Table 6) for all animals were negative, indicating that animal semen should not interfere with interpretation of results.

Table 6. Specificity using animal semen. - refers to negative p30 result at 10 mins. Neat samples of Possum and Koala semen were not available for testing, aliquots of previously diluted semen used.

Animal	Neat	1/10	1/100
Kangaroo	-	-	
Possum	N/A	-	
Koala	N/A	-	-
Cat	-	-	-
Bull	-	-	-
Pig	-	-	-

Conclusions and recommendations

This validation has determined that the ABA card p30 test kits using the modified method of extracting into 200 µL Nanopure water for 10 mins are suitable for routine case work samples within the DNA Analysis Laboratory for the detection of the possible presence of semen. An extraction into 200 µL allows for the initial test using 150 µL, leaving 50 µL available for dilution.

Human male urine is known to show cross-reactivity with the ABA card p30 test however the levels of p30 in human semen is significantly higher that urine should not considerably affect result interpretation.

The high dose hook effect is a possible issue that may affect result interpretation. It is recommended that clear information is obtained from Queensland Police Service about possible semen sample submissions, such as times for positive presumptive AP screenings or whether a sample may be neat semen. This way, informed decisions can be made regarding sampling techniques, result interpretation and the potential requirement for dilutions of samples in order not to miss positive results.

With the high dose hook effect in mind, it is also recommended that, if the effect is suspected, an initial dilution factor of 1:10 be utilised and a possible second dilution of 1:100 to be considered if necessary. Samples that produce strong acid phosphatase reactions, i.e. less than 20 secs and a negative ABA card result should be diluted and retested. The faster the acid phosphatase reaction, the greater the possibility that the high dose hook effect has occurred.

Another consideration is that each sample will require an addition of a minimum of 200 µL Nanopure water in order to perform the test and to allow for possible dilutions. This will also dilute the DNA within each sample. Nevertheless p30 results can give important information to aid in interpreting results for case work samples and it is recommended that test be implemented while bearing this in mind.

Whilst considering the above, it is recommended that the ABA card p30 test kits be introduced in the DNA Analysis Laboratory as a replacement for the PSA Biosign kits. An added benefit of introducing the ABA card kits is the potential monetary savings. At \$7.50 per test, even performing two tests in the case of dilutions using the ABA card kits is less expensive than the Biosign kits at approximately \$30 per test.



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References

- 1. ABA card® p30 Test for the Forensic Identification of Semen, 2001, Technical Information Sheet, Abacus Diagnostics
- 2. Validation of the Onestep ABA card p30 Test Card for the Detection of p30 or Prostate Specific Antigen (PSA) in Semen Stains and Sexual Assault Kit Swabs, 2003, R & D Report: #R 84, Government of South Australia
- 3. Validation Guide for the DNA IQ™ Casework Pro Kit for Maxwell® 16, 2010, Reference
- 4. QIS 17185 Detection of Azoospermic Semen in Casework Samples.
- 5. QIS 17189 Examination of and for Spermatozoa
- 6. QIS 22871 Change Management
- 7. QIS 23401 Validation (DNA Analysis)
- 8. Lee, K., Parry, R. "Change Proposal #24: Comparison of the BioSign and the ABA Card PSA test kits for the identification of PSA in Normal, Oligospermic and Azoospermic semen" 2007 (Unpublished)



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Appendix 1 – Sign off sheet

Evidence Recovery Team:

Assessed by: AKL Date: 13/01/11

Comments: Approved. A Semor Scientist

Analytical Team

Assessed by: ALM Date: 13-01-2011

Comments: Approved.

Quality & Projects Team:

Assessed by: KS Date: 13-07-2011

Comments Approved.

Evidence Recovery & Projects Team:

Assessed by: PMT Date: 12-07-11

Comments: approved .

Reporting 1:

Assessed by:

Comments on heave not. able to sign Kos 13/07/11

Reporting 2:
Assessed by: KOR Date: 13/07/11
Comments Approved

Reporting 3:

Date: South 13/01/11

Assessed by:

Comments

Intelligence Team:

Assessed by:

Comments

Date:

On Leave not KDS 13/07/11

Biostatistician:

Assessed by:

Date:

Comments:

On heave not able to sign KDS13/07/11

Forensic Reporting & Intelligence:

Assessed by: AJR Date: 130711

Comment approved.



JUSTIN HONES AMANAGING SCIENTIST

13-07-11

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Forensic and Scientific Services

Examination of Sexual Cases

1 Purpose

The purpose of this procedure is to describe those procedures required for the examination of sexual assault cases by Evidence Recovery scientists and technicians in Forensic DNA Analysis, in addition to those described in QIS document 17142 (Examination of Items).

2 Scope

This procedure applies to all Forensic DNA Analysis staff that examine or interpret examinations of evidentiary items. This standard operating procedure is in conjunction with individual methods for particular screening tests. Interpretations and limitations of reporting are to be found in each method.

3 Definitions

- Refer to QIS document 23849 (Common DNA Analysis Terms and Acronyms) for a comprehensive list of abbreviations.
- All references to microscopy, refer to QIS document 17189 (Examination For & Of Spermatozoa)
- All references to Acid Phosphatase (AP), refer to QIS document 17186 (The Acid Phosphatase Screening Test for Seminal Stains)
- All references to Phadebas, refer to QIS document 17193 (Phadebas Test For Saliva)
- All references to Tetramethylbenzidine, refer to QIS document 17190 (Tetramethylbenzidine Screening Test for Blood)
- All references to p-30, refer to QIS document 17185 (Detection of Azoospermic Semen in Casework Samples)
- A semen negative item is an item which has either tested negative for spermatozoa microscopically and tested negative for acid phosphatase; or tested negative for spermatozoa microscopically, tested positive for acid phosphatase and tested negative for P30.

4 General Principles

Refer to the general principles contained in QIS document 17142 (Examination of Items).

4.1 Examination Strategies

An examination strategy must be prepared for all SAIKs which are examined. This strategy must include:

- For each item to be examined, what biological fluid is to be screened for
- Items which require no further action
- Items which may only require examination pending presumptive/screening results
- Sample submission strategies (i.e. extraction type, pooling, retain supernatant for Phadebas testing etc).



The examination strategy must be reviewed by a scientist competent to perform the examinations contained in the strategy.

The following are general principles which are used to develop examination strategies for SAIKs, however these principles must be considered within the context of the case history:

- Female SAIKs which are semen negative, any external intimate swabs are submitted for cells (i.e. vulval and perianal)
- Where the complainant is a minor or has an intellectual impairment which may mean that the provided case history is unreliable, all possible offence scenarios are considered.
- Where the complainant is an adult who has lost consciousness, has impaired memory or has consumed alcohol or drugs prior to or during the offence which may impact on memory, all possible offence scenarios are considered.
- Consider previous intercourse with same or different partner, prior to the offence.
 For digital only female complainant cases with prior intercourse, submit external swabs for DLYS with no testing. For male SAIK swabs, consider submitting penile swabs for DLYS where previous intercourse with another partner has occurred.
- Consider the number of offenders for male SAIKs consider submitting penile swabs for DLYS (with no testing) to separate epithelial and spermatozoa.
- · For child complainants, treat all vaginal swabs as external swabs for semen or cells.
- Samples taken from areas of biting, licking or kissing (or other oral contact) are submitted for CSUP. This does not include swabs taken from the mouth (internal or external), anal and vaginal areas which may give false positive results.

5 Examination

The general examination procedures documented in QIS document 17142 (Examination of Items) apply to the examination of sexual cases.

6 Specific Examination Strategies

Refer to Section 6.2 of QIS document 17189 (Examination for and of Spermatozoa) for procedures relating to making a suspension and preparing, staining and reading microscope slides.

6.1 Sexual Assault Investigation Kits

Appendix 1 describes the workflow for presumptive/screening testing of SAIKs. Before commencing the examination of a SAIK an examination strategy must be devised and reviewed in accordance with Section 4.1 of this document.

If there are issues related to the collection or documentation of a SAIK this must be fed back to the relevant FMO or FNE using the SAIK issues log. Examples of issues may include:

- Serum coated, charcoal swabs or other unsuitable swabs/media are submitted (these should tested regardless of the swab type or media and a specimen note must be added to Auslab)
- Insufficient case history
- Labelling issues/inconsistencies
- Smears have been prepared by the FMO/FNE

If a smear has not been received, one will need to be made and tested, refer to QIS document 17189 (Examination for & of Spermatozoa - Section 6).

The following principles should be applied to the submission of SAIK samples:



- Where an amount of spermatozoa which is considered likely to give a DNA profile (i.e. 2+ or more) are seen on multiple swabs from the same internal location (e.g. vagina), and there is no history of sexual contact with another person within the previous seven days, or multiple offenders, then only one of these swabs should be submitted for full analysis. When selecting which swab to submit for testing, preference should be given to the highest internal swab (i.e. submit a high vaginal swab over a low vaginal swab). Other swabs which would otherwise be submitted for full testing based on their presumptive/screening test results must be submitted but with a 'POLD' test code rather than 9plex/Xplex. These samples are then stored in the black box labelled 'SAIK swabs on hold'. This enables the Case Manager to view the results of the first swab, before assessing whether additional samples require processing.
- Submission of swabs for cells (where presumptive and screening tests are negative for semen and spermatozoa) should be considered based on the case history. If more than several days have passed since the offence, it may be unlikely that foreign DNA will be located, particularly if the subject person has bathed. Consult with the Senior Scientist for direction in these matters, however the following scenarios would justify the submission of samples for cells:
 - Child complainant
 - Complainant with mental impairment, or other impairment which may influence reliability of provided offence history
 - Complainant with loss of consciousness or drug/alcohol use which has impaired their recollection of events
 - Other circumstances as deemed appropriate by the QPS or Senior Scientist.

6.2 Acid Phosphatase (AP) Positive Fabrics

Appendix 2 describes the workflow to be used for presumptive/screening testing of AP positive fabrics.

AP positive fabrics are submitted by QPS. The AP positive area should be clearly marked on the fabric. If the fabric is not marked then the entire sample should be tested, including both sides of fabric.

AP positive fabrics should be submitted with sufficient additional area surrounding the circled AP positive area to enable the examining scientist to safely hold the fabric if/when taking a scraping. Where insufficient additional area has been provided a FERRO should be created so that it may be fed back to the QPS.

Where a large piece of AP positive fabric is to be tested, divide the item into sections and test each section separately. If spermatozoa are located microscopically on one section, but are not located on other sections, P30 testing is not required to be performed on all negative sections. All sections are submitted for analysis as DLYS.

If semen is not detected there is no further action and the item is returned.

Generally fabrics are scraped or excised (extreme care to be taken to stop needle stick injuries).

6.3 Semen in-tubes

Appendices 2 and 3 describes the workflow for presumptive/screening testing of semen in tubes based on whether the QPS have conducted AP testing. If semen in tubes are stored

in an in tube registration box, they must be transferred to an items box so that they can be examined by a scientist.

6.4 Condoms

Appendix 3 describes the workflow for presumptive/screening testing of Condoms.

When a condom is received it should be described in terms of "O/S surface as received" and "I/S surface as received". Describe any fluid that may be present on or within the condom. Describe length and diameter, colour, patterning and translucency of condom.

Take one wet and one dry swab from the O/S and I/S surfaces of the condom. Sample and combine I/S wet and dry swabs into one tube and O/S wet and dry swabs into another tube. N.B. When sampling the swabs, to ensure that there is not excess substrate submitted, sample the entire of the wet swab, but only the outer layer of the dry swab.

6.5 Sanitary Pads and Tampons

Appendix 4 describes the workflow to be used for presumptive/screening testing of sanitary pads and tampons.

Sanitary pads are AP tested on the side worn in contact with the skin.

Tampons are cut through the middle and splayed out. The outer sides of the tampon are then AP tested.

6.6 Post Mortem Samples

Appendix 5 describes the workflow for presumptive/screening testing of Post Mortem samples.

The examining scientist assigns an EXH barcode to the PM samples as a whole, which is passed onto QPS DNA results management (DRMU). All other samples submitted will be subsamples of the PM samples EXH (as per SAIK submissions).

The receipt under which the samples are submitted usually has an associated Coronial case number. Before any subsamples are registered this Coronial case number needs to be changed to the associated QP number by an AUSLAB Corrections Officer. If subsamples are registered under a Coronial case number the EXH lines will not be transmitted to QPS

PM samples may include sexual assault swabs and/or slides (high vaginal, low vaginal, vulval etc), pubic hair, head hair, fingernail clippings or scrapings.

Intimate sexual assault swabs which are semen positive are submitted for DLYS. Intimate sexual assault swabs which are semen negative are submitted for cells.

Sometimes the fingernail clippings include a portion of tissue or part of the finger. In this case a moistened swab can be used to sample potential foreign DNA from the underside of the nail, taking care not to sample the deceased person's tissue (i.e. targeting the distal end of the nail).

When PM samples are complete, send an E-mail to the Senior Scientist with the EXH barcode so that that information can be passed onto DRMU to facilitate electronic transfer of results from Auslab to the Forensic Register.

6.7 Clothing and Bedsheets

For large items, an examination strategy should be formulated based on the case history and if necessary consultation with the QPS. This must be recorded in the UR notes for the case.

If the case history suggests that the item has been washed then it may be necessary to perform microscopy only considering the water soluble nature of Acid Phosphatase and P30. Use the case history and if necessary communicate with the investigating officer to establish an area to target.

6.8 Wet and Dry swabs - QPS submitted

When wet and dry swabs are received from the same site (e.g. in a SAIK, or from one item) submit each of the swabs separately.

6.9 Multiple Presumptive/Screening Tests

Consideration should be given to the order in which screening tests are conducted based on the type of tests to be performed and the conservation of sample on the item. Where both AP and Phadebas screening tests are required, Phadebas the exhibit first (using commercial paper). Once the Phadebas test is complete the Phadebas paper can be sprayed with AP reagent.

6.10 Analytical Slides

If reading of differential slides is requested by a case scientist, retrieve slides from storage box and put a borrowed comment in AUSLAB. Stain slides and perform microscopy.

Read slides and fill out QIS document 17037 (Microscopy of Smears)

N.B. Old slides- DLYS step 10 slide may have both sperm and epithelial cells, whereas step 22 slides may have sperm only. Currently only one slide is made- should have sperm only.

Return slides and add a returned comment in AUSLAB.

6.11 Penile Swabs

The presence of spermatozoa on penile swabs is not unexpected. These swabs are generally submitted for Cells, however where the case history indicates multiple offenders, or previous sexual contact, they should be submitted for DLYS. Appendix 1 describes the workflow to be used for presumptive/screening testing of SAIK swabs.

6.12 Lubricant Testing

If an item is required for lubricant testing consult with Forensic Chemistry before any examinations are conducted

7 Associated Documentation

QIS: 16004	AUSLAB Users Manual – DNA Analysis
QIS: 17033	General Examination Record (Unruled)
QIS: 17034	General Examination Record (Ruled)
QIS: 17117	Procedure for Case Management
QIS: 17119	Procedure for Release of Results
QIS: 17135	Handling and Sampling of Syringes and Needles



QIS: 17140 QIS: 17185	Procedure for the Identification and Examination of Hairs Detection of Azoospermic Semen in Casework Samples
QIS: 17186	The Acid Phosphatase Screening Test for Seminal Stains
QIS: 17189	Examination For & Of Spermatozoa
QIS: 17190	Tetramethylbenzidine Screening Test for Blood
QIS: 17193	Phadebas Test for Saliva
QIS: 20080	Digital imaging in DNA Analysis
QIS: 22846	General Swab Exam Record
QIS: 22857	Anti Contamination Procedure
QIS: 22870	Forensic DNA Analysis Outer Packaging Record
QIS: 23008	Explanations of EXR/EXHs
QIS: 23014	Cigarette Butt General Examination Record
QIS: 23055	General Examination Record
QIS: 23849	Common Forensic DNA Analysis terms and Acronyms
QIS: 23898	SAIK Details Record
QIS: 26071	Examination of in-tube samples
QIS: 31286	SAIK form no semen testing

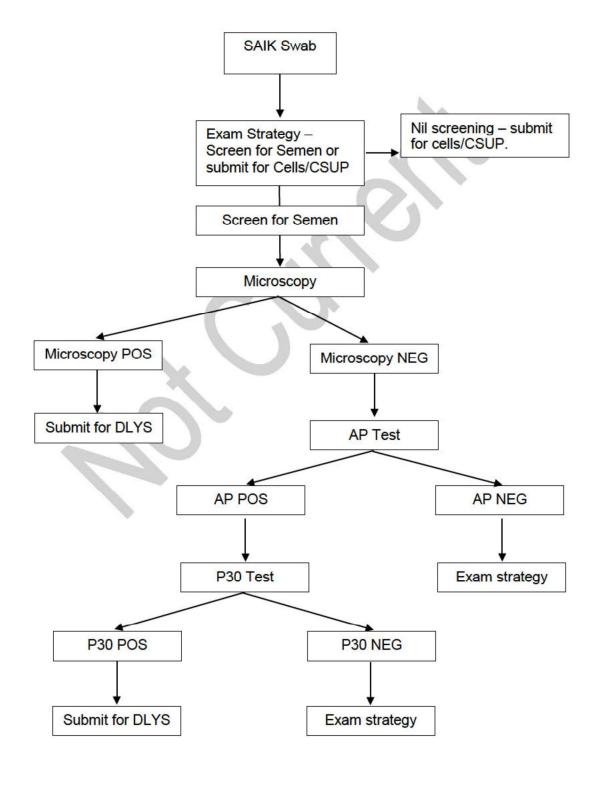
8 Amendment History

Version	Date	Author/s	Amendments
1	23/10/2013	L Ryan A Houlding J Seymour-Murray	Document created (content split from Examination of Items)
2	05/12/13	A Houlding	Update for XPlex
3	03/11/2014	A Houlding	New template, 6.2 title changed, header changed, added POLD test code for SAIK samples on hold, also apply the SAIK on hold procedure to samples with a micro result of 2+ (changed from 1+), fixed hyperlinks Changed wet and dry swabs to be submitted separately. Formatted flowcharts. Added lubricant testing section

9 Appendices

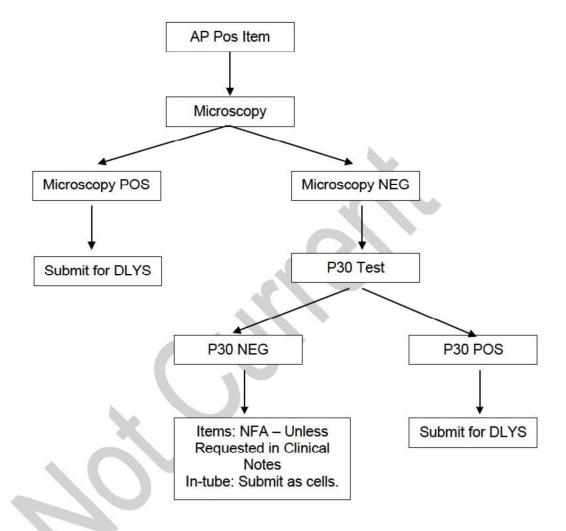
9.1 Appendix 1: SAIK Examination Workflow

This workflow is intended to demonstrate the testing of one sample from a SAIK. The submission of samples should be considered within the context of the Examination Strategy, taking into consideration the case history as well as the presumptive and screening results of other SAIK swabs, particularly those from the same location.

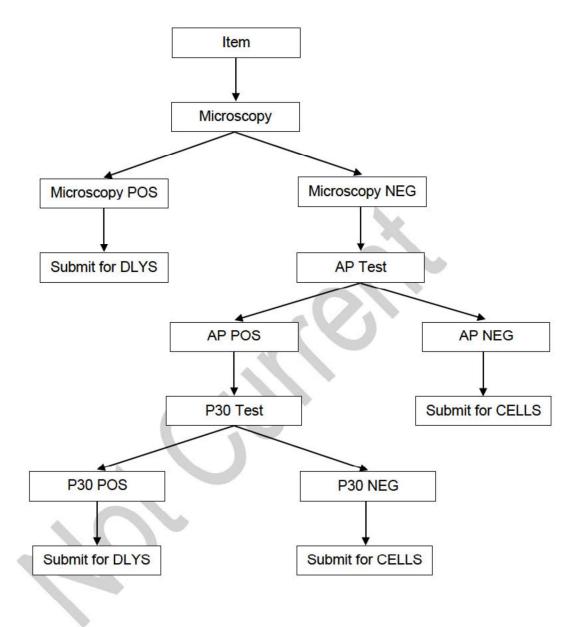


9.2 Appendix 2: QPS AP Tested Items (including Semen in-tubes) Workflow

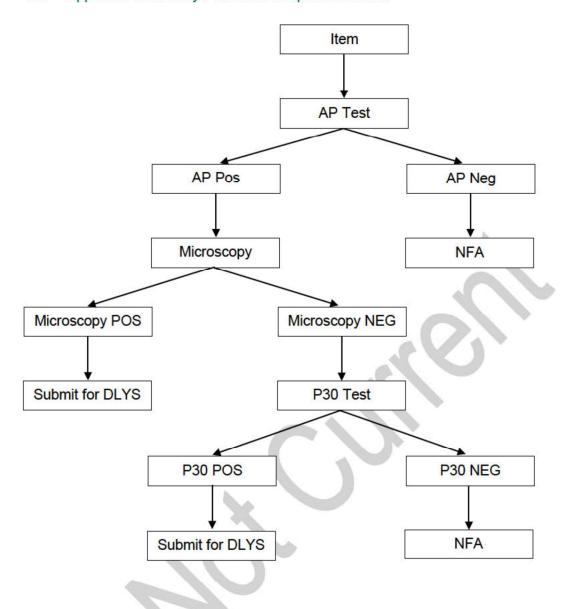
This workflow is to be used for all items which have previously tested positive using the AP test by the QPS.



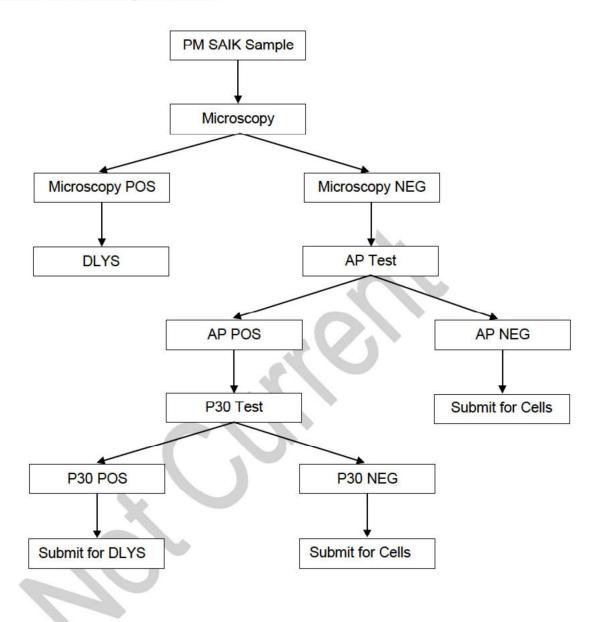
9.3 Appendix 3: Condoms and Semen in Tubes (not AP tested by QPS) Workflow



9.4 Appendix 4: Sanitary Pads and Tampons Workflow



9.5 Appendix 5: PM SAIK Samples Workflow



A CLINICAL AND STATEWIDE SERVICE

Project #92 - Procedural change for Differential Lysis extracts obtained from intimate SAIK swabs

Report – Ch Fractions from

Procedural change for Epithelial amples

Alicia Quarterma DNA Analysis Ur

12

1 Abstrac

Currently within I process to gener quantification, ar

nate SAIK swabs undergo a Differential Lysis Extraction actions. These fractions are then submitted for ectrophoresis. Given the majority of epithelial fraction

DNA profiles do not provide additional information to that which is provided in the spermatozoa fraction DNA profile, epithelial fractions generated after a Differential Lysis Extraction can be stored indefinitely. These epithelial fractions can then be profiled if the case scientist decides that these DNA profiles may aid in result interpretation.

2 Background

Relevant samples which undergo a Differential Lysis Extraction process produce two fractions, one epithelial fraction and one spermatozoa fraction per swab. All DNA profiles generated from these fractions require interpretation by a scientist, and are then reviewed by a second scientist. Only the results generated from certain fractions are reported back to QPS via AUSLAB i.e. not every sperm and epithelial fraction result is entered in an EXH.

From 01/01/2011 to 31/08/2011, 128 intimate SAIK samples were submitted for a Differential Lysis Extraction, generating 128 Sperm Fractions and 128 Epithelial Fractions. Of these 128 Epithelial Fractions, none were shown to produce a DNA profile which was either: useable (e.g. NSD, partial unsuitable, mixed partial unsuitable), or not otherwise demonstrated in the Sperm Fraction (i.e. the Sperm Fraction DNA profile had the same information/additional information – see below).

- 5 had conditioned interpretations done which were not required given the Sperm Fraction result

- 3 had to be upleaded then removed from NCIDD
- 15 had EXH lines entered which were not necessarily required (3 of which had be updated once an evidence sample was received)
- 7 were submitted for nucleospin after the initial run
- 30 were submitted for microcon after the initial run
- 10 were re-amplified
- 13 were submitted for re-genescan after the initial run

3 Purpose

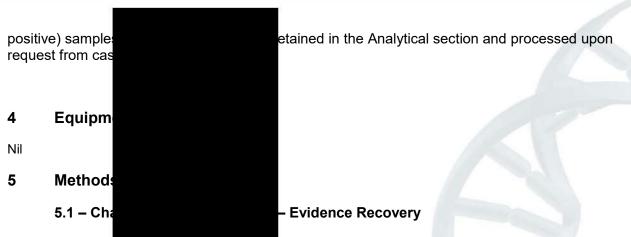
The aim of this management, reachieved through

uce the amount of time and funds spent on case ocessing (initial and reworks). This aim is proposed to be rocessing of epithelial fractions for routine SAIK (sperm



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Project #92 - Procedural change for Differential Lysis extracts obtained from intimate SAIK swabs



For SAIK intimate samples being submitted for a Differential Lysis Extraction – these samples will be registered as per current procedure with the addition of processing comments. There will be processing comments for samples that are <u>not</u> required to proceed through the analytical processes i.e. a HVS sample that is sperm pos will have its sperm fraction registered as per normal, the Epithelial fraction will have a processing comment added to say 'ext&hold'.

For SAIK intimate samples where the examination findings were of AP/p30 positive in the absence of detectable sperm, these epithelial fractions should be processed fully in an attempt to obtain DNA from male epithelial cells (vis vasectomised males). No processing comment is required in this situation. For all other samples (eg. Fabric, condoms) submitted for a Differential Lysis Extraction – no processing comment required as the epithelial fractions require submission.

Priority 1 Epithelial fractions will be submitted for Analytical processing, and will therefore follow the same process as all other samples being submitted for a Differential Lysis Extraction (as above).

5.2 - Change to current process - Analytical Section

The current method of Differential Lysis Extraction will remain unchanged. After the extraction, the Analytical section will alter their current process slightly to ensure that only epithelial lysates mandat processing comments proceed onto quantification (details of procedural change to be confirmed).

5.3 Change to current process - FRIT

Processes remain largely unchanged.

Extra considerations to be made at time of case management and reporting:

- Assess whether all relevant epithelial fractions have been submitted by Evidence Recovery i.e. those from fabric, condoms etc.
- Assess whether epithelial fraction results are required to assist in DNA profile interpretation (if required, this can be done by ordering a 'REQC' on the epithelial fraction and ensure DNA priority is correct).
- Change to star entry of the star entry of the



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Project #92 - Procedural change for Differential Lysis extracts obtained from intimate SAIK swabs

6 Results

Given the figures Reporting Scient consumables an epithelial fraction fractions), it is pr have previously, proposed to impliand interpretation kground), implementation of this change will save iew, reporting), as well as departmental funds used on I in the analytical processing of SAIK (sperm positive) he Analytical Section (through holding epithelial prity samples could be batched earlier than they would bund time for these samples. The turnaround time is as the epithelial fractions and any associated reworks by processed to end stage.

This change is o Processing epith SAIK swabs undergoing a Differential Lysis Extraction. d other substrates (eg condoms) is useful as it is often

important to obtain DNA information in the epithelial fractions; for example, DNA that could have come from a female complainant may be obtained in the epithelial fraction from samples taken from condoms, and DNA that could have come from 'touch' or from vasectomised males could be found in epithelial fractions of samples from other substrates.

Small changes/adjustments will need to be made by each team, however the impact should be minimal. These changes will need to be fine-tuned by the individual teams through discussions with staff.

There is considered to be little to no risk associated with this change. If a sample mistakenly does not have a 'processing comment' added at the Evidence Recovery stage, this can be rectified at case management by the ordering of REQC testcode. The only risk is the time lost.

7 Conclusion and recommendations

Given the minimal risk associated with this change and the positive impact it will have on the DNA Analysis Unit, it is recommended that this change be implemented as soon as possible. Linking hanges to processes can occur at the same time.

Appendix 1 - Sign off sheet

Evidence Recovery Team:

Date:

Assessed by:

Comments:

Analytical Team

Assessed by: Date:

Comments:

Quality

Assesse Commer



Project #92 – Procedural change for Differential Lysis extracts obtained from intimate SAIK swabs

Evidence	Recover	y & Pro	jects	Team:
----------	---------	---------	-------	-------

Date:

Assessed by:

Comments:

Reporting 1:

Assessed by: Date:

Comments:

Reporting 2:

Assessed by: Date:

Comments:

Intelligence Team:

Assessed by:

Date:

Comments:

Forensic Reporting & Intelligence:

Assessed by: Date:

Comments:

Bio-Statistician:

Assessed by: Date:

Comments:

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Managing Scientist DNA Analysis Forensic and Scientific Services

Or by telephone



JH-21

Erin Shearer

From: Amanda Reeves

Sent: Friday, 4 March 2016 2:11 PM

To: Justin Howes
Subject: RE:

Hi JAH

My thoughts are that a further investigation into this is warranted – perhaps looking at how the smear was prepared etc – with the view to widening the investigation if a more systemic issue is observed. I am happy to free up some of JMW's time if you think this is necessary?

Thanks

Amanda Reeves Dip Mgt BSc MSc For Sci Senior Reporting Forensic Scientist Forensic DNA Analysis | Police Services Stream Forensic and Scientific Services Health Support Queensland Department of Health | Queensland Government



From: Jacqui Wilson

Sent: Friday, 4 March 2016 1:58 PM

To: Amanda Reeves
Cc: Justin Howes
Subject:

Hi Amanda

Here's another example of where the initial slide assessment has differed greatly from the DLYS slide – initial screening was 0 sperm seen however upon examination of the DLYS slide it was 3+.

Whilst I expect that where we get AP/PSA +ve screening results we have the opportunity to further investigate the samples following DLYS extraction and profiling; what I'm concerned about is where the timeframe between deposition and recovery is such that it falls outside the window where we would expect to see the +ve AP/PSA results however sperm may still persist, so between the 4 – 7 day mark. In these cases, and I'm worried there may be some, the HVS/LVS swabs would not be tested and it's quite possible that there are sperm present. And we know that our Maxwells have been optimised to work well with low sperm/high epithelial samples.

I personally think we have an issue with the preparation of the slide itself, not in the reading of the slides; Janine phoned me to tell me about the 3+ and indicated that she had gone back to the original slide and still could not find sperm so, in my opinion, there's something wrong there.

Perhaps we could run a bit of an investigation on some mock up samples, looking at the different methods of slide preparation going back to making slides directly from the swabs, making slides from the suspensions, as is currently done, and comparing those results to the DLYS slides.

I'll leave that one with you ©

Jax

Jacqui Wilson B App Sc M Sc Reporting Scientist, Forensic DNA Analysis Police Services Stream | Forensic & Scientific Services | Health Support Queensland Department of Health | Queensland Government





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JH-22

Erin Shearer

From: **Justin Howes**

Friday, 4 March 2016 2:16 PM Sent:

To: Luke Rvan Subject: FW:

Sorry, meant to CC you here.

jah

Justin Howes BSc BA MSc (For Sci) Team Leader - Forensic DNA Analysis

Police Services Stream | Forensic & Scientific Services | Health Support Queensland

Department of Health I Queensland Government





From: Justin Howes

Sent: Friday, 4 March 2016 2:15 PM To: Jacqui Wilson; Amanda Reeves

Subject: RE:

Hi Jacqui,

Great timing in catching Luke and I together on this! We are also together on the fact that these two reads being vastly different is worth looking into further.

Thanks for raising your concern – if that wasn't done, there wouldn't be anything we could do to find out and action this outside of audit schedules.

Good work and we will follow things up here.

jah

Justin Howes BSc BA MSc (For Sci) Team Leader - Forensic DNA Analysis Police Services Stream | Forensic & Scientific Services | Health Support Queensland









Health Support Queensland

From: Jacqui Wilson

Sent: Friday, 4 March 2016 1:58 PM

To: Amanda Reeves Cc: Justin Howes Subject:

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Jax

Jacqui Wilson B App Sc M Sc Reporting Scientist, Forensic DNA Analysis Police Services Stream | Forensic & Scientific Services | Health Support Queensland Department of Health | Queensland Government















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JH-23

Erin Shearer

From: Justin Howes

Sent: Friday, 4 March 2016 2:13 PM

To: Amanda Reeves

Subject: RE: 5

Thanks and agree, will reply to other email.

Justin Howes BSc BA MSc (For Sci)
Team Leader - Forensic DNA Analysis
Police Services Stream | Forensic & Scientific

Police Services Stream | Forensic & Scientific Services | Health Support Queensland

Department of Health | Queensland Government









Health Support

Queensland

From: Amanda Reeves

Sent: Friday, 4 March 2016 2:11 PM

To: Justin Howes

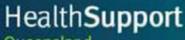
Subject: RE:



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Amanda Reeves Dip Mgt BSc MSc For Sci Senior Reporting Forensic Scientist Forensic DNA Analysis | Police Services Stream Forensic and Scientific Services Health Support Queensland Department of Health | Queensland Government



Queensland

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Sent: Friday, 4 March 2016 1:58 PM

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I'll leave that one with you ©

Jax

Jacqui Wilson B App Sc M Sc Reporting Scientist, Forensic DNA Analysis Police Services Stream | Forensic & Scientific Services | Health Support Queensland Department of Health | Queensland Government















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JH-24

Erin Shearer

From: Paula Brisotto

Sent: Tuesday, 15 August 2017 1:28 PM

To: Justin Howes **Subject:** Data analysis report

Attachments: Data Analysis report (2)KDR.doc; Data Analysis report_draft1 - LBR track changes_ MOH track

changes.doc; Data Analysis report_draft1 - LBR_ MOH_PMB.doc

Follow Up Flag: Follow up Flag Status: Flagged

Hi Justin,

I've had a meeting with Cathie about the feedback received and where to from here.

We are on the second round of feedback, and both Matt, Kylie and Luke have provided theirs. From here, Cathie has requested your involvement, so we can try and take on board some feedback, while trying to keep the data analysis within scope. I am hoping from your experience with sexual assault examination and interpretations, you can provide some wording within the document, so we can all get to a point where we are happy with the content, the assessment and the summary. I think I am too close/getting fatigued with this, and I would like a fresh set of eyes/approach to looking at this report.

Please find attached:

- 1 Kylie's feedback
- 2 Matt's feedback
- 3 my updates from Matt's feedback.

Unfortunately, I didn't get to spend much time doing this today. If you are able to have a look over these documents, and provide wording suggestions/improvements or alternative words, please have at it ©

I have kept in 'systemic failure', as I still feel that this was the concern on my return, and is an evaluation of the data – similar to what we did with the MPII investigation, with the outcome not showing a systemic failure of the instrument.

I am also not sure if the scope is clear, and if the assessment of the case as a whole (e.g. submit any NFA samples or read diff slides if DNA profile indicates male in sperm fraction etc) is clear enough.....

Many many thanks, Paula



Paula Brisotto

Team Leader – Evidence Recovery and Quality Team, Forensic DNA Analysis, Police Services Stream Forensic & Scientific Services, Health Support Queensland, **Department of Health**

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Data Analysis of modified sexual assault process for zero sperm at Evidence Recovery

Paula Brisotto, Matthew Hunt, Kylie Rika

May 2017

Introduction

In August 2017, a modified process was implemented in the Evidence Recovery Team of Forensic DNA Analysis for samples where zero sperm were observed at the evidence recovery stage. This modification meant that every microscopy negative result was submitted for differential lysis DNA extraction, and the microscope slides made during the extraction process were read by the Evidence Recovery Team for final reporting of sperm findings.

This data analysis looks at the differential lysis slide reads for those evidence recovery slide reads of zero sperm seen. For any differential slides with sperm observed, this data analysis attempts to assess what extraction process pre-modification would have occurred, and if this could have impacted on the DNA results obtained for the case.

Results

In August 2016, sexual assault slides that were microscopy negative for the presence of sperm at Evidence Recovery (ER) stage were sent for differential lysis, and the differential lysis slides read for the presence of sperm.

Since August 2016 up to and including 28/03/2017, 738 differential slides have been read from ER microscopy negative slides. From these 738 slides, sperm were observed on 148 slides at differential lysis stage (see Table 1 below).

Table 1 Resulting Differential Lysis slide reads for the presence of sperm.

Diff Lysis Slide read	Number of samples	Percentage of Total
0 sperm	591	80.08
<+1 sperm	104	14.09
+1 sperm	36	4.88
+2 sperm	7	0.95
	0 sperm <+1 sperm +1 sperm	0 sperm 591 <+1 sperm 104 +1 sperm 36

A better Data Analysis would be to take the 104 <+1sp samples and plot them to see how many of the 104 samples were interpretable. This would tell us how many sp we need to see to get an interpretable profile. If



<+1sp doesn't give us interpretable profiles then <+1sp is our limit of detection (almost like a quant value). Therefore if we are missing <1+sp we could live with that because it's not going to give us anything interpretable anyway. This type of data analysis would take away the bias of case info.

We have to bear in mind that if we had another sample of 738 slides, the findings could be completely different because the cases could all be quite different.

Each of the 147 slides which had sperm observed at differential lysis stage were assessed to determine what submission would have occurred prior to the modified process.

Based on the presumptive test results and the examination strategy, **72** (48.1%) of these samples would have undergone differential lysis extraction, **47** (31.9%) of these samples would have undergone cell extraction, and **28** (19%) would have no further action (no submission for DNA extraction).

Those samples which would have undergone differential lysis extraction were not assessed further, as the extraction process would have been the same irrespective of the sperm microscopy results at Evidence Recovery.

All of the remaining samples (those which would have gone through cell extraction and those which would have no further action) were assessed to determine if the pre-modified processes would have impacted on the DNA results obtained. This was looked at from an exhibit (if other items in the SAIK or exhibit would have provided the relevant information to the QPS) and case perspective (if other items in the case would have provided the relevant information to the QPS).

13 samples gave results of a single source DNA profile matching the assumed known contributor; therefore a cell extraction or no further action would not have impacted on the DNA results for these exhibits.

29 samples gave results of *no DNA detected (this doesn't mean we wouldn't get a profile)* at quantitation, *no DNA profile*, or *complex DNA profile*, *unsuitable for DNA interpretation* generally due to partial nature of the DNA profile. For each of these, a cell extraction or no further action would not be expected to impact on the results reported for these exhibits and/or the case. What about 4p that may have been 3p if they had gone through diff lysis? Using the argument in this paragraph negates the need to even do diff lysis in the first place.

22 samples which underwent a cell extraction would have extracted any DNA present in the sample, including any sperm which may have been present. From the results obtained through the modified process, these samples would be expected to have obtained a mixed DNA profile through cell extraction, which could be conditioned on the known contributor (what about in cases where we don't have ref sample and can't condition?) where appropriate, compared to any reference DNA profiles for statistical interpretation, or loaded to NCIDD where possible. Again, what about 4p that may have been 3p if they had gone through diff lysis?

Of these above 22 samples, 10 had other items within the SAIK, exhibit or case which would have undergone differential lysis extraction based on sperm presence or presumptive results, and obtained results that would have provided the relevant information to the QPS.

10 samples which would not have progressed to DNA extraction had other items within the SAIK or exhibit which would have undergone differential lysis extraction based on sperm presence or presumptive results, and obtained the same or similar DNA results. As DNA results from a foreign source of DNA would have been reported to the QPS through pre-modified processing, it is considered

there would be no impact on the DNA results provided for the case. This is very much our opinion, we don't know what QPS's opinion is ie. We don't know what they were looking for as we don't have that info necessarily. Consider also what if say the vag swab and thigh swab gave 2 different males for example?

Of the above 10 samples, 2 were in-tube samples from the same case which would have had no further action, and did provide DNA profile results able to be interpreted. From the sample descriptions, photographs and numbering of these items, it appears as though they are two items from the same parent exhibit, of which a further 4 samples were also submitted as in-tube samples. 3 of these samples would have undergone differential lysis extraction based on sperm presence or presumptive results, and one was sperm negative and no DNA detected throughout the process. Based on this information, it is considered there would be no impact on the DNA results provided for the case.

1 sample which would not have progressed to DNA extraction obtained the only DNA result in the case indicating the presence of foreign DNA (DNA other than the known contributor). The SAIK associated which this case had no sperm detected at evidence recovery or differential lysis, and any cell extraction obtained a DNA profile from the known contributor.

Table 2 Breakdown of	data by sperm seen at	differential ly	vsis (DLYS)

Sperm seen at DLYS	DLYS extraction	Known DNA profile	DNA result *	CELL extraction	Other samples/DNA results**	Impacted
+2	6			1 (1 with other samples/DNA results**)		
+1	20		4	7 (4 with other samples/DNA results**)	4	1
<+1	46	13	25	14 (5 with other samples/DNA results**)	6	

^{*} no DNA detected at quantitation, no DNA profile, or complex DNA profile, unsuitable for DNA interpretation

Or in words.....

A further breakdown of the data by sperm seen at differential lysis as follows:

- +2 sperm seen at differential lysis: of the 7 samples, 6 would have been submitted for DLYS extraction, and 1 would have undergone cell extraction (also with other results providing relevant information), all with nil impact on DNA results.
- +1 sperm seen at differential lysis: of the 36 20 would have been submitted for DLYS extraction, 4 with unsuitable DNA results, 7 would have undergone cell extraction (4 with other results providing relevant information), 4 with other results providing relevant information, and 1 sample impacted.

^{**} other items within the SAIK, exhibit or case which obtained results that would have provided the relevant information

<+1 sperm seen at differential lysis: – of 104 – 46 would have been submitted for DLYS extraction, 13 obtained a DNA profile from the known contributor, 25 with unsuitable DNA results (no DNA detected, no DNA profile, or complex unsuitable for DNA interpretation), 14 would have undergone cell extraction (5 with other results providing relevant information), and 6 with other results providing relevant information.

Discussion

The aim of this data analysis was to assess the impact on information provided to the QPS for the 738 samples had they undergone processing prior to the implementation of the modified process, by looking at what extraction process pre-modification would have occurred, what other results were obtained from other samples in the case, and if this could have impacted on the DNA results obtained and reported to the QPS for the case.

This data did not look at any other slide reads from either evidence recovery or differential lysis other than those that were originally zero sperm at ER, and underwent the modified process from August 2016.

Explanation of why things were determine to be impacted or not? Especially for cells extraction – i.e. cell extraction would co-extract DNA from any cells present, including sperm....?

In summary:

- 1. In the majority of samples (80.08%) the Evidence Recovery slide read of zero was reproduced at the differential lysis stage.
- 2. Where sperm were observed on the differential slide, almost half of all samples (48.1%) would have undergone differential lysis extraction based on previous examination processes, therefore no change to the DNA profiling outcome.
- 3. In all samples where +2 sperm were seen at differential lysis (from zero sperm at ER) DNA results would not have been impacted.
- 4. In one sample where +1 sperm were seen at differential lysis (from zero sperm at ER) DNA results would have been impacted (see point 6 below).
- 5. In all samples where <+1 sperm were seen at differential lysis (from zero sperm at ER) DNA results for the case would not have been impacted.
- 6. 1 sample would have failed to provide critical information to the QPS for a case. From the 738 samples of zero sperm seen at ER, this equates to 0.14% of samples. Of the 147 samples where sperm was detected at differential lysis stage from zero sperm seen at ER, this equates to 0.7% of samples.

Whilst the conclusions drawn from this data analysis suggest that overall, the risk of missing critical evidence is very small in the specific cases contained in this data set, there are limitations to this data including the inability to "extrapolate" back to other cases not within this time period and prior to the modified process. We cannot predict the effect on future cases. In other words whilst one could surmise that in general the chance of missing critical evidence is less than 1%, in reality this rate could be higher or lower by an indeterminate/unquantifiable amount.

This data analysis, in isolation, should not be used to perform a cost/benefit analysis on investigating all cases examined where zero sperm were seen at the evidence recovery stage.

This data shows that there is not a systemic failure in the sexual assault examination process? - but a possible sub-optimisation in the ER slide making process (that 20% means something).



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Data Analysis of modified sexual assault process for zero spermatozoa detected at Evidence Recovery

Paula Brisotto, Matthew Hunt, Kylie Rika, Luke Ryan

May 2017

Introduction

Prior to August 2016, exhibits which gave negative results for spermatozoa and seminal fluid were typically not submitted for DNA testing, using the differential lysis extraction protocol (depending on case circumstances and examination strategy these samples may still be submitted for differential lysis extraction or for cell extraction). In August 2016, the Evidence Recovery Team implemented a modified sample submission process for all samples whereby all samples screened for seminal fluid were submitted for DNA testing, using differential lysis extraction, irrespective of the presumptive screening results (i.e. even those samples for which spermatozoa were not located microscopically and P30 tests were negative were still submitted for DNA testing using a differential lysis extraction). The microscopy slides created during the differential lysis extraction were then read by Evidence Recovery Team scientists for final reporting of spermatozoa detection results.

This modified process was introduced after it was raised that the initial slide microscopy conducted during ERT examinations may have a lower sensitivity than that slides produced during the differential lysis extraction process.

This modification process was introduced in response to concerns that the initial microscopy conducted during ERT examinations may be detecting fewer spormatezea than were seen by subsequent microscopy of slidos producod after the differential lysis extraction process.

This data analysis examines the set of samples which had no spermatozoa or seminal fluid detected during the initial Evidence Recovery examination, and which were then submitted for differential lysis extraction. The results from this data set will be used to assess the difference in the pre and post August 2016 examination and sample submission strategies based on the final DNA results and attempt to determine what, if any, impact this may have had on the DNA results reported for the case.

In assessing the potential implications for the DNA results reported, relevant aspects of casemanagement were taken into account, including presumptive screening test results, existing examination strategies for different sample types and other results within the case.



Results

Since August 2016, the sample submission strategy was changed for samples which gave negative presumptive screening results for spermatozoa and seminal fluid (i.e. no spermatozoa observed on microscopy and P30 negative test). All samples which were tested for the presence of spermatozoa and seminal fluid were submitted for DNA extraction using the differential lysis protocol, regardless of the presumptive screening results.

During the period, 8 August 2016 to 28 March 2017, there were 738 samples for which the presumptive screening in Evidence Recovery did not identify spermatozoa during microscopy which were submitted for differential lysis extraction. For each of these 738 samples a microscopy slide was created during the Evidence Recovery examination (the Evidence Recovery slide), and a second slide was created during the differential lysis extraction (the Differential Lysis slide). Both slides were read by Evidence Recovery scientists.

The differences in the methods for creating the Evidence Recovery and Differential Lysis slides are a relevant context to the results of this data mining. Evidence Recovery slides are created by suspending the sample substrate (i.e. swab, scraping, material etc.) in $200\mu L$ to $300\mu L$ of nanopure water. This suspension is then vortexed and agitated with a disposable pipette. One drop (using a disposable pipette) of the suspension is then spotted onto a microscopy slide. The Differential Lysis slides are created by taking $3\mu L$ from the male fraction (post separation from the female fraction) during the differential lysis protocol. The total volume for the male fraction from which the Differential Lysis slide is created is approximately $50\mu L$. It is expected that the Differential Lysis slide would be more sensitive (in terms of spermatozoa detection) for one main reason: the Differential Lysis slide is created by sampling $3\mu L$ from a total volume of $50\mu L$ of male fraction, therefore any spermatozoa present in the male fraction are at a higher concentration than the Evidence Recovery suspension which has a total volume of $200\mu L$. Suggest could also briefly cover the categorisation of sperm numbers (how many meant by <1+, 1+ etc.)

Table 1 Differential Lysis slide reads for the presence of sperm.

Original ER read	Diff Lysis Slide read	Number of samples	Percentage of Total
0 sperm	0 sperm	591	80.08
0 sperm	<+1 sperm	104	14.09
0 sperm	+1 sperm	36	4.88
0 sperm	+2 sperm	7	0.95

For 591 of these 738 samples, no spermatozoa were observed on either the Evidence Recovery or the Differential Lysis slides. For these results we can infer that either there were no spermatozoa present, or if they were present they were below the limit of detection for both the Differential Lysis and Evidence Recovery slide preparation techniques.

The remaining 147 slides, for which there were no spermatozoa observed on the Evidence Recovery slide, but where spermatozoa were observed on the Differential Lysis slide, were assessed to determine the impact on final DNA results for that sample.

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Of the 7 samples for which no spermatozoa were located on the Evidence Recovery slide, but +2 spermatozoa were located on the Differential Lysis slide: 6 would have been submitted for differential lysis extraction pre-August 2016 based on a positive P30 result SUGGEST use our standard wording for all of the below interpretations/stats: for example: Formatted: Indent: Left: 1.27 cm, No bullets or numbering The-remaining sample was a vulval swab sample Formatted: Highlight be used to identify case details) from a SAIK. The Formatted: Space After: 10 pt A mixed DNA profile was obtained from this vulval-sample-swab, spermatozoa fraction which Formatted: Indent: Left: 0.63 cm, Space After: 10 pt, No bullets or numbering indicated the presence of DNA from 3 contributors, one of whom could be the complainant. Based on statistical analysis it was estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if the nominated suspect has contributed DNA, along with the complainant, rather than if he has not. gave a 3P mixed DNA profile (net yet reported). Both Tihe high vaginal and low vaginal samples Formatted: Font color: Custom Color(RGB(33,23,233)), swabs from this SAIK -had spermatozoa observed on the Evidence Recovery slides. Mixed DNA profiles were obtained from the high vaginal swab, spermatozoa fraction and the low vaginal swab, spermatozoa fraction which each indicated the presence of DNA from 2 contributors, one of whom could be the complainant. Based on statistical analysis it was estimated that the each of these mixed DNA profiles obtained is greater than 100 billion times more likely to have occurred if the suspect has contr buted DNA, along with the complainant, rather than if he has not. The high vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. The low vaginal sample gave a similar result to the high vaginal sample. Formatted: Indent: Left: 1.27 cm, Space After: 10 pt, No bullets or numbering Therefore failure to submit the vulval sample would not have altered the final result for the. Formatted: Highlight SAIK. ?True statement. – the presence of a possible third contributor in the vulval swab may still be considered relevant for the case. Defence could use this evidence of a third person to cast doubt on the complainants story / character - who is this third person? - could this be the true offender etc. - think we should acknowledge that there could be limited impact on the results reported for this SAIK For the 36 samples which gave no spermatozoa on the Evidence Recovery slide but +1 spermatozoa on the Differential Lysis slide: 19 would have been submitted for differential lysis extraction pre-August 2016 based on positive Seven of these 36 samples would have been submitted for cell extraction rather than differential lysis extraction pre-August 2016. Submission of these seven samples for cell extraction rather than for differential lysis would not have altered the final results for these SAIKs because: (As above, too definitive a statement? - A strong contribution from the suspect was obtained for each sample, however there may still be an impact if enough DNA from the female epithelial cells is present following a cell extraction as this could potentially swamp out the contribution of male DNA from the spermatozoa.) The spermatozoa fractions of two of these samples gave single source profiles which

consistent with matched the nominated suspect.

The spermatozoa fractions of four of the samples gave either two/three person mixtures

with >100 billion support for suspect contribution. (Use standard wording...)

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- o The spermatozoa fraction of the final ene sample (was this swab) gave a 2P mixture where the known contributor (SAIK complainant) and the suspect were represented (use std wording). -The vulval swab from this SAIK had +1 spermatozoa observed on the Evidence Recovery slide and the spermatozoa fraction gave a single source final result consistent with that matched the suspect.
- The remaining ten of these 36 samples would not have been submitted for DNA testing (either by cell or differential lysis extraction protocols) pre-August 2016. Of these ten samples:
 - o High vaginal sample-S.frac gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect.—From the same SAIK, the second high vaginal, low vaginal, vulval and perianal swabs samples (suggest change throughout) all had spermatozoa detected on the Evidence Recovery slides. The low vaginal and vulval samples gave single source profiles which were consistent with the suspect. The second high vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore failure to submit the first high vaginal sample would not have 2 significantly altered the final results reported for the SAIK.

Low vaginal sample SFrac) gave a complex final result which was not interpreted. The vulval and rectal samples-Sfracs from this SAIK had spermatozoa detected on the Evidence Recovery slide. The rectal swab gave a single source DNA result which was consistent with the suspect. The vulval gave a complex final result which was not interpreted. Given the results of the rectal sample, and vulval sample, failure to submit the low vaginal sample would not have altered the final DNA results for this SAIK.

- o Low vaginal sample SFrac) gave a complex final result which was not interpreted suitable for interpretation. The high vaginal sample SFrac from this SAIK was AP and P30 positive and therefore would have been submitted for differential lysis extraction pre-August 2016. This high vaginal swab gave a 2 person mixture with >100 billion support for contribution from the suspect. The perianal swab was also p30 positive and therefore would have also been submitted for a differential lysis extraction pre-August 2016. The perianal swab gave a single source profile consistent with the suspect. The left nipple sample from this SAIK was also submitted for testing and have a 3 person mixture with >100 billion support for contr bution from the suspect. Therefore failure to submit the low vaginal sample would not have altered the final result for this SAIK.
- Low vaginal sample_SFrac(7

 deconvoluted based on other results from the SAIK gave a 2person mixture conditioned on the complainant (LR >100billion favouring support for contribution by suspect).—The cervical, high vaginal and posterior formix SFracs all gave 1+ spermatozoa on the Evidence Recovery slide and were submitted for differential lysis extraction (consistent with pre-August process). These cervical sample Sfracs each gave a two person conditioned mixtures with >100 billion support for contribution from the suspect. Based on this other samples from this SAIK were not interpreted further.—Therefore failure to submit the low vaginal sample would not have significantly altered the final reported results for this SAIK.
- Low vaginal sample <u>SFrac</u> (690131930) gave a complex final result which was not interpreted (indicates poss 4p). The high vaginal and vulval samples from this SAIK gave >1+ spermatozoa on the Evidence Recovery slide. The vulval sample (Sfrac) gave a complex final result which was not interpreted. The vulval Efrac give a 3p conditioned

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The high vaginal (Sfrac) gave a 2 person mixture, from which the complainant was excluded. The high vaginal mixture appears to be an --1:1 2 person mixture from two males. No suspect reference samples have been submitted, but if they were they could be compared to this mixture. Therefore failure to submit the low vaginal sample would not have altered the final result for this SAIK.

- Tapelift from inside crotch (a) gave a complex result for the spermatozoa fraction which was not interpreted. The epithelial fraction also gave a complex result which was not interpreted. Therefore submission of this sample for cellular or differential lysis extraction would not have altered the final DNA result for this sample.
- A piece of fabric spermatozoa fraction. The epithelial fraction gave a three person mixture with >100 billion support for contr bution from the suspect. Submission of this sample for cell extraction would not have altered the final result for this sample (i.e. suspect DNA located), albeit from cells extraction rather than a spermatozoa fraction. We would be able to talk about probable biological source in court though given the SFrac is single source and attempting to isolate spermatozoa, then 'in my opinion highly likely at least some of DNA matching suspect is from semen'.
- o Low vaginal sample Sfrac) gave a 2p conditioned mixed DNA profile >100billion for suspect which indicates centribution from a male person, but has not been interpreted based on the high vaginal sample result...The high vaginal sample was AP and P30 positive and therefore submitted for differential lysis and the SFrac gave a two person mixture which was conditioned on the complainant, and gave>100billion favouring contribution by the suspect. a UKM1 remaining profile (n.b. reference sample for suspect does not have a final result). The vulval sample had spermatozoa observed on the Evidence Recovery slide and the SFrac gave a 3p mixed DNA profile conditioned on the complainant, and gave>100billion favouring contribution by the suspect with indications of contribution from a male person, but this result has not been interpreted based on the high vaginal sample result. (Note the Vulval Efrac is yet to be interpreted/reviewed do not include in report until result finalised). Therefore based on the high vaginal sample Sfrac result, failure to submit the low vaginal sample would not have significantly altered the final result for this SAIK.
- o High vaginal sample Sfrac) gave a two person mixture which was conditioned on the complainant and the remaining profile was UKM1 (n.b. offender is unknown). The low vaginal and vulval samples both had spermatozoa observed on the Evidence Recovery slide. The low vaginal SFrac gave a similar result to the high vaginal sample SFrac and was-has-not-been interpreted further at this stage. The vulval sample-SFrac was a three person mixture which was conditioned on the complainant, and the remaining profile was reported sa "not suitable for NCIDD load". The <a href="wulval EFrac gave a partial single source DNA profile consistent with the complainant. Therefore based on the low vaginal samples-SFrac result, failure to submit the high vaginal sample would not have significantly altered the final results reported for this SAIK.
- A sanitary pad SFrac (Wait until interp finalised-maybe 3p with repro) with contribution from a male person (not interpreted further as yet, but apparent major is a male contribution). The high vaginal and rectal samples both had no spermatozoa observed on the Evidence Recovery slide and were P30 negative. The high vaginal sample-Sfrac gave a two person mixed DNA profile which was conditioned on the complainant, and the remaining profile was reported as 'unsuitable for NCIDD'. The rectal sample Sfrac and Efrac both-gave a single source profiles which was were consistent with the complainant. Therefore failure to submit the sanitary pad for DNA testing would

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have impacted on the final result of this SAIK, as the sanitary pad was the only sample which gave a profile with a male contribution.

There were 104 samples which for which no spermatozoa were <u>detected located</u> on the <u>Evidence</u>
Recovery slide, but >1+ spermatozoa were <u>detected located</u> on the <u>Differential Lysis slide</u> (i.e. less than
10 spermatozoa observed on the <u>Differential Lysis slide</u>). The results of these 104 samples should be
considered within the context described previously, i.e. that it is expected that the <u>Differential Lysis slides</u>
are more sensitive than the <u>Evidence Recovery slide</u>, and that an change in microscopy result from zero
spermatozoa detected to between one and ten spermatozoa detected may be representative of this
difference in sensitivity. Of these 104 samples:

- 46 would have been submitted for differential lysis extraction pre-August 2016 based on a
 positive P30 result or examination strategy.
- 39 would have been submitted for cell extraction (rather than differential lysis extraction) pre-August 2016. Of these 39 samples:
 - 17 samples-<u>Sfracs</u> gave final results which were complex unsuitable for comparison, partial unsuitable for comparison or no DNA detected. Therefore submission for cell extraction would be unlikely to <u>alter the final result ?and provide more probative</u> information from for these samples.
 - 8 samples-<u>Sfracs</u> gave final results which were single source and were consistent with an
 assumed known contributor. Therefore submission of these samples for cell extraction
 would be unlikely to have <u>significantly</u> altered the final result.
 - Vaginal and anal swab <u>SFrac</u>
 gave a three person mixed profile which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Given that this sample would have been submitted for cell extraction pre-August 2016, it is I kely that a similar result would have been obtained via a cell extraction.
 - vulval sample (695322140) gave a final DNA result which was not interpreted. Sfrac gave 2p conditioned, >100 bill for suspect. The high vaginal and low vaginal samples from this SAIK had spermatozoa observed on the Evidence Recovery slide. The high vaginal gave a three person mixture, the low vaginal gave a two person mixture. Both mixtures were conditioned on the complainant, and gave a remaining profile with >100 billion support for contr bution from the suspect. Therefore submission of the vulval sample for cell extraction would not have altered the final result for this SAIK.
 - o Rectal sample SFrac gave a single source profile consistent with the suspect. The cervical, high vaginal, low vaginal, vulval and perianal samples from this SAIK all had spermatozoa observed on the Evidence Recovery slide and each SFrac sample gave a single source profile consistent with the suspect. Therefore submission of the rectal sample for cell extraction would not have altered the final result for this SAIK.
 - o Vulval and rectal samples <u>Sfracs</u> (6 gave three person mixtures which were conditioned on the complainant, and gave a remaining profile with >100 billion support for contr bution from the suspect. <u>(Vulval Efrac = 3p, conditioned, 6300 LR favouring suspect, Rectal Efrac 3p, cond and LR of 2 for susp.) LVS was AP/p30 positive Other samples in this SAIK (perianal and low vaginal <u>sfracs</u>) gave <u>(2p and 3p)</u>mixtures which were conditioned on the complainant, and gave remaining profiles with support for contr bution from the suspect. It is possible that had the vulval and rectal samples been submitted for cell extraction they could have given a similar final result to that obtained from the differential lysis extraction.</u>

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- Vulval sample (727184992) <u>Sfrac</u> gave a partial profile consistent with the suspect. <u>(Efrac was 3p cond, LR ~140 for susp.)</u> The high and low vaginal samples from this SAIK had spermatozoa observed on the Evidence Recovery slides. The high vaginal sample <u>Sfrac</u> gave a single source profile consistent with the suspect. The low vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore submission of the vulval sample for cell extraction would not have altered the final result for this SAIK.
- Two fabric samples (581798138 and 581798151) Sfracs gave two person mixtures for which no statistical interpretation was performed. The Efrac from gave a 3p, no statistical interpretation was performed. A third scraping from the same fabric gave a single source profile from an UKF1 from the epithelial fraction. It is poss ble that had these two fabrics been submitted for cell extraction that the final result would not be different.
- Vulval sample Sfrac) gave a two person mixture which was conditioned on the complainant, and gave a remaining profile which was consistent with UKM1. Efrac was SS AKC comp't. The low vaginal sample gave an AP/ P30 positive result and was submitted for a differential lysis extraction and Sfrac gave 2p mixed DNA profile which was conditioned on the complainant and gave a remaining profile which is consistent with UKM1. Given the low vaginal Sfrac sample result, and the possibility that the vulval sample would have given a similar result if submitted for a cell extraction, the final result for the SAIK is unlikely to be different if the vulval sample was submitted for cell extraction. (Vulval may have female swamping male if cell ext (given Efrac result)
- Vulval sample Sfrac gave a partial single source profile from designated as UKM1. Vulval EFrac has not been interpreted (Matter withdrawn) but indicates a male/female mixture (would require rework to determine # contr butors. I wouldn't include this sample in report) The high vaginal swab had spermatozoa observed on the Evidence Recovery slide and the SFrac gave a mixed DNA profile with a male contr butor (consistent with UKM1 although not reported). Given the high vaginal sfrac result and the possibility that the vulval sample would have given a similar result if submitted for cell extraction, the final result for the SAIK is unl kely to be different if the vulval sample was submitted for cell extraction.
- o Mouth sample Sfrac gave a single source female profile (I kely the complainant but FTA does not yet have a final result). 2p mix cond compl't, LR supports non-contrib for sus't. EFrac gave SS AKC comp't. The high vaginal sample from the SAIK had spermatozoa observed on the Evidence Recovery slide and Sfrac gave a 2p mixed DNA profile cond on compl't, susp >100bill with a major male contributor. Therefore given the high vaginal sample result and submission of the mouth sample for cell extraction would not have altered the final result for the SAIK.
- 19 samples would not have been submitted for DNA extraction (either cell or differential lysis extraction). Of these 19 samples:
 - 8 samples gave complex unsuitable, partial unsuitable or no DNA detected final results.
 Failure to submit these samples for DNA extraction would not have altered the final result.
 - 5 samples gave single source profiles from an assumed known contributor. Failure to submit these samples for DNA extraction would not have altered the final result.
 - Low vaginal sample <u>SFrac</u> (727539497) gave a <u>2p</u> mixed profile <u>cond on compl't</u> with major contribution from the complainant (which was not interpreted or reported). The high vaginal sample from the SAIK gave a P30 positive result and spermatozoa were detected on the vulval sample on the Evidence Recovery slide. The vulval sample <u>and HV Sfracs</u> <u>both</u> gave a two person mixture which was conditioned on the complainant and gave a

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remaining profile UKM1 (which was loaded to NCIDD). Therefore given the vulval result, and the www.high?vaginal result, failure to submit the low vaginal sample for testing would not have altered the final result for this SAIK.

gave two person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contribution from the suspect (this result was the same as for the perianal <u>EFrac</u> sample and was not reported via EXH). Spermatozoa were detected on the Evidence Recovery slides for the high vaginal 2, low vaginal, vulval, and perianal samples. The high vaginal (2? - need to specify, also applies to HVS 1) gave a similar result to the perianal <u>EFrac</u> and was not reported via EXH. The low vaginal and vulval <u>Sfracs</u> samples both gave single source profiles consistent with the suspect. (Vulval Efrac = 2p cond, LR susp't low support contribn) Given the results of the other samples for this case, and the fact that the

contribn) Given the results of the other samples for this case, and the fact that the endocervix sample was not reported via EXH, (I'd leave this out – only true because no statement request received to date) failure to submit the endocervix sample for testing

would not have altered the final result of the SAIK.

gave a two person mixture which was conditioned on the complainant and gave a remaining male profile which was compared to two suspects for this case but both were excluded. The cervical sample for this case (which it should be noted had no spermatozoa detected on the Evidence Recovery or Differential Lysis slides) Sfrac gave a similar result. The low vaginal sample gave a AP/P30 positive result and the LVS Sfrac gave a similar final result to the cervical and high vaginal Sfrac, Spermatozoa were detected on the rectal sample on the Evidence Recovery slide, but Sfrac gave a complex final result (+ EFrac SS AKC complition. Therefore given the results of the low vaginal sample Sfrac, failure to submit the high vaginal sample for testing would not have altered the final result for this SAIK.

High vaginal sample

) Sfrac gave a two person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contribution from the suspect. The vulval had no spermatozoa detected on the Evidence Recovery slide, but which pre-August 2016 would have been submitted for cell extraction, Vulval Sfrac gave a three person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contr bution from the suspect. Given that the vulval sample may have given a similar result if submitted for cell extraction (rather than differential lysis) failure to submit the high vaginal sample for testing may not have altered the final result for this SAIK. Or including the female DNA in the epi fraction

may have swamped the male DNA
o

o High vaginal sample (a) Sfrac gave two person mixture which was conditioned on the complainant and the remaining profile was used to compare against nominated suspects. The low vaginal sample was AP/P30 positive and the Sfrac gave a three person cond mixture which was also used to compare against suspects. Given the result of the low vaginal sample, failure to submit the high vaginal is not likely to have altered the final result for this SAIK. Change in # contributors may impact (as above)

o Fabric sample Sfrac gave a two person mixture which had >100 billion support for contr bution from the suspect. (EFRAC = complex). This was the only result for this sample, however there are a large number of exhibits in this case with >100 billion support for contr bution from the suspect. Therefore although failure to submit this

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sample would have changed the final result of this sample, there are a number of other exhibits in this case linked to the suspect.

Discussion

The aim of this data analysis was to assess the 738 samples which had no spermatozoa or seminal fluid detected during the initial Evidence Recovery examination, and which were then submitted for differential lysis extraction, and compare these to pre August 2016 examination and sample submission strategies to determine what, if any, impact this may have had on the DNA results reported for the case as a whole.

738 samples has been considered a sufficiently large dataset for the purposes of drawing some general conclusions, although this relies on the particular cases processed during this period, and therefore sampling variability may show a greater or lesser impact by assessing another dataset. Sug. 'due to sampling variation there may have been a greater or lesser impact if another dataset had been assessed. It was beyond the scope of this data analysis to assess slides other than those that were originally zero spermatozoa detected at examination, and were submitted for differential lysis extraction since 8 August 2016.

The focus of this data analysis has been largely from a whole case perspective and several results were considered not to be impacted upon because of other samples/ similar results within the case. Assessing results on a whole case basis is part of standard case management practice, and is a process utilised across all case and sample types. It is acknowledged that the impact on individual samples may be considered significant if semen is not observed at examination, the presumptive screening is also negative and no further action was taken for that sample. There is a risk that if spermatozoa were consequently detected on the differential lysis slide and provided an interpretable DNA result, then potentially a valuable DNA profile for the case may not be obtained. What this data analysis shows is that this risk is mitigated when considering the typical case submission as a whole. The majority of SAIKs/sexual assault cases contain multiple swabs and items, which provide several opportunities to locate semen and subsequently obtain foreign DNA profile that may support an allegation of sexual assault.

Examination strategies are formulated to try and maximise the chances that even if one sample has no spermatozoa observed and the sample truly contains spermatozoa, then the DNA profile information can be obtained through other means. The presumptive screening for seminal fluid and examination strategies for submitting samples for differential lysis or cell extraction (including but not limited to: submission of external swabs/swabs from minors for cell extraction; submitting all areas from an item if one obtains a positive sperm or presumptive result) and also the capacity of STRmix to interpret even mixtures of up to 3 contributors (and including conditioning) all contribute to minimising the overall case impact for a particular sample.

It is acknowledged the slide read at both examination and differential lysis is a detection step, and the sample used to make the slide is a very small amount from the prepared suspension (a drop and 3uL respectively), which is a representation of the spermatozoa that may be present in the sample. For very low levels of spermatozoa, if a second slide is prepared from the sample, lower or higher levels of spermatozoa may be observed, as is to be expected as a consequence of from sampling variability.

The aim of the differential lysis process is to attempt to separate any spermatozoa from any epithelial cells in order to aid in the interpretation of the DNA profiles obtained. While complete separation of the

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spermatozoa fractions and epithelial fractions is the ideal, this is net-often not-the case, and carry-over of epithelial cells into the sperm fraction is commonly observed. The advantage of using STRmix for mixture interpretation helps mitigate the consequences of failing to obtain the ideal separation of spermatozoa and epithelial fractions, which is the aim for differential lysis. In cases where a sample undergoes a cell extraction and the sample does contain spermatozoa, it is reasonable to assume that this extraction process will extract any DNA present in the sample, including from any spermatozoa present. STRmix will similarly aid in the interpretation of any mixed DNA profiles obtained from this process.

As described previously, there is a degree of concentration of spermatozoa in the differential lysis process, and the number of spermatozoa present to give a slide read of <+1 is very low (defined as 'very hard to find spermatozoa'), therefore to go from zero to <+1 after differential lysis may not be unexpected. Similarly a slide read of +1 ('hard to find spermatozoa') after differential lysis, following a zero slide read at examination may not be too concerning or necessarily need to be taken as symptomatic of a problem with ?inefficiencies in the examination slide read process.

Where a zero sperm read has produced a +2 sperm read of the slide after differential lysis, then this is harder to rationalise, even allowing for some variation in the subsample taken for the slide, and the differential lysis concentrating step. In this data there were 7 samples of the 738 total which showed this degree of difference ie. Zero to +2 (easy to find spermatozoa) which equates to 0.95% of this sample set. 6 of these samples would have undergone differential lysis extraction based on the presumptive result, and therefore the DNA results would are expected to have been unchanged. The one sample remaining was a vulval swab, which and would have been submitted for a cell extraction. Within this particular SAIK, the high vaginal and low vaginal swabs both had sperm observed 22t and examination, and from these samples provided a mixed DNA profiles were obtained that STRmix gave Likelihood Ratios of greater than 100 billion favouring with a contribution >100 billion for by the suspect. Given the reasons listed above, for all samples within this data analysis where +2 spermatozoa were detected at differential lysis, the DNA profiling results for the case were not considered to be negatively impacted.

Conclusions

Therefore in summary:

- Of the 738 total samples for which no spermatozoa were detected on the Evidence Recovery slide, 591 also had no spermatozoa detected on the Differential Lysis slide.
- 147 of the 738 samples had spermatozoa detected on the Differential Lysis slide (>1+, 1+ or 2+).

The results of the analysis of this data set have shown that the difference in sensitivity of the Evidence Recovery and Differential Lysis swabs, although acknowledged, has not resulted in a systemic failure with regards to final reported results. (I wouldn't include this phrase-don't think a 'systemic failure' of reported results is the concern.)

For a small proportion of sexual assault samples containing low numbers of spermatozoa, the difference in the sensitivity between the ERT and Diff Microscopy methods had previously caused samples to be

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reported as semen negative when spermatozoa may have been detectable by microscopy following the differential lysis extraction procedure. As the ERT microscopy was previously used as a key determinant as to which extraction method was employed, and indeed whether the samples were submitted for DNA analysis at all, there is a potential impact on a small subset of reported results. Depending on the case circumstances, the ability to report that semen was detected may or may not be critical, in the context of the allegation. For sexually active adults, confirming the presence of spermatozoa on intimate swabs, may perhaps not be considered as critical as it is for sexual assault allegations involving complainants to whom limited opportunities for the transfer of spermatozoa may make such findings more pertinent (for example minors, the elderly or individuals with disabilities).

In many instances we have found that this issue is mitigated by the common practice of submitting multiple swabs within a typical SAIK, thus increasing the chances of detecting semen. The fact that this data analysis has found that the methods in use prior to August 2016 would have had in only a relatively limited impact on the particular reported results for these SAIKS should be reassuring, however we should continue to strive to eliminate impacts of this type for all cases, and seek to improve the sensitivity of our methods, including by selecting the most effective point in the process to conduct microscopy. It may also be the case that if one swab from a SAIK is affected by this issue, then other swabs in the SAIK could also have an increased chance of being similarly affected, as microscopy examinations are not genuinely independent events. A SAIK containing a High Vaginal Swab with low numbers of

There was one sample in the 738 sample data set which would not have been submitted for DNA testing pre-August 2016, and which gave 1+ spermatozoa on the Differential Lysis swab and a final DNA result consistent with the suspect. This was the only DNA result for this case. Pre-August 2016 this sample would have been reported to the QPS as "Semen not detected" and no further action taken. It should be noted that this presumptive EXH advised the QPS that "Spermatozoa were not observed..." rather than advising that there were no spermatozoa present. If deemed critical, the QPS could request further processing of this sample.

spermatozoa. The microscopy process is a manual one, which is in a large measure dependent on the technique and ability of the examining scientist. If the slide created from one SAIK swab is affected by an issue which decreases the chance of observing spermatozoa, then this would also tend to affect the chance of detecting spermatozoa on a slide from a second swab of the same SAIK, made by the same

Therefore, although some individual samples may be negatively impacted as a consequence of the sensitivity of the examination slide process, overall this is considered to be an acceptable risk as it occurs relatively infrequently, and from a case perspective the risk is mitigated by the established practices of multiple sample submissions, examination submission and interpretation strategies.

The results of this study did not demonstrate a systemic failure in the examination of exhibits for seminal fluid. The examination processes described throughout this report, as well as the resulting DNA profile, the assessment of the whole case, and the ability to submit for processing any samples not actioned, aims to mitigate the risk that may arise when spermatozoa is not detected at the examination step. Continuous process improvements are imbedded in Forensic DNA Analysis and are part of our quality management system, and improvements to the examination of sexual assault process will continue, as they will with all processes within the unit, to ensure any risks are mitigated as much as practical. (???)

Commented [LR1]: Not sure if this para is required given previous paras.

General points:

scientist.

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Suggest all wording used to describe stats/ interpretations should be as per standard wording for statements - see Procedure for Release of Results SOP 17119.

e.g. and gave a remaining profile with >100 billion support for contribution from the suspect."

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Suggested wording: "Based on statistical analysis it is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if the suspect has contributed DNA along with the complainant, rather than if he has not."

Suq. leave out identifiers (sample barcodes) where discussing interps/case details, ?Could number the samples based on the order in the xls. and refer to Sample 1, 2 etc. within the report – add appendix/notes to explain.

Sug. leave out any interpretations which have not been finalised (reviewed), and may be subject to change.

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Queensland Health

HealthSupport Queensland

Data Analysis of modified sexual assault process for zero spermatozoa detected at Evidence Recovery

Paula Brisotto, Matthew Hunt, Kylie Rika, Luke Ryan

May 2017

Introduction

Prior to August 2016, exhibits which gave negative results for spermatozoa and seminal fluid were typically not submitted for DNA testing, using the differential lysis extraction protocol (depending on case circumstances and examination strategy these samples may still be submitted for differential lysis extraction or for cell extraction). In August 2016, the Evidence Recovery Team implemented a modified sample submission process for all samples whereby all samples screened for seminal fluid were submitted for DNA testing, using differential lysis extraction, irrespective of the presumptive screening results (i.e. even those samples for which spermatozoa were not located microscopically and P30 tests were negative were still submitted for DNA testing using a differential lysis extraction). The microscopy slides created during the differential lysis extraction were then read by Evidence Recovery Team scientists for final reporting of spermatozoa detection results.

This modified process was introduced after it was raised that the initial slide microscopy conducted during ERT examinations may have a lower sensitivity than that slides produced during the differential lysis extraction process.

This modification process was introduced in response to concerns that the initial microscopy conducted during ERT examinations may be detecting fewer spormatezea than were seen by subsequent microscopy of slidos producod after the differential lysis extraction process.

This data analysis examines the set of samples which had no spermatozoa or seminal fluid detected during the initial Evidence Recovery examination, and which were then submitted for differential lysis extraction. The results from this data set will be used to assess the difference in the pre and post August 2016 examination and sample submission strategies based on the final DNA results and attempt to determine what, if any, impact this may have had on the DNA results reported for the case.

In assessing the potential implications for the DNA results reported, relevant aspects of casemanagement were taken into account, including presumptive screening test results, existing examination strategies for different sample types and other results within the case.



Results

Since August 2016, the sample submission strategy was changed for samples which gave negative presumptive screening results for spermatozoa and seminal fluid (i.e. no spermatozoa observed on microscopy and P30 negative test). All samples which were tested for the presence of spermatozoa and seminal fluid were submitted for DNA extraction using the differential lysis protocol, regardless of the presumptive screening results.

During the period, 8 August 2016 to 28 March 2017, there were 738 samples for which the presumptive screening in Evidence Recovery did not identify spermatozoa during microscopy which were submitted for differential lysis extraction. For each of these 738 samples a microscopy slide was created during the Evidence Recovery examination (the Evidence Recovery slide), and a second slide was created during the differential lysis extraction (the Differential Lysis slide). Both slides were read by Evidence Recovery scientists.

The differences in the methods for creating the Evidence Recovery and Differential Lysis slides are a relevant context to the results of this data mining. Evidence Recovery slides are created by suspending the sample substrate (i.e. swab, scraping, material etc.) in 200μ L to 300μ L of nanopure water. This suspension is then vortexed and agitated with a disposable pipette. One drop (using a disposable pipette) of the suspension is then spotted onto a microscopy slide. The Differential Lysis slides are created by taking 3μ L from the male fraction (post separation from the female fraction) during the differential lysis protocol. The total volume for the male fraction from which the Differential Lysis slide is created is approximately 50μ L. It is expected that the Differential Lysis slide would be more sensitive (in terms of spermatozoa detection) for one main reason: the Differential Lysis slide is created by sampling 3μ L from a total volume of 50μ L of male fraction, therefore any spermatozoa present in the male fraction are at a higher concentration than the Evidence Recovery suspension which has a total volume of 200μ L. Suggest could also briefly cover the categorisation of sperm numbers (how many meant by <1+, 1+ etc.)

Table 1 Differential Lysis slide reads for the presence of sperm.

Original ER read	Diff Lysis Slide read	Number of samples	Percentage of Total
0 sperm	0 sperm	591	80.08
0 sperm	<+1 sperm	104	14.09
0 sperm	+1 sperm	36	4.88
0 sperm	+2 sperm	7	0.95

For 591 of these 738 samples, no spermatozoa were observed on either the Evidence Recovery or the Differential Lysis slides. For these results we can infer that either there were no spermatozoa present, or if they were present they were below the limit of detection for both the Differential Lysis and Evidence Recovery slide preparation techniques.

The remaining 147 slides, for which there were no spermatozoa observed on the Evidence Recovery slide, but where spermatozoa were observed on the Differential Lysis slide, were assessed to determine the impact on final DNA results for that sample.

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Of the 7 samples for which no spermatozoa were located on the Evidence Recovery slide, but +2 spermatozoa were located on the Differential Lysis slide:

- 6 would have been submitted for differential lysis extraction pre-August 2016 based on a positive P30 result
- The-remaining sample was a vulval swab support of identify case details) from a SAIK. The vulval swab gave a 3P mixed DNA profile. Both the high vaginal and low vaginal swabs from this SAIK had spermatozoa observed on the Evidence Recovery slides. The high vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. (Please note: this wording would not be used in a Statement of Witness. The wording used is for the purposes of this report only. Standard wording to describe probability/interpretations for a statement of witness is in QIS document #17119 Procedure for Release of Results.

The low vaginal sample gave a similar result to the high vaginal sample.

Therefore failure to submit the vulval swab would have fimited impact on the final DNA result reported for this SAIK.

For the 36 samples which gave no spermatozoa on the Evidence Recovery slide but +1 spermatozoa on the Differential Lysis slide:

- 19 would have been submitted for differential lysis extraction pre-August 2016 based on positive P30 results.
- Seven of these 36 samples would have been submitted for cell extraction rather than differential lysis extraction pre-August 2016.
 Submission of these seven samples for cell extraction rather than for differential lysis would have limited impact on the final results for these SAIKs because:
 - The spermatozoa fractions of two of these samples gave single source profiles which matched the nominated suspect.
 - The spermatozoa fractions of four of the samples gave either two/three person mixtures with >100 billion support for suspect contribution. (Use standard wording...)
 - The spermatozoa fraction of the final one sample (was this was) gave a 2P mixture where the known contributor (SAIK complainant) and the suspect were represented (use std wording). -The vulval swab from this SAIK had +1 spermatozoa observed on the Evidence Recovery slide and the spermatozoa fraction gave a single source final result consistent with that matched the suspect.
- The remaining ten of these 36 samples would not have been submitted for DNA testing (either by cell or differential lysis extraction protocols) pre-August 2016. Of these ten samples:
 - o High vaginal sample S.frac gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect—From the same SAIK, [The second high vaginal, low vaginal, vulval and perianal swabs samples (suggest change throughout) all had spermatozoa detected on the Evidence Recovery slides. The low vaginal and vulval samples gave single source profiles which were consistent with the suspect. The second high vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore failure to submit the first high vaginal sample would have limited impact on the final results reported for the SAIK.

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Low vaginal sample SFrac) gave a complex final result which was not interpreted. The vulval and rectal samples-Sfracs from this SAIK had spermatozoa detected on the Evidence Recovery slide. The rectal swab gave a single source DNA result which was consistent with the suspect. The vulval gave a complex final result which was not interpreted. Given the results of the rectal sample, and vulval sample, failure to submit the low vaginal sample would not have altered the final DNA results for this SAIK.

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- Low vaginal sample SFrac) gave a complex final result which was not interpreted suitable for interpretation. The high vaginal sample SFrac from this SAIK was AP and P30 positive and therefore would have been submitted for differential lysis extraction pre-August 2016. This high vaginal swab gave a 2 person mixture with >100 billion support for contribution from the suspect. The perianal swab was also p30 positive and therefore would have also been submitted for a differential lysis extraction pre-August 2016. The perianal swab gave a single source profile consistent with the suspect. The left nipple sample from this SAIK was also submitted for testing and have a 3 person mixture with >100 billion support for contribution from the suspect. Therefore failure to submit the low vaginal sample would not have altered the final result for this SAIK.
- Low vaginal sample_SFrac gave a final result which was not interpreted/
 decenveluted based on other results from the SAIK gave a 2person mixture conditioned on the complainant (LR >100billion favouring support for contribution by suspect).—The cervical, high vaginal and posterior formix SFracs all gave 1+ spermatozoa on the Evidence Recovery slide and were submitted for differential lysis extraction (consistent with pre-August process). These cervical sample-Sfracs each gave-a two person conditioned mixtures with >100 billion support for contribution from the suspect. Based on this other samples from this SAIK were not interpreted further.—Therefore failure to submit the low vaginal sample would not have significantly altered the final reported results for this SAIK.
- o Low vaginal sample SFrac gave a complex final result which was not interpreted (indicates poss 4p). The high vaginal and vulval samples from this SAIK gave >1+ spermatozoa on the Evidence Recovery slide. The vulval sample (Sfrac) gave a complex final result which was not interpreted. The vulval Effac give a 3p conditioned
 The high vaginal (Sfrac) gave a 2 person mixture, from which the complainant was excluded. The high vaginal mixture appears to be an --1:1 2 person mixture from two males. No suspect reference samples have been submitted, but if they were they could be compared to this mixture. Therefore failure to submit the low vaginal sample would not have altered the final result for this SAIK.

Tapelift from inside crotch gave a complex result for the spermatozoa fraction which was not interpreted. The epithelial fraction also gave a complex result which was not interpreted. Therefore submission of this sample for cellular or differential lysis extraction would not have altered the final DNA result for this sample.

o A piece of fabric gave a single source profile matching the suspect for the spermatozoa fraction. The epithelial fraction gave a three person mixture with >100 billion support for contr bution from the suspect. Submission of this sample for cell extraction would not have altered impact on the final DNA result for this sample (i.e. suspect DNA located), albeit from cells extraction rather than a spermatozoa fraction. We would be able to talk about probable biological source in court though — given the SFrac is single source and attempting to isolate spermatozoa, then 'in my opinion highly likely at least some of DNA matching suspect is from semen'.

Low vaginal sample <u>Sfracture</u> gave a <u>2p conditioned</u> mixed DNA profile
 <u>>100billion for suspect.</u> which indicates contribution from a male person, but has not been

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interpreted based on the high vaginal sample result._-The high vaginal sample was AP and P30 positive and therefore submitted for differential lysis and the SFrac gave a two person mixture which was conditioned on the complainant, and gave->100billion favouring contribution by the suspect. a UKM1 remaining profile (n.b. reference sample for suspect does not have a final result). The vulval sample had spermatozoa observed on the Evidence Recovery slide and the SFrac gave a 3p mixed DNA profile conditioned on the complainant, and gave>100billion favouring contribution by the suspect with indications of contribution from a male person, but this result has not been interpreted based on the high vaginal sample result. Note the Vulval Efrac is yet to be interpreted/reviewed - do not include in report until result finalised) Therefore based on the high vaginal sample Sfrac result, failure to submit the low vaginal sample would not have significantly altered the final result for this SAIK.

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High vaginal sample Sfrac) gave a two person mixture which was conditioned on the complainant and the remaining profile was UKM1 (n.b. offender is unknown). The low vaginal and vulval samples both had spermatozoa observed on the Evidence Recovery slide. The low vaginal SFrac gave a similar result to the high vaginal sample SFrac and was has not been interpreted further at this stage. The vulval sample SFrac was a three person mixture which was conditioned on the complainant, and the remaining profile was reported as 'not suitable for NCIDD load'. The vulval EFrac gave a partial single source DNA profile consistent with the complainant. Therefore based on the low vaginal samples-SFrac result, failure to submit the high vaginal sample would not have

significantly altered the final results reported for this SAIK.

 A sanitary pad <u>SFrac</u> gave a two person mixture (Wait until interp finalised-maybe 3p with repro) with contribution from a male person (not interpreted further as yet, but apparent major is a male contribution). The high vaginal and rectal samples both had no spermatozoa observed on the Evidence Recovery slide and were P30 negative. The high vaginal sample-Sfrac gave a two person mixed DNA profile which was conditioned on the complainant, and the remaining profile was reported as 'unsuitable for NCIDD'. The rectal sampleSfrac and Efrac both-gave a single source profiles which was were consistent with the complainant. Therefore failure to submit the sanitary pad for DNA testing would have impacted on the final result of this SAIK, as the sanitary pad was the only sample which gave a profile with a male contribution.

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There were 104 samples which for which no spermatozoa were detected located on the Evidence Recovery slide, but >1+ spermatozoa were detected located on the Differential Lysis slide (i.e. less than 10 spermatozoa observed on the Differential Lysis slide). The results of these 104 samples should be considered within the context described previously, i.e. that it is expected that the Differential Lysis slides are more sensitive than the Evidence Recovery slide, and that an change in microscopy result from zero spermatozoa detected to between one and ten spermatozoa detected may be representative of this difference in sensitivity. Of these 104 samples:

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- 46 would have been submitted for differential lysis extraction pre-August 2016 based on a positive P30 result or examination strategy.
- 39 would have been submitted for cell extraction (rather than differential lysis extraction) pre-August 2016. Of these 39 samples:
 - o 17 samples-Sfracs gave final results which were complex unsuitable for comparison, partial unsuitable for comparison or no DNA detected. Therefore submission for cell

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extraction would be unl kely to alter the final result ?and provide more probative information from for these samples.

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- 8 samples-<u>Sfracs</u> gave final results which were single source and were consistent with an
 assumed known contributor. Therefore submission of these samples for cell extraction
 would be unlikely to have <u>significantly</u> altered the final result.
- Vaginal and anal swab <u>SFrac</u>
 gave a three person mixed profile which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Given that this sample would have been submitted for cell extraction pre-August 2016, it is I kely that a similar result would have been obtained via a cell extraction.
- vulval sample <u>gave a final DNA result which was not interpreted.</u> <u>Sfrac gave 2p conditioned, >100 bill for suspect.</u> The high vaginal and low vaginal samples from this SAIK had spermatozoa observed on the Evidence Recovery slide. The high vaginal gave a three person mixture, the low vaginal gave a two person mixture. Both mixtures were conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore submission of the vulval sample for cell extraction would not have altered the final result for this SAIK.
- Rectal sample SFrac gave a single source profile consistent with the suspect. The cervical, high vaginal, low vaginal, vulval and perianal samples from this SAIK all had spermatozoa observed on the Evidence Recovery slide and each SFrac sample gave a single source profile consistent with the suspect. Therefore submission of the rectal sample for cell extraction would not have altered the final result for this SAIK.
- o Vulval and rectal samples Sfracs gave three person mixtures which were conditioned on the complainant, and gave a remaining profile with >100 billion support for contr bution from the suspect. (Vulval Efrac = 3p, conditioned, 6300 LR favouring suspect. Rectal Efrac 3p, cond and LR of 2 for susp.) LVS was AP/p30 positive Other samples in this SAIK (perianal and low vaginal sfracs) gave <a href="quad 2p and 3p)mixtures which were conditioned on the complainant, and gave remaining profiles with support for contr bution from the suspect. It is possible that had the vulval and rectal samples been submitted for cell extraction they could have given a similar final result to that obtained from the differential lysis extraction.
- Vulval sample) <u>Sfrac gave a partial profile consistent with the suspect (Efrac was 3p cond, LR ~140 for susp)</u> The high and low vaginal samples from this SAIK had spermatozoa observed on the Evidence Recovery slides. The high vaginal sample <u>Sfrac gave a single source profile consistent with the suspect.</u> The low vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore submission of the vulval sample for cell extraction would not have altered the final result for this SAIK.
- Two fabric samples

) Stracs gave two person mixtures for which no statistical interpretation was performed. The Efrac from gave a 3p, no statistical interpretation was performed. A third scraping from the same fabric gave a single source profile from an UKF1 from the epithelial fraction. It is possible that had these two fabrics been submitted for cell extraction that the final result would not be different
- o Vulval sample Sfrac gave a two person mixture which was conditioned on the complainant, and gave a remaining profile which was consistent with UKM1. Effac was SS AKC compt. The low vaginal sample gave an AP/ P30 positive result and was submitted for a differential lysis extraction and Sfrac gave 2p mixed DNA profile which was conditioned on the complainant and gave a remaining profile which is consistent with UKM1. Given the low vaginal Sfrac sample result, and the possibility that the vulval

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- sample would have given a similar result if submitted for a cell extraction, the final result for the SAIK is unlikely to be different if the vulval sample was submitted for cell extraction. (Vulval may have female swamping male if cell ext (given Efrac result)
- o Vulval sample Sfrac gave a partial single source profile from designated as UKM1. Vulval EFrac has not been interpreted (Matter withdrawn) but indicates a male/female mixture (would require rework to determine # contr butors. I wouldn't include this sample in report) The high vaginal swab had spermatozoa observed on the Evidence Recovery slide and the SFrac gave a mixed DNA profile with a male contr butor (consistent with UKM1 although not reported). Given the high vaginal sfrac result and the possibility that the vulval sample would have given a similar result if submitted for cell extraction, the final result for the SAIK is unlikely to be different if the vulval sample was submitted for cell extraction.
- o Mouth sample Sfrac) gave a single source female profile (I kely the complainant but FTA does not yot have a final result). 2p mix cond compl't, LR supports non-contrib for sus't. EFrac gave SS AKC comp't. The high vaginal sample from the SAIK had spermatozoa observed on the Evidence Recovery slide and Sfrac gave a 2p mixed DNA profile cond on compl't, susp >100bill with a major male contributor. Therefore given the high vaginal sample result and submission of the mouth sample for cell extraction would not have altered the final result for the SAIK.
- 19 samples would not have been submitted for DNA extraction (either cell or differential lysis extraction). Of these 19 samples:
 - 8 samples gave complex unsuitable, partial unsuitable or no DNA detected final results.
 Failure to submit these samples for DNA extraction would not have altered the final result.
 - 5 samples gave single source profiles from an assumed known contributor. Failure to submit these samples for DNA extraction would not have altered the final result.
 - Low vaginal sample SFrac) gave a 2p mixed profile cond on complit with major contribution from the complainant (which was not interpreted or reported). The high vaginal sample from the SAIK gave a P30 positive result and spermatozoa were detected on the vulval sample on the Evidence Recovery slide. The vulval sample and HV Sfracs both gave a two person mixture which was conditioned on the complainant and gave a remaining profile UKM1 (which was loaded to NCIDD). Therefore given the vulval result, and the low-High? vaginal result, failure to submit the low vaginal sample for testing would not have altered the final result for this SAIK.
 - o Endocervix sample SFrac gave two person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contribution from the suspect (this result was the same as for the perianal Frac sample and was not reported via EXH). Spermatozoa were detected on the Evidence Recovery slides for the high vaginal 2, low vaginal, vulval, and perianal samples. The high vaginal (2? need to specify, also applies to HVS 1) gave a similar result to the perianal Frac and was not reported via EXH. The low vaginal and vulval Sfracs samples both gave single source profiles consistent with the suspect. (Vulval Efrac = 2p cond, LR susp't low support contribn) Given the results of the other samples for this case, and the fact that the endocervix sample was not reported via EXH, (I'd leave this out only true because no
 - _ High vaginal sample Sfrac gave a two person mixture which was conditioned on the complainant and gave a remaining male profile which was compared to two suspects for this case but both were excluded. The cervical sample for this case

would not have altered the final result of the SAIK.

statement request received to date) failure to submit the endocervix sample for testing

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(which it should be noted had no spermatozoa detected on the Evidence Recovery or Differential Lysis slides) Sfrac gave a similar result. The low vaginal sample gave a APIP30 positive result and the LVS Sfrac gave a similar final result to the cervical and high vaginal Sfrac spermatozoa were detected on the rectal sample on the Evidence Recovery slide, but Sfrac gave a complex final result (+ EFFac SS AKC complitit). Therefore given the results of the low vaginal sample Sfrac, failure to submit the high vaginal sample for testing would not have altered the final result for this SAIK.

- Sfrac gave a two person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contribution from the suspect. The vulval had no spermatozoa detected on the Evidence Recovery slide, but which pre-August 2016 would have been submitted for cell extraction, Vulval Sfrac gave a three person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contr bution from the suspect. Given that the vulval sample may have given a similar result if submitted for cell extraction (rather than differential lysis) failure to submit the high vaginal sample for testing may not have altered the final result for this SAIK. Or including the female DNA in the epi fraction may have swamped the male DNA
- o High vaginal sample (690132011) <u>Sfrac gave two person mixture which was conditioned on the complainant and the remaining profile was used to compare against nominated suspects. The low vaginal sample was <u>AP/P30</u> positive and <u>the Sfrac gave a three person cond mixture which was also used to compare against suspects. Given the result of the low vaginal sample, failure to submit the high vaginal is not likely to have altered the final result for this SAIK. Change in # contributors may impact (as above)</u></u>
- Fabric sample Sfrac gave a two person mixture which had >100 billion support for contr bution from the suspect. (EFRAC = complex). This was the only result for this sample, however there are a large number of exhibits in this case with >100 billion support for contr bution from the suspect. Therefore although failure to submit this sample would have changed the final result of this sample, there are a number of other exhibits in this case linked to the suspect.

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Discussion

The aim of this data analysis was to assess the 738 samples which had no spermatozoa or seminal fluid detected during the initial Evidence Recovery examination, and which were then submitted for differential lysis extraction, and compare these to pre August 2016 examination and sample submission strategies to determine what, if any, impact this may have had on the DNA results reported for the case as a whole.

738 samples has been considered a sufficiently large dataset for the purposes of drawing some general conclusions, although this relies on the particular cases processed during this period, and therefore due to sampling variation there may have been a greater or lesser impact if another dataset had been assessed. It was beyond the scope of this data analysis to assess slides other than those that were originally zero spermatozoa detected at examination, and were submitted for differential lysis extraction since 8 August 2016.

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The focus of this data analysis has been largely from a whole case perspective and several results were considered not to be impacted upon because of other samples/ similar results within the case. Assessing results on a whole case basis is part of standard case management practice, and is a process utilised across all case and sample types. It is acknowledged that the impact on individual samples may be considered significant if semen is not observed at examination, the presumptive screening is also negative and no further action was taken for that sample. There is a risk that if spermatozoa were consequently detected on the differential lysis slide and provided an interpretable DNA result, then potentially a valuable DNA profile for the case may not be obtained. What this data analysis shows is that this risk is mitigated when considering the typical case submission as a whole. The majority of SAIKs/sexual assault cases contain multiple swabs and items, which provide several opportunities to locate semen and subsequently obtain foreign DNA profile that may support an allegation of sexual assault

Examination strategies are formulated to try and maximise the chances that even if one sample has no spermatozoa observed and the sample truly contains spermatozoa, then the DNA profile information can be obtained through other means. The presumptive screening for seminal fluid and examination strategies for submitting samples for differential lysis or cell extraction (including but not limited to: submission of external swabs/swabs from minors for cell extraction; submitting all areas from an item if one obtains a positive sperm or presumptive result) and also the capacity of STRmix to interpret mixtures of up to 3 contributors (and including conditioning) all contribute to minimising the overall case impact for a particular sample.

It is acknowledged the slide read at both examination and differential lysis is a detection step, and the sample used to make the slide is a very small amount from the prepared suspension (a drop and 3uL respectively), which is a representation of the spermatozoa that may be present in the sample. For very low levels of spermatozoa, if a second slide is prepared from the sample, lower or higher levels of spermatozoa may be observed, as is to be expected as a consequence of sampling variability.

The aim of the differential lysis process is to attempt to separate any spermatozoa from any epithelial cells in order to aid in the interpretation of the DNA profiles obtained. While complete separation of the spermatozoa fractions and epithelial fractions is the ideal, this is often not the case, and carry-over of epithelial cells into the sperm fraction is commonly observed. The advantage of using STRmix for mixture interpretation helps mitigate the consequences of failing to obtain the ideal separation of spermatozoa and epithelial fractions, which is the aim for differential lysis. In cases where a sample undergoes a cell extraction and the sample does contain spermatozoa, it is reasonable to assume that this extraction process will extract any DNA present in the sample, including from any spermatozoa present. STRmix will similarly aid in the interpretation of any mixed DNA profiles obtained from this process.

As described previously, there is a degree of concentration of spermatozoa in the differential lysis process, and the number of spermatozoa present to give a slide read of <+1 is very low (defined as 'very hard to find spermatozoa'), therefore to go from zero to <+1 after differential lysis may not be unexpected. Similarly a slide read of +1 ('hard to find spermatozoa') after differential lysis, following a zero slide read at examination may not be too concerning or necessarily need to be taken as symptomatic of inefficiencies in the examination slide read process.

Where a zero sperm read has produced a +2 sperm read of the slide after differential lysis, then this is harder to rationalise, even allowing for some variation in the subsample taken for the slide, and the differential lysis concentrating step. In this data there were 7 samples of the 738 total which showed this degree of difference ie. Zero to +2 (easy to find spermatozoa) which equates to 0.95% of this sample set. 6 of these samples would have undergone differential lysis extraction based on the presumptive

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result, and therefore the DNA results are expected to have been unchanged. The one sample remaining was a vulval swab, which would have been submitted for a cell extraction. Within this particular SAIK, the high vaginal and low vaginal swabs both had sperm observed at examination, and from these samples mixed DNA profiles were obtained that STRmix gave L kelihood Ratios of greater than 100 billion favouring contribution by the suspect. Given the reasons listed above, for all samples within this data analysis where +2 spermatozoa were detected at differential lysis, the DNA profiling results for the case were not considered to be negatively impacted.

Conclusions

The purposes of the examination process in the Evidence Recovery Team is to attempt to identify areas of biological material for submission to the Analytical team for DNA processing.

The processes for the detection of spermatozoa and seminal fluid within the Evidence Recovery Team......(summary around what is the actual purpose – to detect sperm for submission to Analytical for diff processing.) Whilst the observations of spermatozoa at ER of diff stage is a confirmation of their presence, the absence of sperm is not confirmation of their absence. It means that spermatozoa, if present, where at a level that was not able to be detected.

Etc etc - this could be fit in somewhere below....?

Therefore in summary:

- Of the 738 total samples for which no spermatozoa were detected on the Evidence Recovery slide, 591 also had no spermatozoa detected on the Differential Lysis slide.
- 147 of the 738 samples had spermatozoa detected on the Differential Lysis slide (>1+, 1+ or 2+).
- Of these 147 slides, 1 sample (a sanitary pad 688640090), gave a final result which would not
 have been obtained pre-August 2016. I.e. the decreased sensitivity of the Evidence Recovery
 slide (when compared to the Differential Lysis slide) resulted would have resulted in the sanitary
 pad sample not being submitted for DNA testing pre-August 2016.

The results of the analysis of this data set have shown that the difference in sensitivity of the Evidence Recovery and Differential Lysis swabs, although acknowledged, has not resulted in a systemic failure with regards to final reported results. (I wouldn't include this phrase-don't think a 'systemic failure' of reported results is the concern.)

For a small proportion of sexual assault samples containing low numbers of spermatozoa, the difference in the sensitivity between the ERT and Diff Microscopy methods had previously caused samples to be reported as semen negative when spermatozoa may have been detectable by microscopy following the differential lysis extraction procedure. As the ERT microscopy was previously used as a key determinant as to which extraction method was employed, and indeed whether the samples were submitted for DNA analysis at all, there is a potential impact on a small subset of reported results. Depending on the case circumstances, the ability to report that semen was detected may or may not be critical, in the context of the allegation. For sexually active adults, confirming the presence of spermatozoa on intimate swabs, may perhaps not be considered as critical as it is for sexual assault allegations involving complainants to

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whom limited opportunities for the transfer of spermatozoa may make such findings more pertinent (for example minors, the elderly or individuals with disabilities).

In many instances we have found that this issue is mitigated by the common practice of submitting multiple swabs within a typical SAIK, thus increasing the chances of detecting semen. The fact that this data analysis has found that the methods in use prior to August 2016 would have had in only a relatively limited impact on the particular reported results for these SAIKS should be reassuring, however we should continue to strive to eliminate impacts of this type for all cases, and seek to improve the sensitivity of our methods, including by selecting the most effective point in the process to conduct microscopy. It may also be the case that if one swab from a SAIK is affected by this issue, then other swabs in the SAIK could also have an increased chance of being similarly affected, as microscopy examinations are not genuinely independent events. A SAIK containing a High Vaginal Swab with low numbers of spermatozoa may be more likely to also contain a Low Vaginal Swab with low numbers of spermatozoa. The microscopy process is a manual one, which is in a large measure dependent on the technique and ability of the examining scientist. If the slide created from one SAIK swab is affected by an issue which decreases the chance of observing spermatozoa, then this would also tend to affect the chance of detecting spermatozoa on a slide from a second swab of the same SAIK, made by the same scientist.

There was one sample in the 738 sample data set which would not have been submitted for DNA testing pre-August 2016, and which gave 1+ spermatozoa on the Differential Lysis swab and a final DNA result consistent with the suspect. This was the only DNA result for this case. Pre-August 2016 this sample would have been reported to the QPS as "Semen not detected" and no further action taken. It should be noted that this presumptive EXH advised the QPS that "Spermatozoa were not observed..." rather than advising that there were no spermatozoa present. If deemed critical, the QPS could request further processing of this sample.

Therefore, although some individual samples may be negatively impacted as a consequence of the sensitivity of the examination slide process, overall this is considered to be an acceptable risk as it occurs relatively infrequently, and from a case perspective the risk is mitigated by the established practices of multiple sample submissions, examination submission and interpretation strategies.

The results of this study did not demonstrate a systemic failure in the examination of exhibits for seminal fluid. The examination processes described throughout this report, as well as the resulting DNA profile, the assessment of the whole case, and the ability to submit for processing any samples not actioned, aims to mitigate the risk that may arise when spermatozoa is not detected at the examination step. Continuous process improvements are imbedded in Forensic DNA Analysis and are part of our quality management system, and improvements to the examination of sexual assault process will continue, as they will with all processes within the unit, to ensure any risks are mitigated as much as practical. (???)

Commented [LR6]: Not sure if this para is required given previous paras

General points:

Suggest all wording used to describe stats/ interpretations should be as per standard wording for statements - see Procedure for Release of Results SOP 17119.

e.g. "and gave a remaining profile with >100 billion support for contribution from the suspect."

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Suggested wording: "Based on statistical analysis it is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if the suspect has contributed DNA along with the complainant, rather than if he has not."

Sug. leave out identifiers (sample barcodes) where discussing interps/case details. ?Could number the samples based on the order in the xls. and refer to Sample 1, 2 etc. within the report – add appendix/notes to explain.

Sug. leave out any interpretations which have not been finalised (reviewed), and may be subject to change. Will check on their status

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Data Analysis of modified sexual assault process for zero spermatozoa detected at Evidence Recovery

Paula Brisotto, Matthew Hunt, Kylie Rika, Luke Ryan

May 2017

Introduction

Prior to August 2016, exhibits which gave negative results for spermatozoa and seminal fluid were typically not submitted for DNA testing, using the differential lysis extraction protocol (depending on case circumstances and examination strategy these samples may still be submitted for differential lysis extraction or for cell extraction). In August 2016, the Evidence Recovery Team implemented a modified sample submission process for all samples whereby all samples screened for seminal fluid were submitted for DNA testing, using differential lysis extraction, irrespective of the presumptive screening results (i.e. even those samples for which spermatozoa were not located microscopically and P30 tests were negative were still submitted for DNA testing using a differential lysis extraction). The microscopy slides created during the differential lysis extraction were then read by Evidence Recovery Team scientists for final reporting of spermatozoa detection results.

This modification process was introduced in response to concerns that the initial microscopy conducted during ERT examinations may be detecting fewer spermatozoa than were seen by subsequent microscopy of slides produced after the differential lysis extraction process.

This data analysis examines the set of samples which had no spermatozoa or seminal fluid detected during the initial Evidence Recovery examination, and which were then submitted for differential lysis extraction. The results from this data set will be used to assess the difference in the pre and post August 2016 examination and sample submission strategies based on the final DNA results and attempt to determine what, if any, impact this may have had on the DNA results reported for the case.

In assessing the potential implications for the DNA results reported, relevant aspects of casemanagement were taken into account, including presumptive screening test results, existing examination strategies for different sample types and other results within the case.

Results

Since August 2016, the sample submission strategy was changed for samples which gave negative presumptive screening results for spermatozoa and seminal fluid (i.e. no spermatozoa observed on microscopy and P30 negative test). All samples which were tested for the presence of spermatozoa and



seminal fluid were submitted for DNA extraction using the differential lysis protocol, regardless of the presumptive screen results.

During the period, 8 August 2016 to 28 March 2017, there were 738 samples for which the presumptive screening in Evidence Recovery did not identify spermatozoa during microscopy which were submitted for differential lysis extraction. For each of these 738 a microscopy slide was created during the Evidence Recovery examination (the Evidence Recovery slide), and a second slide was created during the differential lysis extraction (the Differential Lysis slide). Both slides were read by Evidence Recovery scientists.

The differences in the methods for creating the Evidence Recovery and Differential Lysis slides are a relevant context to the results of this data mining. Evidence Recovery slides are created by suspending the sample substrate (i.e. swab, scraping, material etc.) $200\mu L$ to $300\mu L$ of nanopure water. This suspension is then vortexed and agitated with a disposable pipette. One drop (using a disposable pipette) of the suspension is then spotted onto a microscopy slide. The Differential Lysis slides are created by taking $3\mu L$ from the male fraction (post separation from the female fraction) during the differential lysis protocol. The total volume for the male fraction when the Differential Lysis slide is created is approximately $50\mu L$. It is expected that the Differential Lysis slide would be more sensitive (in terms of spermatozoa detection) for one main reason: the Differential Lysis slide is created by sampling $3\mu L$ from a total volume of $50\mu L$ of male fraction, therefore any spermatozoa present in the male fraction are at a higher concentration than the Evidence Recovery suspension which has a total volume of $200\mu L$.

Table 1 Differential L	ysis slide reads for the	presence of sperm.

Original ER read	Diff Lysis Slide read	Number of samples	Percentage of Total
0 sperm	0 sperm	591	80.08
0 sperm	<+1 sperm	104	14.09
0 sperm	+1 sperm	36	4.88
0 sperm	+2 sperm	7	0.95

Of these 738 samples, no spermatozoa were observed on both the Evidence Recovery and Differential Lysis slides for 591 samples. This means that for 591 samples, either there were no spermatozoa present, or if they were present they were below the limit of detection for both the Differential Lysis and Evidence Recovery slide preparation techniques.

The remaining 147 slides, for which there were no spermatozoa observed on the Evidence Recovery slide, but where spermatozoa were observed on the Differential Lysis slide, were assessed to determine the impact on final DNA results for that sample.

Of the 7 samples for which no spermatozoa were located on the Evidence Recovery slide, but +2 spermatozoa were located on the Differential Lysis slide:

- 6 would have been submitted for differential lysis extraction pre-August 2016 based on a positive P30 result.
- The remaining sample was a vulval sample (makes a from a SAIK. The vulval sample gave a 3P mixed DNA profile (not yet reported). The high vaginal and low vaginal samples had spermatozoa observed on the Evidence Recovery slides. The high vaginal sample gave a two

person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. The low vaginal sample gave a similar result to the high vaginal sample. Therefore failure to submit the vulval sample would not have altered the final result for the SAIK.

For the 36 samples which gave no spermatozoa on the Evidence Recovery slide but +1 spermatozoa on the Differential Lysis slide:

- 19 would have been submitted for differential lysis extraction pre-August 2016 based on positive P30 results.
- Seven of these 36 samples would have been submitted for cell extraction rather than differential lysis extraction pre-August 2016. Submission of these seven samples for cell extraction rather than for differential lysis would not have altered the final results for these SAIKs because:
 - o two gave single source profiles consistent with the suspect.
 - o four gave either two/three person mixtures with >100 billion support for suspect contribution.
 - one sample (a perianal SAIK swab) gave a 2P mixture where the known contributor (SAIK complainant) and the suspect were represented. The vulval swab from this SAIK had +1 spermatozoa observed on the Evidence Recovery slide and gave a single source final result consistent with the suspect.
- The remaining ten of these 36 samples would not have been submitted for DNA testing (either by cell or differential lysis extraction protocols) pre-August 2016. Of these ten samples:
 - High vaginal sample () gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. The second high vaginal, low vaginal, vulval and perianal samples all had spermatozoa detected on the Evidence Recovery slides. The low vaginal and vulval samples gave single source profiles which were consistent with the suspect. The second high vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore failure to submit the first high vaginal sample would not have altered the final result for the SAIK.
 - o Low vaginal sample gave a complex final result which was not interpreted. The vulval and rectal samples from this SAIK had spermatozoa detected on the Evidence Recovery slide. The rectal swab gave a single source DNA result which was consistent with the suspect. The vulval gave a complex final result which was not interpreted. Given the results of the rectal sample, and vulval sample, failure to submit the low vaginal sample would not have altered the final DNA results for this SAIK.
 - O Low vaginal sample gave a complex final result which was not interpreted. The high vaginal sample from this SAIK was P30 positive and therefore would have been submitted for differential lysis extraction pre-August 2016. This high vaginal swab gave a 2 person mixture with >100 billion support for contribution from the suspect. The perianal swab was also p30 positive and therefore would have also been submitted for a differential lysis extraction pre-August 2016. The perianal swab gave a single source profile consistent with the suspect. The left nipple sample from this SAIK was also submitted for testing and have a 3 person mixture with >100 billion support for contribution from the suspect. Therefore failure to submit the low vaginal sample would not have altered the final result for this SAIK.
 - be Low vaginal sample gave a final result which was not interpreted/deconvoluted based on other results from the SAIK. The cervical, high vaginal and

posterior fornix all gave 1+ spermatozoa on the Evidence Recovery slide and were submitted for differential lysis extraction (consistent with pre-August process). The cervical sample gave a two person mixture with >100 billion support for contribution from the suspect. Based on this other samples from this SAIK were not interpreted further. Therefore failure to submit the low vaginal sample would not have altered the final result for this SAIK.

- o Low vaginal sample gave a complex final result which was not interpreted. The high vaginal and vulval samples from this SAIK gave >1+ spermatozoa on the Evidence Recovery slide. The vulval sample gave a complex final result which was not interpreted. The high vaginal gave a 2 person mixture, from which the complainant was excluded. The high vaginal mixture appears to be an 1:1 2 person mixture from two males. No suspect reference samples have been submitted, but if they were they could be compared to this mixture. Therefore failure to submit the low vaginal sample would not have altered the final result for this SAIK.
- o Tapelift from inside crotch gave a complex result for the spermatozoa fraction which was not interpreted. The epithelial fraction also gave a complex result which was not interpreted. Therefore submission of this sample for cellular or differential lysis extraction would not have altered the final DNA result for this sample.
- o A piece of fabric gave a single source profile matching the suspect for the spermatozoa fraction. The epithelial fraction gave a three person mixture with >100 billion support for contribution from the suspect. Submission of this sample for cell extraction would not have altered the final result for this sample (i.e. suspect DNA located), albeit from cells extraction rather than a spermatozoa fraction.
- o Low vaginal sample gave a mixed DNA profile which indicates contribution from a male person, but has not been interpreted based on the high vaginal sample result. The high vaginal sample was P30 positive and therefore submitted for differential lysis and gave a two person mixture which was conditioned on the complainant, and gave a UKM1 remaining profile (n.b. reference sample for suspect does not have a final result). The vulval sample had spermatozoa observed on the Evidence Recovery slide and gave a mixed DNA profile with indications of contribution from a male person, but this result has not been interpreted based on the high vaginal sample result. Therefore based on the high vaginal sample would not have altered the final result for this SAIK.
- o High vaginal sample gave a two person mixture which was conditioned on the complainant and the remaining profile was UKM1 (n.b. offender is unknown). The low vaginal and vulval samples both had spermatozoa observed on the Evidence Recovery slide. The low vaginal gave a similar result to the high vaginal sample and was not interpreted further. The vulval sample was a three person mixture which was conditioned on the complainant, and the remaining profile was not suitable for NCIDD load. Therefore based on the low vaginal samples result, failure to submit the high vaginal sample would not have altered the final result for this SAIK.
- A sanitary pad grave (a) gave a two person mixture with contribution from a male person (not interpreted further as yet, but apparent major is a male contribution). The high vaginal and rectal samples both had no spermatozoa observed on the Evidence Recovery slide and were P30 negative. The high vaginal sample gave a two person mixed DNA profile which was conditioned on the complainant, and the remaining profile was unsuitable for NCIDD. The rectal sample gave a single source profile which was consistent with the complainant. Therefore failure to submit the sanitary pad for DNA testing would have impacted

on the final result of this SAIK, as the sanitary pad was the only sample which gave a profile with a male contribution.

There were 104 samples which for which no spermatozoa were located on the Evidence Recovery slide, but >1+ spermatozoa were located on the Differential Lysis slide (i.e. less than 10 spermatozoa observed on the Differential Lysis slide). The results of these 104 samples should be considered within the context described previously, i.e. that it is expected that the Differential Lysis slides are more sensitive than the Evidence Recovery slide, and that an change in microscopy result from zero spermatozoa detected to between one and ten spermatozoa detected may be representative of this difference in sensitivity. Of these 104 samples:

- 46 would have been submitted for differential lysis extraction pre-August 2016 based on a positive P30 result or examination strategy.
- 39 would have been submitted for cell extraction (rather than differential lysis extraction) pre-August 2016. Of these 39 samples:
 - 17 samples gave final results which were complex unsuitable for comparison, partial unsuitable for comparison or no DNA detected. Therefore submission for cell extraction would be unlikely to alter the final result for these samples.
 - 8 samples gave final results which were single source and were consistent with an assumed known contributor. Therefore submission of these samples for cell extraction would be unlikely to have altered the final result.
 - Vaginal and anal swab () gave a three person mixed profile which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Given that this sample would have been submitted for cell extraction pre-August 2016, it is likely that a similar result would have been obtained via a cell extraction.
 - Vulval sample (gave a final DNA result which was not interpreted. The high vaginal and low vaginal samples from this SAIK had spermatozoa observed on the Evidence Recovery slide. The high vaginal gave a three person mixture, the low vaginal gave a two person mixture. Both mixtures were conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore submission of the vulval sample for cell extraction would not have altered the final result for this SAIK.
 - o Rectal sample) gave a single source profile consistent with the suspect. The cervical, high vaginal, low vaginal, vulval and perianal samples from this SAIK all had spermatozoa observed on the Evidence Recovery slide and each sample gave a single source profile consistent with the suspect. Therefore submission of the rectal sample for cell extraction would not have altered the final result for this SAIK.
 - o Vulval and rectal samples (gave three person mixtures which were conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Other samples in this SAIK (perianal and low vaginal) gave mixtures which were conditioned on the complainant, and gave remaining profiles with support for contribution from the suspect. It is possible that had the vulval and rectal samples been submitted for cell extraction they could have given a similar final result to that obtained from the differential lysis extraction.
 - Vulval sample (a) gave a partial profile consistent with the suspect. The high and low vaginal samples from this SAIK had spermatozoa observed on the Evidence Recovery slides. The high vaginal sample gave a single source profile consistent with the suspect. The low vaginal sample gave a two person mixture which was conditioned on

the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore submission of the vulval sample for cell extraction would not have altered the final result for this SAIK.

- Two fabric samples () gave two person mixtures for which no statistical interpretation was performed. A third scraping from the same fabric gave a single source profile from an UKF1 from the epithelial fraction. It is possible that had these two fabrics been submitted for cell extraction that the final result would not be different.
- Vulval sample () gave a two person mixture which was conditioned on the complainant, and gave a remaining profile which was consistent with UKM1. The low vaginal sample gave a P30 positive result and was submitted for a differential lysis extraction and gave mixed DNA profile which was conditioned on the complainant and gave a remaining profile which is consistent with UKM1. Given the low vaginal sample result, and the possibility that the vulval sample would have given a similar result if submitted for a cell extraction, the final result for the SAIK is unlikely to be different if the vulval sample was submitted for cell extraction.
- Vulval sample () gave a single source profile from UKM1. The high vaginal had spermatozoa observed on the Evidence Recovery slide and gave a mixed DNA profile with a male contributor (consistent with UKM1 although not reported). Given the high vaginal result and the possibility that the vulval sample would have given a similar result if submitted for cell extraction, the final result for the SAIK is unlikely to be different if the vulval sample was submitted for cell extraction.
- Mouth sample () gave a single source female profile (likely the complainant but FTA does not yet have a final result). The high vaginal sample from the SAIK had spermatozoa observed on the Evidence Recovery slide and gave a mixed DNA profile with a major male contributor. Therefore given the high vaginal sample result and submission of the mouth sample for cell extraction would not have altered the final result for the SAIK.
- 19 samples would not have been submitted for DNA extraction (either cell or differential lysis extraction). Of these 19 samples:
 - 8 samples gave complex unsuitable, partial unsuitable or no DNA detected final results.
 Failure to submit these samples for DNA extraction would not have altered the final result.
 - 5 samples gave single source profiles from an assumed known contributor. Failure to submit these samples for DNA extraction would not have altered the final result.
 - o Low vaginal sample gave a mixed profile with major contribution from the complainant (which was not interpreted or reported). The high vaginal sample from the SAIK gave a P30 positive result and spermatozoa were detected on the vulval sample on the Evidence Recovery slide. The vulval sample gave a two person mixture which was conditioned on the complainant and gave a remaining profile UKM1 which was loaded to NCIDD. Therefore given the vulval result, and the low vaginal result, failure to submit the low vaginal sample for testing would not have altered the final result for this SAIK.
 - Endocervix sample gave two person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contribution from the suspect (this result was the same as for the perianal sample and was not reported via EXH). Spermatozoa were detected on the Evidence Recovery slides for the high vaginal 2, low vaginal, vulval, and perianal samples. The high vaginal gave a similar result to the perianal and was not reported via EXH. The low vaginal and vulval samples both gave single source profiles consistent with the suspect. Given the results of the other samples for this case, and the fact that the endocervix sample was not reported via EXH, failure to

- submit the endocervix sample for testing would not have altered the final result of the SAIK.
- o High vaginal sample gave a two person mixture which was conditioned on the complainant and gave a remaining male profile which was compared to two suspects for this case but both were excluded. The cervical sample for this case (which it should be noted had no spermatozoa detected on the Evidence Recovery or Differential Lysis slides) gave a similar result. The low vaginal sample gave a P30 positive result and gave a similar final result to the cervical and high vaginal. Spermatozoa were detected on the rectal sample on the Evidence Recovery slide, but gave a complex final result. Therefore given the results of the low vaginal sample, failure to submit the high vaginal sample for testing would not have altered the final result for this SAIK.
- o High vaginal sample gave a two person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contribution from the suspect. The vulval had no spermatozoa detected on the Evidence Recovery slide, but which pre-August 2016 would have been submitted for cell extraction, gave a three person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contribution from the suspect. Given that the vulval sample may have given a similar result if submitted for cell extraction (rather than differential lysis) failure to submit the high vaginal sample for testing may not have altered the final result for this SAIK.
- o High vaginal sample gave two person mixture which was conditioned on the complainant and the remaining profile was used to compare against nominated suspects. The low vaginal sample was P30 positive and gave a three person mixture which was also used to compare against suspects. Given the result of the low vaginal sample, failure to submit the high vaginal is not likely to have altered the final result for this SAIK.
- o Fabric sample gave a two person mixture which had >100 billion support for contribution from the suspect. This was the only result for this sample, however there are a large number of exhibits in this case with >100 billion support for contribution from the suspect. Therefore although failure to submit this sample would have changed the final result of this sample, there are a number of other exhibits in this case linked to the suspect.

Discussion

The aim of this data analysis was to assess the 738 samples which had no spermatozoa or seminal fluid detected during the initial Evidence Recovery examination, and which were then submitted for differential lysis extraction, and compare these to pre August 2016 examination and sample submission strategies to determine what, if any, impact this may have had on the DNA results reported for the case as a whole.

738 samples has been considered a sufficiently large dataset for the purposes of drawing some general conclusions, although this relies on the particular cases processed during this period, and therefore sampling variability may show a greater or lesser impact by assessing another dataset. It was beyond the scope of this data analysis to assess slides other than those that were originally zero spermatozoa detected at examination, and were submitted for differential lysis extraction since 8 August 2016.

The focus of this data analysis has been largely from a whole case perspective and several results were considered not to be impacted upon because of other samples/ similar results within the case. Assessing results on a whole case basis is standard case management practice, and is a process utilised across all

case and sample types. It is acknowledged that the impact on individual samples may be considered significant if semen is not observed at examination, the presumptive screening is also negative and no further action was taken for that sample. The risk if spermatozoa were consequently detected on the differential lysis slide and provided an interpretable DNA result, then potentially a valuable DNA profile for the case may not be obtained. What this data analysis shows is that this risk is mitigated when considering the typical case submission as a whole. The majority of SAIKs/sexual assault cases contain multiple swabs and items, which provide several opportunities to locate semen and subsequently obtain foreign DNA profile that may support an allegation of sexual assault.

Examination strategies are formulated to try and maximise the chances that even if one sample has no spermatozoa observed and the sample truly contains spermatozoa, then the DNA profile information can be obtained through other means. The presumptive screening for seminal fluid and examination strategies for submitting samples for differential lysis or cell extraction (including but not limited to: submission external swabs/swabs from minors for cell extraction; submitting all areas from an item if one obtains a positive sperm or presumptive result) and also the capacity of STRmix to interpret even mixtures of up to 3 contributors (and including conditioning) all contribute to minimising the overall case impact for a particular sample.

It is acknowledged the slide read at both examination and differential lysis is a detection step, and the sample used to make the slide is a very small amount from the prepared suspension (a drop and 3uL respectively), which is a representation of the spermatozoa that may be present in the sample. For very low levels of spermatozoa, if a second slide is prepared from the sample, lower or higher levels of spermatozoa may be observed, as is expected from sampling variability.

The aim of the differential lysis process is to attempt to separate any spermatozoa from any epithelial cells in order to aid in the interpretation of the DNA profiles obtained. While complete separation of the spermatozoa fractions and epithelial fractions is the ideal, this is not often the case, and carry-over of epithelial cells into the sperm fraction is common. The advantage of using STRmix for mixture interpretation helps mitigate the consequences of failing to obtain the ideal separation of spermatozoa and epithelial fractions, which is the aim for differential lysis. In cases where a sample undergoes a cell extraction and the sample does contain spermatozoa, it is reasonable to assume that this extraction process will extract any DNA present in the sample, including from any spermatozoa present. STRmix will similarly aid in the interpretation of any mixed DNA profiles obtained from this process.

As described previously, there is a degree of concentration of spermatozoa in the differential lysis process, and the number of spermatozoa present to give a slide read of <+1 is very low (defined as 'very hard to find spermatozoa'), therefore to go from zero to <+1 after differential lysis may not be unexpected. Similarly a slide read of +1 ('hard to find spermatozoa') after differential lysis, following a zero slide read at examination may not be too concerning or necessarily need to be taken as symptomatic of a problem with the examination slide read process.

Where a zero sperm read has produced a +2 sperm read of the slide after differential lysis, then this is harder to rationalise, even allowing for some variation in the subsample taken for the slide, and the differential lysis concentrating step. In this data there were 7 samples of the 738 total which showed this degree of difference ie. Zero to +2 (easy to find spermatozoa) which equates to 0.95% of this sample set. 6 of these samples would have undergone differential lysis extraction based on the presumptive result, and therefore the DNA results would have been unchanged. The one sample remaining was a vulval swab, and would have been submitted for a cell extraction. Within this particular SAIK, the high vaginal and low vaginal swabs both had sperm observed and examination, and provided a DNA profile with a contribution >100 billion for the suspect. Given the reasons listed above, all samples where +2

spermatozoa were detected at differential lysis, the results for the case were not considered to be negatively impacted.

Conclusions

Therefore in summary:

- Of the 738 total samples for which no spermatozoa were detected on the Evidence Recovery slide, 591 also had no spermatozoa detected on the Differential Lysis slide.
- 147 of the 738 samples had spermatozoa detected on the Differential Lysis slide (>1+, 1+ or 2+).
- Of these 147 slides, 1 sample (a sanitary pad have been obtained pre-August 2016. I.e. the decreased sensitivity of the Evidence Recovery slide (when compared to the Differential Lysis slide) resulted would have resulted in the sanitary pad sample not being submitted for DNA testing pre-August 2016.

The results of the analysis of this data set have shown that the difference in sensitivity of the Evidence Recovery and Differential Lysis swabs, although acknowledged, has not resulted in a systemic failure with regards to final reported results. There was one sample in the 738 sample data set which would not have been submitted for DNA testing pre-August 2016, and which gave 1+ spermatozoa on the Differential Lysis swab and a final DNA result consistent with the suspect. This was the only DNA result for this case. Pre-August 2016 this sample would have been reported to the QPS as "Semen not detected" and no further action taken. It should be noted that this presumptive EXH advised the QPS that "Spermatozoa were not observed..." rather than advising that there were no spermatozoa present. If deemed critical, the QPS could request further processing of this sample.

Therefore, although some individual samples may be negatively impacted as a consequence of the sensitivity of the examination slide process, overall this is considered to be an acceptable risk as it occurs relatively infrequently, and from a case perspective the risk is mitigated by the established practices of multiple sample submissions, examination submission and interpretation strategies.

The results of this study did not demonstrate a systemic flaw in the examination of exhibits for seminal fluid. The examination processes described throughout this report, as well as the resulting DNA profile, the assessment of the whole case, and the ability to submit for processing any samples not actioned, aims to mitigate the risk that may arise when spermatozoa is not detected at the examination step. Continuous process improvements are imbedded in Forensic DNA Analysis and are part of our quality management system, and improvements to the examination of sexual assault process will continue, as they will with all processes within the unit, to ensure any risks are mitigated and as much as practical. (????)

Queensland Health



Data Analysis of modified sexual assault process for zero spermatozoa detected at Evidence Recovery

Paula Brisotto, Matthew Hunt, Kylie Rika, Luke Ryan

May 2017

Introduction

Prior to August 2016, exhibits which gave negative results for spermatozoa and seminal fluid were typically not submitted for DNA testing, using the differential lysis extraction protocol (depending on case circumstances and examination strategy these samples may still be submitted for differential lysis extraction or for cell extraction). In August 2016, the Evidence Recovery Team implemented a modified sample submission process for all samples whereby all samples screened for seminal fluid were submitted for DNA testing, using differential lysis extraction, irrespective of the presumptive screening results (i.e. even those samples for which spermatozoa were not located microscopically and P30 tests were negative were still submitted for DNA testing using a differential lysis extraction). The microscopy slides created during the differential lysis extraction were then read by Evidence Recovery Team scientists for final reporting of spermatozoa detection results.

This modified process was introduced after it was raised that the initial slide microscopy conducted during ERT examinations may have a lower sensitivity than that slides produced during the differential lysis extraction process.

This modification process was introduced in response to concerns that the initial microscopy conducted during ERT examinations may be detecting fewer spermatezea than were seen by subsequent microscopy of slides produced after the differential lysis extraction process.

This data analysis examines the set of samples which had no spermatozoa or seminal fluid detected during the initial Evidence Recovery examination, and which were then submitted for differential lysis extraction. The results from this data set will be used to assess the difference in the pre and post August 2016 examination and sample submission strategies based on the final DNA results and attempt to determine what, if any, impact this may have had on the DNA results reported for the case.

In assessing the potential implications for the DNA results reported, relevant aspects of casemanagement were taken into account, including presumptive screening test results, existing examination strategies for different sample types and other results within the case.



Results

Since August 2016, the sample submission strategy was changed for samples which gave negative presumptive screening results for spermatozoa and seminal fluid (i.e. no spermatozoa observed on microscopy and P30 negative test). All samples which were tested for the presence of spermatozoa and seminal fluid were submitted for DNA extraction using the differential lysis protocol, regardless of the presumptive screening results.

During the period, 8 August 2016 to 28 March 2017, there were 738 samples for which the presumptive screening in Evidence Recovery did not identify spermatozoa during microscopy which were submitted for differential lysis extraction. For each of these 738 samples a microscopy slide was created during the Evidence Recovery examination (the Evidence Recovery slide), and a second slide was created during the differential lysis extraction (the Differential Lysis slide). Both slides were read by Evidence Recovery scientists.

The differences in the methods for creating the Evidence Recovery and Differential Lysis slides are a relevant context to the results of this data mining. Evidence Recovery slides are created by suspending the sample substrate (i.e. swab, scraping, material etc.) in 200μ L to 300μ L of nanopure water. This suspension is then vortexed and agitated with a disposable pipette. One drop (using a disposable pipette) of the suspension is then spotted onto a microscopy slide. The Differential Lysis slides are created by taking 3μ L from the male fraction (post separation from the female fraction) during the differential lysis protocol. The total volume for the male fraction from which the Differential Lysis slide is created is approximately 50μ L. It is expected that the Differential Lysis slide would be more sensitive (in terms of spermatozoa detection) for one main reason: the Differential Lysis slide is created by sampling 3μ L from a total volume of 50μ L of male fraction, therefore any spermatozoa present in the male fraction are at a higher concentration than the Evidence Recovery suspension which has a total volume of 200μ L. Suggest could also briefly cover the categorisation of sperm numbers (how many meant by <1+, 1+ etc.)

Table 1 Differential Lysis slide reads for the presence of sperm.

Original ER read	Diff Lysis Slide read	Number of samples	Percentage of Total
0 sperm	0 sperm	591	80.08
0 sperm	<+1 sperm	104	14.09
0 sperm	+1 sperm	36	4.88
0 sperm	+2 sperm	7	0.95

For 591 of these 738 samples, no spermatozoa were observed on either the Evidence Recovery or the Differential Lysis slides. For these results we can infer that either there were no spermatozoa present, or if they were present they were below the limit of detection for both the Differential Lysis and Evidence Recovery slide preparation techniques.

The remaining 147 slides, for which there were no spermatozoa observed on the Evidence Recovery slide, but where spermatozoa were observed on the Differential Lysis slide, were assessed to determine the impact on final DNA results for that sample.

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Of the 7 samples for which no spermatozoa were located on the Evidence Recovery slide, but +2 spermatozoa were located on the Differential Lysis slide:

- 6 would have been submitted for differential lysis extraction pre-August 2016 based on a positive P30 result
- The-remaining sample was a vulval swab to identify case details) from a SAIK. The vulval swab gave a 3P mixed DNA profile. Both the high vaginal and low vaginal swabs from this SAIK had spermatozoa observed on the Evidence Recovery slides. The high vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. (Please note: this wording would not be used in a Statement of Witness. The wording used is for the purposes of this report only. Standard wording to descr be probability/interpretations for a statement of witness is in QIS document #17119 Procedure for Release of Results.

The low vaginal sample gave a similar result to the high vaginal sample.

Therefore failure to submit the vulval swab would have fimited impact on the final DNA result reported for this SAIK.

For the 36 samples which gave no spermatozoa on the Evidence Recovery slide but +1 spermatozoa on the Differential Lysis slide:

- 19 would have been submitted for differential lysis extraction pre-August 2016 based on positive P30 results.
- Seven of these 36 samples would have been submitted for cell extraction rather than differential lysis extraction pre-August 2016.
 Submission of these seven samples for cell extraction rather than for differential lysis would have limited impact on the final results for these SAIKs because:
 - The spermatozoa fractions of two of these samples gave single source profiles which matched the nominated suspect.
 - The spermatozoa fractions of four of the samples gave either two/three person mixtures with >100 billion support for suspect contribution. (Use standard wording...)
 - The spermatozoa fraction of the final one sample (was this swab) gave a 2P mixture where the known contributor (SAIK complainant) and the suspect were represented (use std wording). -The vulval swab from this SAIK had +1 spermatozoa observed on the Evidence Recovery slide and the spermatozoa fraction gave a single source final result consistent with that matched the suspect.
- The remaining ten of these 36 samples would not have been submitted for DNA testing (either by cell or differential lysis extraction protocols) pre-August 2016. Of these ten samples:
 - o High vaginal sample S.frac gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect—From the same SAIK, [The second high vaginal, low vaginal, vulval and perianal swabs samples (suggest change throughout) all had spermatozoa detected on the Evidence Recovery slides. The low vaginal and vulval samples gave single source profiles which were consistent with the suspect. The second high vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore failure to submit the first high vaginal sample would have limited impact on the final results reported for the SAIK.

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Low vaginal sample SFrac) gave a complex final result which was not interpreted. The vulval and rectal samples Sfracs from this SAIK had spermatozoa detected on the Evidence Recovery slide. The rectal swab gave a single source DNA result which was consistent with the suspect. The vulval gave a complex final result which was not interpreted. Given the results of the rectal sample, and vulval sample, failure to submit the low vaginal sample would not have altered the final DNA results for this SAIK.

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- Low vaginal sample SFrac) gave a complex final result which was not interpreted suitable for interpretation. The high vaginal sample SFrac from this SAIK was AP and P30 positive and therefore would have been submitted for differential lysis extraction pre-August 2016. This high vaginal swab gave a 2 person mixture with >100 billion support for contribution from the suspect. The perianal swab was also p30 positive and therefore would have also been submitted for a differential lysis extraction pre-August 2016. The perianal swab gave a single source profile consistent with the suspect. The left nipple sample from this SAIK was also submitted for testing and have a 3 person mixture with >100 billion support for contribution from the suspect. Therefore failure to submit the low vaginal sample would not have altered the final result for this SAIK.
- Low vaginal sample-SFrac pawer a final result which was not interpreted/deconvoluted based on other results from the SAIK gave a 2person mixture conditioned on the complainant (LR >100billion favouring support for contribution by suspect). The cervical, high vaginal and posterior formix SFracs all gave 1+ spermatozoa on the Evidence Recovery slide and were submitted for differential lysis extraction (consistent with pre-August process). These cervical sample Sfracs each gave a two person conditioned mixtures with >100 billion support for contribution from the suspect. Based on this other samples from this SAIK were not interpreted further. Therefore failure to submit the low vaginal sample would not have significantly altered the final reported results for this SAIK.
- o Low vaginal sample SFrac) gave a complex final result which was not interpreted (indicates poss 4p). The high vaginal and vulval samples from this SAIK gave >1+ spermatozoa on the Evidence Recovery slide. The vulval sample (Sfrac) gave a complex final result which was not interpreted. The vulval Effac give a 3p conditioned
 The high vaginal (Sfrac) gave a 2 person mixture, from which the complainant was excluded. The high vaginal mixture appears to be an --1:1 2 person mixture from two males. No suspect reference samples have been submitted, but if they were they could be compared to this mixture. Therefore failure to submit the low vaginal sample would not have altered the final result for this SAIK.

Tapelift from inside crotch gave a complex result for the spermatozoa fraction which was not interpreted. The epithelial fraction also gave a complex result which was not interpreted. Therefore submission of this sample for cellular or differential lysis extraction would not have altered the final DNA result for this sample.

A piece of fabric) gave a single source profile matching the suspect for the spermatozoa fraction. The epithelial fraction gave a three person mixture with >100 billion support for contr bution from the suspect. Submission of this sample for cell extraction would not have altered impact on the final DNA result for this sample (i.e. suspect DNA located), albeit from cells extraction rather than a spermatozoa fraction. We would be able to talk about probable biological source in court though — given the SFrac is single source and attempting to isolate spermatozoa, then 'in my opinion highly likely at least some of DNA matching suspect is from semen'.

Low vaginal sample <u>Sfrac.</u>
) gave a <u>2p conditioned mixed DNA profile</u>
 >100billion for suspect, which indicates contribution from a male person, but has not been

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interpreted based on the high vaginal sample result._-The high vaginal sample was AP and P30 positive and therefore submitted for differential lysis and the SFrac gave a two person mixture which was conditioned on the complainant, and gave->100billion favouring contribution by the suspect. a UKM1 remaining profile (n.b. reference sample for suspect does not have a final result). The vulval sample had spermatozoa observed on the Evidence Recovery slide and the SFrac gave a 3p mixed DNA profile conditioned on the complainant, and gave>100billion favouring contribution by the suspect with indications of contribution from a male person, but this result has not been interpreted based on the high vaginal sample result. Note the Vulval Efrac is yet to be interpreted/reviewed - do not include in report until result finalised) Therefore based on the high vaginal sample Sfrac result, failure to submit the low vaginal sample would not have significantly altered the final result for this SAIK.

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High vaginal sample Sfrac) gave a two person mixture which was conditioned on the complainant and the remaining profile was UKM1 (n.b. offender is unknown). The low vaginal and vulval samples both had spermatozoa observed on the Evidence Recovery slide. The low vaginal SFrac gave a similar result to the high vaginal sample SFrac and was has not been interpreted further at this stage. The vulval sample SFrac was a three person mixture which was conditioned on the complainant, and the remaining profile was reported as 'not suitable for NCIDD load'. The vulval EFrac gave a partial single source DNA profile consistent with the complainant. Therefore based on the low vaginal samples-SFrac result, failure to submit the high vaginal sample would not have

significantly altered the final results reported for this SAIK.

o A sanitary pad SFrac gave a two person mixture (Wait until interp finalised- maybe 3p with repro) with contribution from a male person (not interpreted further as yet, but apparent major is a male contribution). The high vaginal and rectal samples both had no spermatozoa observed on the Evidence Recovery slide and were P30 negative. The high vaginal sample-Sfrac gave a two person mixed DNA profile which was conditioned on the complainant, and the remaining profile was reported as 'unsuitable for NCIDD'. The rectal sampleSfrac and Efrac both-gave a single source profiles which was were consistent with the complainant. Therefore failure to submit the sanitary pad for DNA testing would have impacted on the final result of this SAIK, as the sanitary pad was the only sample which gave a profile with a male contribution.

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There were 104 samples which for which no spermatozoa were detected located on the Evidence Recovery slide, but >1+ spermatozoa were detected located on the Differential Lysis slide (i.e. less than 10 spermatozoa observed on the Differential Lysis slide). The results of these 104 samples should be considered within the context described previously, i.e. that it is expected that the Differential Lysis slides are more sensitive than the Evidence Recovery slide, and that an change in microscopy result from zero spermatozoa detected to between one and ten spermatozoa detected may be representative of this difference in sensitivity. Of these 104 samples:

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- 46 would have been submitted for differential lysis extraction pre-August 2016 based on a positive P30 result or examination strategy.
- 39 would have been submitted for cell extraction (rather than differential lysis extraction) pre-August 2016. Of these 39 samples:
 - o 17 samples-Sfracs gave final results which were complex unsuitable for comparison, partial unsuitable for comparison or no DNA detected. Therefore submission for cell

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extraction would be unl kely to alter the final result ?and provide more probative information from for these samples.

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- 8 samples-<u>Sfracs</u> gave final results which were single source and were consistent with an
 assumed known contributor. Therefore submission of these samples for cell extraction
 would be unlikely to have <u>significantly</u> altered the final result.
- Vaginal and anal swab <u>SFrac</u> gave a three person mixed profile which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Given that this sample would have been submitted for cell extraction pre-August 2016, it is I kely that a similar result would have been obtained via a cell extraction.
- o Vulval sample gave a final DNA result which was not interpreted. Sfrac gave 2p conditioned, >100 bill for suspect. The high vaginal and low vaginal samples from this SAIK had spermatozoa observed on the Evidence Recovery slide. The high vaginal gave a three person mixture, the low vaginal gave a two person mixture. Both mixtures were conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore submission of the vulval sample for cell extraction would not have altered the final result for this SAIK.
- Rectal sample SFrac gave a single source profile consistent with the suspect. The cervical, high vaginal, low vaginal, vulval and perianal samples from this SAIK all had spermatozoa observed on the Evidence Recovery slide and each SFrac sample gave a single source profile consistent with the suspect. Therefore submission of the rectal sample for cell extraction would not have altered the final result for this SAIK.
- o Vulval and rectal samples Sfracs
) gave three person mixtures which were conditioned on the complainant, and gave a remaining profile with >100 billion support for contr bution from the suspect. (Vulval Efrac = 3p, conditioned, 6300 LR favouring suspect. Rectal Efrac 3p, cond and LR of 2 for susp.) LVS was AP/p30 positive Other samples in this SAIK (perianal and low vaginal sfracs) gave <a href="mailto:(2p and 3p) mixtures which were conditioned on the complainant, and gave remaining profiles with support for contr bution from the suspect. It is possible that had the vulval and rectal samples been submitted for cell extraction they could have given a similar final result to that obtained from the differential lysis extraction.
- Vulval sample Sfrac gave a partial profile consistent with the suspect (Ffrac was 3p cond, LR ~140 for susp) The high and low vaginal samples from this SAIK had spermatozoa observed on the Evidence Recovery slides. The high vaginal sample Sfrac gave a single source profile consistent with the suspect. The low vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore submission of the vulval sample for cell extraction would not have altered the final result for this SAIK.
 - Two fabric samples

 (a) Sfracs gave two person mixtures for which no statistical interpretation was performed. The Efrac from gave a 3p, no statistical interpretation was performed. A third scraping from the same fabric gave a single source profile from an UKF1 from the epithelial fraction. It is possible that had these two fabrics been submitted for cell extraction that the final result would not be different
- o Vulval sample Sfrac gave a two person mixture which was conditioned on the complainant, and gave a remaining profile which was consistent with UKM1. Effac was SS AKC compt. The low vaginal sample gave an AP/ P30 positive result and was submitted for a differential lysis extraction and Sfrac gave 2p mixed DNA profile which was conditioned on the complainant and gave a remaining profile which is consistent with UKM1. Given the low vaginal Sfrac sample result, and the possibility that the vulval

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- sample would have given a similar result if submitted for a cell extraction, the final result for the SAIK is unlikely to be different if the vulval sample was submitted for cell extraction. (Vulval may have female swamping male if cell ext (given Efrac result)
- Vulval sample <u>Sfrac</u>) gave a <u>partial</u> single source profile from-designated as UKM1. <u>Vulval EFrac</u> has not been interpreted (Matter withdrawn) but indicates a male/female mixture (would require rework to determine # contr butors. I wouldn't include this sample in report) The high vaginal <u>swab</u> had spermatozoa observed on the Evidence Recovery slide and <u>the SFrac</u> gave a mixed DNA profile with a male contr butor (consistent with UKM1 although not reported). Given the high vaginal <u>sfrac</u> result and the possibility that the vulval sample would have given a similar result if submitted for cell extraction, the final result for the SAIK is unlikely to be different if the vulval sample was submitted for cell extraction.
- o Mouth sample Sfrac) gave a single source female profile (I kely the complainant but FTA does not yot have a final result). 2p mix cond compl't, LR supports non-contrib for sus't. EFrac gave SS AKC comp't. The high vaginal sample from the SAIK had spermatozoa observed on the Evidence Recovery slide and Sfrac gave a 2p mixed DNA profile cond on compl't, susp >100bill with a major male contributor. Therefore given the high vaginal sample result and submission of the mouth sample for cell extraction would not have altered the final result for the SAIK.
- 19 samples would not have been submitted for DNA extraction (either cell or differential lysis extraction). Of these 19 samples:
 - 8 samples gave complex unsuitable, partial unsuitable or no DNA detected final results.
 Failure to submit these samples for DNA extraction would not have altered the final result.
 - 5 samples gave single source profiles from an assumed known contributor. Failure to submit these samples for DNA extraction would not have altered the final result.

 - o Endocervix sample SFrac

) gave two person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contribution from the suspect (this result was the same as for the perianal Frac sample and was not reported via EXH). Spermatozoa were detected on the Evidence Recovery slides for the high vaginal 2, low vaginal, vulval, and perianal samples. The high vaginal (2? need to specify, also applies to HVS 1) gave a similar result to the perianal Frac and was not reported via EXH. The low vaginal and vulval Sfracs samples both gave single source profiles consistent with the suspect. (Vulval Efrac = 2p cond, LR susp't low support contribn) Given the results of the other samples for this case, and the fact that the endocervix sample was not reported via EXH, (I'd leave this out only true because no statement request received to date) failure to submit the endocervix sample for testing would not have altered the final result of the SAIK.
 - High vaginal sample <u>Sfrac</u> gave a two person mixture which was conditioned on the complainant and gave a remaining male profile which was compared to two suspects for this case but both were excluded. The cervical sample for this case

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(which it should be noted had no spermatozoa detected on the Evidence Recovery or Differential Lysis slides) Sfrac gave a similar result. The low vaginal sample gave a APIP30 positive result and the LVS Sfrac gave a similar final result to the cervical and high vaginal Sfrac spermatozoa were detected on the rectal sample on the Evidence Recovery slide, but Sfrac gave a complex final result (+ EFFac SS AKC complition. Therefore given the results of the low vaginal sample Sfrac, failure to submit the high vaginal sample for testing would not have altered the final result for this SAIK.

- High vaginal sample Sfrac gave a two person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contribution from the suspect. The vulval had no spermatozoa detected on the Evidence Recovery slide, but which pre-August 2016 would have been submitted for cell extraction, Vulval Sfrac gave a three person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contr bution from the suspect. Given that the vulval sample may have given a similar result if submitted for cell extraction (rather than differential lysis) failure to submit the high vaginal sample for testing may not have altered the final result for this SAIK. Or including the female DNA in the epi fraction may have swamped the male DNA
- o High vaginal sample

 Sfrac gave two person mixture which was conditioned on the complainant and the remaining profile was used to compare against nominated suspects. The low vaginal sample was AP/P30 positive and the Sfrac gave a three person cond mixture which was also used to compare against suspects. Given the result of the low vaginal sample, failure to submit the high vaginal is not likely to have altered the final result for this SAIK. Change in # contributors may impact (as above)
- Fabric sample Sfrac gave a two person mixture which had >100 billion support for contr bution from the suspect. (EFRAC = complex). This was the only result for this sample, however there are a large number of exhibits in this case with >100 billion support for contr bution from the suspect. Therefore although failure to submit this sample would have changed the final result of this sample, there are a number of other exhibits in this case linked to the suspect.

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Discussion

The aim of this data analysis was to assess the 738 samples which had no spermatozoa or seminal fluid detected during the initial Evidence Recovery examination, and which were then submitted for differential lysis extraction, and compare these to pre August 2016 examination and sample submission strategies to determine what, if any, impact this may have had on the DNA results reported for the case as a whole.

738 samples has been considered a sufficiently large dataset for the purposes of drawing some general conclusions, although this relies on the particular cases processed during this period, and therefore due to sampling variation there may have been a greater or lesser impact if another dataset had been assessed. It was beyond the scope of this data analysis to assess slides other than those that were originally zero spermatozoa detected at examination, and were submitted for differential lysis extraction since 8 August 2016.

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The focus of this data analysis has been largely from a whole case perspective and several results were considered not to be impacted upon because of other samples/ similar results within the case. Assessing results on a whole case basis is part of standard case management practice, and is a process utilised across all case and sample types. It is acknowledged that the impact on individual samples may be considered significant if semen is not observed at examination, the presumptive screening is also negative and no further action was taken for that sample. There is a risk that if spermatozoa were consequently detected on the differential lysis slide and provided an interpretable DNA result, then potentially a valuable DNA profile for the case may not be obtained. What this data analysis shows is that this risk is mitigated when considering the typical case submission as a whole. The majority of SAIKs/sexual assault cases contain multiple swabs and items, which provide several opportunities to locate semen and subsequently obtain foreign DNA profile that may support an allegation of sexual assault

Examination strategies are formulated to try and maximise the chances that even if one sample has no spermatozoa observed and the sample truly contains spermatozoa, then the DNA profile information can be obtained through other means. The presumptive screening for seminal fluid and examination strategies for submitting samples for differential lysis or cell extraction (including but not limited to: submission of external swabs/swabs from minors for cell extraction; submitting all areas from an item if one obtains a positive sperm or presumptive result) and also the capacity of STRmix to interpret mixtures of up to 3 contributors (and including conditioning) all contribute to minimising the overall case impact for a particular sample.

It is acknowledged the slide read at both examination and differential lysis is a detection step, and the sample used to make the slide is a very small amount from the prepared suspension (a drop and 3uL respectively), which is a representation of the spermatozoa that may be present in the sample. For very low levels of spermatozoa, if a second slide is prepared from the sample, lower or higher levels of spermatozoa may be observed, as is to be expected as a consequence of sampling variability.

The aim of the differential lysis process is to attempt to separate any spermatozoa from any epithelial cells in order to aid in the interpretation of the DNA profiles obtained. While complete separation of the spermatozoa fractions and epithelial fractions is the ideal, this is often not the case, and carry-over of epithelial cells into the sperm fraction is commonly observed. The advantage of using STRmix for mixture interpretation helps mitigate the consequences of failing to obtain the ideal separation of spermatozoa and epithelial fractions, which is the aim for differential lysis. In cases where a sample undergoes a cell extraction and the sample does contain spermatozoa, it is reasonable to assume that this extraction process will extract any DNA present in the sample, including from any spermatozoa present. STRmix will similarly aid in the interpretation of any mixed DNA profiles obtained from this process.

As described previously, there is a degree of concentration of spermatozoa in the differential lysis process, and the number of spermatozoa present to give a slide read of <+1 is very low (defined as 'very hard to find spermatozoa'), therefore to go from zero to <+1 after differential lysis may not be unexpected. Similarly a slide read of +1 ('hard to find spermatozoa') after differential lysis, following a zero slide read at examination may not be too concerning or necessarily need to be taken as symptomatic of inefficiencies in the examination slide read process.

Where a zero sperm read has produced a +2 sperm read of the slide after differential lysis, then this is harder to rationalise, even allowing for some variation in the subsample taken for the slide, and the differential lysis concentrating step. In this data there were 7 samples of the 738 total which showed this degree of difference ie. Zero to +2 (easy to find spermatozoa) which equates to 0.95% of this sample set. 6 of these samples would have undergone differential lysis extraction based on the presumptive

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result, and therefore the DNA results are expected to have been unchanged. The one sample remaining was a vulval swab, which would have been submitted for a cell extraction. Within this particular SAIK, the high vaginal and low vaginal swabs both had sperm observed at examination, and from these samples mixed DNA profiles were obtained that STRmix gave L kelihood Ratios of greater than 100 billion favouring contribution by the suspect. Given the reasons listed above, for all samples within this data analysis where +2 spermatozoa were detected at differential lysis, the DNA profiling results for the case were not considered to be negatively impacted.

Conclusions

The purposes of the examination process in the Evidence Recovery Team is to attempt to identify areas of biological material for submission to the Analytical team for DNA processing.

The processes for the detection of spermatozoa and seminal fluid within the Evidence Recovery Team......(summary around what is the actual purpose – to detect sperm for submission to Analytical for diff processing.) Whilst the observations of spermatozoa at ER of diff stage is a confirmation of their presence, the absence of sperm is not confirmation of their absence. It means that spermatozoa, if present, where at a level that was not able to be detected.

Etc etc - this could be fit in somewhere below....?

Therefore in summary:

- Of the 738 total samples for which no spermatozoa were detected on the Evidence Recovery slide, 591 also had no spermatozoa detected on the Differential Lysis slide.
- 147 of the 738 samples had spermatozoa detected on the Differential Lysis slide (>1+, 1+ or 2+).
- Of these 147 slides, 1 sample (a sanitary pad have been obtained pre-August 2016. I.e. the decreased sensitivity of the Evidence Recovery slide (when compared to the Differential Lysis slide) resulted would have resulted in the sanitary pad sample not being submitted for DNA testing pre-August 2016.

The results of the analysis of this data set have shown that the difference in sensitivity of the Evidence Recovery and Differential Lysis swabs, although acknowledged, has not resulted in a systemic failure with regards to final reported results. (I wouldn't include this phrase-don't think a 'systemic failure' of reported results is the concern.)

For a small proportion of sexual assault samples containing low numbers of spermatozoa, the difference in the sensitivity between the ERT and Diff Microscopy methods had previously caused samples to be reported as semen negative when spermatozoa may have been detectable by microscopy following the differential lysis extraction procedure. As the ERT microscopy was previously used as a key determinant as to which extraction method was employed, and indeed whether the samples were submitted for DNA analysis at all, there is a potential impact on a small subset of reported results. Depending on the case circumstances, the ability to report that semen was detected may or may not be critical, in the context of the allegation. For sexually active adults, confirming the presence of spermatozoa on intimate swabs, may perhaps not be considered as critical as it is for sexual assault allegations involving complainants to

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whom limited opportunities for the transfer of spermatozoa may make such findings more pertinent (for example minors, the elderly or individuals with disabilities).

In many instances we have found that this issue is mitigated by the common practice of submitting multiple swabs within a typical SAIK, thus increasing the chances of detecting semen. The fact that this data analysis has found that the methods in use prior to August 2016 would have had in only a relatively limited impact on the particular reported results for these SAIKS should be reassuring, however we should continue to strive to eliminate impacts of this type for all cases, and seek to improve the sensitivity of our methods, including by selecting the most effective point in the process to conduct microscopy. It may also be the case that if one swab from a SAIK is affected by this issue, then other swabs in the SAIK could also have an increased chance of being similarly affected, as microscopy examinations are not genuinely independent events. A SAIK containing a High Vaginal Swab with low numbers of spermatozoa may be more likely to also contain a Low Vaginal Swab with low numbers of spermatozoa. The microscopy process is a manual one, which is in a large measure dependent on the technique and ability of the examining scientist. If the slide created from one SAIK swab is affected by an issue which decreases the chance of observing spermatozoa, then this would also tend to affect the chance of detecting spermatozoa on a slide from a second swab of the same SAIK, made by the same scientist.

There was one sample in the 738 sample data set which would not have been submitted for DNA testing pre-August 2016, and which gave 1+ spermatozoa on the Differential Lysis swab and a final DNA result consistent with the suspect. This was the only DNA result for this case. Pre-August 2016 this sample would have been reported to the QPS as "Semen not detected" and no further action taken. It should be noted that this presumptive EXH advised the QPS that "Spermatozoa were not observed..." rather than advising that there were no spermatozoa present. If deemed critical, the QPS could request further processing of this sample.

Therefore, although some individual samples may be negatively impacted as a consequence of the sensitivity of the examination slide process, overall this is considered to be an acceptable risk as it occurs relatively infrequently, and from a case perspective the risk is mitigated by the established practices of multiple sample submissions, examination submission and interpretation strategies.

The results of this study did not demonstrate a systemic failure in the examination of exhibits for seminal fluid. The examination processes described throughout this report, as well as the resulting DNA profile, the assessment of the whole case, and the ability to submit for processing any samples not actioned, aims to mitigate the risk that may arise when spermatozoa is not detected at the examination step. Continuous process improvements are imbedded in Forensic DNA Analysis and are part of our quality management system, and improvements to the examination of sexual assault process will continue, as they will with all processes within the unit, to ensure any risks are mitigated as much as practical. (???)

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General points:

Suggest all wording used to describe stats/ interpretations should be as per standard wording for statements - see Procedure for Release of Results SOP 17119.

e.g. and gave a remaining profile with >100 billion support for contribution from the suspect."

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Suggested wording: "Based on statistical analysis it is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if the suspect has contributed DNA along with the complainant, rather than if he has not."

Sug. leave out identifiers (sample barcodes) where discussing interps/case details. ?Could number the samples based on the order in the xls. and refer to Sample 1, 2 etc. within the report – add appendix/notes to explain.

Sug. leave out any interpretations which have not been finalised (reviewed), and may be subject to change. Will check on their status

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Data Analysis of modified sexual assault process for zero spermatozoa detected at Evidence Recovery

Paula Brisotto, Matthew Hunt, Kylie Rika, Luke Ryan

May 2017

Introduction

Prior to August 2016, exhibits which gave negative results for spermatozoa and seminal fluid were typically not submitted for DNA testing, using the differential lysis extraction protocol (depending on case circumstances and examination strategy these samples may still be submitted for differential lysis extraction or for cell extraction). In August 2016, the Evidence Recovery Team implemented a modified sample submission process for all samples whereby all samples screened for seminal fluid were submitted for DNA testing, using differential lysis extraction, irrespective of the presumptive screening results (i.e. even those samples for which spermatozoa were not located microscopically and P30 tests were negative were still submitted for DNA testing using a differential lysis extraction). The microscopy slides created during the differential lysis extraction were then read by Evidence Recovery Team scientists for final reporting of spermatozoa detection results.

This modified process was introduced after it was raised that the initial slide microscopy conducted during ERT examinations may have a lower sensitivity than that slides produced during the differential lysis extraction process.

This modification process was introduced in response to concerns that the initial microscopy conducted during ERT examinations may be detecting fewer spermatezea than were seen by subsequent microscopy of slides produced after the differential lysis extraction process.

This data analysis examines the set of samples which had no spermatozoa or seminal fluid detected during the initial Evidence Recovery examination, and which were then submitted for differential lysis extraction. The results from this data set will be used to assess the difference in the pre and post August 2016 examination and sample submission strategies based on the final DNA results and attempt to determine what, if any, impact this may have had on the DNA results reported for the case.

In assessing the potential implications for the DNA results reported, relevant aspects of casemanagement were taken into account, including presumptive screening test results, existing examination strategies for different sample types and other results within the case.



Results

Since August 2016, the sample submission strategy was changed for samples which gave negative presumptive screening results for spermatozoa and seminal fluid (i.e. no spermatozoa observed on microscopy and P30 negative test). All samples which were tested for the presence of spermatozoa and seminal fluid were submitted for DNA extraction using the differential lysis protocol, regardless of the presumptive screening results.

During the period, 8 August 2016 to 28 March 2017, there were 738 samples for which the presumptive screening in Evidence Recovery did not identify spermatozoa during microscopy which were submitted for differential lysis extraction. For each of these 738 samples a microscopy slide was created during the Evidence Recovery examination (the Evidence Recovery slide), and a second slide was created during the differential lysis extraction (the Differential Lysis slide). Both slides were read by Evidence Recovery scientists.

The differences in the methods for creating the Evidence Recovery and Differential Lysis slides are a relevant context to the results of this data mining. Evidence Recovery slides are created by suspending the sample substrate (i.e. swab, scraping, material etc.) in 200μ L to 300μ L of nanopure water. This suspension is then vortexed and agitated with a disposable pipette. One drop (using a disposable pipette) of the suspension is then spotted onto a microscopy slide. The Differential Lysis slides are created by taking 3μ L from the male fraction (post separation from the female fraction) during the differential lysis protocol. The total volume for the male fraction from which the Differential Lysis slide is created is approximately 50μ L. It is expected that the Differential Lysis slide would be more sensitive (in terms of spermatozoa detection) for one main reason: the Differential Lysis slide is created by sampling 3μ L from a total volume of 50μ L of male fraction, therefore any spermatozoa present in the male fraction are at a higher concentration than the Evidence Recovery suspension which has a total volume of 200μ L. Suggest could also briefly cover the categorisation of sperm numbers (how many meant by <1+, 1+ etc.)

Table 1 Differential Lysis slide reads for the presence of sperm.

Original ER read	Diff Lysis Slide read	Number of samples	Percentage of Total
0 sperm	0 sperm	591	80.08
0 sperm	<+1 sperm	104	14.09
0 sperm	+1 sperm	36	4.88
0 sperm	+2 sperm	7	0.95

For 591 of these 738 samples, no spermatozoa were observed on either the Evidence Recovery or the Differential Lysis slides. For these results we can infer that either there were no spermatozoa present, or if they were present they were below the limit of detection for both the Differential Lysis and Evidence Recovery slide preparation techniques.

The remaining 147 slides, for which there were no spermatozoa observed on the Evidence Recovery slide, but where spermatozoa were observed on the Differential Lysis slide, were assessed to determine the impact on final DNA results for that sample.

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Of the 7 samples for which no spermatozoa were located on the Evidence Recovery slide, but +2 spermatozoa were located on the Differential Lysis slide: 6 would have been submitted for differential lysis extraction pre-August 2016 based on a positive P30 result SUGGEST use our standard wording for all of the below interpretations/stats: for example: Formatted: Indent: Left: 1.27 cm, No bullets or numbering The-remaining sample was a vulval swab sample-Formatted: Highlight be used to identify case details) from a SAIK. The Formatted: Space After: 10 pt A mixed DNA profile was obtained from this vulval-sample-swab, spermatozoa fraction which Formatted: Indent: Left: 0.63 cm, Space After: 10 pt, No bullets or numbering indicated the presence of DNA from 3 contributors, one of whom could be the complainant. Based on statistical analysis it was estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if the nominated suspect has contributed DNA, along with the complainant, rather than if he has not. gave a 3P mixed DNA profile (net yet reported). Both Tihe high vaginal and low vaginal samples Formatted: Font color: Custom Color(RGB(33,23,233)), swabs from this SAIK -had spermatozoa observed on the Evidence Recovery slides. Mixed DNA profiles were obtained from the high vaginal swab, spermatozoa fraction and the low vaginal swab, spermatozoa fraction which each indicated the presence of DNA from 2 contributors, one of whom could be the complainant. Based on statistical analysis it was estimated that the each of these mixed DNA profiles obtained is greater than 100 billion times more likely to have occurred if the suspect has contr buted DNA, along with the complainant, rather than if he has not. The high vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. The low vaginal sample gave a similar result to the high vaginal sample. Formatted: Indent: Left: 1.27 cm, Space After: 10 pt, No bullets or numbering Therefore failure to submit the vulval sample would not have altered the final result for the. Formatted: Highlight SAIK. ?True statement. – the presence of a possible third contributor in the vulval swab may still be considered relevant for the case. Defence could use this evidence of a third person to cast doubt on the complainants story / character - who is this third person? - could this be the true offender etc. - think we should acknowledge that there could be limited impact on the results reported for this SAIK For the 36 samples which gave no spermatozoa on the Evidence Recovery slide but +1 spermatozoa on the Differential Lysis slide: 19 would have been submitted for differential lysis extraction pre-August 2016 based on positive Seven of these 36 samples would have been submitted for cell extraction rather than differential lysis extraction pre-August 2016. Submission of these seven samples for cell extraction rather than for differential lysis would not have altered the final results for these SAIKs because: (As above, too definitive a statement? - A strong contribution from the suspect was obtained for each sample, however there may still be an impact if enough DNA from the female epithelial cells is present following a cell extraction as this could potentially swamp out the contribution of male DNA from the spermatozoa.) The spermatozoa fractions of two of these samples gave single source profiles which consistent with matched the nominated suspect. The spermatozoa fractions of four of the samples gave either two/three person mixtures Formatted: Highlight with >100 billion support for suspect contribution. (Use standard wording...) Formatted: Highlight - 3 of 12 -

- The spermatozoa fraction of the final ene sample (was this swab) (a perianal SAIK swab) gave a 2P mixture where the known contributor (SAIK complainant) and the suspect were represented (use std wording). The vulval swab from this SAIK had +1 spermatozoa observed on the Evidence Recovery slide and the spermatozoa fraction gave a single source final result consistent with that matched the suspect.
- The remaining ten of these 36 samples would not have been submitted for DNA testing (either by cell or differential lysis extraction protocols) pre-August 2016. Of these ten samples:
 - o High vaginal sample-S frac gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect.—From the same SAIK, the second high vaginal, low vaginal, vulval and perianal swabs samples (suggest change throughout) all had spermatozoa detected on the Evidence Recovery slides. The low vaginal and vulval samples gave single source profiles which were consistent with the suspect. The second high vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore failure to submit the first high vaginal sample would not have 2 significantly altered the final results reported for the SAIK.

Low vaginal sample SFrac gave a complex final result which was not interpreted. The vulval and rectal samples-Sfracs from this SAIK had spermatozoa detected on the Evidence Recovery slide. The rectal swab gave a single source DNA result which was consistent with the suspect. The vulval gave a complex final result which was not interpreted. Given the results of the rectal sample, and vulval sample, failure to submit the low vaginal sample would not have altered the final DNA results for this SAIK.

- o Low vaginal sample<u>SFrac</u>) gave a complex final result which was not interpreted suitable for interpretation. The high vaginal sample<u>SFrac</u> from this SAIK was AP and P30 positive and therefore would have been submitted for differential lysis extraction pre-August 2016. This high vaginal swab gave a 2 person mixture with >100 billion support for contribution from the suspect. The perianal swab was also p30 positive and therefore would have also been submitted for a differential lysis extraction pre-August 2016. The perianal swab gave a single source profile consistent with the suspect. The left nipple sample from this SAIK was also submitted for testing and have a 3 person mixture with >100 billion support for contr bution from the suspect. Therefore failure to submit the low vaginal sample would not have altered the final result for this SAIK.
- o Low vaginal sample_SFrac)-gave a final result which was not interpreted/deconvoluted based on other results from the SAIK gave a 2person mixture conditioned on the complainant (LR >100billion favouring support for contribution by suspect).—The cervical, high vaginal and posterior formix SFracs all gave 1+ spermatozoa on the Evidence Recovery slide and were submitted for differential lysis extraction (consistent with pre-August process). These cervical sample Sfracs each gave a two person conditioned mixtures with >100 billion support for contribution from the suspect. Based on this other samples from this SAIK were not interpreted further.—Therefore failure to submit the low vaginal sample would not have significantly altered the final reported results for this SAIK.
- Low vaginal sample <u>SFract</u>) gave a complex final result which was not interpreted <u>(indicates poss 4p)</u>. The high vaginal and vulval samples from this SAIK gave >1+ spermatozoa on the Evidence Recovery slide. The vulval sample <u>(Sfrac)</u> gave a complex final result which was not interpreted. The vulval Efrac give a 3p conditioned

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The high vaginal (Sfrac) gave a 2 person mixture, from which the complainant was excluded. The high vaginal mixture appears to be an --1:1 2 person mixture from two males. No suspect reference samples have been submitted, but if they were they could be compared to this mixture. Therefore failure to submit the low vaginal sample would not have altered the final result for this SAIK.

- Tapelift from inside crotch (a) gave a complex result for the spermatozoa fraction which was not interpreted. The epithelial fraction also gave a complex result which was not interpreted. Therefore submission of this sample for cellular or differential lysis extraction would not have altered the final DNA result for this sample.
- A piece of fabric) gave a single source profile matching the suspect for the spermatozoa fraction. The epithelial fraction gave a three person mixture with >100 billion support for contr bution from the suspect. Submission of this sample for cell extraction would not have altered the final result for this sample (i.e. suspect DNA located), albeit from cells extraction rather than a spermatozoa fraction. We would be able to talk about probable biological source in court though given the SFrac is single source and attempting to isolate spermatozoa, then 'in my opinion highly likely at least some of DNA matching suspect is from semen'.
- o Low vaginal sample Sfrac gave a 2p conditioned mixed DNA profile >100billion for suspect. which indicates centribution from a male person, but has not been interpreted based on the high vaginal sample result...The high vaginal sample was AP and P30 positive and therefore submitted for differential lysis and the SFrac gave a two person mixture which was conditioned on the complainant, and gave>100billion favouring contribution by the suspect. a UKM1 remaining profile (n.b. reference sample for suspect does not have a final result). The vulval sample had spermatozoa observed on the Evidence Recovery slide and the SFrac gave a 3p mixed DNA profile conditioned on the complainant, and gave>100billion favouring contribution by the suspect with indications of contribution from a male person, but this result has not been interpreted based on the high vaginal sample result. (Note the Vulval Efrac is yet to be interpreted/reviewed do not include in report until result finalised). Therefore based on the high vaginal sample Sfrac result, failure to submit the low vaginal sample would not have significantly altered the final result for this SAIK.
- high vaginal sample strac) gave a two person mixture which was conditioned on the complainant and the remaining profile was UKM1 (n.b. offender is unknown). The low vaginal and vulval samples both had spermatozoa observed on the Evidence Recovery slide. The low vaginal SFrac gave a similar result to the high vaginal sample SFrac and was-has not been interpreted further at this stage. The vulval sample-SFrac was a three person mixture which was conditioned on the complainant, and the remaining profile was reported as 'not suitable for NCIDD load'. The vulval EFrac gave a partial single source DNA profile consistent with the complainant. Therefore based on the low vaginal samples-SFrac result, failure to submit the high vaginal sample would not have significantly altered the final results reported for this SAIK.
- A sanitary pad SFrac SFrac Sgave a two person mixture (Wait until interp finalised- maybe 3p with repro) with contribution from a male person (not interpreted further as yet, but apparent major is a male contribution). The high vaginal and rectal samples both had no spermatozoa observed on the Evidence Recovery slide and were P30 negative. The high vaginal sample-Sfrac gave a two person mixed DNA profile which was conditioned on the complainant, and the remaining profile was reported as 'unsuitable for NCIDD'. The rectal sample Sfrac and Efrac both-gave a single source profiles which was were consistent with the complainant. Therefore failure to submit the sanitary pad for DNA testing would

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have impacted on the final result of this SAIK, as the sanitary pad was the only sample which gave a profile with a male contribution.

There were 104 samples which for which no spermatozoa were detected located on the Evidence Recovery slide, but >1+ spermatozoa were detected located on the Differential Lysis slide (i.e. less than 10 spermatozoa observed on the Differential Lysis slide). The results of these 104 samples should be considered within the context described previously, i.e. that it is expected that the Differential Lysis slides are more sensitive than the Evidence Recovery slide, and that an change in microscopy result from zero spermatozoa detected to between one and ten spermatozoa detected may be representative of this difference in sensitivity. Of these 104 samples:

46 would have been submitted for differential lysis extraction pre-August 2016 based on a
positive P30 result or examination strategy.

- 39 would have been submitted for cell extraction (rather than differential lysis extraction) pre-August 2016. Of these 39 samples:
 - 17 samples-<u>Sfracs</u> gave final results which were complex unsuitable for comparison, partial unsuitable for comparison or no DNA detected. Therefore submission for cell extraction would be unlikely to <u>alter the final result ?and provide more probative</u> information from for these samples.
 - 8 samples-<u>Sfracs</u> gave final results which were single source and were consistent with an
 assumed known contributor. Therefore submission of these samples for cell extraction
 would be unlikely to have <u>significantly</u> altered the final result.
 - Vaginal and anal swab <u>SFract</u> gave a three person mixed profile which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Given that this sample would have been submitted for cell extraction pre-August 2016, it is I kely that a similar result would have been obtained via a cell extraction.
 - vulval sample page 2p conditioned, >100 bill for suspect. The high vaginal and low vaginal samples from this SAIK had spermatozoa observed on the Evidence Recovery slide. The high vaginal gave a three person mixture, the low vaginal gave a two person mixture. Both mixtures were conditioned on the complainant, and gave a remaining profile with >100 billion support for contr bution from the suspect. Therefore submission of the vulval sample for cell extraction would not have altered the final result for this SAIK.
 - o Rectal sample SFrac gave a single source profile consistent with the suspect. The cervical, high vaginal, low vaginal, vulval and perianal samples from this SAIK all had spermatozoa observed on the Evidence Recovery slide and each SFrac sample gave a single source profile consistent with the suspect. Therefore submission of the rectal sample for cell extraction would not have altered the final result for this SAIK.
 - o Vulval and rectal samples Sfracs
) gave three person mixtures which were conditioned on the complainant, and gave a remaining profile with >100 billion support for contr bution from the suspect. (Vulval Efrac = 3p, conditioned, 6300 LR favouring suspect. Rectal Efrac 3p, cond and LR of 2 for susp.) LVS was AP/p30 positive Other samples in this SAIK (perianal and low vaginal sfracs) gave (2p and 3p)mixtures which were conditioned on the complainant, and gave remaining profiles with support for contr bution from the suspect. It is possible that had the vulval and rectal samples been submitted for cell extraction they could have given a similar final result to that obtained from the differential lysis extraction.

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- o Vulval sample Sfrac gave a partial profile consistent with the suspect. (Efrac was 3p cond, LR ~140 for susp.) The high and low vaginal samples from this SAIK had spermatozoa observed on the Evidence Recovery slides. The high vaginal sample Sfrac gave a single source profile consistent with the suspect. The low vaginal sample gave a two person mixture which was conditioned on the complainant, and gave a remaining profile with >100 billion support for contribution from the suspect. Therefore submission of the vulval sample for cell extraction would not have altered the final result for this SAIK.
- e—Two fabric samples

 (a) Sfracs gave two person mixtures for which no statistical interpretation was performed. The Efrac from gave a 3p, no statistical interpretation was performed. A third scraping from the same fabric gave a single source profile from an UKF1 from the epithelial fraction. It is possible that had these two fabrics been submitted for cell extraction that the final result would not be different.
- o Vulval sample Sfrac) gave a two person mixture which was conditioned on the complainant, and gave a remaining profile which was consistent with UKM1. Efrac was SS AKC comp't. The low vaginal sample gave an AP/ P30 positive result and was submitted for a differential lysis extraction and Sfrac gave 2p mixed DNA profile which was conditioned on the complainant and gave a remaining profile which is consistent with UKM1. Given the low vaginal Sfrac sample result, and the possibility that the vulval sample would have given a similar result if submitted for a cell extraction, the final result for the SAIK is unlikely to be different if the vulval sample was submitted for cell extraction. (Vulval may have female swamping male if cell ext (given Efrac result)
- O Vulval sample Sfrac) gave a partial single source profile from-designated as UKM1. Vulval EFrac has not been interpreted (Matter withdrawn) but indicates a male/female mixture (would require rework to determine # contr butors. I wouldn't include this sample in report) The high vaginal swab had spermatozoa observed on the Evidence Recovery slide and the SFrac gave a mixed DNA profile with a male contr butor (consistent with UKM1 although not reported). Given the high vaginal sfrac result and the possibility that the vulval sample would have given a similar result if submitted for cell extraction, the final result for the SAIK is unlikely to be different if the vulval sample was submitted for cell extraction.
- o Mouth sample Sfrac (6 gave a single source female profile (I kely the complainant but FTA does not yet have a final result). 2p mix cond compl't, LR supports non-contrib for sus't. EFrac gave SS AKC comp't. The high vaginal sample from the SAIK had spermatozoa observed on the Evidence Recovery slide and Sfrac gave a 2p mixed DNA profile cond on compl't, susp >100bill with a major male contributor. Therefore given the high vaginal sample result and submission of the mouth sample for cell extraction would not have altered the final result for the SAIK
- 19 samples would not have been submitted for DNA extraction (either cell or differential lysis extraction). Of these 19 samples:
 - 8 samples gave complex unsuitable, partial unsuitable or no DNA detected final results.
 Failure to submit these samples for DNA extraction would not have altered the final result.
 - 5 samples gave single source profiles from an assumed known contributor. Failure to submit these samples for DNA extraction would not have altered the final result.
 - Low vaginal sample <u>SFracture</u>) gave a <u>2p</u> mixed profile <u>cond on compl't with major contribution from the complainant (which was not interpreted or reported). The high vaginal sample from the SAIK gave a P30 positive result and spermatozoa were detected on the vulval sample on the Evidence Recovery slide. The vulval sample <u>and HV Sfracs both gave</u> a two person mixture which was conditioned on the complainant and gave a</u>

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remaining profile UKM1 (which was loaded to NCIDD). Therefore given the vulval result, and the https://www.high?vaginal result, failure to submit the low vaginal sample for testing would not have altered the final result for this SAIK.

Endocervix sample <u>SFrac</u> gave two person mixture which was conditioned on the complainant and gave a remaining profile with >100 billion support for contribution from the suspect (this result was the same as for the perianal <u>EFrac</u> sample and was not reported via EXH). Spermatozoa were detected on the Evidence Recovery slides for the

high vaginal 2, low vaginal, vulval, and perianal samples. The high vaginal (2? – need to specify, also applies to HVS 1) gave a similar result to the perianal EFrac and was not reported via EXH. The low vaginal and vulval Sfracs samples both gave single source profiles consistent with the suspect. (Vulval Efrac = 2p cond, LR susp't low support contribn) Given the results of the other samples for this case, and the fact that the

endocervix sample was not reported via EXH, (I'd leave this out – only true because no statement request received to date) failure to submit the endocervix sample for testing would not have altered the final result of the SAIK.

High vaginal sample Sfrac) gave a two person mixture which was conditioned on the complainant and gave a remaining male profile which was compared to two suspects for this case but both were excluded. The cervical sample for this case (which it should be noted had no spermatozoa detected on the Evidence Recovery or Differential Lysis slides) Sfrac gave a similar result. The low vaginal sample gave a AP/P30 positive result and the LVS Sfrac gave a similar final result to the cervical and high vaginal Sfracs. Spermatozoa were detected on the rectal sample on the Evidence Recovery slide, but Sfrac gave a complex final result (+ EFrac SS AKC compl't). Therefore given the results of the low vaginal sample Sfrac, failure to submit the high vaginal sample for testing would not have altered the final result for this SAIK.

o High vaginal sample () Sfrac gave two person mixture which was conditioned on the complainant and the remaining profile was used to compare against nominated suspects. The low vaginal sample was AP/P30 positive and the Sfrac gave a three person cond mixture which was also used to compare against suspects. Given the result of the low vaginal sample, failure to submit the high vaginal is not likely to have altered the final result for this SAIK. Change in # contributors may impact (as above)

Fabric sample support of contr bution from the suspect. (EFRAC = complex), This was the only result for this sample, however there are a large number of exhibits in this case with >100 billion support for contr bution from the suspect. Therefore although failure to submit this

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sample would have changed the final result of this sample, there are a number of other exhibits in this case linked to the suspect.

Discussion

The aim of this data analysis was to assess the 738 samples which had no spermatozoa or seminal fluid detected during the initial Evidence Recovery examination, and which were then submitted for differential lysis extraction, and compare these to pre August 2016 examination and sample submission strategies to determine what, if any, impact this may have had on the DNA results reported for the case as a whole.

738 samples has been considered a sufficiently large dataset for the purposes of drawing some general conclusions, although this relies on the particular cases processed during this period, and therefore sampling variability may show a greater or lesser impact by assessing another dataset. Sug. 'due to sampling variation there may have been a greater or lesser impact if another dataset had been assessed. It was beyond the scope of this data analysis to assess slides other than those that were originally zero spermatozoa detected at examination, and were submitted for differential lysis extraction since 8 August 2016.

The focus of this data analysis has been largely from a whole case perspective and several results were considered not to be impacted upon because of other samples/ similar results within the case. Assessing results on a whole case basis is part of standard case management practice, and is a process utilised across all case and sample types. It is acknowledged that the impact on individual samples may be considered significant if semen is not observed at examination, the presumptive screening is also negative and no further action was taken for that sample. There is a risk that if spermatozoa were consequently detected on the differential lysis slide and provided an interpretable DNA result, then potentially a valuable DNA profile for the case may not be obtained. What this data analysis shows is that this risk is mitigated when considering the typical case submission as a whole. The majority of SAIKs/sexual assault cases contain multiple swabs and items, which provide several opportunities to locate semen and subsequently obtain foreign DNA profile that may support an allegation of sexual assault.

Examination strategies are formulated to try and maximise the chances that even if one sample has no spermatozoa observed and the sample truly contains spermatozoa, then the DNA profile information can be obtained through other means. The presumptive screening for seminal fluid and examination strategies for submitting samples for differential lysis or cell extraction (including but not limited to: submission of external swabs/swabs from minors for cell extraction; submitting all areas from an item if one obtains a positive sperm or presumptive result) and also the capacity of STRmix to interpret even mixtures of up to 3 contributors (and including conditioning) all contribute to minimising the overall case impact for a particular sample.

It is acknowledged the slide read at both examination and differential lysis is a detection step, and the sample used to make the slide is a very small amount from the prepared suspension (a drop and 3uL respectively), which is a representation of the spermatozoa that may be present in the sample. For very low levels of spermatozoa, if a second slide is prepared from the sample, lower or higher levels of spermatozoa may be observed, as is to be expected as a consequence of from sampling variability.

The aim of the differential lysis process is to attempt to separate any spermatozoa from any epithelial cells in order to aid in the interpretation of the DNA profiles obtained. While complete separation of the

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spermatozoa fractions and epithelial fractions is the ideal, this is net-often not the case, and carry-over of epithelial cells into the sperm fraction is commonly observed. The advantage of using STRmix for mixture interpretation helps mitigate the consequences of failing to obtain the ideal separation of spermatozoa and epithelial fractions, which is the aim for differential lysis. In cases where a sample undergoes a cell extraction and the sample does contain spermatozoa, it is reasonable to assume that this extraction process will extract any DNA present in the sample, including from any spermatozoa present. STRmix will similarly aid in the interpretation of any mixed DNA profiles obtained from this process.

As described previously, there is a degree of concentration of spermatozoa in the differential lysis process, and the number of spermatozoa present to give a slide read of <+1 is very low (defined as 'very hard to find spermatozoa'), therefore to go from zero to <+1 after differential lysis may not be unexpected. Similarly a slide read of +1 ('hard to find spermatozoa') after differential lysis, following a zero slide read at examination may not be too concerning or necessarily need to be taken as symptomatic of a problem with ?inefficiencies in the examination slide read process.

Where a zero sperm read has produced a +2 sperm read of the slide after differential lysis, then this is harder to rationalise, even allowing for some variation in the subsample taken for the slide, and the differential lysis concentrating step. In this data there were 7 samples of the 738 total which showed this degree of difference ie. Zero to +2 (easy to find spermatozoa) which equates to 0.95% of this sample set. 6 of these samples would have undergone differential lysis extraction based on the presumptive result, and therefore the DNA results would are expected to have been unchanged. The one sample remaining was a vulval swab, which and would have been submitted for a cell extraction. Within this particular SAIK, the high vaginal and low vaginal swabs both had sperm observed retain-not-step: retain-not-step: retain-not-st

Conclusions

Therefore in summary:

- Of the 738 total samples for which no spermatozoa were detected on the Evidence Recovery slide, 591 also had no spermatozoa detected on the Differential Lysis slide.
- 147 of the 738 samples had spermatozoa detected on the Differential Lysis slide (>1+, 1+ or 2+).

The results of the analysis of this data set have shown that the difference in sensitivity of the Evidence Recovery and Differential Lysis swabs, although acknowledged, has not resulted in a systemic failure with regards to final reported results. (I wouldn't include this phrase-don't think a 'systemic failure' of reported results is the concern.)

For a small proportion of sexual assault samples containing low numbers of spermatozoa, the difference in the sensitivity between the ERT and Diff Microscopy methods had previously caused samples to be

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reported as semen negative when spermatozoa may have been detectable by microscopy following the differential lysis extraction procedure. As the ERT microscopy was previously used as a key determinant as to which extraction method was employed, and indeed whether the samples were submitted for DNA analysis at all, there is a potential impact on a small subset of reported results. Depending on the case circumstances, the ability to report that semen was detected may or may not be critical, in the context of the allegation. For sexually active adults, confirming the presence of spermatozoa on intimate swabs, may perhaps not be considered as critical as it is for sexual assault allegations involving complainants to whom limited opportunities for the transfer of spermatozoa may make such findings more pertinent (for example minors, the elderly or individuals with disabilities).

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In many instances we have found that this issue is mitigated by the common practice of submitting multiple swabs within a typical SAIK, thus increasing the chances of detecting semen. The fact that this data analysis has found that the methods in use prior to August 2016 would have had in only a relatively limited impact on the particular reported results for these SAIKS should be reassuring, however we should continue to strive to eliminate impacts of this type for all cases, and seek to improve the sensitivity of our methods, including by selecting the most effective point in the process to conduct microscopy. It may also be the case that if one swab from a SAIK is affected by this issue, then other swabs in the SAIK could also have an increased chance of being similarly affected, as microscopy examinations are not genuinely independent events. A SAIK containing a High Vaginal Swab with low numbers of spermatozoa may be more likely to also contain a Low Vaginal Swab with low numbers of spermatozoa. The microscopy process is a manual one, which is in a large measure dependent on the technique and ability of the examining scientist. If the slide created from one SAIK swab is affected by an issue which decreases the chance of observing spermatozoa, then this would also tend to affect the chance of detecting spermatozoa on a slide from a second swab of the same SAIK, made by the same scientist.

There was one sample in the 738 sample data set which would not have been submitted for DNA testing pre-August 2016, and which gave 1+ spermatozoa on the Differential Lysis swab and a final DNA result consistent with the suspect. This was the only DNA result for this case. Pre-August 2016 this sample would have been reported to the QPS as "Semen not detected" and no further action taken. It should be noted that this presumptive EXH advised the QPS that "Spermatozoa were not observed..." rather than advising that there were no spermatozoa present. If deemed critical, the QPS could request further processing of this sample.

Therefore, although some individual samples may be negatively impacted as a consequence of the sensitivity of the examination slide process, overall this is considered to be an acceptable risk as it occurs relatively infrequently, and from a case perspective the risk is mitigated by the established practices of multiple sample submissions, examination submission and interpretation strategies.

The results of this study did not demonstrate a systemic failure in the examination of exhibits for seminal fluid. The examination processes described throughout this report, as well as the resulting DNA profile, the assessment of the whole case, and the ability to submit for processing any samples not actioned, aims to mitigate the risk that may arise when spermatozoa is not detected at the examination step. Continuous process improvements are imbedded in Forensic DNA Analysis and are part of our quality management system, and improvements to the examination of sexual assault process will continue, as they will with all processes within the unit, to ensure any risks are mitigated as much as practical. (???)

Commented [LR1]: Not sure if this para is required given previous paras.

General points:

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Suggest all wording used to describe stats/ interpretations should be as per standard wording for statements - see Procedure for Release of Results SOP 17119.

e.g. and gave a remaining profile with >100 billion support for contribution from the suspect."

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Suggested wording: "Based on statistical analysis it is estimated that the mixed DNA profile obtained is greater than 100 billion times more likely to have occurred if the suspect has contributed DNA along with the complainant, rather than if he has not."

Suq. leave out identifiers (sample barcodes) where discussing interps/case details, ?Could number the samples based on the order in the xls. and refer to Sample 1, 2 etc. within the report – add appendix/notes to explain.

Sug. leave out any interpretations which have not been finalised (reviewed), and may be subject to change.

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JH-28

Erin Shearer

From: Paula Brisotto

Sent: Thursday, 4 May 2017 3:13 PM

To: Justin Howes **Subject:** Proof read

Attachments: Sexual assault modified process data review_updated PMB.xls

Hi Justin,

How does this sound? I'm hoping if I put into word what the intention is, it might help Kylie with reviewing the spreadsheet.

Thanks, Paula

Hi Matthew and Kylie,

I've gone through the spreadsheet with your comments and feedback. Again, thanks for doing this as it can be quite mind numbing ©

In the attached spreadsheet, there are rows highlighted blue and one yellow.

The yellow one is a sample from a case which, through the pre-modified process, wouldn't have provided QPS with the critical DNA profile information for the case.

The blue highlighted rows are ones which were flagged in the previous versions of spreadsheets, and I've updated them, or would like your review/discussion on. Matt, I believe I have discussed some of these with you.

I've started to put some wording in a report.

The scope of this is to look and see if any cases from the 738 zero sperm reads at ER would have been impacted in relation to DNA profile information reported to the QPS. I'm summarising the data as simple as I can.

- Any samples that would have gone through DLYS based on exam strategy/presump, I've not delved into further, as the resulting extraction would be the same.
- Any samples with NDNAD, No profile, complex unsuitable, based on DNA result do not impact the case.
- Samples the obtained the known contributor do not impact the case.
- Samples that have other items in the SAIK/item which were submitted for DLYS through the pre-modified process (some of these would have obtained similar results, or would themselves have still undergone CELL extraction) which have not impacted the results for the case.
- Samples that would have undergone CELL extraction (which theoretically would have co-extracted DNA from sperm present in the sample)
- Samples that would be NFA, and no other samples were submitted or obtained DNA results useful to the QPS

I would really appreciate your input on this, and will send you a draft once I have the bones of it together.

Thanks, Paula



Paula Brisotto

A/Managing Scientist
Police Services Stream
Forensic & Scientific Services,
Health Support Queensland, Department of Health

HSQ's vision | Delivering the best health support services and solutions for a safer and healthier Queensland.

Queensland Health acknowledges the Traditional Owners of the land, and pays respect to Elders past, present and future.

E-3p

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Notes								HVS and LVS both sperm pos at ER. Yulval submitted for CELLS	-					6 swabs (HV8, LV8, vulval, perlanal, rectal, oral) - Vulval and rectal	micro pos at ER. HV3 +2, LV3 +1, pertanal <+1. HV3 would have	change in outcome of DNA results.		CELL on expect have given a mixed profile given on 1 of ER.	samples (thich neck/ear) \$5 such, NVS as assumed known. Ext varies	CMPU				HVS gave some result. Vulnal swab would have gone through cell	extraction most likely (based on DNA profile result and epi micro	from ER) would have obtained 2P mixture. "major" known, Could have conditioned and remaining companied to current.	Conditioned and remaining compared to support	Cell extraction would have obtained same DNA result. No epi seen on	ER side.	Cell extraction would have obtained same DNA result. No epi seen on	ER side.		Vuhal swab sperm seen at ER. SS matching suspect. swab would have gone brough cell extraction most likely (based on DNA profile result and ep micro from ES) would have obtained 2P mixture. Could have	conditioned and remaining compared to suspect.	Vulval swab sperm seen at ER. 55 matching suspect, swab would have gone through cell extraction most likely (based on DNA profile result.	and ep mido from EK would have obtained 2º mixture. Could have conditioned and remaining compared to suspect.		Sperm seen on all other SAIK swabs. All DNA results the same.				From piece of fabric from teatowel. Note: seems to be from same	Results support out from both such and compt.				Vulval sperm pas at ER. HVS p30 pos submitted for DLYS. Similar DNA	results	Vulval and IVS sperm seen at ER.	HVS 2 was pos for sperm, therefore HVS 1 would have been	Submitte for a ri.	HV3 and Rectal swabs both 0 sperm at diff, presump neg. HV3 NFA.	Rectal as CELLS would have given known profile.		_				CLOYEDANA	See 72/334423		Vulval swab would have zone for cels - sp fraction cusp x1008	_
DNA Results impacted?		No Contraction		No-DLT3	No-DLYS	No-Orac		No-Other results		No-DLYS	No-DLY3		No-DLY3		No-DNA resut		No-DLYS	NO-CELLS		No-other samples/CELLS	No-DLY8	No-DLYS	No-DNA result			Sillar samue (CEII	No-DLY3		No-CELLS		NO-CELLS	No-D-48	Section 200	NO-OMET RESURD LELLS		No-other results/CELLS	Neorts	No-other results	No-DNA result	No-DNA result No-DLYS			No-other results*	No-DLYS	No-DLYS	N-D-VS		No-other results	No - other results No-DLYS		No-DLYS No-DLYS		Yes	No-DLYS		No-DNA result	No-DNA resut	No-Known	No-DNA result	No-DLY B	No-DNA result	No? other results	No-DNA result No-DLY8
DNA results		ompres discussions			22		בן בתומ: (נסו ובלים ובמ	72p mix		30 mix		2p mix	2p mix			_	SS - susp	_		P cond known remaining >1006 support for suspt	2		Complex unsuitable			De count borones annairing of 0.08 encount for event	Containment remaining states support for suspic		52 - susp		SS - susp			Ar company remaining major states support to suspen		2. P known and susp represented	to upload UtiM1 and UKP1			Complex unsuitable 2P?			S. sura			SS suspect		2P 'major' susp 'minor' known	LP major unknown male 'minor known		2P 'major' susp 'minor' known		PP: "Major" male minor unsuitable - compl?			complex unsuitable PP	complex unsuitable PP			Itable		rts contrib susp. 18 - 1008	PP unsuitable SS assumed inown
Submission (pre modified)		DIVE	Ī			DLYS	Ī	CELLS		DLYS	DLYS		DLYS		NFA		DLY8			CELLS	DLY8		NFA				DLYS		CELLS			DLYS				CELLS				NFA C			NFA	Ī		DLYS		NIA Z	NFA DLY8		DLYS		NFA 2	DLYS		Ĭ				DLY8			CELLS P
A.		200 000	Т	AP pos 10s	Т	AP pos 80s	т	AP neg							AP pos 110s			Ar neg		AP neg		AP pos 10s	AP pos 30s			200	AP DOS 90s		AP neg		AP neg			Ar pos 1maos		AP neg	A* pos 205	AP neg	AP pos 40s	AP neg				AP pos 43s	П	Т	Ar pos 305	AP meg	AP pos 1m15s	Γ	AP neg		AP pos 30s					П	AP neg	П	AP neg		AP neg
D20		and hos	pso pos	p30 pos	P30 pos	p30 pos	and non			030 008		pag bes	p30 pos		p30 neg		p30 pos				p30 pos	p30 pos	p30 neg				030 003					p30 pos		Sauned			pag pag		p30 neg	p30 pos	and and		DEC DATE	p30 pos	p30 pos	p30 pos	and ned	Sau neg	p30 meg	and and	030 D0s	and and	p30 neg	pag bos	and hos	p30 neg	pad neg			p30 pos		030 neg	03D 005
Diff clide sperm mioro recult	1					2 sperm		2+ sperm		1+ sp h	1+ 3p H		1+ 3p H		1+3pH		1+ 3p	1 sperm				1 sperm	Ι			- toem			1 sperm	No. Charles	1 sperm	1 sperm		T sperm		1 sperm	I	1 sperm	П	1 sperm			1 soem	Γ	1 sperm		1 sperm	1 sperm	1+ sperm		1+ sperm			1+ sperm		<1+3pH		<1+3p H	<1+3pH			ct sperm	ct sperm
	Date diff slide read	40/00/2016	13/03/5010	20/12/2016	13/01/2017	13/01/2017	- Inches	13/03/2017		24/08/2016		18/08/2016	18/08/2016			29/08/2016	2/03/2016	3/03/2016		19/09/2016	32/03/2016	23/09/2016	23/09/2016			33/09/2016	23/09/2016		12/10/2016		12/10/2016	19/10/2016	200(14)00	48/11/4016		28/11/2016	28/11/2016	7/12/2016	7/12/2016	14/12/2016	2000		23/12/2016	23/12/2016	20/01/2017	20/01/2017	43/05/5027	23/02/2017	13/03/2017		15/03/2017		21/03/2017	77/03/2017		18/08/2016	19/08/2016	29/08/2016	29/08/2016 39/08/2016	2/05/2016	2/09/2016	3/03/2016	3/09/2016
Decoription	1	An februar	AD IBOTIC	low veginal	Fabric KG40	SA.	- All	Vulvai		AP fabric	AP fabric		AP fabric		low vaginal swab		AP fabric	vulvalismao		oral swab	Scraping A4	high veginal swab	low vegnel swab			Contrast Course.	verval swab		midpubic area swab	Charles and Charles and Charles	top public area swab	fabric		21		perional	ngn vagna	low veginal	low veginal	Item V1b T/L fabric			Pabric	vulval	fabric KG44	fabric KG46	CAH	IVS	e2 fabric		LVS 1		Sanitary Ped	Fabric 7		AP fabric	AP fabric	low veginal swab	vulval swab	perianal swab AP fabric	vulvalswab	hich verinal swab	wet anal swab
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Item (EXH) barcode QP/CA Number									
	Description	Date diff slide read	Diff slide sperm micro result	р30	AP	Submission (pre modified)	DNA results	DNA Results impacted?	Notes
	External Vagina	19/09/2016	<1 sperm	p30 pos	AP pos 60s DI	TAS comb	ex unsuitable	No-DLYS	 This sample would have undergone cell ext. From this DNA rpofiel
		a portion						51110	susp is major' therefore Cell ext likely to give results based on < 1
	Scraping A3	22/09/2016		p30 neg	Ar neg	DLYS compl	nem-1006 susp unsuitable	No-DNA result	בלו אפנו עו בא
	scraping AP pos fabric	27/09/2016		p30 pos			detected	No-DLYS	_
	perianal wet swab	29/09/2016		030 neg	40c	S S	No DNA detected	No-DNA result	
	blind vaginal 1	13/10/2016		p30 pos	AP pos 10s DI	DLYS 2P - cc	rem unsuit	No-DLYS	
	blind vaginal 2	13/10/2016			AP pos 15s DI	LYS 2P - cc	rem unsuit	No-DLYS	
	Dlind vaginal 3	13/10/2016		p30 pos	AP pos 20s DI	17 55	- cond known rem consistent	No-DLYS	
	fabric	19/10/2016			П	DLYS compl	9	No-DLYS	
	fabric	24/10/2016		p30 pos		No	detected	No-DLYS	
	LVS Iow vaginal	24/10/2016	<1 sperm	p30 pos p30 pos	AP pos 20s DI	LYS 3P condition	cnown rem unsuitable -' known 'minor' unsuitable	No-DLYS No-DLYS	
	ledes	98/10/2016	mars.		and do	SI DE	mond? Bam cites	No.CELIS/otherresults	This sample would have undergone cell ext. Additionally HVS and LVS corm coon at EP DIVS
			-						All actions of the second control of the sec
	rectal	28/10/2016	<1 sperm		AP neg CE	CELLS SS susp		No-CELLS -other results	have given a mixed profile which could have been conditioned.
									Item would have gone for cells expected to have obtained mixed
	lucione inco	24/14/2016	7		70 av	3113	and the second s	No. CELLS	profile which could have been conditioned. Sperm micro on rectal
	peri-vaginal peri-anal	25/11/2016	₹ ₹			No		No-DNA result	swab - complex unsuitable. All other swabs NDNAD
	high vaginal	29/11/2016	<1 sperm	P30 neg	15.5	CELLS SS ass	u	No-Known	
	low vaginal	29/11/2016	<1 sperm		П	SS		No-Known	
	vulval	29/11/2016	<1 sperm	P30 neg	AP pos 45s CE	CELLS SS ass	known	No-known No-DLYS	_
	fabric	1/12/2016	<1 sperm	p30 neg	Z	FA PP uns	unsuftable	No-DNA result	_
	low vaginal	29/11/2016	<1 sperm		AP neg NI	FA SS - as		No-Known	Should have been included
	vulval	1/12/2016	<1 sperm	p30 pos	AP pos 40s DI	LYS SS ass	assumed known psti	No-DLYS	_
	rectal	6/12/2016	<1 sperm	030 000	8	SS		No-Known	
	lvs1	6/12/2016			AP pos 50s NFA	SS		No-Known	
	inner labial	6/12/2016	<1 sperm			S SS	lown psti	No-Known	
	density leaders daily	21/13/2016	of more	20 000	AD not 15c	NEA 30 mg	Trade Chamber manner by MCDD Proposed Section 1997	Mo2 other country	LVS would have gone for DLYS based on p30 - 3P 'major' known. C
	low vaginal	1/12/2016			T	LYS 3p conc	Contraction (contraction)	No-DLYS	
									Zero sperm on diff slide too - incorrect as confirmed < 1 seen on diff
	condom K a/s	7/12/2016	<1 sperm		AP neg CE	CELLS No DN	No DNA detected	No-DNA result	slide
	hvs	14/12/2016		p30 pos	.m 30s	SS	yn psti	No-DLYS	_
	4A T/L	14/12/2016			AP neg CE	03	ole .	No-DNA result	
	scraping A1 fabric 4D	14/12/2016	<1 sperm	p30 pos	IQ	LYS compl	ex unsuitable	No-DLYS	_
	perianal	22/12/2016	< sperm		AP neg	FA SS acc	ex unsurrable	No-Dina resuit	
					0				Perianal swab 0 sperm seen at Diff. Obtained 2P cond known
	vulval	22/12/2016	<1 sperm	p30 neg		æ	'Major' known	No-other results/CELLS	remaining susp >100B. Would have been submitted for CELLS - notentially giving the same result.
	rectal	22/12/2016	<1 sperm		AP pos 90s CE	CELLS 3P cor	wn/susp	No-other results/CELLS	0
	fabric	23/12/2016	<1 sperm	p30 neg	IQ	DLYS 2P sup	supports cont susp. No support for compt.	No-DLYS	(note - piece of fabric has two areas a and b - b was p30 pos therefore both areas submitted for DLYS)
									From piece of fabric from teatowel. Note: seems to be from same teatowel as as 5 other samples two of which were sperm pos at ER.
	fabric	23/12/2016	<1 sperm	p30 neg	Z	VFA 2P sup	ports cont susp. No support for compt.	No-other results?	Results support cont from both susp and compt.
	vulval	23/12/2016	<1 sperm		AP neg CE	CELLS 3P cor	3P cond remaining? (Known 'major')	No - cells?	Sample would be submitted as CELLS - epi may have swamped the other contrib (note complainant 4yo)
	fabric2	6/01/2017	<1 sperm	p30 neg	2 2	NFA No DN	ected	No-DNA result	
	fabric 4	6/01/2017	<1 sperii	p30 neg	Z	FA No DN		No-DNA result	
	perineal wet	6/01/2017	<1 sperm	p30 neg	AP pos 90s CE	CELLS compl	:omplex unsuitable (PP)	No-DNA result	_
									HVS and LVS both pos for sperm at ER. Cell extraction would have
	vulval	11/01/2017	<1 sperm	200000	AP neg CE	CELLS PP susp?	400	No-other results/CELLS	obtained possible mix which could have been conditioned.
	90000	12/07/2021	made 44				naden.		HVS would have undergone DLYS ext and not obtained any different
	IVS	13/01/2017	<1 sperm	p30 neg	AP pos 80s NI	NFA COMPLEX		No-DNA result	result.
	mouth swab 1	20/01/2017	<1 sperm		AP neg	CELLS 3p cond	D	No - cells?	Sample would be submitted as CELLS - epi may have swamped the other contrib
	fabric 3	20/01/2017		p30 pos	IQ		complex unsuitable (1pk)	No-DLYS	
	hve 2	31/01/2017		230 050	AD mos 30s	July S	20 cond ram 1000-10 000 alim	O NICON	HVS 1 sperm seen at ER - both HVS would have been submitted for prive
	fabric 2	30/01/2017		p30 neg			detected	No-DNA resut	
	fabric 3 A2 fabric A - area 2	30/01/2017	<1 sperm	p30 pos n30 neg	0 0	LYS compl	ex unsuitable (PP)	No-DLYS No-DLYS	
									A third piece of fabric from same tem, which tested neg, no sperm seen obtained minor with more information. All would have been
	fabric	13/02/2017	rm.	p30 neg	ō	CELLS 2P 'ma	'major' known minor unsuitable (RW?)	No-DNA resut	submitted for CELL
	fabric Item 9 subsmple 1	13/02/2017	<1+ sperm	p30 neg	0 0	ELLS ZP 'mz	minor unsuitable (RW?)	No-DNA resut	

motorioge CEXH) harrode OD/CA Nimber	Description		Diff slide sperm micro	030	ΔÞ	Submission (pre	DNAresults	DNA Recults impacted?	Notes	
		Date diff slide read	result	Ŀ	:	modified)				
	Item 9 subsmple 2	15/02/2017	<1+ sperm	P30 Pos		DLYS		No-DLYS	-	
	Item 11 Sect 1	15/02/2017	<1+ sperm	P30 Pos		DLYS		No-DLYS		
	Item 13 Section 3	17/02/2017	<1+ sperm	P30 Pos		DLYS		No-DLYS		
	Item 13 Section 8	17/02/2017		P30 Pos		DLYS		No-DLYS		
	Fabric 1C - section B	23/02/2017		P30 Pos		DLYS		No-DLYS		
	Fabric KG11	1/03/2017	<1+ sperm		AP neg	CELLS	No DNA detected	No-DNA result	-	
	Semen in-tube	28/02/2017	<1+ sperm		AP neg	CELLS	No DNA profile	No-DNA result		
	Vulval	28/02/2017	<1+ sperm		AP neg	CELLS	SS Assumed known	No-Known		
	Fabric 7j	10/03/2017	<1+ sperm	P30 Pos		DLYS		No-DLYS		
	8	13/03/2017	<1+ sperm	1	AP ned	NFA	Sp. 'maior' ass known, Minor?	No-other results	HVS submitted for Diff as p30 pos (no sperm seen at ER, 2+ at Diff) obtained 2P even mix, cond on ass known	
	perianal	13/03/2017	<1+ sperm			CELLS	SS assumed known	No-Known		
	s1 fabric	10/03/2017	<1+ sperm	b30 pos		DLYS		No-DLYS		
	s 10 fabric	10/03/2017	<1+ sperm	930 pos		DLYS		No-DLYS		
	Semen in-tube	13/03/2017	<1+ sperm		AP neg	CELLS	No DNA detected	No-DNA result	-	
	Semen in-tube	13/03/2017	<1+ sperm		AP neg	CELLS	Complex unsuitable - PP	No-DNA result		
	hvs	13/03/2017	<1+ sperm	/ sod 0£d	AP pos 30s	DLYS		No-DLYS		
	lvs	13/03/2017	<1+ sperm		AP pos 40s	DLYS		No-DLYS		
	endocervix	15/03/2017	<1+ sperm			NFA	2P	No-other results	HVS, LVS, Vulva and Perianal a I gave micro pos at ER.	
									Rectal swab sperm pos at ER. Rectal complex unsuitable. LVS p30 pos- would have undergone DLYS. Vulval would have gone through CELL	
	HVS	21/03/2017	<1+ sperm	p30 neg	AP pos 55s	NFA	2P(?) cond assumed known. Remaining male (as for LVS) (not CM No-other results	Mo-other results	extraction	
	TAS	21/03/2017	<1+ sperm	y sod 06 d	AP pos 60s	DLYS	2P(?) 'major' assumed known. 'Minor' male (not CM)	No-DLYS	< 1 Epi at ER for all 3 swabs. Cervical (0 sperm at ER and d ff) gave mixed profile rem. UKM loaded to NCIDD.	
	Vulval	21/03/2017	<1+ sperm	/ bau 06d	AP pos 65s	CELLS	2P(?) 'major' assumed known. 'Minor' male (not CM)	No-other results/CELLS	Sample would have undergone CELL extraction	
	HVS	22/03/2017	<1+ sperm		AP pos 60s	DLYS		No-DLYS	-	
	FAS	28/03/2017	<1+ sperm		AP neg	NFA	Complex unsuitable PP (not CM)	No-DNA result/other results	HVS sperm seen at ER.	
	vulval	28/03/2017	<1+ sperm		AP neg	CELLS	SS -uk m1 (PP)	No - CELLS - other results	Would have undergone cell extraction. HVS sperm seen at ER.	
	Vulval	27/03/2017	<1+ sperm	1	AP neg	CELLS	No DNA detected	No-DNA resut		
	Mouth	22/03/2017	<1+ sperm	1	AP neg	CELLS	2P 'major' known 'minor'	No-DNA result/other results	HVS sperm seen at ER.	
	Fabric 8 Excised	27/03/2017	<1+ sperm	sod 0£d		DLYS		No-DLYS		
	Fabric 8 Area 1	27/03/2017	<1+ sperm	b30 bos		DLYS		No-DLYS		
	Fabric 8 Area 2	27/03/2017	<1+ sperm	sod 0£d		DLYS		No-DLYS		
	Fabric 5 Area 1	27/03/2017	<1+ sperm			DLYS	No DNA detected	No-DLYS		
	Fabric 5 Area 2	27/03/2017	<1+ sperm			DLYS	No DNA detected	No-DLYS		
	Fabric 5 Area 3	27/03/2017	<1+ sperm				No DNA detected	No-DLYS		
	Fabric 5 Area 5	27/03/2017	<1+ sperm			DLYS	No DNA detected	No-DLYS		
	Fabric 5 Area 9	27/03/2017	<1+ sperm				No DNA detected	No-DLYS		
	Fabric 5 Area 10	27/03/2017	<1+ sperm			DLYS	No DNA detected	No-DLYS		

No-DIX5 - not impact as sample would have gone through diff lysis based on proung results and exam strategy
No-DMA result. Volk results were nor meaningful - No DMA detect. No DMA profile chained or complex/PP
No-CAMO-ON TO MA results see the assumed confination or contributor or not one such as the No-CAMO-ON CAMO results were the satured contributor or not set EV and or when through DLYS and the

Notes	_		HVS, LVS, Vulva and Perianal all gave micro pos at ER.	Rectal swab sperm pos at ER. Rectal complex unsuitable.	Cells from assumed known may swamp out male profle in cell extraction?	HVS sperm seen at E.R.	0 epi seen at ER		HVS snerm seen at FR
DNA Results impacted?	No-DNA result	No-DNA result	No-other results	No-other results	No-other results/CELLS	No-DNA result/other results	No-DNA result	No-DNA result	No-DNA requit/other require
DNA results	No DNA detected	Complex unsuitable - PP	dč	2P(?) cond assumed known. Remaining male (as for LVS) (not CM)	2P(?) 'major' assumed known. 'Minor' male (not CM)	Complex unsultable PP (not CM)	PP-unsuitable (not CM)	No DNA detected	28 'mainr' known 'minor' uncuitable
Submission (pre modified)	CELLS	CELLS C	NFA 2	NFA 2	CELLS 2	NFA G	CELLS P	CELLS N	CELIS
АР	AP neg	AP neg (AP neg	AP pos 55s	AP pos 65s	AP neg	AP neg (AP neg (AP next
p30				p30 neg	p30 neg				
Diff silde sperm micro result	<1+ sperm	<1+ sperm	<1+ sperm	<1+ sperm	<1+ sperm	<1+ sperm	<1+ sperm	<1+ sperm	<1+ enerm
Description	Semen in-tube	Semen in-tube	endocervix	HVS	Vulval	FAS	vulval	Vulval	Mouth
QP/CA Number Description									
Item (EXH) barcode									
Subsample Barcode									



JH-29

Health Support

Queensland

Forensic and Scientific Services

Forensic DNA Analysis - Management Team Meeting

Kylie Rika (KDR)

Date: 12 May 2016 Time: 9.30am – 11.45am

Venue: FSS CR611

1. Present

Allan McNevin (ARM)

Amanda Reeves (AJR) Kerry-Anne Lancaster (KAL)

Cathie Allen (CJA) Luke Ryan (LBR)

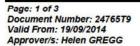
Kirsten Scott (KDS) Sharon Johnstone (SMJ)
Justin Howes (JAH) Wendy Harmer (WAH)

2. Apologies -

3. Guests - Nil

Agenda items

Item	Topic	
1.0	Confirmation of previous minutes - SMJ	
2.0	Conflicts of Interest - Nil	
3.0	Action Register – See below	
4.0	4.1 Workplace Health & Safety Issues – Annual safety audit of lab space to be done – allocated to a staff member on campus to complete. Vicky has completed a risk assessment of the freezer area – will be loaded to QIS soon. Details within the risk assessment. 4.2 Analytical Issues of Note - LBR 7500B has dye calibrations completed, 7500A to be done this morning. Hamilton instruments have been purchased and soon to be delivered to campus. No update on mastermix separation issue – other than LBR's email sent during this week. No comment back yet from PP21 BSAG labs. 4.3 AUSLAB Working Party Update – KDS A request for change regarding QIASymphony will need to be submitted (Quant Report Analyser Mask work).	





4.4 Project Updates

Project #152 – Y-Filer Plus – LBR.

No update from last meeting.

Project #168 - Validation of QIAsymphony - LBR

Running some final AS integrated runs (ie overnight & different temps). Report anticipated to be completed in the next week. Training in the next few weeks.

Project #170 – Reassessment of in-house stutter thresholds and stutter file used in STRmix – JAH.

Linked with v2.4 of STRmix. AAP and EJC looking for samples that can be re-used for v2.4. Need to coordinate any changes with implementation with v2.4 being brought online.

Project #171 Verification of the Internal Lane Standard from CC5 to WEN (Promega) – LBR

Presentation and discussion ensued. LBR to send out an email with clarifying email and a voting question to respond to.

Project #172 – Phadebas testing from suspension in ERT – ARM

Report has been provided to Management Team for review. ARM would like to investigate the way that suspensions are made, so this project will be placed on hold until this has been completed. ACTION: ARM to look at the way in which suspensions are made.

Projects on-hold

Project #146 - GlobalFiler - LBR

On hold until new size standard has been completed (17.03.2016)

5.0 New business

5.1 LOD and LOR 3130x/ PP21 WEN (LBR)

As above. Presentation saved here: G:\ForBiol\DNA Analysis Team Meetings\DNA Analysis Management Team\2016\Jan - Jun

5.2 HR – part day absences (time claimed) (CJA)

Please ask team members to ensure their leave taken is to the 15min, 30mins, 45mins or the hour as this requires resources to check on.

5.3 OQI 41850 re: Link not being associated and reported (SMJ)

Link #12541 was found in 2012, however not placed into Auslab and reported to QPS. QPS have been notified. This was found due to the daily audit which was put in place after the last incident was found. Link was crime scene to crime scene, and when updated recently it was to add a Person Sample and audit discovered the missing crime scene profile – refer to OQI for detail.

5.4 Sperm seen on Diff Lysis extraction slide vs ER suspension slide - ARM

This has been raised as a potential issue. First step to look at this is to compare a number of Diff Lysis extraction slides against ER suspension slide to see if the trend shows that more are seen on the extraction slides as a norm. This first step project is being monitored by ARM and KDS.

6.0 New business – for noting

Nil

Next Meeting

Date: 26 May 2016, 9.30am

Venue: CR611

ACTION REGISTER

Minutes Reference	Item Number	Subject	Action	Action Officer	Status
14/04/2016	5.2	Validation Baseline Methods	To be prepared - one page on software, benefits, costs, if more than 1 product etc. Once software has been looked at, then this will affect the SOP to be created – Found STR-validator program (G:\ForBiol\DNA Analysis Team Meetings\DNA Analysis Management Team\2016\Jan – Jun) – SMJ to complete a project initiation document	SMJ	Closed
29/04/2016	4.1	Change in access for retrieving liquid nitrogen	Add a comment to the SOP	KDS	Closed

JH-30

Erin Shearer

From: Allan McNevin

Sent: Friday, 27 May 2016 12:23 PM

To: Deborah Whelan; Allan McNevin; Amanda Reeves; Cathie Allen; Justin Howes; Kerry-Anne

Lancaster; Kirsten Scott; Kylie Rika; Luke Ryan; Sharon Johnstone

Subject: FYI - Project proposal #181 - Sperm microscopy sensitivity

Hi all,

Just as an FYI – As discussed in the management team meeting today, the further investigation into the differences between ER microscopy and Diff Lysis slide microscopy will not be a formal change proposal and I am giving it proposal # 181 and calling it "Investigation into sensitivity of spermatozoa microscopy"

Cheers

Αl

Allan McNevin

Forensic Scientist - Advanced, Evidence Recovery Team Forensic DNA Analysis | Police Services Stream Forensic and Scientific Services | Health Support Queensland

JH-31

Erin Shearer

From: Allan McNevin

Sent: Thursday, 1 September 2016 12:55 PM

To: Allan McNevin; Amanda Reeves; Deborah Whelan; Justin Howes; Kirsten Scott; Kylie Rika; Luke

Ryan; Paula Brisotto; Timothy Gardam; Emma Caunt; Rhys Parry

Subject: Project #181 - project plan and experimental design

Hello all,

Please review the following documents and complete the risk assessment in the project plan document

I:\Change Management\Proposal#181 - Sperm microscopy sensitivity
Proposal#181 Investigation of sperm micro sensitivity - Experimental design v1.0.docm
Proposal#181 Investigation of sperm micro sensitivity - Project plan v1.0.docm

I understand there is a change over of acting and when it's time to put pen to paper I will update who is in what seat, and with that in mind, please provide feedback to Emma and myself by COB next Friday

Thanks

Αl

Allan McNevin

Forensic Scientist - Advanced, Evidence Recovery Team Forensic DNA Analysis | Police Services Stream Forensic and Scientific Services | Health Support Queensland Department of Health | Queensland Government

JH-32

Erin Shearer

From: Paula Brisotto

Sent: Thursday, 9 February 2017 5:08 PM

To: Justin Howes; Cathie Allen Subject: Microscopy stats FYI

Attachments: Sexual Assault modified process data review.doc

Hi Cathie and Justin,

Please find attached a quick number crunch I did on the ER slides and subsequent Diff slides since the implementation of the modified sexual assault process in August 2016.

Thanks, Paula



Paula Brisotto

Team Leader – Evidence Recovery and Quality Team, Forensic DNA Analysis, Police Services Stream Forensic & Scientific Services, Health Support Queensland, **Department of Health**



HSQ's vision | Delivering the best health support services and solutions for a safer and healthier Queensland.

Queensland Health acknowledges the Traditional Owners of the land, and pays respect to Elders past, present and future.

Sexual Assault modified process data review

02/02/2017

In August 2016, sexual assault slides that were microscopy negative for the presence of sperm at Evidence Recovery stage were sent for differential lysis, and the differential lysis slides read for the presence of sperm.

Since August 2016, 528 differential slides have been read from ER micro negative slides.

Of the 528 slides:

428 differential lysis slides read as 0 sperm (81.1%)

66 differential lysis slides read as <+1 sperm (12. 5%)

27 differential lysis slides read as +1 sperm (5.1%)

6 differential lysis slides read as +2 sperm (1.1%)

1 differential lysis slide read as +3 sperm (0.2%)





Pathology & Scientific Services Biomedical Technology Services

PART A: REQUES	T FOR CHANGE	Proposal #27
TITLE: Further validation	n of BioSign PSA cards and refine	ement of standard operating procedure
PROPOSED BY: Rhys I	Parry and Kate Lee	
DATE: 19 th November 2	007	URGENT
Predicted Scope (Tick bo	AND VIEW PORTS	
THE PROBLEM: //ssue	s driving the project / Justification for	or change: (approx 100 words)
absence of sperm is be relying on the literature for cross-reactivity and was Our case load means the near future due to the fathe effects of washing,	eing challenged/questioned mor for theoretical knowledge of PSA hing of exhibits). How much bett at Biosign based PSA determina act that most cases are not resolute.	mechanism of detection of semen in the revigorously. At present we are largely in a wide variety of situations (especially ter to be able to quote in-house studies? ations will be relevant for the foreseeable ved through to trial for at least two years of optimal elution volumes, and direct verse.
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Authorised by V lentile

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Page 1 of 8

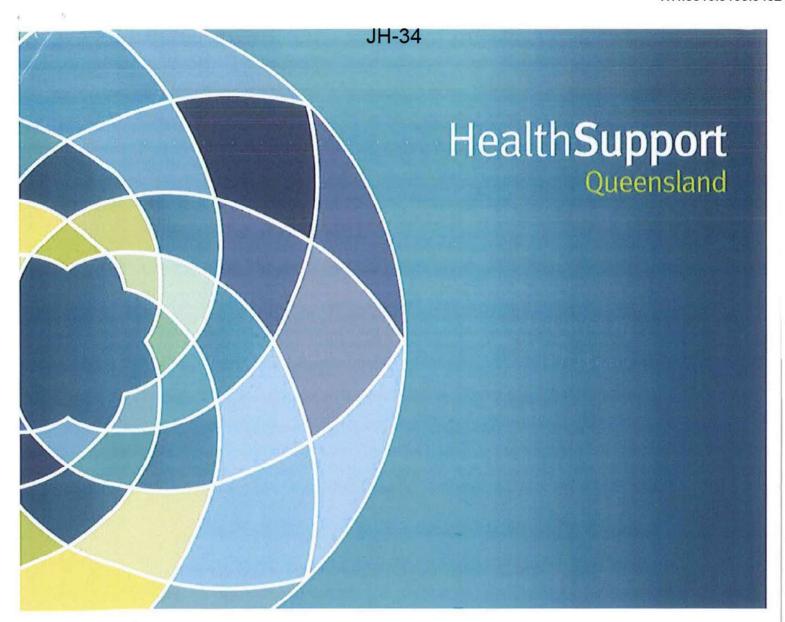
RECOMMENDATIONS from Forensic Biology Management Team

Proposal directed to Minor Change Register

22872R1.doc

(Date: 23/11/07)

No. of the contract of the con	Change Proposal Template
	Part B planning process (Internal Change) art C planning process (Project Planning) ne
REASON: In-house studies / valide confirm concepts report	
literature.	
DOCUMENTATION REQUIRED by Forensic Bi	ology Management Team (Please tick):
Internal Change process (Complete Part E	3, D, E) Due: 6/12/07 (date)
☐ Project (Complete Part C, D, E)	Due:(date)
FB Management Representative: V (ent.)	
FBMT request completion of Part B (further info	
Start Date:	
Proposed Implementation Date:	
What is changing? (approximately ½ page)	
Attachment:	
Who is involved? Change Leader, Change To	eam and Sponsor
VALIDATION PLAN:	Required: YES NO
Attachment:	
DOCUMENTATION AFFECTED:	
Document#	Document Title



Project Proposal #181

Investigation into the sensitivity of spermatozoa microscopy

August 2016 Allan McNevin, Emma Caunt and Cathie Allen



Project Proposal #181 - Investigation into the sensitivity of spermatozoa microscopy

Published by the State of Queensland (Queensland Health), September 2016



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Document Details

Contact for enquiries and proposed changes

If you have any questions regarding this document or if you have a suggestion for improvements, please contact:

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Senior Scientist - Evidence Recovery

Phone:



Version history

Version	Date	Changed by	Description
1.0	16/08/2016	Allan McNevin	Document Created.
1.1	28/09/2016	Allan McNevin	Incorporated feedback post review

Document sign off

This document has been approved by:

Name	Position	Signature	Date	6M
Cathie Allen	Managing Scientist Police Services		13/10/16.	13/0/2016

The following officers have endorsed this document

Name	Position	Signature	Date
Justin Howes	Team Leader FRIT		07-10-2016
Paula Brisotto	Team Leader ER & Q		07-10-2016
Luke Ryan	Senior Scientist Analytical		07-10-20
Allan McNevin	Senior Scientist ER		06.10.206
Kirsten Scott	Senior Scientist Q & P		07/10/2016
Sharon Johnstone	Senior Scientist Intel		07/10/2016
Amanda Reeves	Senior Scientist Reporting 1		10.10.2016.
Kylie Rika	Senior Scientist Reporting 2		11/10/201

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1. Introduction

1.1. Background

Concerns were raised by the Forensic Reporting and Intelligence Team around the difference in spermatozoa microscopy counts observed at the time of examination and the numbers of spermatozoa observed on slides made from the same sample during the differential lysis extraction procedure. Namely, examples where nil or <1+ spermatozoa were observed during item examination and 3+ or 4+ spermatozoa were observed on differential lysis slide microscopy.

Within the Evidence Recovery team, spermatozoa numbers are graded on microscopy using a semi-quantitative scale of 0 (nil seen), <1+ (<10 cells seen on the whole slide, very hard to find), 1+ (10 or more cells

seen, hard to find), 2+ (easy to find); 3+ (very easy to find) and 4+ (abundant). Results from earlier investigations (refer 1.1.1 below) showed examples of casework samples had failed to show the presence of spermatozoa by microscopy during processing in evidence recovery and the presence of spermatozoa from microscopy slides made during differential lysis DNA extraction. The concern therefore is the sensitivity of the original slide microscopy:

- i. Is the current suspension method resulting in slides made from overly diluted material such that a sample may be called negative for spermatozoa at the point of examination when there are sufficient numbers present to produce a DNA profile from differential lysis extraction?
- ii. Is there a potential problem associated with the slide staining procedure such that spermatozoa are potentially being "lost" and are therefore not visualised on microscopy?

1.1.1. Previous investigations

An initial analysis of a selection of differential lysis slides from samples extracted in 2014 (N=31), 2015 (N=11) and 2016 (N=37) was conducted. Only those samples that had been reported but no statement required of the case, or those samples that had yet to be case managed were included in the selection. Selection was conducted by checking for samples across a number of differential lysis extraction batches in order to capture a range of both evidence recovery and analytical personnel involved in processing. For any sample where the differential lysis slide had not been examined, the slide was stained and microscopy results were recorded.

Results for initial microscopy, DNA quantification of the sperm lysate and differential lysis slide microscopy were tabulated. A total of 79 results were collated. There was a consistent trend towards more spermatozoa observed on the differential lysis slide than what was observed on initial microscopy (N=52), compared to samples where the microscopy was concordant (N=17) and samples where more spermatozoa were seen on initial microscopy (N=10). Average quantification values obtained from sperm lysate samples correlated well with differential lysis extraction slide microscopy, but not so well with initial microscopy. Data available in (G:\ForBiol\AAA Evidence Recovery\Projects and Datamining\2016 comparison of original v diff micro\ 2016 - Diff Lysis slide micro v original micro.xls). There were 7 instances where the original microscopy was negative for spermatozoa but positive from differential lysis microscopy as follows: 2+ (N=2), 3+ (N=2) or 4+ (N=3) were observed from the differential lysis slide. It is worth noting there were also 7 instances no spermatozoa seen on differential lysis slide whilst spermatozoa were observed on original microscopy (all graded at <1+). Results shown in Figure 1 below

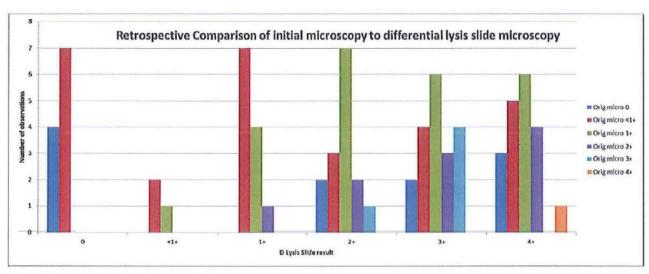


Figure 1 Retrospective data analysis

A review of previously obtained in-house data suggests that current AP and p30 methods have a sensitivity of detecting semen at a dilution of approximately 1/100. A dilution of approximately 1/20 of semen is used for making in-house extraction positive control samples and these samples will yield a microscopy result of approximately 1+ to 2+, with quantification values approximating 0.01 ng/μL (according to positive control log) up to 0.07 ng/μL (according to average positive control results post processing).

Purpose and scope

This project aims to investigate the performance of the current method as outlined in standard operating procedures. This project should then inform directions for further investigations.

This project should also then fill a knowledge gap that currently exists within the department. Recording of AP and p30 presumptive testing results compared directly with microscopy results aims to assist in providing indicative information, as there is no current in-house experimental data comparing the sensitivity of sperm microscopy, AP and p30 detection and DNA profiling.

Governance

3.1. Project Personnel

Project Manager: Allan McNevin – Senior Scientist, Evidence Recovery Team

Senior Project Officer: Emma Caunt, Scientist, Reporting Team

3.2. Decision Making Group

The Management Team and the Senior Project Officer, are the decision making group for this project and may use the defined acceptance criteria in this project to cease part or all of the experimentation at any stage. The Decision Making Group may also make modifications to this Experimental Design as required, however this must be documented and retained with the original approved Experimental Design.

The Senior Project Officer is included in the Decision Making Group in their capacity as an expert user.

3.3. Reporting

The Project Manager will provide a weekly project status update to the Team Leader, Evidence Recovery and Quality who will in turn advise the Decision Making Group at the Management Team meetings and by exception as required.

4. Resources

The following resources are required for this validation/project:

4.1. Reagents

- 5% v/v Bleach White N Bright (Ecolab, NSW, AU)
- 5% v/v Trigene Advance (CEVA DEIVET Pty. Ltd. Seven Hills, NSW, AU)
- Ethanol (Recochem Incorporated, Wynnum, QLD, AU)
- Nanopure water (Forensic DNA Analysis, Brisbane, QLD, AU)
- Proteinase K (20mg/mL) (Sigma-Aldrich® Corporation, St Louis, MO, US)
- Brentamine Fast blue B (Sigma Aldrich, Castle Hill, NSW, Australia)
- Anhydrous Sodium Acetate (Sigma Aldrich, Castle Hill, NSW, Australia)
- Glacial acetic acid (Univar AJAX Finechem Pty. Ltd., Taren Point, NSW, Australia)
- Sodium α-naphthyl phosphate (Sigma Aldrich, Castle Hill,, NSW, Australia)
- Nanopure water (Millipore Milli-Q® Advantage A10 system)
- ABA card p30 test kits (Abacus Diagnostics)
- Haematoxylin and Eosin stains (Forensic DNA Analysis, Brisbane, QLD, AU)
- Dithiothreitol (Sigma-Aldrich® Corporation, St Louis, MO, US)
- Sarcosyl (Sigma-Aldrich® Corporation, St Louis, MO, US)
- Positive controls (Forensic DNA Analysis, Brisbane, QLD, AU)
- TNE (Forensic DNA Analysis, Brisbane, QLD, AU)

 DNA IQ[™] Casework Pro Kit for Maxwell® 16 (Promega Corp., Madison, WI, US)

4.2. Materials

- Sterile 1.5 and 2 mL screw-cap tubes (Axygen Scientific Inc., Union City, CA, US)
- Sterile 5 mL screw-cap tubes (Axygen Scientific Inc., Union City, CA, US)
- ART Filtered 1000 μL, 300 μL & 20p pipette tips (Molecular BioProducts Inc., San Diego, CA, US)
- F1-ClipTip pipette tips 20μL, 50μL, 200μL & 1000 μL (Thermo Fisher Scientific Inc,)
- Nunc[™] Bank-It[™] tubes (Nunc A/S DK-4000 Roskilde, Denmark)
- Rediwipes (Cello Paper Pty. Ltd., Fairfield, NSW, AU)
- Petri dishes (Starstedt Australia Pty. Ltd., Mawson Lakes, SA, AU)
- Sterile rayon swabs (Copan Diagnostics Inc., Murrieta, CA, USA)
- Grale HDS SureFrost™ Microscope slides (Trajan Scientific, Milton Keynes, United Kingdom)

4.3. Equipment

- Biological safety cabinets class II (ESCO, Lytton, QLD, AU)
- Vortex Mixer VM1 (Ratek Instruments Pty Ltd, Melbourne, VIC, AU)
- MixMate (Eppendorf AG, Hamburg, DE)
- Micro centrifuge (Tomy, Tokyo, JP)
- Eppendorf 5424 centrifuge and Eppendorf 5804 centrifuge (Eppendorf, North Ryde, NSW, Australia)
- Dry Block Heater (Ratek, Boronia, NSW, Australia)
- Milli-Q® Integral 3 (A10) System with Q-POD™ (Millipore™, Billerica, MA, USA)
- Pipettes (Eppendorf, Hamburg, DE and Thermo Fisher Scientific (Finnpipette), Waltham, MA, US)
- ClipTip Pipettes (Thermoscientific)
- Promega Maxwell® 16 MDx 1 and 2 Instruments (Promega Corp., Madison, WI, USA)
- Milli-Q® Integral 3 (A10) System with Q-POD™ (Millipore™, Billerica, MA, US)
- Minifuge (CS Bio Co. (ex-Tomy Tech US Inc.), Menlo Park, CA, US)
- Tube Centrifuge (Eppendorf South Pacific Pty. Ltd., North Ryde, NSW, AU)
- BX41 Microscope (Olympus Corporation, Tokyo, Japan)

Forensic DNA Analysis Analytical Staff, Computer and instrument time, as well as bench space in DNA Analysis Analytical Laboratory will also be used in the duration of this project.

5. Methods

5.1. Mock Sample Creation

Mock samples will be created following processes outlined within standard operating procedure 25874V7 Preparation of DNA Quantification Standards & In-house Quality Controls section 5.5 with noted exceptions as follows:

- Instead of dilutions of positive semen control as outlined in the procedure, the following dilutions of neat semen will be used:
 - 0 1/5; 1/10; 1/20; 1/50; 1/100; 1/200; 1/500
- Approximately 3 x the amount of epithelial cells will be added to each swab
- 4 replicates of each semen dilution will be made resulting in 28 mock samples in total.

5.2. Evidence recovery processing

All mock samples will be processed by a single operator following current inhouse procedures (17142V12 Examination of items; 171894V13 Examination for & of Spermatozoa), with the exception that regardless of results of microscopy for spermatozoa, samples will also be tested for the presence of AP and p30 (17185V10 Detection of Azoospermic Semen in Casework Samples; 17186V12 The Acid Phosphatase Screening Test for seminal stains).

5.3. DNA extraction

Each swab will undergo a differential lysis extraction process and a slide prepared according to current routine procedure (29344V5 DNA IQ Extraction using the Maxwell 16). The extracts will be held pending further investigations.

6. Experimental Design

6.1. Experiment 1: Investigation of current process

i. Intent

To approximately quantify the difference in the number of sperm observed on microscopy slides made from cell suspensions during the evidence recovery process compared to those made during differential lysis DNA extraction.

Additionally, this experiment may identify the approximate sensitivity of detection of sperm at each of these stages of the process.

ii. Experimental Design

The mock samples created as per 4.1 above will be processed through evidence recovery by a single operator. For differential lysis DNA extraction, the mock samples will be split into two batches, each containing duplicates of each sperm sample dilution. Both batches will be processed by the same operator. Layout of extraction batches is shown in Table 1 below.

Table 1 Extraction batches - Experiment 1

Extraction batch 1		Extraction Batch 2	
Position	Sample	Position Sample	
1	Positive Control	1	Positive Control
2	Negative Control	2	Negative Control
3	1/5 semen dilution	3	1/5 semen dilution
4	1/5 semen dilution	4	1/5 semen dilution
5	1/10 semen dilution	5	1/10 semen dilution
6	1/10 semen dilution	6	1/10 semen dilution
7	1/20 semen dilution	7	1/20 semen dilution
8	1/20 semen dilution	8	1/20 semen dilution
9	1/50 semen dilution	9	1/50 semen dilution
10	1/50 semen dilution	10	1/50 semen dilution
11	1/100 semen dilution	11	1/100 semen dilution
12	1/100 semen dilution	12	1/100 semen dilution
13	1/200 semen dilution	13	1/200 semen dilution
14	1/200 semen dilution	14	1/200 semen dilution
15	1/500 semen dilution	15	1/500 semen dilution
16	1/500 semen dilution	16	1/500 semen dilution

iii. Acceptance Criteria

This experiment has no specific acceptance criteria as it is being used as a baseline upon which further experimentation will be compared.

7. Results and Data Compilation

The results of Evidence recovery presumptive testing, microscopy and differential slide microscopy will be collated and tabulated. This information will formulate decisions on the direction of any further experimentation.

If the Project Team forms the opinion that additional experiments are required before a final assessment can be made, application will be made to the Decision Making Group for a modification to this Experimental Design. The Decision Making Group is responsible for assessing this application and approving or rejecting it.

A final report will be produced which will compile all analyses, conclusion and recommendations. The final report will be prepared by the Project Group.



HealthSupport Queensland

Forensic and Scientific Services

Project Plan

Stage 2			
		Project #:	181
Name/s of Project	Allan McNevin, Emma Caunt	Start Date:	
Staff:	7 mail Mortovini, Elimia Gadin	Due Date:	
Name Project Team Leader:	Allan McNevin	Contact Phone Number:	
Technical Reviewer/s	Forensic DNA Analysis manag	gement team	
Project Title:	Investigation into the sensitivit	y of spermatozoa	microscopy
Project type	Administration IT/LIMS Data mining/analysis External		
Project Background (r	nay include a literature review):		
spermatozoa micros spermatozoa observextraction procedure item examination an microscopy. Within the Evidence semi-quantitative scandant, An initia 2014 (N=31), 2015 (spermatozoa observed microscopy (N=52), samples where more quantification values microscopy, but not recovery\Projects a slide micro v original negative for spermato differential lysis slide differential lysis slide <1+).	ed by the Forensic Reporting and copy counts observed at the time of an slides made from the same of t	ne of examination ne sample during or <1+ spermatozo bserved on different numbers are grades seen on the whole find); 3+ (very eatential lysis slides do a consistent treathan what was obtained a microscopy was tial microscopy was tial microscopy (Imples correlated Data available in (In of original v diffices where the original or 4+ (N=3) which is the original or	and the numbers of the differential lysis on were observed during ential lysis slide ed on microscopy using a ole slide, very hard to find), asy to find) and 4+ from samples extracted in and towards more observed on initial sconcordant (N=17) and N=10). Average well with Diff Lysis slide G:\ForBiol\AAA Evidence micro\ 2016 - Diff Lysis iginal microscopy was ere observed from the observed from the observed at microscopy (all graded at
that a sample m	spension method resulting in sli ay be called negative for sperment numbers present to produce	atozoa at the poir	nt of examination when

Page: 1 of 5 Document Number: 22872V8 Valid From: 16/12/2015 Approver/s: Cathle ALLEN ii. Is there a potential problem associated with the slide staining procedure such that spermatozoa are potentially being "lost" and are therefore not visualised on microscopy?

This project aims to investigate (i) above, as there is no current in-house experimental data comparing the sensitivity of sperm microscopy, AP and p30 detection and DNA profiling. However, if discrepant results are obtained from replicates of the same sample, this project may identify problems related to (ii) above.

A review of previously obtained in-house data suggests that current AP and p30 methods have a sensitivity of detecting semen at a dilution of approximately 1/100. A dilution of approximately 1/20 of semen is used for making in-house extraction positive control samples and these samples will yield a microscopy result of approximately 1+ to 2+, with quantification values approximating 0.01 ng/µL (according to positive control log) up to 0.07 ng/µL (according to average positive control results post processing).

Benefit of Project:

Given that no formal validation of the making of cell suspensions was recorded at the time the procedure was introduced (possibly around 2008, details not found), an investigation into the effectiveness of current procedures will fill the gap in departmental records.

Additionally, the determination of the sensitivity of microscopy and presumptive testing compared to profiling results is worth investigating since this has not been done since the introduction of the PowerPlex21 amplification kit which has a greater level of sensitivity compared to Profiler Plus.

Proposed Methodology:

Mock casework samples will be prepared using a modification of the method used to prepare inhouse differential lysis positive control swabs (QIS 25874V7). Initial investigations will consist of:

- Decreasing amounts of semen from a single donor will be applied to a swab in the presence of constant amounts of epithelial cells.
- Proposed semen dilutions are 1/5; 1/10; 1/20; 1/50; 1/100; 1/200; 1/500
- Proposed level of Epithelial cells present on initial microscopy will be in the 2+ to 3+ range
- Each swab will be tested using current in-house procedures (17142V12; 171894V13), with the exception that samples that are microscopically positive for spermatozoa will also be tested for the presence of AP and p30 (17186V12; 17185V10).
- Each swab will undergo a differential lysis extraction process and a slide prepared. The
 extracts will be held pending further investigations.
- Results for sperm microscopy, AP and p30 presumptive tests, and microscopy from each differential lysis slide will all be collated and compared.

Dependant on the results obtained, further investigations will then be proposed; this may involve (for example) testing of samples in an identified critical range, testing of samples from a range of donors and / or various reproducibility or repeatability tests.

Expected Outcome:

- Characterisation of the sensitivity of current procedures as they relate to DNA profiling outcomes. This will fill a gap in departmental records, provide greater insight into current procedures, and may identify an area for procedural improvement.
- Assessment of the sensitivity of the detection of spermatozoa on evidence recovery slides.

	ct Milestones: (Ensure that the Char ement\Change Management Milesto		tone Register	is filled out
Description of Outpo	uts/Milestones:		Expected due date:	Completed date:
1.				
2.				
3.				
4.				
5.				
If expected due da	te/s not met - explanation of reaso	n required:		
Project Budget:		Total Project Budget		
Prepare using QIS 3105	2 (and attach to Project Plan)	\$ Not yet suppl	ied.	
Gantt Chart (for la	rge projects): If required, refer to Quality	,		t Plan)
RISK ASSESSMENT If a risk is identifie identification and	d: Refer to QIS document <u>29100</u> and	l <u>29106</u> for further info	rmation on ris	sk
Team:	Details of Risk/s Identified		Туре	of Risk/s:
	nd identified.			siness Risk I&S
Evidence Recovery :			Signate Line M	ure anager
	Wil identified			siness Risk I&S
Analytical:	Occ march fred		Signate Line M	anager

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Intel:	Nonsk to intel	Business Risk OH&S Signature Line Manager
Reporting 1:	Nil further risks -	Business Risk OH&S Signature Line Manager
Reporting 2:	This project covers testing to address concerns raised by reporting teams the extra testing . No risks identified.	Business Risk OH&S Signature Line Manager
Quality and Projects (includes OO) :	Given concern with process efficiency only risk is if an assessment of process is not undertaken. This project is an initial assessment to rutigate this risk.	Business Risk OH&S Signature Line Manager
Admin :	Nil risk to Admin.	Business Risk OH&S Signature Line Manager
Team Leader ER &Quality:	Minimal risks as project is initially assessing current processes. Further investigations or projects may be proposed as an outcome of this assessment.	Business Risk OH&S Signature Team Leader PMB
Team Leader FRIT :	No risks to this project.	Business Risk OH&S Signature Team Leader)

	JAH

Signature Team Leader ER and Quality:	Date:	13/10/2016
Signature Team Leader FRIT:	Date:	12/10/2016
Signature Managing Scientist:	Date:	13/10/16

Comments:

Please send to Quality Team

JH-35

Erin Shearer

From: Justin Howes

Sent: Friday, 21 May 2021 2:59 PM

To: Adrian Pippia; Alicia Quartermain; Allan McNevin; Allison Lloyd; Angela Adamson; Angelina

Keller; Anne Finch; Cassandra James; Cathie Allen; Claire Gallagher; Deborah Nicoletti; Emma Caunt; Ingrid Moeller; Jacqui Wilson; Josie Entwistle; Kerry-Anne Lancaster; Kylie Rika; Luke Ryan; Matthew Hunt; Paula Brisotto; Penelope Taylor; Rhys Parry; Sharon Johnstone; Tegan Dwyer;

Thomas Nurthen

Cc: Kirsten Scott; Chelsea Savage

Subject: RE: Slight Appendix change relating to semen testing

Attachments: APPENDIX_current_JAH21052021.doc

Hi all

Some further minor changes have been made to the current Appendix for statements. As there is more than one minor change, I am supportive of a new version to be put into use. From here, I would prefer not to make further changes until there is a significant need and that could be with use of VFP or varNOC or other ySTRs for example.

Attached is the latest version for use in the next statement you are drafting where practicable. This doc is located in G:\ForBiol\AAA Forensic Reporting & Intel\AAA Reporting guidelines\Appendix.

I will edit the recent comment added to SOP 34006.

Regards

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

Please note that I may be working from a different location during the COVID-19 Pandemic. The best contact method is via email.



Queensland Health acknowledges the Traditional Owners of the land, and pays respect to Elders past, present and emerging.

From: Justin Howes

Sent: Wednesday, 28 April 2021 12:02 PM

To: Adrian Pippia; Alicia QuartermainAllan McNevinAllison Lloyd; AngelaAdamson; Angelina KellerAnne Finch; Cassandra James; Cathie Allen



Subject: Slight Appendix change relating to semen testing

Hi all

A slight modification to the wording for the semen paragraph has been suggested and consultation has occurred. The modification relates to the change in process as a result of Project #181.

Attached is the latest version for use in the next statement you are drafting where practicable. This doc is located in G:\ForBiol\AAA Forensic Reporting & Intel\AAA Reporting guidelines\Appendix.

I will add a comment to SOP 34006.

Regards Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

Please note that I may be working from a different location during the COVID-19 Pandemic. The best contact method is via email.



APPENDIX

<u>Procedural and technical overview of DNA profiling at Forensic DNA Analysis,</u> <u>Forensic and Scientific Services, Health Support Queensland</u>

Forensic Biologist

It is a forensic biologist's role to:

- 1. Report on the examination of items submitted in relation to a case for the presence of possible biological material. If identified, a sample of the biological material is analysed in an attempt to obtain a DNA profile.
- 2. Report on the DNA profiles obtained from samples submitted by the Queensland Police Service (QPS) in relation to a case.

Any DNA profiles that are obtained from these samples are compared with the DNA profile obtained from an individual's reference sample to assess whether or not the individual may be a contributor of DNA. Where multiple reference DNA profiles are analysed for a case, they are all genetically different to each other unless otherwise specified. That is, they all possess differing allelic designations – see the *DNA Profiling* section below.

Examinations

Unless otherwise stated, the examinations of items for biological material were conducted by staff within the QPS. Sub-samples from these items were forwarded to Forensic and Scientific Services (FSS), Health Support Queensland, for the purposes of conducting DNA analysis.

The descriptions of items and/or samples submitted to Forensic DNA Analysis by the QPS, and listed within this Statement, are derived from the descriptions entered by the QPS into the Forensic Register.

Receipt details are listed for items reported in this statement, including reference samples where relevant. The period of testing on these items is from the date of the first submission, to the date of this Statement. Details of specific dates of testing are retained within the Laboratory Information Management System (LIMS). Where relevant, any items that are still in progress at the time of issuing this Statement are identified and final results will be reported in an addendum Statement.

Forensic DNA Analysis operates under the agreement that the QPS are responsible for item prioritisation, sample selection, selection of screening/sampling methods, anti-contamination procedures and the application of Standard Operating Procedures (SOPs) on work undertaken on the items/samples prior to submission to the Forensic DNA Analysis laboratory. As such, forensic biologists may not be able to provide information or opinion on possible biological origin of DNA profiles that may be obtained from these samples.

At the discretion of the QPS, some items may be submitted to this laboratory for the purposes of both examination and DNA profiling. These examinations are performed in accordance with the SOPs of this laboratory. For these items, contemporaneous notes are made during the examination by the examining scientist and these notes form part of the casefile.

Forensic DNA Analysis casefiles and any samples remaining are available for independent examination and / or testing upon request.

As a representative of the laboratory, I am only able to comment on the processes performed within Forensic DNA Analysis.

Chain of Custody

All Forensic DNA Analysis case files and exhibits are electronically tracked, monitored, and securely stored to ensure that appropriate chain of custody and continuity measures are maintained. The QPS case number and sample submission information is provided from the QPS via an electronic interface to FSS, and this information is cross-checked against labelling on the exhibit packaging prior to processing.

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Entry into Forensic DNA Analysis is restricted to authorised persons only, via electronic proximity access cards. Forensic DNA Analysis forms part of a Health Support Queensland campus site wherein access is controlled and monitored by a security team. Records of visitors to Forensic DNA Analysis are retained.

Accreditation

Forensic DNA Analysis first achieved accreditation by the National Association of Testing Authorities (NATA) to conduct forensic DNA analyses in 1998, and has continuously maintained NATA accreditation since this date. NATA ensures continued compliance with the accreditation requirements through routine reassessment (every 3 years) and an intermediate surveillance visit (at approximately 18 months between reassessment surveys).

NATA accredited facilities are assessed against best international practices based on the ISO/IEC 17025 standard. Laboratories are deemed able to competently perform testing and analysis activities if the laboratory demonstrates that it meets compliance with the standard within the scope of their accreditation.

The parameters assessed during accreditation include:

- Organisation and management
- Quality management system
- Personnel
- Evidence management
- Methods and procedures
- Quality control and Proficiency Testing
- Equipment
- Reporting of results
- Procurement of services and supplies
- Accommodation and safety
- Security and access

For details of the current ISO/IEC 17025 Standard, refer to Standards Australia.

http://www.nata.com.au

DNA Profiling

Deoxyribonucleic acid (DNA) is a complex chemical found in almost all cells of the human body. It carries genetic information that determines the physical and chemical characteristics of a person. Forensic DNA Analysis uses two main systems for the generation of DNA profiling results. In this case, the PowerPlex® 21 System was used, which examines 21 regions (loci) of DNA, 20 of which contain highly variable short tandem repeats (STRs). The 21st region gives an indication as to the genotypic sex of the donor (for details see Table 1). The generation of a DNA profile involves a method known as the polymerase chain reaction (PCR), which is used to produce numerous copies of these specific regions of the DNA. In this way, minimal amounts of DNA isolated from small or degraded samples can be increased to a level where the DNA can be detected, profiled, and compared with DNA profiles from other samples.

The individual components (alleles) of a DNA profile are represented by a series of peaks on a graph that are measured and given a designation, using standard sizing ladders. A person will have two alleles (represented graphically as peaks) for each locus, one inherited from the biological mother and one inherited from the biological father. However, if the same allele is inherited from both parents, only one peak will be graphically represented at that locus.

A DNA profile obtained from biological material such as blood, semen, saliva, hair or cellular material (eg. touch DNA) can be compared with the DNA profile obtained from the reference sample from any person.

Table 1: PowerPlex® 21 system, list of loci

Abbreviated Name	Scientific Name	Chromosomal Name
Amel	AMELOGENIN	Sex (X and Y)
D3	D3S1358	3
D1	D1S1656	1
D6	D6S1043	6
D13	D13S317	13
Penta E	Penta E	15
D16	D16S539	16
D18	D18S51	18
D2	D2S1338	2
CSF	CSF1PO	5
Penta D	Penta D	21
TH01	TH01	11
νWA	HUMVWAFA31/A	12
D21	D21S11	21
D7	D7S820	7
D5	D5S818	5
TPOX	TPOX	2
D8	D8S1179	8
D12	D12S391	12
D19	D19S433	19
FGA	HUMFIBRA	4

Statistical Analysis of DNA Profiles

STRmix[™] is an expert statistical DNA profile analysis system developed and validated in Australia and New Zealand. Forensic DNA Analysis uses the STRmix[™] software to assist in the interpretation of DNA profiles and the calculation of likelihood ratios for DNA profiles generated using the PowerPlex® 21 system.

DNA profiles are initially assessed to determine the number of contributors. This value will be the minimum number of people that are required to reasonably explain the observed profile, however, it is noted that there is always the possibility that the profile is a result of a different number of contributors.

As such, if there is no indication of a contribution by more than one person, then a DNA profile is described as being from a "single contributor". If less than 40 alleles present in a DNA profile, this is referred to as a "partial" or "incomplete" DNA profile. If there are indications of two or more contributors, then a DNA profile is described as being a "mixed" DNA profile.

DNA Profiles Assumed To Originate From One Person (Single Source)

A person can be excluded as a possible source of the biological material if the alleles of the crime-scene DNA profile are different from the corresponding alleles of the person's reference DNA profile. If the corresponding alleles of the crime-scene DNA profile contain the same information, then that person, along with any other person who has the same corresponding alleles as the crime-scene DNA profile, can be considered as a potential contributor of the DNA.

The evidential significance of such a finding is assessed by considering two competing propositions:

Proposition 1: The crime-scene DNA originated from the person of interest.

Proposition 2: The crime-scene DNA originated from someone other than, and unrelated to, the person of interest.

The resultant figure (termed the 'Likelihood Ratio') compares the two opposing propositions. The likelihood ratio describes how likely the DNA profile obtained from the biological material is to have occurred if Proposition 1 were true (the DNA originated from the person of interest) rather than if Proposition 2 were true (the DNA originated from someone other than, and unrelated to, the person of interest).

The likelihood ratio is calculated by taking into account the characteristics of the DNA profile and the frequency of occurrence of the individual alleles that make up the DNA profile.

If less than the 20 STR regions of DNA are observed in a DNA profile, the likelihood ratio will be smaller than the likelihood ratio calculated for a full DNA profile obtained from the same individual. In other words, the more incomplete a DNA profile is, the greater the likelihood that the obtained DNA profile could have come from someone other than, and unrelated to the person of interest.

DNA Profiles Assumed To Originate From More Than One Person (Mixed DNA Profiles)

In order to assess whether a person may or may not have contributed to a mixed DNA profile, a set of competing propositions (similar to the single source DNA profile example) are considered. For example, for a two-person mixture:

Proposition 1: The crime-scene DNA originated from the person of interest and an unknown person, unrelated to the person of interest.

Proposition 2: The crime-scene DNA originated from two unknown people, both unrelated to the person of interest.

The likelihood ratio provides a statistical assessment of the possibility that the DNA profile obtained was the result of a DNA contribution by the person of interest.

The likelihood ratio will not always favour Proposition 1 (the DNA originated from the person of interest and an unknown person unrelated to the person of interest). The likelihood ratio could favour Proposition 2 (the DNA originated from two unknown people unrelated to the person of interest).

In certain circumstances, if the ownership of an item is established or if the sample was collected from an identified person's body, then it is possible to make the reasonable assumption that the identified person has contributed DNA to the resultant mixed DNA profile. In these cases, a mixed DNA profile can be 'conditioned' on the DNA profile of the assumed contributor, such that the presence of the alleles corresponding with the assumed contributor's reference DNA profile can be factored into the statistical interpretation. This may facilitate a more meaningful statistical analysis of potential second and/or third contributors to the DNA profile. In this situation, the likelihood ratio is based on the following propositions:

Proposition 1: the DNA has originated from the assumed contributor and the person of interest.

Proposition 2: the DNA has originated from the assumed contributor and an unknown individual, unrelated to the person of interest.

DNA profiles with an uncertain number of contributors

Forensic DNA Analysis is currently validated for the interpretation of DNA profiles with 1-4 contributors. Once the assessment of the number of contributors has been made, this information is used in the STRmix™ analysis.

When a DNA profile is deemed unsuitable for interpretation it may be described as 'complex'. This can occur under the following circumstances:

- When a DNA profile could have a large number of contributors and therefore it is difficult to exclude individuals and/or determine whether a person could be a potential contributor to the DNA profile
- When there is very limited information available and/or the quality of the profile is poor.

It is important to note that should further information be received that renders the assumptions used in an analysis invalid, the DNA profile will require additional statistical interpretation.

Datasets Used in Statistical Analyses

Three validated datasets consisting of DNA profiles obtained from individuals of the Australian Caucasian, Aboriginal, and South-East Asian populations are used to calculate the likelihood ratio. A population correction factor, θ (theta), is applied to all likelihood ratio calculations in order to correct for the common genetic ancestry of people within a particular population (sharing of DNA components inherited from a common ancestor). In Forensic DNA Analysis, a likelihood ratio that represents a stratified population statistic is reported irrespective of whether the DNA profile of interest is single source or a mixed DNA profile.

The likelihood ratio is calculated using all three datasets and the relative proportions of each ethnic grouping in the overall Australian population based on data sourced from the Australian Bureau of Statistics. This calculation, which is known as stratification, allows for the possibility that a random unknown person, unrelated to the person of interest, from anywhere in Australia could have contributed to the DNA profile obtained. Therefore, the likelihood ratio is not dependent on the ethnicity of the person of interest, but it is representative of the relative proportion of the person of interest's ethnic population within the larger Australian population.

Theta cannot account for close blood relatives. Closely related people, such as siblings, will have a greater chance of sharing similar components within their DNA profiles. However, due to the nature of parental DNA recombination during conception, the probability that two siblings would share the same 40 alleles would be very small. As this relationship becomes more distant, the probability of two relatives having the same DNA profile becomes smaller still. If it were thought that a close blood relative might have been a contributor of DNA, the most meaningful approach to interpretation would be to submit the reference sample from the relative in question for DNA analysis and subsequent direct comparison to the crime-scene DNA profile.

Often the calculated likelihood ratio produces numbers of thousands (1000s) or even millions (1000,000s) of billions. To avoid the use of potentially confusing mathematical terminology, a "ceiling figure" for the likelihood ratio of 100 billion is used (this is called "truncation"). For example, a calculated likelihood ratio of "150 000 billion times more likely", would be reported as "greater than 100 billion times more likely". The actual calculated figure can be provided upon request.

Parentage Testing and Statistical Calculations

In a disputed paternity matter, DNA profiles are obtained from the foetus/child, the biological mother, and the putative father(s). Based on the assumption that the nominated mother is indeed the biological mother of the foetus/child, it is possible to determine which alleles within the DNA profile of the child could have originated from her. Therefore, the remaining alleles within the foetus/child's DNA profile must have originated from the biological father. These are called *obligate paternal alleles*.

If the DNA profile of a putative father <u>does not</u> contain the obligate paternal alleles at three or more of the DNA loci tested, then he is *excluded* as a potential biological father of the foetus/child.

If the DNA profile of a putative father <u>does</u> contain the obligate paternal alleles at each of the DNA regions tested, then he is <u>not excluded</u> as a potential biological father of the foetus/child. This means that this putative father could be the biological father.

A statistical analysis is performed to calculate a *Paternity Index (PI)*. The PI is a ratio of two probabilities conditional upon different competing propositions.

Proposition 1: The alleged father is the true father (and the mother is the true mother).

Proposition 2: A random person who is not related to the alleged father is the true father (and the mother is the true mother).

The PI reflects how many times more likely it is to see the evidence (i.e. the child's DNA profile) under the first proposition compared to the second proposition.

For an inclusion of paternity/maternity the PI must not be less than 1000, according to ISO/IEC 17025 Application Document, Legal (including Forensic Science) – Annex, Parentage Testing for the Australian Family Law Act. See www.nata.com.au

Touch DNA / Transfer of DNA

When a person touches a surface, it is possible for their DNA to be transferred onto that surface. This transferred DNA can often be recovered (sampled) by a swab, tape lift, or excision depending upon the nature of the surface in question, and the sample can then be subjected to DNA profiling.

This transferred DNA can originate from cells or cellular material within sweat and/or oils on the skin. The generation of a DNA profile will depend on many factors. These include the amount of DNA transferred, the nature of the surface being touched, and the amount of genetic material available for transfer. The persistence of any transferred genetic material to a surface will depend largely upon the nature of the surface and the conditions the surface has been subjected to in the time between deposition and recovery of the DNA. For example, DNA could be lost from the surface by washing or mechanical action such as abrasion.

It therefore follows that the inability to obtain a DNA profile from a touched surface does not necessarily mean that a person has not had contact with the surface. It is possible for a person to contact a surface without a detectable amount of their DNA being transferred or subsequently recovered for analysis.

Blood Stains

Potential bloodstains are located in the laboratory by means of their visual appearance and the use of a presumptive chemical test (Tetramethylbenzidine – TMB). A positive result with this test is a good indication that blood may be present, however it does not provide proof as other substances are known to give the same result.

Semen Stains

Semen is the collective name for the mixture of spermatozoa (sperm) and seminal fluid. The presence of semen on an item can be indicated by using a presumptive chemical test that detects a major constituent of seminal fluid, namely Prostate Specific Antigen (PSA / p30). This constituent may exist in other body fluids, such as urine, faecal material, sweat, breast milk and blood, albeit usually at much lower concentrations.

The location or presence of possible semen on items may also be indicated by using a presumptive chemical test that detects another major constituent of seminal fluid (Acid Phosphatase – AP). This constituent exists in other body fluids, such as vaginal secretions, albeit usually at much lower concentrations.

The presence of semen can be confirmed via the microscopic identification of spermatozoa.

Samples where semen may be present undergo a differential lysis extraction process that aims to separate spermatozoa and epithelial cells into separate fractions. This separation is not always completely effective, and a mixing of fractions can occur. This is often referred to as cellular carryover.

The current practice within Forensic DNA Analysis is for epithelial fractions from internal female sexual assault investigation kit (SAIK) samples to be stored following a differential lysis extraction process. This is because when these fractions are profiled, they are generally found to be a single contributor match to the person from whom the sample was taken. Given the nature of these samples, this finding is not unexpected. These epithelial fractions are stored indefinitely, and can be sent for DNA profiling at a future date if required.

Semen Staining on Items

The presence of semen on an item is normally the result of either direct ejaculation or contact with an item wet with semen (transfer). Any semen that may have been transferred / deposited can subsequently be lost by actions such as washing.

Persistence of Semen in the Vagina

The presence of semen in the vagina is normally the result of vaginal intercourse with internal ejaculation. The chance of detecting semen on a vaginal swab depends upon a number of factors such as:

• the effectiveness of the sampling process;

- the delay between deposition of the semen and sampling during the medical examination;
- · the biochemical conditions within the vagina;
- any physiological factors that may affect semen production in the donor.

The greater the delay between deposition and sampling, the less chance there is of finding semen. Although highly variable, semen is likely to be found on vaginal swabs if they were taken 1-2 days after the act of vaginal intercourse. Semen is sometimes found on swabs taken between 2-7 days afterwards, but it is unlikely to be detected after 7 days. This is due to a number of factors that can include the following:

- · drainage of semen from the vagina;
- loss of semen by bathing or washing (especially on external sites);
- degradation of the spermatozoa and seminal fluid constituents.

Saliva

A presumptive chemical test (Phadebas) may be used to detect the possible presence of saliva. This test exploits the enzyme activity of a constituent of saliva called amylase. Amylase is usually present at a relatively high concentration in saliva, though this can vary considerably between individuals. Amylase may also be detected in other body fluids such as sweat, vaginal fluid and anal secretions, although usually at much lower concentration than that found in saliva.

The presence of saliva on a surface may be the result of spitting or direct oral contact. Saliva may subsequently be transferred onto other items such as clothing or other areas of the body. Possible saliva stains may then be detected on skin swabs or items of clothing by the Phadebas test, as long as the clothing or skin has not been washed. Cellular material within the saliva, if present in sufficient quantities, can be used to obtain a DNA profile.

JUSTICES ACT 1886

I acknowledge by virtue of Section 110A (6C) (c) of the Justices Act 1886 that:

- (i) This written statement by me dated 4 October 2022 and contained in the pages numbered 1 to 7 is true to the best of my knowledge and belief; and
- (ii) I make it knowing that, I would be liable to prosecution for stating anything that I know is false.

Reporter

Signed at BRISBANE on 4 October 2022

9/12/2020 12:33:05 PM Chelses SAVAGE:

Comments

- The diff slides are stained by HP2s. Once stained, the HP3s get a number of slides. They are to check whether these slides need reading. If they don't (GMO smear was sperm pos, or sample doesn't requirence analytical note is to be entered. For all samples requiring the slide to be read - read slide and enter results. - All micro neg slides are to have p30 testing performed (except for AP fabrics where one section is sperm positive). Perform p30 testing and enter p30 results against the subsample.

- Enter relevant result lines (see appendix).

- Submit sperm pos or sperm neg, p30 pos samples for quant. Testing on semen negative samples is to be ceased.

- Submit sperm pos or sperm neg, p30 pos samples for quant. Testing powerpoint (1:\Change Management\Proposal\#151 to \#200 (completed projects)\Proposal\#181 - Sperm microscopy sensitiv (complete) and 2 new appendices created. Please see training powerpoint (1:\Change Management\Proposal\#151 to \#200 (completed project\#181 Training for ER). Slide 9 shows the information required in the first appendix and slide 10 shows the information required in the second appendix. - Validator to add to quant worklist if micro pos / micro neg and p30 pos. (or other relevant scenarios - see page 9 and 10 of training presentation - I.\Change Management\Proposal#151 to #200 (complet Section 5.10: Diff lysis slides are read for all samples unless a GMO slide was read originally and it was sperm positive, or when semen analysis is not required. Paragraph 2: HP2/HP3 staff are to track slid. Under the HP3 section - make a note that if a slide cannot be read (e.g. broken) then the 'no result' box is to be ticked and detailed notes are to be written in the notes section. P30 is to be performed and References to appendices throughout the SOP will need to be fixed as the appendices themselves will need updating (see suggestions further down regarding which appendices need editing/deleting). Section 3 last dot point: change to 'A semen negative item is an item which has tested negative for spermatozoa microscopically and tested negative for p30'. Section 5.1: dot point starting with 'All samples examined for spermatozoa' is no longer correct. - Note: p30 testing not required on AP fabrics, sectioned, one or more sections spermatozoa positive. Add notation 'p30 not performed as alternate section

- barcode> is micro pos for sperm'. - If a p30 kit is faulty (control line doesn't appear): Enter a presumptive but don't select result, enter batch number of p30 kit and add a comment 'faulty p30 kit, control line did not appear' Section 5.1: Second last paragraph regarding smears is now slightly incorrect. Slides are no longer created from swabs.
Section 5.2: Slight wording changes required. Fourth paragraph last sentence: All items are submitted for quant. 5th paragraph: If semen is not detected, all testing on SFRACs is ceased.
Section 5.3: No difference in processing in-tubes regardless of whether QPS have conducted AP testing. Just one appendix will need to be linked here. - The sample is then validated. Validator to put on 'Differential Lysis' (no saliva testing required) or 'Differential Lysis Retain Supernatant' (saliva testing required) extraction worklist.
- Analytical will process EFRACs as per our current procedure (Ext and hold for internal swabs - adult females. Adult external swabs, child swabs and p1 samples are processed). - HP2s to obtain blue slide box from Analytical each day and stain all slides. They are to enter a microscopic, including the diff slide number and the H+E stain. Section 5.11: Make note that if these are sent for diff lysis, the diff slide does not need to be read and presumptive testing does not need to be performed. Section 6.1.6 step 2: Screenshot needs updating (amount of water added is now 400ul and last sentence should be removed). Delete Section 6.2. A new section should be added detailing how to create a subsample for the retain p30.
Section 6.5 can be deleted as the addition of an SRP can be included in section 6.2. More sections need to be added after 6.6 to detail how to enter microscopy and p30 results and regarding result lines. In Short: Section 5.6: Paragraph 3 - Change first sentence to ... 'submitted for quant'. Change second sentence to '... testing is ceased'. projects)\Proposal#181 - Sperm microscopy sensitivity (complete)\Implementation Project#181 Training for ER) Once slides are read, the scientist to p30 test any spermatozoa negative samples. Note: No AP testing Ext and hold on EFRAC for internal swabs - adult females, p1 samples do not need this analytical note. - Track p30 supernatants to the new 'p30 supernatant' box. These will be stored in the walk-in freezer. If slide cannot be read (e.g. broken), tick 'no result' and enter notes. P30 testing to be performed. Appendix 7: first paragraph – remove information regarding microscopy neg and the diff slide list. This supernatant is for p30 testing and is to be registered as a 'retain' sub-sample. The item is sampled and child exhibits created as per our current procedure SFRACs – Analytical will stop work after extraction (P1s won't be stopped). Steps needed to be added in: Create a subsample. Add an SRP line. Discard supernatant after testing (spermatozoa negative samples)
 Send subsample to 'destroyed' and discard in yellow waste bin incubate on a hot block at 30deg for 15 minutes Enter a 'ext and hold' analytical note as per current procedures Scientist to add relevant result lines (see table on next page) regardless of p30 result the sample is to proceed to quant. Scientists to read slides and enter microscopic results The following changes will need to be made to the SOP: - Untested supernatants will be retained for 3 months. to permanent slide storage once the slides are read. 200ul supernatant into 2ml tube and store frozen. Step 1: The diff slide list is no longer in use. Step 2. Retrieve blue slide box from Analytical. Appendix 5 needs to be modified. See below: Vortex mix and centrifuge for 3 minutes 15/12/2020 9:04:54 AM Allan MCNEVIN: Dot point 2 - no slide is created Implementation of Project #181. Transfer sample to an ERT box Enter a 'SRP' result line Response

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Response	15/12/2020 9:06:59 AM Allan MCNEVIN:	lan MCNEVIN:				389
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JH-38

Experimental designs for #181.

Name 33017V41_FSS RD short form for low value projects_181 Brainstorming 21-01-2019 initial Request #181 - signed Project #181 - Investigation into spermatozoa microscopy - Part 2 project #181 - Investigation into the sensitivity of spermatozoa microscopy - Part 2 Project #181 - Signed project proposal part 4 (Version 2) Project #181 - Signed project proposal Project proposal and project plan #181 - signed Project#181 Investigation of sperm micro sensitivity - project budget v1.0 Proposal#181 Investigation of sperm micro sensitivity - Experimental design Part 1 v1.1 Proposal#181 Investigation of sperm micro sensitivity - Experimental design Part 2 v1.1 Proposal#181 Investigation of sperm micro sensitivity - Experimental design Part 3 v1.2 Proposal#181 Investigation of sperm micro sensitivity - Experimental design Part 4 v1.2 🙀 Proposal#181 Investigation of sperm micro sensitivity - Experimental design Part 4 v2.1 additional testing Proposal#181 Investigation of sperm micro sensitivity - Experimental design v1.1 Proposal#181 Investigation of sperm micro sensitivity - Part 3 outline ver1.2 Proposal#181 Investigation of sperm micro sensitivity - Project plan v1.0 Signed Project Proposal #181 Part 1 13-Oct-2016 Signed Project Proposal #181 Part 2 04-05-2017 Signed Project Proposal #181 Part 3 05-Jun-2018 Signed Project Proposal #181 Part 4 (Ver 2) 01-Jul-2019 Signed Project Proposal #181 Part 4 08-Mar-2019 Signed Project Propsal #181 Part 3 5th June 2018 Spermatozoa microscopy experiment design part 1

JH-39

Erin Shearer

From: Cathie Allen

Sent: Thursday, 4 January 2018 9:26 AM **To:** Justin Howes; Paula Brisotto

Subject: Report

Attachments: ESR report 11 April 2017 - admin release.pdf

Sensitivity: Confidential

Hi Paula & Justin

I'll come over for a chat regarding this.

Cheers Cathie



Cathie Allen

Managing Scientist - Police Services Stream

Forensic & Scientific Services, Health Support Queensland, **Department of Health**

HSQ's vision | Delivering the best health support services and solutions for a safer and healthier Queensland.

Queensland Health acknowledges the Traditional Owners of the land, and pays respect to Elders past, present and future.



SCIENTIFIC REVIEW

Forensic DNA Analysis

Forensic and Scientific Services

Overall Findings

In my opinion the protocols and methods used at Queensland Health for the examination of SAIKs and other items relating to alleged sexual assaults as detailed in the documents listed above are fit for purpose and in line with best practice for this type of examination.

The methods are clearly written, and would enable a worker to follow them accurately. Appropriate background information and reading sources are given. While some of the references are a little old (eg those given on p11 of Examination for and of Spermatozoa) this is inevitable with well-established techniques, and more recent references are generally included. It may be useful to do a literature search to ensure that the reference lists are up-to-date.

The information supplied on the SAIK Medical Form should allow correct examination and analysis decisions to be made.

I have made comments on specific aspects of the information and documents below. None of these comments should be taken as contradicting my general findings. In addition, when I state that information is missing, it may be in a document that mave not read.

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1 of 3

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The Acid Phosphatase Screening Test for Seminal Stains QIS#17186v12;

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11 Apr 17

"AP paper - False Positive Investigation"

This investigation was occasioned by the finding of a negative control used in the AP test giving a positive result. The investigation clearly established that the effect was repeatable and an appropriate outcome, that is the removal and replacement of the paper used as the negative control, determined.

Procedure for Examination of Sexual Assault cases QIS #32106v4

- Section 3 Care should be taken over the use of phrases like "semen negative", as we are
 actually saying that none was detected. You do this in your AP screening information.
- · Section 4 is excellent, and very helpful to the analyst.
- Although the triaging process as laid out in 6.1 is quite restrictive, it is clear and assists the
 analyst in decision-making. Nothing is disposed of at this stage.
- Section 6.2 I would recommend that you consider stopping scraping stains. You run the risk
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 contamination.

Examination for and of Spermatozoa QIS #17189v13

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- Appendix 2 although you give details of the various studies considering the persistence of
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 anything that says how an analyst uses this data. There are also no details regarding
 persistence in the anus or mouth.

Detection of Azoospermic Semen in Casework Samples QIS17185v10

- Section 6.2 I don't know what "ambiguous" means.
- There does not appear to be detail on how to report positive or negative results
- There is quite a lot of work in the literature about p30 being found in breast milk, urine, tears and so on, which may be useful regarding interpretation and reporting.

Examination of items QIS #17142v13

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3 of 3

11 Apr 17.



JH-40

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3 of 3

11 Apr 17.

JH-41

Cass | Forensic and Scientific Services

DNA IQ™ Method of Extracting DNA from Blood and Cell Substrates

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1 PURPOSE AND SCOPE

This method describes the routine automated DNA extraction of cell and blood samples on the PerkinElmer MultiPROBE® II PLUS HT EX with Gripper™ Integration platforms using the Promega DNA IQ™ system. The manual method has been included as a back-up method should it be required.

This method applies to all Forensic Biology staff that is required to extract cell and blood samples.

Reference samples and Casework samples must be extracted separately. If Casework and Reference samples are to be extracted on the same instrument, the instrument must be decontaminated between operations.

2 DEFINITIONS

Samples Samples awaiting DNA extraction

DNA Extracts Samples that had DNA extraction processes performed

DNA IQ™ Resin Magnetic Resin Beads used to bind DNA

MP II MultiPROBE® II Platform

DTT 1,4 Dithiothreitol Pro K Proteinase K

SDS Sodium Dodecyl Sulphate
TNE Tris, NaCl and EDTA buffer
EDTA Ethylenediaminetetraacetate

EP-A Extraction Platform A – back wall platform EP-B Extraction Platform B – side wall platform

3 PRINCIPLE

Sample Pre-lysis

The Extraction Buffer used in the pre-lysis of cellular material contains Proteinase K (Pro K), TNE (Tris, NaCl, EDTA) and SDS. TNE acts as a basic buffer with EDTA chelating ions in solution. SDS is a detergent that lyses open cell membranes. Proteinase K is added to digest protein and cellular material that interferes with the DNA binding capacity of the resin. It is also added to rapidly inactivate enzymatic activity that could potentially degrade DNA (e.g.nucleases).

Proteinase K (also known as Endoproteinase K) is a fungal enzyme from *Engyodontium album* (formerly *Tritirachium album*). It is a stable S8 family serine alkaline protease that has broad specificity and is inactivated by diisopropylfluorophosphate, phenyl-methane sulfonyl fluoride and Hg²⁺ ions. As a highly active endopeptidase, it cleaves peptide bonds adjacent/next to the carboxyl group of N-substituted (blocked alpha amino) hydrophobic, aliphatic and aromatic amino acids. Therefore, it actively hydrolyses proteins, glycoproteins, peptides and esters of amino acids. The smallest peptide that can be hydrolysed with Proteinase K is a tetrapeptide.

DNA IQ™ Kit

The DNA IQ ™ kit is designed to purify DNA from a number of different substrates and has been optimised for trace DNA samples. An in house validation was performed using a modified version of the PerkinElmer automated protocol. The protocol has been modified to incorporate a number of work practices used in Forensic Biology FSS. These are:

o The use of the Slicprep™ 96 device (Promega) for removing substrate from lysate.



- The increase of extraction buffer volume to 500µL for use with the Slicprep™ 96 device.
- The increase of Lysis Buffer volume to 957μL proportional to the increase of Extraction Buffer volume, according to the manufacturer's instructions.
- o Double Elution step, with an Elution buffer volume of 60μ L for a final volume of 100μ L.
- The use of NUNC Bank-It tubes for storage of final extracts.

Cell lysis is performed with Promega Lysis Buffer containing Dithiothreitol (DTT). 1,4 Dithiothreitol is a reducing agent used in extractions to break disulfide bonds of proteins. The Lysis Buffer used is a proprietary buffer containing chaotropic salts required for binding of DNA to the magnetic beads. According to the MSDS, the lysis buffer contains 50-75% guanidinium thiocyanate, < 2.5% EDTA, < 2% 3-[(3-Choalamidopropryl)dimethylammonio] propanesulfonic and < 2% polyethylene glycol tert-octylphenyl ether.

The basis of the DNA IQ™ kit is a magnetic bead resin which contains novel paramagnetic particles covered with silica. The magnetic bead resin usually has a DNA binding capacity of 100ng but the addition of Pro K will increase the binding capacity. Samples with small amounts of DNA are more efficient than samples with large amounts of DNA, and as a result a small sample size is critical to ensure efficient recovery of DNA.

The magnetic beads with silica have a negative charge at basic and near neutral pH. The Lysis Buffer changes the pH and salt concentration of the solution and the silica on the magnetic beads becomes positively charged which is then able to bind the DNA.

Several washing steps are employed in the protocol to remove inhibitors. The first washing step is performed with Lysis Buffer. This wash ensures the DNA is bound to the magnetic bead resin and washes out inhibitors. The next three washing procedures are with a 1xWash Buffer. This buffer contains an alcohol/aqueous mixture which ensures the DNA is not eluted during washing by keeping the DNA dehydrated, and aqueous phase washes out the inhibitor.

Elution buffer removes the DNA from the magnetic bead resin. The Elution Buffer changes the salt content. Heating the complex to 65°C allows the DNA to be released from the magnetic beads. The Elution Buffer is a low ionic strength buffer that reduces the affinity of the DNA for the silica that covers the magnetic beads, by re-hydration of the phosphate backbone.

The DNA IQ™ kit isolates DNA greater than 80bp, smaller DNA is removed selectively to prevent PCR inhibition.

MultiPROBE® II HT EX Plus with Gripper™ Integration Platform

Within Forensic Biology, blood and cell extractions are performed using 2 MultiPROBE® II PLUS HT EX with Gripper™ Integration platforms (Extraction Platform A, EP-A) and (Extraction Platform B, EP-B) perform casework or reference samples. Each platform uses a computer – controlled Cartesian X-Y-Z liquid handling system designed for the efficient automation of sample preparation. Liquid transfer is performed by an 8-tip System with VersaTip® and VariSpan™ options. The VersaTip® option allows the use of fixed and/or disposable tips (both clear and conductive). The VariSpan™ option permits variable probe spacing between each of the individual probes so that a wide range of labware such as micro plates, tubes, vials and reagent troughs can be accessed. Each sample probe is capable of independent motion in the Z direction due to independent Z drives.

The 8-tip System is also capable of multichannel liquid-level sensing by utilising Accusense™ technology. This technology works by each probe detecting a change in capacitance within the liquid. This capacitive mode of detection is also possible when using conductive disposable tips. Accusense™ also permits the detection of low or non-ionic polar solutions and solvents. Pipetting on the platforms is driven by 8 individually controlled syringes via the positive displacement of the system liquid (nanopure water) when transferring liquid.

The Gripper™ Integration on all the platforms (except for the Post – PCR MPII) allows for automated identification of labware via the scanning of barcodes and also allows for automated transfer of plates from one position on the deck to another. To increase deck space and capacity, all platforms include a left deck extension.

In this program a platemap is utilised to provide the necessary information for correct volumes and locations for pipetting. It also contains information regarding positions where pipetting must take place at various locations, the number and IDs of samples. Without a platemap the program will fail to work.

4 REAGENTS AND EQUIPMENT

4.1 Reagents

- DNA IQ™ System Kit 400 sample Kit
 - o Resin
 - Lysis Buffer (LB)
 - o 2x Wash Buffer (2xWB)
 - Elution Buffer (EB)
- Tris/Sodium chloride/EDTA Buffer (TNE)
- 3. Proteinase K (Pro K) 20mg/mL
- Dithiothreitol (DTT) 1M
- 5. 5% TriGene
- 6. 70% Ethanol
- 7. 1% Amphyl
- 8. 0.2% Amphyl
- 9. Isopropyl alcohol
- 10. AnalR 100 %Ethanol
- 11, 20% SDS
- 12. Decon® 90 solution
- 13. Nanopure H₂O

Table 1. Reagent storage locations.

Reagent	Device	Storage Location
Pro K	Freezer	Room 6120
DTT	Freezer	Room 6120
20% SDS	Shelf	Room 6122
Isopropyl alcohol	Shelf	Room 6122
TNE pH 8 Buffer	Shelf	Room 6122
DNA IQ™ Kit	Shelf	Room 6122
Amphyl (1% and 0.2%)	Shelf	Room 6127
Nanopure Water	Shelf	Room 6127
5% TriGene	Shelf	Room 6127
AnalR 100 %Ethanol	Shelf	Room 6127

Please see Table 2 for the volume of reagents for a full plate or half plate. See QIS <u>17165</u> (Receipt, Storage and Preparation of Chemicals, Reagents and Kits) for preparation of the TNE buffer. All reagents, except for the Lysis Buffer with DTT (in fume hood), can be made on the bench in Room 6122. DNA IQ reagents are prepared by staff performing the DNA IQ method.

Table 2. Table of reagent volumes.

Reagent (volume per sample)	Constituent (volume per sample)	Volume req'd for 96 Samples (mL)	Volume req'd for 48 Samples (mL)
Extraction Buffer	TNE buffer 462.5µL	54	27
Extraction Buffer (500 µL/sample	Prot K (20 mg/mL)25.0 μL	2.9	1.5
(300 μL/sample	SDS (20 %) 12.5µL	1.5	0.7
Lysis buffer (with DTT)	Lysis buffer (no DTT)	130	66
(1.127mL/sample)	DTT (add to Lysis buffer)	1.3	0.66
Lysis Buffer (with DTT) Reagent Trough	As above	125	63
DNA IQ RESIN Solution	Lysis buffer (with DTT) (from above) 43µL	6	3
(50μL/sample)	DNA IQ RESIN 7µL	1	0.5
DNA IQ 1X Wash Buffer (300µl/sample)	See Below for preparation	35	18
DNA IQ Elution Buffer (120µl/sample)	Use directly from Kit	14	8

NOTE: For batches not equal to either 96 or 48 samples samples, refer to Appendix Reagents Calculation Tables. Table 1 for batches of <48 samples and Table 2 for <96 (but >48)

Extraction Buffer

Prepare the buffer just before extraction in a 100mL sterile glass bottle according to the table 2. Use one aliquot of 20mg/mL Proteinase K (1.5mL) for a 48 sample run and two aliquots of Proteinase K for a 96 sample run. Remove aliquot/s of Proteinase K from the freezer, defrost, vortex and centrifuge before use. Ensure that the 20% (v/v) SDS is completely dissolved (clear) in the stock solution before making the extraction buffer. If not dissolved invert the container a few times and leave longer at room temperature.

Lysis Buffer with DTT

Lysis buffer is supplied with the kit. Lysis buffer with DTT is prepared at the start of each run. Remove the DTT from the freezer, defrost, vortex and centrifuge before use. Into a sterilised glass bottle, add 1.3mL of DTT to 130ml of Lysis buffer for 96 samples. If 48 samples are to be run, use 660µl of DTT to 66ml of Lysis buffer, again, made up in a sterile glass bottle. Make up the Lysis buffer with DTT within the Laminar Flow and ensure that full PPE is worn, including face shield. Warning: Lysis Buffer and DTT are toxic.

DNA IQ™ Resin

DNA IQ™ Resin is supplied with the kit. The resin is prepared at the start of each run in a 10mL sterile tube. Ensure the resin is properly mixed by vortexing before pipetting. Look for calculations in table 2 for the correct volumes of resin and lysis buffer (with DTT). In the 10mL tube, mix by inversion before adding to column 4 in the 12-channel reagent plate.

1X Wash buffer

2X Wash buffer is supplied with the kit. Once a new kit has been opened, add 35mL of **AnalR** Ethanol and 35mL of Isopropyl alcohol to the 2X wash buffer. Once the reagents have been added, label the lid and side of the bottle with "1X Wash buffer," initial and date.

4.2 Equipment

The following equipment (Table 3) and consumables (Table 4) are required for the DNA IQ extraction.

Table 3. Equipment used and location.

Equipment	Asset No.	Location
STORstar (B system)	10238493	6122
MultiPROBE® II PLUS HT EX with Gripper™ Integration Platform (Ext A Platform)	10076438	6127
MultiPROBE® II PLUS HT EX with Gripper™ Integration Platform (Ext B Platform)	10076437	6127
DPC Shaker (Ext A Platform)	N/A	6127
DPC Shaker (Ext B Platform)	N/A	6127
Automated Temperature Controller, Heat Block tiles and heat block adapters (Ext A Platform)	N/A	6127
Automated Temperature Controller, Heat Block tiles and heat block adapters (Ext B Platform)	N/A	6127
Eppendorf 5804 centrifuge	10238416	6127
Vortex	30087015	6127
Fridge	30433424	6127
Micro centrifuge	None	6127
Decapper	None	6127

Table 4. Consumables used for extraction

Consumables	Location
175uL Clear Non-Conductive Filter Tips – Pre-sterilized	6127
MβP Pure 1000uL Tips – Pre-Sterilized	6127
SlicPrep™ 96 device plate	6122
Axygen 2mL Deep Well storage plate	6127
12 Channel plate	6127
Nunc tubes	6120
Nunc Caps	6127
Sterile 50mL Falcon tubes	6122
Sterile 10mL tubes	6122
Autoclaved 100mL glass bottles	6122
Autoclaved 250mL glass bottles	6122
Aluminium sealing film	6127

5 SAFETY

As per the procedures in the QIS document "Operational Practices in the DNA Dedicated Laboratories" (QIS 17120), PPE is to be worn by all staff when performing this procedure.

The automated platforms, labware and associated equipment should be wiped with 5% TriGene™ followed by 70% ethanol before and after use.

While the MPII is running a procedure, refrain from placing any part of the body inside the cabinet. Either pause the program or use the emergency STOP button located at the front of the instrument before touching anything within the cabinet or on the deck surface.

Warning: Tris base, EDTA, SDS and Lysis Buffer are irritants. DTT is a reducing agent that destroys disulphide bonds. Chaotropic reagents such as guanidinium thiocyanate (GuSCN) are toxic. Do not breathe alcohol fumes. Handle carefully and wear appropriate PPE.

Lysis Buffer contains guanidinium thiocyanate which can be harmful if inhaled, swallowed or comes in contact with skin. Any left over Lysis Buffer with DTT is to be disposed of in a brown Winchester bottle in the fume hood. Never dispose down the sink. If spillage occurs, absorb with liquid-binding material (such as sand, kitty litter, etc) and dispose in a biohazard bin. Again, handle carefully and wear appropriate PPE, including face shield.

6 SAMPLING AND SAMPLE PREPARATION

Samples waiting to be extracted are stored in freezers as described in Table 5.

Table 5. Sample storage locations.

Sample type	Storage Device	Storage Location
Urgent/High/Medium Priority Samples Medium Priority Samples	Freezer Walk in Freezer or Freezer in 6117	6117-2 6109 or 6117-5
Low Priority Samples	Walk in Freezer or Freezer in 6117	6109 or 6117-5

QC samples

All extraction batches require two controls to be registered. These controls, UR numbers and positions are listed in Table 6.

Table 6. Extraction Quality Controls

QC Name	UR Number	Description
Negative Control	FBOT33	Negative Extraction control – Empty well
Positive Control	FBOT35	Positive extraction control - Known Donor dried blood swab

Registration of QC

- 1. Log into the AUSLAB Main Menu.
- Select 1. Request Registration.
- 3. Select 2. Full Reception Entry.
- Scan in barcode of control.
- 5. Enter the UR number as per Table 4 and press [Enter].
- 6. Enter the appropriate Specimen type (e.g. Blood for blood control).
- Request a 9PLEX test, when prompted to enter the processing comment, enter EXTP (Positive extraction control) or EXTN (Negative extraction control).
- Enter LAB in the Billing code field.
- Press [F7] Save to save the Billing details.
- 10. Press [F4] Save twice to save the registration details.

N.B Quality controls should not have a DNA priority allocated at time of registration to ensure they are included in the top positions of a batch

Create the Extraction Batch

- Log into the AUSLAB Main Menu.
- Select 5. Workflow management.
- 3. Select 1. DNA workflow table.
- 4. Highlight the appropriate Extraction batch type and press [F5] Batch Allocation.
- 5. Press [F6] Create batch.
- 6. Press [F8] Print menu.
- 7. Press [F6] Print Batch label. (print 7)
- 8. Press [F7] Print Sample Label. (print 3 sets)
- 9. Press [F8] Print Worksheet. (print 2)
- 10. Press [SF5] Main menu.
- 11. Press [SF11] Print.
- 12. Press [SF6] Accept batch.
- 13. Press [Pause/Break] to exit to the Main Menu.
- 14. Obtain worksheets (FBLASER3) and labels (FBLABEL13-16) from the Analytical Section printing bench (Room 6117).

Locating Samples

To locate samples refer to "Analytical Sample Storage" (QIS 24255).

Checking Samples

Check that appropriately sized portions of sample (eg swab, fabric, cigarette butts) have been submitted. If samples are not sized correctly they are to be sub-sampled please refer to "Examination of Items" (QIS 17142)

Label 1.5mL tubes removed from inside the original 5mL tube with sample labels if required. Label empty Nunc tubes ready for sequence checking.

Sequence Check the Sample substrates and Nunc Bank-It™ tubes

To sequence check sample substrates and storage tubes please refer to method "Procedure for the Use of the STORstar unit for automated sequence checking" (QIS 24256).

ENSURE the Slicprep™ 96 device is labelled, with the AUSLAB Batch ID label on the left side of the plate and the barcode on the right hand side of the plate.

ENSURE the Nunc tube rack is labelled with the AUSLAB Batch ID and barcode on the front of the plate.

7 PROCEDURE

Automated setup of DNA IQ extractions in 96-deep well format is performed using the MultiPROBE® II PLUS HT EX EP-A and EP-B platforms located in Room 6127.

Refer to "Operation and Maintenance of the MultiPROBE® II PLUS HT EX and MultiPROBE® II PLUS HT EX with Gripper™ Integration Platform" (QIS 23939) for instructions on the use and maintenance of the MultiPROBE® II PLUS HT EX platforms.

Summary of DNA IQ EXTRACTION winprep program (v 1.3)

- Lysis of the biological material on solid support: Add prepared Extraction Buffer (500μL) to Slicprep plate wells. Cover the Slicprep plate & Spin baskets with Aluminium seal and incubate 45 min @ 37 °C. (this occurs at steps 8-12 of the protocol)
- Remove the Slicprep plate & Spin baskets: add the collar and centrifuge for 2 min.
 Remove the collar and discard it. Remove the Spin baskets part and keep it in a clean
 container. Return the Slicprep plate to the deck. (this occurs at step 14 of the protocol)
- Binding of paramagnetic resin to DNA and further Lysis: add Resin solution (50μL) and Lysis Buffer (957μL). Automated mixing and shaking @ room temperature for 5 min. (this occurs at steps 17-22 of the protocol)
- 4. Removing lysis reagents: Slicprep plate is moved to the PKI Magnet to separate beads. Removing of supernatant (1600μL) without disturbing resin, dispense this solution in the storage plate. (this occurs at steps 25-27 of the protocol)
- 5. Washing of the resin-DNA complex: To remove any inhibitors in solution. The first wash is with Lysis buffer (125µL), shaking @ room temperature for 1 min. The plate is moved to the PKI Magnet and the supernatant is removed into the storage plate.

The next three washes are with 1X Wash buffer (100µL), shaking @ room temperature for 1 min. During each wash cycle, the plate is moved to the PKI Magnet and the supernatant is discarded. (this occurs at steps 30-68 of the protocol)

- Removing any excess of 1X Wash buffer: air dry @ room temperature for 5 min. (this occurs at step 69 of the protocol)
- 7. Elution of DNA from the Resin-DNA complex: Add Elution buffer (60μL) and incubate @65 °C for 6 minutes (3 min no shaking and 3 min shaking). The plate is moved to the PKI Magnet. The eluted solution (supernatant) is removed to the NUNC tubes. Step 7 is repeated twice. (this occurs at steps 71-92 of the protocol)
- 8. Flushing of capillaries: The capillaries are washed with Amphyl and nanopure water.

Preparation of Reagents prior to extraction

- 1. Defrost Prot K and DTT
- Refer to table 2 for reagent volumes to make up the required amount of Extraction Buffer, Lysis buffer (with DTT) and Resin solution. Also measure the required amount of 1X Wash buffer.
- Record the Lot numbers of all the reagents used onto the AUSLAB worksheet.

Setting up the EP-A or EP-B MPIIs

These steps are to be carried out in the Automated extraction Room (Room 6127)

- 4. Turn on the instrument PC.
- Log onto the network using the Robotics login.
- 6. Double click the WinPrep® icon on the computer desktop (Figure 1).
- 7. Log onto the WinPrep® software by entering your username and password, then press "Enter".



- 8. Ensure the System Liquid Bottle is FULL before every run and perform a Flush/Wash.
- Ensure that the daily/weekly start-up has been performed before running any program.
 If WinPrep® has been closed or been idle for a long period of time initialise the MP II platform as described in QIS 23939.
- 10. Open the Extraction setup MP II test file in WinPrep® by selecting:
 - File
 - Open, navigate to C:\Packard\MultiPROBE\Bin\QHSS PROTOCOLS
 - Select "DNA IQ Extraction Ver1.3.mpt."
 - Click the "Open" button
- 11. Check the whole program there are no bold fonts (e.g. plates may lose gripability and the program will not run). See the line manager.
- 12. Decontaminate the required labware by wiping with 5% TriGene followed by 70% ethanol before placing the labware on to the designated grid positions as outlined in the virtual deck view within WinPrep® (Figure 2).

- The white WALLAC Isoplates (catalogue #1450-514) that are used to support the SlicPrep™ 96 device plate must be placed into positions E13, D16 and C19.
- Ensure that the PKI Magnet at F16 is stable in its insert. This is critical so that beads and DNA are not lost during pipetting.

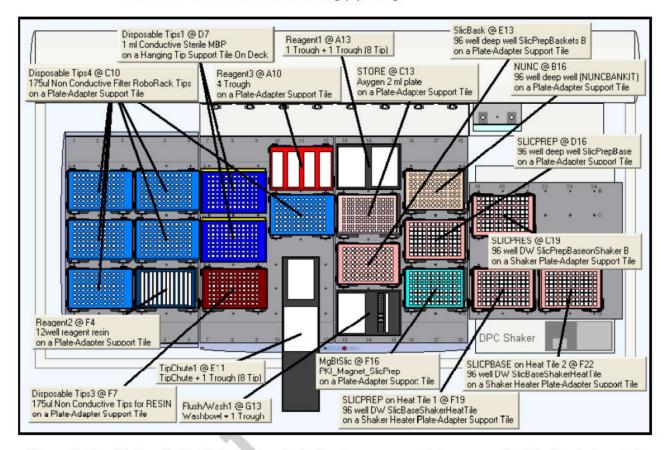


Figure 2. The WinPrep[®] virtual deck view displaying the necessary labware required for the Automated DNA IQ™ Method of Extraction on Extraction Platform A.

- 13. Ensure that the DPC Shaker and Heater Controller Box are switched on. For EP-A: Tile 3 should be at F19 (50°C), Tile 1 at F22 (85°C). For EP-B: Tile 1 should be at F19 (50°C), Tile 2 at F22 (85°C). Note: Press the start/stop button twice at the front of the DPC Shaker to ensure that it displays zero on the screen.
- 14. To the Amphyl wash station at A10, add fresh 1% Amphyl to the trough on the far left hand side, 0.2% diluted Amphyl to the middle left and place an empty reagent tough in the middle right position of the station. The nanopure water goes to position G13 into a 160mL trough in the Flush-Wash station.
- 15. Pour the required amounts of Extraction Buffer and Lysis Buffer into the labelled 150mL reagent troughs. Place Lysis Buffer on the left hand side and the Extraction buffer on the right hand side of the 2 trough holder located in position A13.
 Note: Ensure that full PPE is worn, including face shield when handling these reagents
- 16. Nunc tube rack: Check that is the same Auslab batch number as the platemap to be run. Ensure that the barcode of the first (A1) and last tube (H12) are the respective ones. Attach batch label and batch barcode label to front side of rack. Add B1-Lite

- generated "NUNC" barcode to the right side of the nunc tube rack. Then place nunc rack into position B16
- 17. On an Axygen 2ml deep well Storage plate, label the left side of the plate with both the Batch ID and barcode. Label the right side of the plate with a B1-Lite generated "STORE" barcode. Then place in position C13.
- 18. **Slicprep™ 96 device**: Gently remove septa mat from Slicprep™ 96 device and check that substrates are at the bottom of the Spin baskets, if not push them down with a sterile disposable tip and place the Slicprep™ 96 device into position **E13**.
- 19. In I drive from Extraction folder open the required plate map. Check that the plate map is complete. If there are blanks (samples that have been removed), make all volumes to 0. If it is not a full batch, delete the rows of blank samples at the end of the platemap, then save the changes. Copy the plate map to the following file path: C:\Packard\Ext Plate MAPS
- 20. After ensuring that all the necessary labware has been positioned in the correct grid position as displayed within WinPrep®, click the "EXECUTE TEST" button. While the test is loading, record all run information in the Run Log book.
- 21. Click "Reset Tip Boxes" and ensure that the number of tips displayed in this window match those as available on the deck platform. Fill new tips if necessary. Click "Close" to accept the tip count, followed by clicking "Next"
- 22. Select the correct platemap by browsing to C:\PACKARD\EXT PLATE MAPS. Ensure that the platemap selected matches the batch ID affixed to the 96-well Slicprep™ 96 device in position D16. Once this has been done, click "Start", to continue.
- 23. After the barcodes have been read, a user prompt will appear as a reminder to: "Ensure
 - 1. Shaker and heat box are on.
 - 2. Deck has been populated correctly.
 - 3. The Lysis buffer is on the left side and Extraction buffer is on the right at A13." Click "OK" to continue.
- 24. Once the extraction buffer has been added to the plate, a message will appear waiting for the heating tile to reach 50°C (real temp 37°C). When current temperature reaches 50°C click "Continue".
- 25. The next prompt that appears will request the following: "Cover Slicprep with the Aluminium sealing film, then place in position F19. Press "OK."
- 26. After shaking, a User Prompt will appear with the following directions:
 - "Remove plate, add white plastic collar and centrifuge 5mins at 3021rpm, then in the cabinet, remove the spin basket part and place it in the empty 1 ml tip container."
 - Place the Slicprep[™] 96 device into the plate centrifuge and ensure the correct balance plate is used. Once the plate has been centrifuged, carry the plate to the hood and remove the basket of the Slicprep[™] 96 device, storing the basket in an empty 1mL tip box, discard the Collar. Complete the step by clicking "**OK**".
- 27. Add the Resin to the 12 Channel plate. Pipette mix thoroughly. Then dispense 50uL of resin into each well of the Slicprep™ 96 device.

- 28. Place the 12 channel plate into position **F4** then add the Elution buffer to the plate by splitting the amount of elution buffer in half between channels 11 and 12.
- 29. Place the Wash buffer in the far right hand side of the 4 well reagent trough holder (Amphyl wash station at A10)
- 30. The next User prompt will appear with the following directions: "Place the Slicprep in position D16. Ensure wash buffer has been added. Manually add 50uL of Resin. Ensure Elution Buffer has been added." Press "OK" when steps 23-25 have been performed.
- 31. Once the lysis and wash steps have been completed, a message will appear waiting for the heating tile to reach 85°C (real temp 65°C). DO NOT PRESS CONTINUE it will continue automatically when temperature has reached 85°C.
- 32. After the first elution step when the plate is moved to the PKI Magnet, a User message will appear requesting:

"Check Nunc tubes are uncapped at position B16
Push down the Slicprep on the PKI Magnet then press OK."

Note: Allow the plate to cool slightly before pushing down on all four corners of the plate to lock it into place on the PKI Magnet.

- 33. After the second elution step, the above prompt will appear again. Note: Allow the plate to cool slightly before pushing down on all four corners of the plate to lock it into place on the PKI Magnet
- 34. Once the program is completed, a final User Message prompt appears asking to: "Remove all the plates starting with the Slicprep plate, place the Spin Basket into the Slicprep plate.

Cover the Storage plate with the aluminium sealing film."

Recap the NUNC tubes

Once all plates are removed from the deck and sealed, place into small clipseal plastic bags. Click "**OK**" to proceed to the Amphyl wash step to decontaminate the system tubing.

Finalising the MP II run

- 35. Discard the 12-channel plate used for Resin and Elution Buffer in the biohazard waste bin.
- 36. Remove Lysis buffer with DTT (wear face shield) and dispose of left over reagent into the glass bottle used previously. Bring this bottle to the room 6122 and dispose in the brown Winchester bottle located in the fume hood.
- 37. Empty the contents of the Amphyl wash station into the sink and rinse the troughs with nanopure H₂O upon completion of the run.
- 38. Remove all labware from the deck and clean with 5% TriGene™ followed by 70% ethanol, and setup for the next run if necessary.
- 39. Remove the tip chute and funnel, rinse with warm tap water to help loosen and remove any residue inside the chute before cleaning with 5% TriGene and 70% ethanol. Hang the chute to dry on the rack beside the sink. If running another extraction straight after, replace the tip chute with another spare, clean chute.
- Move the platemap to C:\Packard\Ext PLATE MAPS to the "Completed Extractions" folder.

Recording Reagent Details and other information in AUSLAB

- 41. To record reagent lot numbers, log into the AUSLAB Main Menu.
- 42. Select 5. Workflow Management.
- 43. Select 2. DNA Batch Details.
- 44. Scan in the Extraction Batch ID.
- 45. Press [F6] Reagents.
- 46. Press [SF8] Audit.
- 47. Press **[F5]** *Insert Audit Entry*, enter the lot number details, operator name and Extraction platform the batch was run on and press **[Enter]**.

Importing the MP II log file into AUSLAB

- 48. To extract the MP II log file, click on the Microsoft Access icon in the WinPrep® main menu to open the MultiPROBE log database.
- Click on the relevant run Test ID in the Test Selection box. In the Report/Query/Action Selection dropdown menu, select "Report: Test Summary (Sorted by Destination Rack ID)"
- 50. In the Output Selection dropdown menu, select "File". Save the output file as *.csv format to C:\Packard\Ext Plate MAPS\Ext Logs with the same name as the AUSLAB batch ID and click "Apply". (refer to figure 4. below)

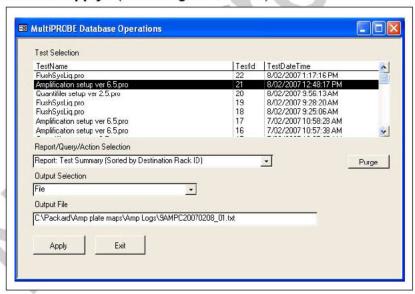


Figure 4. The MultiPROBE log database for collecting MP II run information

- 51. Open the log file and check for any errors that may have arisen during the Extraction process. Compare the listed errors to any that were encountered during the run. Report any critical errors to the line manager.
- 52. Copy the log file to I:\EXTRACTION\EXT A MPII\Logs or I:\ EXTRACTION\EXT B MPII\Logs for uploading to AUSLAB.
- 53. Log into the AUSLAB Main Menu.
- Select 5.Workflow Management.
- 55. Select 2. DNA Batch Details.
- Scan in the Extraction Batch ID barcode.
- 57. Press [SF6] Files.
- 58. Press [SF6] Import Files.
- 59. AUSLAB prompts "*Enter filename*"; enter the filename and extension and press [Enter]. (e.g. I:\EXTRACTION\EXT A MPII\Logs\CWIQEXT20071115_01.csv)
- AUSLAB prompts "Is this a result file Y/N?" enter N and press [Enter].

61. Press [Esc].

Importing Extraction "Results" into AUSLAB

- 62. Log into the AUSLAB Main Menu.
- 63. Select 5. Workflow Management.
- 64. Select 2. DNA Batch Details.
- 65. Scan the Extraction batch ID barcode located on the worksheet.
- 66. Press [SF6] Files.
- 67. Press [SF6] Import Files.
- 68. AUSLAB prompts "*Enter filename*"; enter batch name and extension and press [Enter].(e.g. CWIQEXT20071115_01.txt)
- 69. AUSLAB prompts "Is this a results file y/n?" enter "y" and press [Enter].
- 70. The file will be imported into AUSLAB and appear in the DNA file table.
- 71. Highlight entry and press [Enter], for access to the DNA results table.
- 72. Page down through the table and check that all sample results have been imported.
- 73. Press [SF8] *Table Sort Order*, this sorts the table so any samples that have failed Autovalidation are sorted to the top of the table.
- 74. For all samples that have failed check the **Processing Comments**, by entering into the sample.
- 75. a) If processing comments state sample is to be sent to another batch type other than quant. Return the sample to the correct next batch type – e.g. microcon, nucleospin and pooling
 - b) Press [Esc] to exit back to the DNA results table.
 - c) Do not toggle accept.
- 76. a) If processing comment does not state next step for sample the sample will be processed as normal.
 - b) Press [Esc] to exit back to the DNA results table.
 - c) Highlight any entries to be changed and press [SF7] Toggle Accept
- 77. Press [F7] Complete Batch, all of the samples with a Yes in the Accept column will be transferred to the outstanding Quant Casework or Quant Reference lists.
- 78. File the Extraction worksheet into the relevant folder in Room 6117.

8 SAMPLE STORAGE

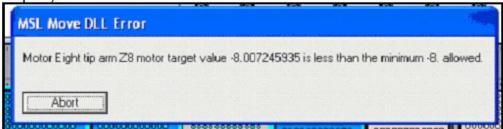
Please refer to "Analytical Sample Storage" (QIS <u>24255</u>) for how to store the old original 5 mL sample tubes, the DNA extract NUNC tubes, Slicprep with Basket and Axygen store plates.

9 TROUBLESHOOTING

- 1. If the barcode reader is not reading the barcodes of the Nunc tube rack, or the Slicprep Plate or the Store plate, manually scan the appropriate barcodes.
- When reading the Nunc tube rack barcode, if the Gripper is not picking up or placing the Nunc tube rack properly on the deck, just manually place the rack properly on the plate adapter support tile.
- When reading the Store plate barcode, if the Gripper is not picking up or placing the Store plate properly on the deck, just manually place the plate properly on the plate adapter support tile.
- 4. When reading the Slicprep plate barcode if the Gripper is not picking up the plate properly:
 - a. if the plate was not properly placed on the plate adapter support tile with the Wallac Isoplate, just manually place the plate properly.
 - b. if the plate was properly placed on the plate adapter support tile with the Wallac Isoplate on it: it means that the gripper needs to be initialised. Abort the



- run, Initialise the instrument and restart the run. If problem persists, shutdown the MPII and PC, restart and then initialise the whole instrument. Otherwise, contact your line manager.
- c. Calibrate relevant labware using the SlicPrep Calibration plate. This has preset standardised positions that need to be the same on all labware where the Slicprep plate is being moved. The same plate is used on both extraction platforms A and B.
- d. Check the calibrations against the run program DNAIQGripperTest.pro. This program moves the Slicprep across all the labware the gripper moves across. Start with the Slicprep at D16.
- 5. In steps 18 or 26, if a message is stating that the instrument is having a motor problem when picking up 1 mL tips and the only option is to Abort, abort, initialise and open program version 1.3a (if the problem is in step 18) or version 1.3b (if the problem is in step 26).



As the program will start the gripper will pick up the plates, it is not necessary that the Nunc tube rack is in position (B16), only ensure that it is reading the correct barcode. It is **important not** to place the Slicprep in the original position (E13) as the Slicprep plate has the Spin basket part removed (ie keep at D16), ensure it will scan the correct batch barcode. The Store plate remains in the original position. If the problem persists even after restarting, replace the rack of disposable 1 mL conductive sterile MBP tips for a new one.

- 6. If the program has already started step 18 and the previous message is appearing, you need to abort. Initialise the instrument and open program version 1.3a.

 Go to the plate map in C:\PACKARD\ EXT PLATE MAPS and change volume to 0 for all the samples that the Lysis Buffer have been dispensed (Column 6), ensure that the number of samples where the Lysis buffer was added is the same as the ones where the volume needs to be changed.
 - Save the changes and use this plate map for the restart of the program. Please read troubleshooting 5 for barcode reading of plates.
 - If the problem persists even after restarting, replace the rack of disposable 1 mL conductive sterile MBP tips for a new one.
- 7. If the program has already started step 26 and the previous message is appearing, you need to abort. Initialise the instrument and open program version 1.3b.

 Go to the plate map in C:\PACKARD\ EXT PLATE MAPS and change volume to 0 in all the samples that the Lysis Buffer and Ext buffer have been removed (Column 9), ensure that the number of samples where the solution was removed is the same that the ones the volume need to be changed.
 - Save the changes and use this plate map for the restart of the program. Please read troubleshooting 5 for barcode reading of plates.
 - If the problem persists even after restarting, replace the rack of disposable 1 mL conductive sterile MBP tips for a new one.
- 8. If a disposable tip gets stuck on the 8 tip arm during disposal of tips a user message will appear. Remove and press retry and then continue.
- 9. If the message:



has appeared, press OK and the program will be aborted automatically. Check that all the connections to the instrument (shaker, heater and computer) are properly plugged in. If everything is OK, you need to close WinPrep, shut down the instrument, shaker, heater and PC. After 2 min restart everything. Once Winprep has been opened, reinitialise the instrument and start the program (check version number according to which step the message has came up). Please read troubleshooting 5 for barcode reading of plates.

10 VALIDATION

- Nurthen, T., Hlinka, V., Muharam, I., Gallagher, B., Lundie, G., Iannuzzi, C. "Project 9. Automation: Report on the Evaluation of DNA Extraction Chemistries." June 2007.
- Nurthen, T., Hlinka, V., Muharam, I., Gallagher, B., Lundie, G., Iannuzzi, C. "Project 11: Report on the Validation of the Automated Extraction Chemistry Kit using the MultiPROBE® II PLUS HT EX with Gripper™ Integration Casework Platform." 2007.
- Nurthen, T., Hlinka, V., Muharam, I., Gallagher, B., Lundie, G., Iannuzzi, C. "Project 13: Report on the Verification of the Automated Extraction Chemistry Kit using the MultiPROBE® II PLUS HT EX with Gripper™ Integration Casework Platform." 2007.

11 QUALITY ASSURANCE/ACCEPTANCE CRITERIA

- A Negative control (also known as the reagent blank) is included with each batch of
 extractions. This reagent blank is processed as a normal sample through to completion.
 If any results are obtained from this sample, either at the quantitation step or the
 Genescan analysis step, then the possible source of the contamination is investigated.
 The samples extracted with this control are thoroughly checked and repeated if
 possible.
- Positive and negative controls are included in each extraction batch as per table 4.

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- Vandenberg, N., van Oorchot., R. A. H., & Mitchell, R. J., An evaluation of selected DNA extraction strategies for short tandem repeat typing. Electrophoresis, 1997. 18: p. 1624-1626.

13 STORAGE OF DOCUMENTS

All worksheets are stored in the Analytical area (Room 6117).

14 ASSOCIATED DOCUMENTS

- QIS 17120 Operational Practices in the DNA Dedicated Laboratories
- QIS 17142 Examination of Items
- QIS 17171 Method for Chelex Extraction
- QIS 17165 Receipt, Storage and Preparation of Chemicals, Reagents and Test Kits
- QIS <u>23939</u> Operation and Maintenance of the MultiPROBE® II PLUS HT EX and MultiPROBE® II PLUS HT EX with Gripper™ Integration Platform
- QIS 24255 Analytical Sample Storage
- QIS 24256 Sequence Checking with the STORstar Instrument
- QIS 24469 Batch functionality in AUSLAB

15 AMENDMENT HISTORY

Revision	Date	Author/s	Amendments
0	23 Oct 2007	B. Gallagher, T. Nurthen,	First Issue
		C. lannuzzi, V. Hlinka,	
		G. Lundie, I Muharam.	
1	12 Dec 2007	M Harvey, C Iannuzzi, A McNevin	Reviewed and updated after initial training

16 APPENDIX

16.1 Reagents Calculation Tables

1. Table for less than 48 samples (note difference is in DNA IQ RESIN Solution)

Lysis-DTT buffer		Volume (in mL)
Lysis buffer	(Nx1.35)+0.75	
DTT (1M)	Lysis Buffer/100	
900 80		
Extraction Buffer		
TNE buffer	Nx0.56	
Prot K (20 mg/L)	Nx0.03	
SDS (20 %)	Nx0.015	
DNA IQ RESIN Solution		
LYSIS buffer	0.054x(N+8)	
DNA IQ RESIN	0.009x(N+8)	
DNA IQ 1X Wash buffer	Nx0.36	
DNA IQ Elution buffer	Nx0.144	

N = number of samples on batch including positive and negative controls, round up to nearest multiple of 8)

2. Table for more than 48 samples (note difference is in DNA IQ RESIN Solution)

Lysis-DTT buffer	Volume (in mL)		
Lysis buffer	(Nx1.35)+0.75		
DTT (1M)	Lysis Buffer/100		
Extraction Buffer			
TNE buffer	Nx0.56		
Prot K (20 mg/L)	Nx0.03		
SDS (20 %)	Nx0.015		
DNA IQ RESIN Solution			
LYSIS buffer	0.054x(N+16)	5	
DNA IQ RESIN	0.009x(N+16)		
DNA IQ 1X Wash buffer	Nx0.36		
DNA IQ Elution buffer	Nx0.144		

N = number of samples on batch including positive and negative controls, round up to nearest multiple of 8)

16.2 Manual method for extraction using DNA IQ™

16.2.1 Sampling and Sample Preparation

Samples waiting to be extracted are stored in freezers as described in Table 3.

Table 3. Sample storage locations.

Sample type	Storage Device	Storage Location
Urgent/High/Medium Priority Samples	Freezer	6117-2
Medium Priority Samples	Wak in Freezer	6109
Low Priority Samples	N/A	

QC samples

All extraction batches require two controls to be registered. These controls, UR numbers and positions are listed below in Table 4.

Table 4. Extraction Quality Controls

QC	UR Number	Extraction types	
Neg Control	FBOT33	All	
QC swab (blood)	FBOT35	Blood	

- 1. Log into the AUSLAB Main Menu.
- Select 1. Request Registration.
- Select 2. Full Reception Entry.
- 4. Scan in barcode of control.
- Enter the UR number as per Table 4 and press [Enter].
- Enter the appropriate Specimen type (e.g. Blood for blood extraction).
- Request a 9PLEX test, when prompted to enter the processing comment, enter EXTP (Positive extraction control) or EXTN (Negative extraction control).
- 8. Press [F7] Enter LAB in the Billing code field.
- 9. Press [F4] Save to save the Billing details.
- 10. Press [F4] Save to save the registration details.

N.B Quality controls should not have a DNA priority allocated at time of registration to ensure they are included in the top positions of a batch

Create the Extraction Batch

- 15. Log into the AUSLAB Main Menu.
- Select 5. Workflow management.
- Select 1. DNA workflow table.
- 18. Highlight the appropriate Extraction batch type and press [F5] Batch Allocation.
- 19. Press [F6] Create batch.
- 20. Press [F8] Print menu.
- 21. Press [F6] Print Batch label. (for the deep well plate)
- 22. Press [F7] Print Sample labels. (print four sets of labels for all extractions)
- 23. Press [F8] Print Worksheet.
- 24. Press [SF5] Main menu.
- 25. Press [SF11] Print.
- Press [SF6] Accept batch.
- 27. Press [Pause/Break] to exit to the Main Menu.
- 28. Obtain worksheets (FBLASER3) and labels (FBLABEL13-16) from the Analytical Section printing bench (Room 6117).

Locating Samples

Determine the storage locations of the required samples using the Batch Creation table/Batch details table print out. The columns *Rack* and *Pos* respectively identify the rack and the grid location where the sample has been stored. Remove the samples from the storage rack and place in an orange rack (12x8).

When all samples have been located:

- Log into the AUSLAB Main Menu.
- 2. Select 2. Sample Processing.
- 3. Select 7. Search Sample storage.
- 4. Scan in the sample barcode that is affixed to the sample tube.
- 5. Press [F6] Remove Sample.
- AUSLAB prompts "Are you sure you want to remove XXXX-XXXX? (Y/N)", Enter Y and press [Enter].
- AUSLAB prompts "Please enter remove comment", No comment is required. Press [Enter].
- Press [Scroll lock] to clear.
- 9. Repeat steps 5 8 until all of the samples have been removed from their rack.

Sequence Check the tubes

- Thaw samples at room temperature and label 1.5mL sample tubes.
- 2. Sequence check the tubes.
- Add the sequence check details into AUSLAB.
- 4. Log into AUSLAB Main Menu.
- 5. Select 5. Workflow Management.
- 6. Select 2. DNA Batch Details.
- 7. Scan in the appropriate extraction batch ID barcode.
- 8. Press [F5] Sequence Check.
- 9. Scan in the appropriate extraction batch ID barcode.
- 10. Press [Pause/Break] to exit to Main Menu.

16.2.2 Procedure

- Enter the number of samples to be extracted (including controls) into the "No of Samples" column of the DNA IQ Reagent Calculations Table to calculate the volumes of each reagent to be measured out for the extraction. Aliquot regents into either 5ml tubes or 50ml Falcon tubes. Note: The volume of Lysis buffer calculated includes the volume used in the resin-lysis solution
- 2. Turn on the Eppendorf Thermo mixer and set the temperature to 37°C.
- 3. Remove 1.5ml tube and retain the 5mL tube. Prepare the Spin baskets by placing a DNA IQ™ Spin basket into a 1.5mL Microtube. Label the spin baskets (for every tube except Ext. control), 2mL SSI tubes and Nunc storage tubes (for every sample) with the sample barcodes. Have a second operator perform a sequence check of all tubes. This person must Press [F5] Sequence Check against the batch in AUSLAB
- 4. Using the Reagents table, prepare Extraction Buffer, Lysis buffer with DTT, & Resin Solution. Reagents need to be prepared fresh before each run.
- Add 300 μL of Extraction buffer to each tube. Vortex each tube before incubating the tubes at 37°C on the Thermomixer at 1000 rpm for 45 minutes.

- Remove the tubes from the Thermo mixer and add to a rack, increase the temperature on the Thermomixer to 65°C (for use in the Elution steps).
- 7. Transfer the substrate from the original tube to a DNA IQ™ Spin Basket using autoclaved twirling sticks. Centrifuge the Spin basket for 2 minutes at room temperature at its maximum speed. Once completed, remove the spin basket & retain in the original 5ml tube. Ensuring minimal contamination and transfer the extract to a labelled 2mL SSI sterile screw tube.
- 8. Transfer the remaining extract from the original tube to the corresponding 2mL tube. Vortex the tube gently.
- Add 550 μL of Lysis Buffer to each tube.
- 10. Into a separate, clean 2mL SSI tube, aliquot the required amount of lysis buffer for the Resin solution. Ensure that the DNA IQ™ Resin solution has been thoroughly mixed by vortexing the resin bottle before adding the required resin volume to the lysis buffer. Pipette mix the solution to clear the tip of any lingering resin beads. Mix the solution by inverting the tube to prevent bubbles forming.
- 11. Add 50µL of DNA IQ™ Resin-Lysis solution into each tube. Invert the resin-lysis tube at regular intervals to keep the resin suspended within the solution to ensure uniform results.
- 12. Vortex each tube for 3 seconds at high speed before placing the tubes in the Multitubeshaker set at 1200 rpm to incubate at room temperature for 5 minutes.
- 13. Remove from the Multitubeshaker and vortex the tubes for 2 seconds at high speed before placing the tubes in the magnetic stand. Separation will occur instantly.

Note: If resin does not form a distinct pellet on the side of the tube, or if the pellet has accidentally mixed with the solution while in the stand, vortex the tube and quickly place back in the stand.

14. Carefully remove all of the solution in the tube into the original 1.5mL tube (i.e. the tube which originally contained the substrate), ensuring that the resin is not disturbed from its place on the side of the tube.

Note: If some resin is drawn up in tip, gently expel resin back into tube to allow reseparation.

- 15. Add 125µL of prepared Lysis Buffer and vortex for 2 seconds at high speed. Return the tubes to the magnetic stand and allow for separation to occur. Once separation has occurred again remove the Lysis Buffer into the original 1.5mL tube (i.e. the tube which originally contained the substrate).
- 16. Add 100µL of prepared 1X Wash Buffer and vortex for 2 seconds at high speed. Return tube to the magnetic stand and once separation has occurred remove and discard all Wash Buffer.
- 17. Repeat Step 16 another two times for a total of three washes. Ensure that all of the solution has been removed after the last wash.
- 18. Wipe down a Biohazard hood with bleach followed by ethanol. Uncap the tubes, placing the lids inside down onto a clean rediwipe in consecutive order and place the tubes in the same order into a clean plastic rack. Air-dry the resin in the hood for 15

- minutes at Room temperature. Do not dry for more than 20 minutes, as this may inhibit removal of DNA during the elution phase.
- 19. Once the resin is dry, replace the lids on the corresponding tubes and remove from the hood. Add 50µl of Elution Buffer to each of the samples by carefully pipetting the liquid to the side of the tube, above the pellet. Do not mix.
- 20. With the lids on, incubate the tubes in the Thermomixer at 65°C for 3 minutes. After the three minutes are up, continue to incubate for a further 3 minutes shaking at 1100 rpm.
- 21. Remove the tubes and vortex for 2 seconds at high speed. Immediately place the tube in the magnetic stand while hot to ensure maximum DNA yield during elution.
- Carefully transfer the DNA containing supernatant to the corresponding labelled Nunc tubes.
- 23. Remove tubes from the magnetic stand and add carefully another 50 μ L of Elution Buffer above the magnetic pellet.
- 24. Repeat step 30 to 32. The final volume after this elution should be approximately of 95 μL of DNA solution.
- 25. DNA can be stored at 4°C for short-term storage or at -20 or -70°C for long-term storage.

16.2.3 Sample storage

- Log into AUSLAB Main Menu.
- Select 2. Sample Processing.
- Select 6. Sample Storage.
- 4. Scan in Rack barcode.
- 5. Press [SF5] Fill Rack.
- 6. Scan in sample barcode and place in rack in scanned position.
- Repeat for all samples.
- 8. Press [Esc].
- 9. Press [Pause/Break] to return to the Main Menu.
- 10. Select 3. Patient Enquiry.
- 11. Scan in Rack barcode.
- 12. Tab down to the next blank **DNA Batch No** field and press **[F2] Edit**.
- 13. Scan in the Batch ID of the samples stored.
- 14. Press [Pause/Break] to return to the Main Menu.

JH-42

Issue Identified	Barcode(s) affected (new cell for each barcod please)	Suggested cause	Date	Initials (of person making entry)	Outcome
Some reagents were made and left on the bench in the kit room. As they were unlabelled and the autoclave tape was not intact, and they were still there over 24 hrs later, they have been discarded.		Please label reagents made on the day you make them	24/04/2008	OF	See Operational Issues Log
I've been coming across a lot of samples that have been reamped at the same volume but with significantly less alleles being obtained in the reamp. For example, 285012671 was originally amped at 6uL and was a full profile with an extra peak (NR) at D8. It was then reamped at 6uL to confirm the extra peak but only 6 alleles were obtained.	More barcodes are available for egsamples.	Based on this, and on the numerous other ones I've seen, it would seem that the samples aren't being resuspended when reworks are being ordered.	21/05/2008	ON	More barcodes have been provided: These barcodes relate to FTA's - these appear to be punching inconsistencies These will be investigated as soon as possible 22/9/08 Sufficient further examples have been provided are currently being sorted and results will be fed back once investigation is complete 20-10-08 Investigation being written up and will be submitted to management team for each team leader to distrubution to teams 11-12-08 Report has been emailed out to management team for distribution to teams
For ReGS, specimen notes should inlcude the amp batch and position - not the 3100/3130 or Genotyper batch an position.				MA	Issue has been raised at relevant team meetings
Thermalcycler program wasn't recognising when E and F were turned on. Check that the cables are connected properly/ push them in a bit			6/08/2008	ВМ	Discussed in Analytical team meeting 8/9/08

Issue Identified	Barcode(s) affected (new cell for each barcod please)	Suggested cause	Date	Initials (of person making entry)	Outcome
The use of forceps for cell extraction, is time consuming, poses contamination risks between samples, requires more area in the DNA suite and OHS consideration with bunsen burners and ethanol in close proximity.			15/09/2008		Discussed in Analytical team meeting 8/9/08 - Forceps not to be used for Chelex batches, however will still be used for off-deck lysis batches in the future
Sample tubes are sometimes not being wiped dry with a rediwipe, or briefly left to dry on the bench before being centrifuged after boiling step. This results in water pooling in the centrifuge columns and wetting any further tubes placed in the centrifuge.			15/09/2008		Discussed in Analytical team meeting 8/9/08
Waterbaths in DNA suite- where is the best place/ configuration			26/09/2008		Hot block will be moved from 6121 to 6120.
Bleach and ethanol stock bottles moved to 6120.			29/09/2008		Discussed in Analytical team meeting 29/09/2008
Test Quants for Kits - the SOP is ambiguous regarding whether points on the std curve are able to be removed to allow the Quant to pass. A consensus needs to be reached and QIS comment added to SOP regarding this.			14/10/2008		OK to remove points when testing kits or a control but NOT standards. Remove points as per std quant SOP. KAL to amend SOP.
Concerns have been raised about incronguent NucleoSpin Clean-up results, some barcodes have been provided. These will be investigated as time permits			20/10/2008	W	Investigation into the efficiency of Nuc Cleanup extractions underway - results to be published when complete

Issue Identified	Barcode(s) affected (new cell for each barcod please)	Suggested cause	Date	Initials (of person making entry)	Outcome
Damage to MPIIs from barcode reader screw. Has stripped of paint and is creating holes in robot frame which are becomming rusty. Needs repaint of section affected to contain rust on Pre-PCR and U-shield ordered from workshop.			27.10.2008	₽	VH to do a CMMS
TE for amps runs out when we have lots of dilutions on an Amp plate. On one plate have had to replace TE two times (3 tubes used instead of one). Can we have more TE aliquoted to the 2mL SSI tubes which program uses? Thanks VH			27.10.2008	Α	to be aliquot into 400uL tubes
When labelling 5mL tubes and sample tubes for "transfer", some people aren't covering the "old" barcode with the new number label. This resulted in a transfer sample being scanned for storage under the old number instead of new number. The new number label should cover the "old" barcode, preventing it from being scanned, with the "old" and "new" numbers still clearly visible.			6/11/2008	П	Noted
When performing Blood chelex extraction noticed that in tubes with large cotton swab and some swabs from QPS, the swabs are stack in the tube and it is hard to make the H2O solution release the stained material, as well as in the last step some solution get trapped between the bottom of the tube and the stained head of the swab. The chelex seems to be on the top of the swab and not in contact with this solution. Very hard or impossible to remove solution. Suggestion sample the large swabs longside			10/11/2008	ō	Interim solution - transfer all blood swabs to an appropraite 2mL screw-cap tube to perform blood extractions. Long term solution - combined blood/cell chelex method (separate change proposal) to be developed
18/11/2008 In workflow when creating batches and locating samples please write in the diary that it has been done. Therfore the OO's won't be looking for something that has already been done.					Noted
When changing/removing spent gas canisters from bunsen burners this must be done in a fume hood. Also initial and date empty canister and leave in fume hood for a few days and dispose in normal waste bin.			19/11/2008	BM	Noted

	ı										
Outcome	Noted / Discussed	Noted	Noted	Noted	Noted	Noted	Noted	Noted	Noted. Please also reply to any email sent to FSS DNAorders	Noted	Noted
Initials (of person making entry)	HA	OF	OF	SES	LDT	MLM	OF	BM	OF	Ą	KML
Date	25/05/2009	16/07/2009	16/07/2009	21/07/2009	5/08/2009	19/08/2009	22/09/2009	2/10/2009	2/10/2009	30/11/2009	4/12/2009
Suggested cause											
Barcode(s) affected (new cell for each barcod please)											
Issue Identified	Desperately need second STORstar working again in Analytical. Now required for DNAIQ process. Interferring with Operational staff pulling samples for Analytical.	Don't put a blue liner in the bins for tip box re-cycling as they are being confused with the blue lined rubbish bins. Tip boxes don't make a mess, so this will save on blue liners.	In the DNA suite Room 6120 there is a VISY re-cyling box for cardboard, so if all the cardboard from the three rooms can be put in this box, it will be easier to collect and neater than piling it up on the floor or under the sinks.	In Workflow when requesting in diary Ref Amps, Amp Refs, Quant Refs, Ref Quants, please indicate correct one.	After an extraction is completed, could Analytical Scientist remember to store there tubes please. ie. CWIQEXT.	Please remember to check processing comments on every worksheet.	With Profiler reagents, please don't take them out of use if they are still being used for FTA's. Otherwise Operational staff are unable to use the AUSLAB functionality to attach	reagents to FTA batches Cleaning the 7500 If anyhody requires information on whether stores items are	on order/coming in/delayed tell them to email FSS DNAorders for this information as the Operational staff do not have access to this information. This is also where requests for stock items are to be directed.	Can plate readers be careful of the comments used when reading REF plates; as these direct samples to rework outstanding batches and samples are going to batches which they cannot be processed on, e.g FTAEVD to FTARUN, HAIR to FTARUN, FTABLD to RUN, FTAEVD to RPT etc.	Reminder for CE operators - check raw data for NAD samples to see whether it is due to XS or bad rox

- 1	d)		
Outcome	If this occurs you need to let ARM know at the time. Consider the need to work together.	Discussed 28/11/2011, not wearing booties and not using Trigene on the floors anymore. Look into using an appropriate floor cleaner. Just clean with ethanol this week.	Noted
Initials (of person making entry)	a very disappointed MMA :0(MMA	MMA
Date	24/11/2011	24/11/2011	28/11/2011
Suggested cause			
Barcode(s) affected (new cell for each barcod please)			
Issue Identified	Be mindful of other people on the floor when they are asking around for help. In the past, we have had FTEs call in sick and the lab has managed to complete this extra work for that missing FTE. Why is it that when we are looking for staff to help out on one simple process (e.g. put lysate on a maxwell), that it is impossible for the lab to decide to help out. Also, if you are doing other tasks other than what you are rostered on, the roster needs to be changed accordingly to reflect this. There are such columns in the roster like 'other work' which should be used when this happens.	Discuss booties - very slippery at the moment	As suggested before, may we please extend meeting time and booking of meeting room for an extra 1/2 hour so that we are able to discuss all issues. Today, we didn't have time to look at any issues logs which had agendas to discuss which may be considered important to staff.

Outcome	Yes	Noted.	Noted	Noted	Noted	Will use a generic log on. 12/12/2011 Will be done today by BUA 19/12/2011	Buffer x10 is received and we dilute to x1. All OK
Initials (of person making entry)	MMA	MJC	Z L	MMA	BUA	MLM	WIC
Date	30/11/2011	2/12/2011	2/12/2011	5/12/2011	6/12/2011	9/12/2011	16/12/2011
Suggested cause							
Barcode(s) affected (new cell for each barcod please)							
Issue Identified	Discuss electronic instrument logs. Will these continue?	Just a reminder to please ensure you leave your work areas clean after use. I came into the clean room this morning and there were used rediwipes on the bench and on the floor. Pipettes were scattered around rather than on the stand aswell. Also, has anyone noticed that the fume hood in the clean	A couple of boxes have been labelled and are going to have a place in the Orford in Pre-PCR sorting. These will be used for old DNA samples that have been pulled for CWTFR batches and so on. The other box will be for Analytical staff who have finished a batch and the sample needs to be restored in the freezer room.	Good idea to check the tables for the MPIIs daily if you are rostered on them. Both Extraction MPII are now due for their 3-monthly verifications.	I have reading the SOP for our anti-contaimination procedures (22857) and it clearly states that all PPE is to be discarded into the biohazard bin as soon as you leave the lab. There have been several labcoats hanging in the antichambers of late so this is just a reminder to please discard you labcoat EVERY time you leave the lab	There is not enough space on the maxwell's to have everyone's names in as a user. Is it better to have general logins, share logins or do people want to know how to change the user and people just change if their name isn't listed?	The laminated sign on each of the 3130 machines needs to be updated as it says Buffer (x1) - Shouldn't this say Buffer (x10)? If so, does anyone know where I can find the file to reprint it? Thanks

Outcome	Noted. Make sure lid is on bucket.	Noted	Noted
Initials (of person making entry)	MMA	SES	BM
Date	25/01/2012	9/02/2012	30/04/2012
Suggested cause			
Barcode(s) affected (new cell for each barcod please)			
Issue Identified	Hi all, in the clean reagent room the tips container fell spilling all its contents. While playing 3000 pick up, it brought some suggestions to mind: Can we please just use another bucket underneath like we do in manual ext's rather than balancing it on the stool? Can we fasten the lid of the bucket when we start to use a tip bucket so if it does fall in future, maybe its contents may be more contained, Can we maybe make a habit of re-screwing back the lids of mastermix vials before discarding so that there isn't any potential pcr reactions happening on our floors if this occurs in future, and perhaps discarding these as well as all other containers in the biohazard bin bags rather than using space in these tip containers which we seem to run out of fairly regularly? I found falcon tubes, 10ml tubes, 5 ml tubes and IQ elution and lysis buffer containers in this container using too much room. Thanks ;0)	Please keep a check on the bleach bottles in the labs for their dates. If see any out of date ones empty them and place them in the blue container in the antechamber for replacing with up to date bleach. Thanks	Remember to recycle. The purple plastic from the MPB 1ml robotic hanging tips & the racks that the maxwell cartridges sit in are recyclable. Keep finding them in the bins.

Outcome	Noted	Noted	noted	Noted- will review the numbers in this stocktake
Initials (of person making entry)	MMA	SE/SES	TJD/MLG	MM A
Date	28/05/2012	30/05/2012	21/08/2012	27/08/2012
Suggested cause				
Barcode(s) affected (new cell for each barcod please)				
Issue Identified	Please remember to lock fridge/freezers in CE so that the doors don't stay opened causing temps to fluctuate.	Should we have Spill Kits in the lab? SOP for Blood Clothing still mentions having a Spill Kit. (4ml &10ml blood tubes still received) Also put in Op.Issues Log	Please replace full bin bags. If you have to pull the sides of the bin bag up to fit in your waste PPE, you should be emptying the bin and replacing the bag.	Please count stock and do the stocktake list properly before bringing stock into the lab. There have been too many consumables brought into the lab which are not required. E.g. we don't need 12 packets of 12-channel plates (which equals to 100 plates in one cupboard - especially now that we are reusing them, and we need alot more than just the 10 Abgene plates). The next step would be to add a count of each consumable into the spreadsheet as part of stocktake as well as bringing back the second cupboard in auto ext. and not many want that to happen. I will be more than happy to change the stocktake list according to what is decided upon.

						once made e Noted.	Oyrex glass) can board to go into		
Outcome	Noted	Noted. Noted	Noted.	Noted.	Noted.	Store powder in freezer, once made up/aliquoted store in fridge Noted.	Noted - Glass bottles (except Pyrex glass) can go into the recycling bin. Cardboard to go into trolley.	Noted	
Initials (of person making	PA	GSL MMA	ВМ	ВМ	ВМ	TJD	Ō	MMA	
Date	7/09/2012	28/09/2012 28/11/2012	6/12/2012	25/02/2013	25/02/2013	5/03/2013	25/03/2013	12/04/2013	9 0 400 1
Suggested cause									
Barcode(s) affected (new cell for each barcod please)									
Issue Identified	Maxwell cartridges- make sure you check each cartridge before adding your lysate- some have been found with small cracks and the wash buffer has leaked out When mining Extraction MPII A please check the lysis	buffer has been added before sealing- off the lysate plate. Please ensure that we use ARTEL reagents in order of expiry	If you pour the Lysis Buffer from the bottle, you must wipe the lip of bottle with a kimwipe afterwards. Otherwise it goes crusty. Bad lab and WHS practice	Don't place items in any fridge up against the back wall of fridge. Condensation makes things soggy.	In CE, leave the spare (2) frozen plate holders upside down. That way when needed for use the wells are not full of ice.	The new brand of Pro K (Affymetrix USB) has come in and is stored at +4 unlike the old brand which was stored at -15. I have put it on the third shelf of the least used door of the twoway fridge for the meantime, a more permanent home may need to be set up.	The recycling bin in front of CE room is for tip boxes. It is not for cardboard or glass bottles. It was found Artel Rgt bottles.	The vials of Pro K (Affymetrix USB) that are recieved are to be stored at +4 and once prepared, the aliquots are to be stored at -20oC. This information has recently come from the supplier and manufacturer.	Just a reminder to take care when storing samples. There was an instance in Pre-PCR where a sample was miss

Outcome	Noted	Noted	Noted	Noted
Initials (of person making entry)	MMA	SES	WLM	GSL
Date	26/03/2014	4/04/2014	17/04/2014	26/05/2014
Suggested cause				
Barcode(s) affected (new cell for each barcod please)				
Issue Identified	Hi all, the operational staff have asked that when we have emptied any of the fluoro/black reagent boxes used for Quant/Amp reagents to clean them down and pass them through the two way fridge for the next lot of reagents that get received. And those boxes used for Pro K/DTT aliquotes that have been emptied, wipe those down and place back in the pile in the corner and place wiped sign back into reagents sign bag ready for the next lot.	On Friday's before putting reagents (mastermix and primer) back into freezer for end of week, please check with whoever is on punching, BSD, if they have finished with them for the day.	When doing GII's please regulary green out the cell and save when doing the pass status as you have told the operational team, this will avoid the operational team overwriting your pass status, an example of this is today a CE batch had been done by an analytical staff and 40 minutes had passed before the cell was green, yes the operational staff can check before completing the start status but workflow should be saved and updated regulary as the operational team has been told.	Just a gentle reminder to Auto Extraction operators to regulary check and ensure the discarded basket are aligned with the tip chute. It was noted this morning the dirty tips are scattered all around and have to wiped cleaned the floor board as well.

Outcome	Noted	Noted	Noted	Noted	Noted	Noted	Noted
Initials (of person making entry)	MMA	MMA	MMA	MLM	MLG	LBR	ō
Date	3/06/2014	3/06/2014	3/06/2014	10/06/2014	30/06/2014	14/07/2014	25/07/2014
Suggested cause							
Barcode(s) affected (new cell for each barcod please)							
Issue Identified	Hi all, we were experiencing problems with the running of PP21 matrix (lot X) with 3130xlB where 0 capillaries passed and an error would appear to state this although it had passed on 3130A. Plates were continuing to run ok, and P+matrix were passing 16/16. If this PP21 Matrix of 0 caps passing persists try using other PP21 Matrix kit. Or somebody mentioned trying to do a spatial? or array change. PP21 matrix Y then passed 14/16, but please monitor capillary 15 and 16 as the array had also been changed to one of the regenerated arrays. Thanks, Maria.	Arrays to be regenerated are to be stored in the interem away from the 3130xls. Found one that the water had evaporated from next to 3130xlA. I have started storing them above sink upright next to the pile of 3500 arrays until ready to send them off when we have enough to send together. Thanks Maria	Wander from Life Technologies came in to adjust the Z position for the autosampler to try to fix the 16 capillary NAD issue. Please advise him if still not working. Thanks, Maria	Sample tracking has been disabled on the Maxwells. The sample input screen will no longer appear. Gll uploads for CE - pass batches: Some people have been	using the genemapper batch ID instead of the CE batch which results in the "Pass" info not getting entered on the correct line. Please make sure you are entering the correct	Please do not hang jumpers in the lab ante chambers. Please leave them in an office area.	Auto Ext's: 175uL Conductive tips have been verified and passed for EXT MPII A, on 14/07/2014. (non cond tips have been ordered). Pos Ctls of Ext's batches run on Ext MPII A show expected quant values.

Issue Identified	Barcode(s) affected (new cell for each barcod please)	Suggested cause	Date	Initials (of person making entry)	Outcome
Please be careful when creating/moving shortcuts and/folders. The workflow diary folder had a lot of the older diaries disappear, they were found in folders in the I/:AAAoperational folder. A new workflow diary folder was then created in April in I drive (?). This is the second time in the last few months that this has happened. If you think you have moved/deleted something you shouldn't have, please send an email for it to be investigated.			28/05/2019	BM	Noted
FYI - CTRL-Z is undo For the staff who are rostered in stocktake, can they also aliquout Amp water for Pre PCR when you have time please.			5/06/2019	GSL	Noted (HP2s should be taking over stocktake again)
Please communicate to contracters when entering the lab about cleaning equipment they are going to use and if placing anything on a bench or floor to clean to use a blue cloth.			17/06/2019	LMF	Noted
Clean room balance bench- 2 ENVM in a row has large number of peaks. Pls clean regularly Pre-PCR Milli-Q - floodedagain. The screw-in plug on top			30/09/2019	BM	Noted
of unit was excessively tight and the unit flooded water all over the floor. The tool provided is only for UN-SCREWING the plug, not for tightening it. Only ever screw plug in to fingertight.			4/02/2020	AKD	Noted
Please remember to sign and date reagent bottles when newly opened and also write "In Use" on the bottle or lid.			9/11/2020	MLM	Noted
Plastic slide racks not to go through the washer anymore. Some rack were severly warped and not wrth risk of happening again. Please soak in decon bucket then dry on dish rack in the lab.			8/04/2021	MLG	Noted

	teep batches in be processed. If lab needs more eturn that batch the LAs. We will what is required.	.op ⊪	.ll do.
Outcome	Agreed in meeting - aim to keep batches in Ext Sorting until it is ready to be processed. If a batch which is in the main lab needs more samples added to it, we will return that batch to the Ext Sorting Freezer for the LAs. We will also add a Comment stating what is required. HPs will add a batch note if the HP has added extra samples.	Agreed and we will do.	Agreed and we will do.
Initials (of person making entry)	AKD/MLG	AKD/MLG	LBR
Date	7/09/2021	7/09/2021	
Suggested cause			
Barcode(s) affected (new cell for each barcod please)			
Issue Identified	Extraction sorting - adding samples after the batch has already been located by the LAs. When the batch is still in "Prep" and we want the LAs to add more samples we need a clearer process, e.g. return the rack to Ext Sorting and label. Also, if HPs add more samples, we need a batch note to update the sample "located" info. This has been causing confusion for the LAs.	Workflow diary comments - please ensure the comment is removed after the action has been completed.	Please remember to recycle cardboard boxes in the lab and not to but them in the biohazard bins

ù.	Issue identified	initials	date	Notified to whom
	Example only, noticed tube cracked when	AM	22/10/2008	AAP
	starting extraction	300000		
	Hair attached by barcode to outer surface of tube	AAP	4/11/2008	EJC
	Noticed tube cracked when performing			
	evidence recovery	TK	7/11/2008	JM
20	Noticed tube cracked when performing	TK	10/11/2008	AH
	evidence recovery	TIX.	10/11/2000	741
	Noticed tube cracked when performing evidence recovery	AH	11/11/2008	AAP
	Noticed tube cracked when performing	displacement.		
	evidence recovery	AH	13/11/2008	AAP
8	On submission CSSE window had a tear in it		8.00	i.
	and tube had popped open. Swab was still in	AH	13/11/2008	AAP
	tube.		Section and Co.	
**************************************	Noticed tube cracked when performing	AH	17/11/2008	AAP
	evidence recovery	АП	17/11/2006	AAP
	Noticed Tube lid open and swab no longer	TK	21/10/2008	AAP
3	inside tube when photographing CSSE	113	21/10/2000	7 6 11
	Noticed tube cracked when performing	AH	26/11/2008	AAP
	evidence recovery Noticed tube cracked when performing			
	evidence recovery	AH	26/11/2008	AAP
8	Noticed tube cracked when performing			102
	evidence recovery	AH	26/11/2008	AAP
	Noticed tube cracked when performing	AH	26/11/2008	AAP
	evidence recovery	АП	26/11/2006	AAF
	Noticed tube cracked when performing	AH	2/12/2008	AAP
	evidence recovery	5,25(1)		1.2.00
	Noticed tube cracked when performing evidence recovery	AH	5/12/2008	AAP
	Noticed tube cracked when performing			Same Color
	evidence recovery	AH	5/12/2008	AAP
70.	Lid of tube snapped off	AH	5/12/2008	AAP
	Noticed tube cracked when performing	AH	8/12/2008	AAP
	evidence recovery. Lid was also snapped off.	AH	0/12/2000	1
-	Noticed tube cracked when performing	AH	11/12/2008	AAP
10	evidence recovery. Noticed tube cracked when performing		7 V	
	evidence recovery.	AH	16/12/2008	AAP
7	Noticed tube cracked when performing			
	evidence recovery.	AH	16/12/2008	AAP
3	Noticed tube cracked when performing		47/40/0000	
	evidence recovery.	AH	17/12/2008	AAP
	Noticed tube cracked when performing	AH	22/12/2008	AAP
	evidence recovery.	14		
	Lid of tube snapped off	AH	22/12/2008	AAP
	Noticed tube cracked when performing evidence recovery.	AH	7/01/2009	ADA
9	Noticed tube cracked when performing	Total Control		NECTOR CO.
	evidence recovery.	AH	8/01/2009	ADA
	Noticed tube cracked when perfoming	ALL	0/04/0000	454
gic	evidence recovery.	AH	8/01/2009	ADA
	Noticed tube cracked when performing	AH	9/01/2009	ADA
	evidence recovery.		0.0112000	
	Noticed tube cracked when performing	AH	13/01/2009	ADA
	evidence recovery. Noticed tube cracked when performing		9. 2	
	evidence recovery.	AH	15/01/2009	ADA
	Lid of tube snapped off	AH	15/01/2009	ADA

Issue identified	initials	date	Notified to whom
Noticed tube cracked when performing	АН	15/01/2009	ADA
evidence recovery.			
Lid of tube snapped off	AH	15/01/2009	ADA
Noticed tube cracked when performing evidence recovery.	AH	15/01/2009	ADA
Lid of tube snapped off	AH	15/01/2009	ADA
Noticed tube cracked when performing			
evidence recovery.	AH	15/01/2009	ADA
Noticed tube cracked when performing	AH	15/01/2009	ADA
evidence recovery. Lid of tube snapped off	AH	15/01/2009	ADA
Noticed tube cracked when performing	АП	15/01/2009	
evidence recovery.	AH	27/01/2009	ADA
Noticed tube cracked when performing			1
evidence recovery.	AH	30/01/2009	ADA
Noticed tube cracked when performing	A11	0/00/0000	ADA
evidence recovery.	AH	2/02/2009	ADA
Lid of tube snapped off	AH	9/02/2009	AAP
Noticed tube cracked when performing	AH	10/02/2009	AAP
evidence recovery.	АП	10/02/2009	AAP
Noticed tube cracked when performing	AH	11/02/2009	AAP
evidence recovery.	АП	1 1/02/2009	AAP
Noticed tube cracked during extraction.	PA/AH	11/02/2009	AAP
Noticed tube cracked when perfoming	AH	12/02/2009	AAP
evidence recovery.	АП	12/02/2009	AAF
Noticed tube cracked when perfoming	AH	12/02/2009	AAP
evidence recovery. Noticed tube cracked during extraction.	PA/AH	12/02/2009	IAAP
Noticed tube cracked when performing	FAVALL	12/02/2009	AAF
evidence recovery.	AH	13/02/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	AH	16/02/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	AH	16/02/2009	AAP
Noticed tube cracked and lid snapped off when			
performing evidence recovery	AH	16/02/2009	AAP
Noticed tube cracked when performing			1
evidence recovery.	AH	17/02/2009	AAP
Noticed tube cracked when performing		40/00/0000	1
evidence recovery.	AH	18/02/2009	AAP
Lid of tube snapped off	AH	19/02/2009	AAP
Lid of tube snapped off	AH	23/02/2009	AAP
Noticed tube cracked when perfoming	АН	22/02/2000	AAP
evidence recovery.	АП	23/02/2009	AAP
Noticed tube cracked when performing	AH	24/02/2009	AAP
evidence recovery.	АП	24/02/2009	AAF
Noticed tube cracked when performing	AH	24/02/2009	AAP
evidence recovery.		24/02/2009	AAF
Lid of tube snapped off	AH	24/02/2009	AAP
Noticed tube cracked when performing	АН	24/02/2009	AAP
evidence recovery.			
Lid of tube snapped off	AH	24/02/2009	AAP
Noticed tube cracked when performing	АН	24/02/2009	AAP
evidence recovery			
Noticed tube cracked when performing	АН	24/02/2009	AAP
evidence recovery			
Noticed tube cracked when performing	АН	24/02/2009	AAP
evidence recovery.			
Noticed tube cracked under label when	\//.	04/00/0000	
performing extraction. Leaked under label but	VH	24/02/2009	AAP
inside of tube should be OK.	I		

Issue identified	initials	date	Notified to whom
Noticed long crack on down top half of QPS		_	
tube when performing extraction, transferred to	F.II		I _{AAP}
new labelled tube and processed as normal.			
·		24/02/2009	
Noticed tube cracked when performing	AH	05/00/0000	AAP
evidence recovery.	A 1 1	25/02/2009	
Lid of tube snapped off	AH	26/02/2009	
Lid of tube snapped off	AH	26/02/2009	IAAP
Noticed tube cracked when performing	AH	2/03/2009	AAD
evidence recovery. Noticed tube cracked when performing	All	2/03/2009	
evidence recovery.	AH	2/03/2009	ΔΔΡ
Noticed tube cracked when performing	All	2/03/2003	/ · · · · · · · · · · · · · · · · · ·
evidence recovery.	AH	5/03/2009	
Noticed tube cracked when performing	All	3/03/2003	/ · · · · · · · · · · · · · · · · · ·
evidence recovery.	AH	5/03/2009	
Noticed tube cracked when performing	7 (1 1	0/00/2003	7 0 11
evidence recovery.	AH	6/03/2009	
Noticed tube cracked when performing	7 (1 1	0/00/2003	7 0 11
evidence recovery.	AH	9/03/2009	
Noticed tube cracked when performing	7 (1 1	3/00/2003	7 0 11
evidence recovery.	AH	10/03/2009	
Lid of tube snapped off	AH	10/03/2009	
Noticed tube cracked when performing	AH	10/03/2003	/ · · · · · · · · · · · · · · · · · ·
evidence recovery.	AH	10/03/2009	
Noticed tube cracked when performing	All	10/03/2003	
evidence recovery.	AH	13/03/2009	
Noticed tube cracked when performing	All	13/03/2009	
evidence recovery.	AH	18/03/2009	
Lid of tube shattered into a few pieces.	AH	19/03/2009	
Noticed tube cracked when performing	AH	19/03/2009	
evidence recovery.	AH	21/03/2009	
Lid of tube snapped off	AH	21/03/2009	
Noticed tube cracked when performing	7 (1 1	21/00/2000	7 0 11
evidence recovery.	AH	21/03/2009	I _{AAP}
Noticed tube cracked when performing	7 (1 1	21/00/2003	
evidence recovery.	AH	21/03/2009	
Noticed tube cracked when performing	A11	21/03/2003	/\/\l
evidence recovery.	AH	21/03/2009	
Noticed tube cracked and lid snapped off when	7 (1 1	21/00/2000	7 0 11
performing evidence recovery.	AH	21/03/2009	I _{AAP}
Noticed tube cracked when performing	7 (1 1	21/00/2000	7 0 11
evidence recovery.	AH	25/03/2009	AAP
Noticed tube cracked and lid snapped off when		20/00/2000	
performing evidence recovery.	AH	25/03/2009	AAP
Noticed tube cracked and lid snapped off when	7 (1)	20/00/2000	7 0 11
performing evidence recovery.	AH	25/03/2009	I _{AAP}
Noticed tube cracked and lid snapped off when	7 (1)	20/00/2000	7 0 11
performing evidence recovery.	AH	25/03/2009	AAP
Noticed tube cracked when performing	,	20/00/2000	7 0 11
evidence recovery.	AH	25/03/2009	AAP
Noticed tube cracked and lid snapped off when	,	20/00/2000	7 0 11
performing evidence recovery.	AH	25/03/2009	I _{AAP}
Noticed tube cracked when performing		25, 30, 2000	p = #
evidence recovery.	AH	3/04/2009	IAAP
Noticed tube cracked when performing	, 11 1	3,34,2009	/ v 1
evidence recovery.	AH	3/04/2009	IAAP
Noticed tube cracked when performing	. 11 1	3/0-1/2009	
evidence recovery.	AH	6/04/2009	IAAP
Noticed tube cracked when performing	, 11 1	3/0-/2009	, v u
evidence recovery.	AH	6/04/2009	IAAP
OVIGORIOG FOUDVOLY.	, w i	3/0-7/2003	7 4 11

Issue identified	initials	date	Notified to whom
Noticed tube cracked and lid snapped off when			
performing evidence recovery.	AH	6/04/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	AH	6/04/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	AH	6/04/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	AH	6/04/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	AH	6/04/2009	AAP
Noticed tube cracked and lid snapped off when			
performing evidence recovery.	AH	6/04/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	AH	6/04/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	AH	7/04/2009	
Lid of tube snapped off	AH	7/04/2009	AAP
Lid of tube snapped off	AH	7/04/2009	AAP
Lid of tube snapped off	AH	9/04/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	AH	16/04/2009	AAP
Lid of tube snapped off	AH	16/04/2009	
Noticed tube cracked when performing			
evidence recovery.	АН	17/04/2009	AAP
Lid of tube snapped off	AH	17/04/2009	
Noticed tube cracked when performing	,	1170172000	7 0 11
evidence recovery.	AH	24/04/2009	AAP
Noticed tube cracked when performing	7 4 1	21/01/2000	7011
evidence recovery.	reh	30/04/2009	ΔΔΡ
Noticed tube cracked and lid snapped off when	1011	00/04/2000	7 0 11
performing evidence recovery.	reh	30/04/2009	ΔΔΡ
Noticed tube cracked and lid snapped off when	1011	00/04/2003	7 0 11
performing evidence recovery.	reh	6/05/2009	ΔΔΡ
Noticed tube cracked and lid snapped off when	IGII	0/03/2009	
performing evidence recovery.	reh	7/05/2009	ΛΛD
Noticed tube cracked when performing	IGII	110312009	
evidence recovery.	reh	7/05/2009	A A D
Noticed tube cracked when performing	ren	110312009	AAF
	RGM	8/05/2009	KD6
evidence recovery. Noticed tube cracked when performing	KGW	6/03/2009	KD3
· -	RGM	8/05/2009	NDG
evidence recovery.	RGIVI	6/05/2009	KD3
Cracked tube noted by PA in analytical and			
returned to Evidence Recovery for processing	RGM	8/05/2009	KDS
Cracked tube noted by PA in analytical and			
returned to Evidence Recovery for processing	RGM	8/05/2009	KDS
Cracked tube noted by PA in analytical and			
returned to Evidence Recovery for processing	RGM	8/05/2009	KDS
Cracked tube noted by PA in analytical and			
returned to Evidence Recovery for processing	RGM	8/05/2009	
Lid of tube snapped off	REH	13/05/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	REH	14/05/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	mb	15/05/2009	AAP
Noticed tube cracked when performing			
	IMD	18/05/2009	IAAP
evidence recovery	MB	10/03/2009	1
	INIR	18/03/2009	<u> </u>
evidence recovery	MB MB	18/05/2009	
evidence recovery noticed tube cracked when performing			

No.	Issue identified	initials	date	Notified to whom
	Noticed tube cracked when performing			
	evidence recovery	MB	19/05/2009	AAP
	Tube broken upon opening during evid			
	recovery process	МВ	19/05/2009	AAP
	Noticed tube cracked when performing	DELL	04 14	A A D
	evidence recovery.	REH	21-May	IAAP
	tube lid broke off upon opening for processing	mb	22/05/2009	AAP
	Noticed tube cracked when performing			
	evidence recovery.	REH	25/05/2009	AAP
	noticed crack tube during evid recovery		00/05/0000	
	process	MB	28/05/2009	IAAP
	tube lid broke off upon opening for processing	МВ	29/05/2009	AAP
	Noticed tube cracked when performing			
	evidence recovery	REH	2/06/2009	AAP
	Noticed crack near lid of tube when labelling tubes for Cell Ext batch CELEXT20090603_01,			
	swab transferred to new labelled 1.5mL tube			
	and processing continued.	EJL	4/02/2009	AAP
	noticed tube cracked during evid recovery			
	process	MB	5/06/2009	AAP
	Noticed tube cracked and lid snapped off when			
	performing evidence recovery.	reh	9/06/2009	AAP
	noticed small crack near lid during evid			
	recovery process	JSM	10/06/2009	AAP
	noticed small crack near lid during evid			l <u>-</u>
	recovery process	JSM	10/06/2009	AAP
	noticed large crack and lid snapped off during	1014	40/00/0000	
	evid recovery process	JSM	10/06/2009	IAAP
	noticed multiple small cracks near lid during	ICM	10/06/2000	
	evid recovery process noticed small crack near lid during evid	JSM	10/06/2009	AAP
	recovery process	JSM	11/06/2009	AAD
	noticed damaged lid during evid recovery	JOIVI	11/00/2009	
	process	JSM	11/06/2009	I _{AAP}
		CONT	11/00/2000	7 0 11
	tube lid broke off upon opening for processing	REH	15/06/2009	I _{AAP}
	noticed large crack in tube	JSM	2/07/2009	
	noticed crack in tube during evid recovery			
	processing	JSM	3/07/2009	AAP
	crack in tube and broken lid upon opening	mb	3/07/2009	AAP
	Noticed crack near lid of tube when labelling			
	tubes for batch CWIQEXM20090727_02, swab			
	transferred to new labelled 2mL tube and		00/07/0000	A A D
	processing continued.	EJL	28/07/2009	IAAP
	Noticed tube cracked when perfoming	A	42/00/2000	
	evidence recovery.	AH	13/08/2009	IAAP
	Noticed tube cracked when perfoming	AH	13/08/2009	
	evidence recovery. Noticed tube cracked when perfoming	АП	13/06/2009	AAP
		AH	13/08/2009	
	evidence recovery. Noticed tube cracked when perfoming		13/00/2009	
	evidence recovery.	AH	15/08/2009	
	Noticed tube cracked when perfoming	/ 11 1	13/00/2009	ורע <i>ו</i>
	evidence recovery.	AH	15/08/2009	
	Noticed tube cracked when perfoming	7 11 1	13/30/2009	/ V 11
	evidence recovery.	AH	24/08/2009	I _{AAP}
	Noticed tube cracked when perfoming		21,30,2000	,,
	evidence recovery.	AH	27/08/2009	 AAP
	1			· · · · ·

Issue identified	initials	date	Notified to whom
Noticed lid of tube snapped off when			
performing evidence recovery	AH	27/08/2009	AAP
Noticed tube cracked when perfoming			
evidence recovery.	RGM	27/08/2009	AAP
Noticed tube cracked when perfoming			
evidence recovery.	RGM	27/08/2009	AAP
Noticed tube cracked when perfoming			
evidence recovery.	KDS	27/08/2009	AAP
Noticed tube cracked when perfoming			
evidence recovery.	RGM	27/08/2009	AAP
Noticed lid of tube snapped off when			
performing evidence recovery	AH	28/08/2009	AAP
Noticed tube cracked when perfoming			
evidence recovery.	RGM	28/08/2009	AAP
Noticed tube cracked when perfoming			
evidence recovery.	KDS	28/08/2009	AAP
Noticed tube cracked when perfoming			
evidence recovery.	KDS	3/09/2009	AAP
Noticed tube cracked when performing			
evidence recovery.	JSM	3/09/2009	AAP
Noticed tube cracked when performing	-		
evidence recovery	JSM	4/09/2009	AAP
Noticed tube cracked when performing	-		
evidence recovery	МЈС	11/09/2009	AAP
noticed damaged lid when performing		1 1,700,1200	
evidence recovery	JSM	5/10/2009	AAP
noticed cracked tube when performing	-		
evidence recovery	IRGM	13/10/2009	МЈС
noticed cracked tube when performing		10/10/2000	
evidence recovery	RGM	13/10/2009	MJC
noticed cracked tube when performing	110	10/10/2000	
evidence recovery	REH	15/10/2009	AAP
tube lid broke off upon opening for proc		3/12/2009	
cracked tube	MB	4/12/2009	
damaged lid	JSM	9/12/2009	
damaged lid	JSM	20/01/2010	
Cracked tube	KDS	25/01/2010	
Cracked tube Cracked tube. Noticed at start of ODL	1,00	20/01/2010	
procedure.	ВМ	27/01/2010	ARM
damaged lid	JSM	2/02/2010	
noticed cracked tube when performing	JOIVI	2/02/2010	
evidence recovery	RGM	9/04/2010	ΔΔΡ
noticed cracked tube when performing	I (GIVI	3/04/2010	/ V \l
	RGM	11/06/2010	ΔΔΡ
evidence recovery noticed cracked tube and damaged lid v		11/00/2010	
•		22/07/2040	AAB
performing evidence recovery	JSM	22/07/2010	AAP
noticed cracked tube and damaged lid		10/04/0044	I A A D
performing evidence recovery	AKF	10/01/2011	JAAP

Problem Detected	Date Identified	Initials (of person making entry)	Barcode of sample where problem identified	Batch ID(s) or barcodes affected	Specimen notes & UR notes made (Y, N/A, In progress)	Batch audit entries made (Y, N, W/A, In progress)	Causality Identified or proposed	#100	Report written & released (Y, M, N/A, In/A, In draft)	Unknown Profiles added to staff Database?	STATUS
Profile in BSD Validation Performance Qualification Negative Control	19/05/2006	NL		BSD Validation Performance Qualification	Z	N/A	Unknown female profile in negative control that did not match staff or any profiles on NC DD. Unknown source of contamination.	14150	z	٨	
QC Dots Giving Poor Result	29/05/2006	AM,CI		N/A	NA	NIA	No further action was taken because the results were not low enough to be of significant concern, new extraction chemistries are to be investigated in the near future and the potential workload could not be completed under current staffing circumstances.	N/A	*	NA	
Default Run module used on 3100A instead of the settings created by Forensic Biology Staff	19/06/2006	АМ		All batches from the 17/05/2006 - 18/06/2006	z	NIA	Change occurred when the PC used for 3100A had been worked on by both ABI and CHSS-IT staff to rectify networking problems. Problem fixed immediately once identified and all affected samples were re-run.	14180	٨	N/A	
Wrong plate run on 3100 or incorrect naming of Run folders	27/06/2008	АМ		FTA#844 & FTA#842	NA	NIA	Ether incorrect prepping, incorrect linking on 3100 or incorrect naming of ouput files	14274	NA	N/A	
Wrong plate map imported	9002/90/2	PT		FTA#924 FTA#924R	Z	AIN	When FTA#924R was run on 3100, the FTA#924 plate map was wrongly imported.	14459	z	N/A	
DNA Master "cut and paste" error-wiping of sample information	4/07/2006	нм		Not Reported	>	NIA	Details for sample 259692096 were pasted over details for sample 196084629 in DNA Master. A new DNA# was given and the correct details re-entered from printed workbook	14512	Z	NIA	
Two extracted sample tubes with same DNA number-labelling error	18/07/2006	- IL		QF#859	z	NIA	Tube for extracted sample DNA#10621 labelled incorrectly as DNA#10216.	14604	z	NIA	
Wrong profile uploaded to DNA Master	21/07/2006	NO		CW#848 CW#916 Not Reported	zz	NIA NIA	Profile for 209308653 was uploaded incorrectly in DNA Master instead of the profile for 209308635- problem corrected as soon as identified.	14625	z	NIA	
Two different DNA numbers with the same sample number	31/07/2006	PT		CW#1377	Z	NIA	Operator error. DNA# AND the sample ID need to be corrected when fixing errors using the "rework macro"	14695	z	N/A	
Partial Profile in Negative Blood Extraction Control	31/07/2006	PT		CW#1366	z	NIA	Prepping error as re-prepared and re- amped samples both NSD. Staff notified. Plates released after investigation.	14696	z	N/A	
DNA Master "typing" error	2/08/2006	KR		Not Reported	Z	NIA	A "0" had been put in a cell where DNA#96829 was supposed to be.	14718	z	N/A	

Initials (of Barcode of sample Batch IDIs) or barcodes UR notes made (Y, person where problem affected N, NIA, In progress)		UR notes made (Y, N, N/A, In progress)		Batch audit entries made (Y, N, N/A, In progress)	Causality Identified or proposed	#100	Report written & released (Y, N, N/A, In draft)	Unknown Profiles added to staff Database?	STATUS
LR - Not Reported	Not Reported		>	NA NA B	Samples potentially affected could not be repeated so results for these samples were not reported.	15056	z	N/A	
AM CW#1589 CW#1598 REF#264 CW#1594	CW#1589 CW#1598 REF#264 CW#1594		z	N/A II	Profile originated Primer Mix tube P8 that was used to make the PCR master mix in each of the affected runs. How this tube came to be identified is unknown.	15189	⋇	٨	
AM Nucleospin Extraction- 28/09/2006	Nucleospin Extraction- 28/09/2006		٨	N/A III	Origin of profile is unknown. Each team leader was to assess each profile derived from the affected extraction batch on a case-by-case basis.	15285	*	٨	
AM Not Reported	Not Reported		N (extraction controls)	N/A III	Samples were tracked through to completion to ensure that results were consistent with what the correct labelling was assumed to be. For INIA#106375 the case scientist and team leader were notified as soon as discrepancy was picked up.	15289	z	N/A	
TM Not Reported	Not Reported		N/A	N/A F	Profile did not match either of the samplee tested for this coronial case. Possible profile match to DNA#79680 but unifliely due to circumstances. Unknown source of contamination.	15472	z	٨	
AM CW#1793 CW#1837 (rework)	CW#1793 CW#1837 (rework)		,	N/A 8	Partial profile found that did not match any staff members. CW#1793 plate withheld. Staff notfied and decisions on the profiles from this plate made on a case-by-case basis.	15928	*	>	
IM Not Reported	Not Reported		NA	N/A E	Mixed profile was found in one of three crushed bone samples for identification purposes. Unknown female contributor. Did not natch any staff or NC DD profiles. Mixed profile was not used in any interpretations.	15901	z	*	
MLH REF#308	REF#308		٨	γ γ	Contamination from an unkown source. All staff notified. Plate witheld until investigation complete.	16363	z	Z	
MP	N/A		NA	N/A	Mixture partial profile obtained from scanner 5 in volume crime area. Area decontaminated. No match to staff. Project officer has been working in area his profile was taken and he could not be excluded from the mixture. Procedure for Quality Practice will be updated.	16314	z	z	

Problem Detected	Date Identified	Initials (of person making entry)	Barcode of sample where problem identified	Batch ID(s) or barcodes affected	Specimen notes & UR notes made (Y, N, N/A, In progress)	Batch audit entries made (Y, N, WA, In progress)	Causality Identified or proposed	#100	Report written & released (Y, N, N/A, In draft)	Unknown Profiles added to staff Database?	STATUS
REF sample run on a CW plate	4/12/2007	АР		GEN9CW20071130_03	ż	i	Sample was correctly registered and allocated. Moved to CW batch allocated. Moved to CW batch allocation list by operator using AUSLAB "return" function- incorrect action. Staff member involved was notified.	18331	[®] N	N/A	
Staff profile in mixed CW samplie (sampling error)	5/12/2007	¥		GEN9CW20070917_01 GEN9CW20071024_02 GEN9CW20071003_01 GEN9CW20071115_01 GEN9CW2007112_02	۶	<i>≻</i>	Profile was a mixture of 3 contributors with one contributor identified as the staff member who sampled this case. Subsequent sampling and amplification produced a mixed profile with only 2 contributors. Staff contamination in first lot of sampling confirmed.	18341	z	N/A	
Samples stored in incorrect well position during the STORstar of a slicprep	14/12/2007	АМ		CWIQEXT20071211_03	۶	≻	Four samples affected, substrate retained from subsampling process prior to error. We staff to check all processing of samples and checks to be made when adding samples to the wells.	18465	z	N/A	
Slicprep base dropped out from collar and spin basket onto MPII B bench	10/01/2008	BG		CWIQEXT20071219_03	Å	. A	Possible splashing of lysis solution on the bench and into other samples. Staff reminded to always hold slicprep by the base. Continuation of extraction. Case managet to assess usability of each sample.	18580	Z	N/A	Complete - OQI
Rework Proille did not match Original Profile	21/01/2008	Ω		GEN9REF20071031_01 Not Reported Not Reported	۶	*	Only the last 4 digits were checked so sample 308866053 was accidentally repunched instead of sample 325106053. Manual punching has since been superseded and incorrectly sampled specimens will be highlighted fitnough unsuccessful batch creation in AUSI.AB.	18645	z	N/A	Complete - OCI
Profile located on Major Crime Fume Hood Window Handle	22/01/2008	S		VIN	N/A	N/A s	Profile cannot be identified-unknown source. Cleaning procedures to include these areas. All staff notified.	18650	z	*	Closed
Unknown Minor Male Profile- Profile inconsistencies for same sample. Manual Amp mixing	30/01/2008	H		CW#3 CW#24 CW#116	٨	, Y	Mix-up of DNA#'s during manual amplification process in 2004 - mot likely one sample amplified incorrectly with the other	18695	*	N/A	Complete - OQI
Mixed DNA Profile in Reference Sample	11/02/2008	AM		GEN9REF20080128_02 GEN9REF20080211_05 Not Reported	*	×	Mixed profile identified in single source reference sample after extraction process- no mixture on original run. Has occurred between the extraction and amplification process but exact point of contamination is unknown. Aerosol transfer unlikely considering concentration of contaminant.	19330	*	N/A	Complete - OGI

					Г	
STATUS						
Unknown Profiles added to staff Database?						
Report written & released (Y, N, N/A, In draft)						
# 100	20690	21016	20925 & 21050	21062		
Causality Identified or proposed						
Batch audit entries made (Y, N, N/A, In progress)						
Specimen notes & L. UR notes made (Y, N. M.A, In progress)						
Batch ID(s) or barcodes affected					CDNAEXT20170829-08	
Barcode of sample where problem identified						
Initials (of person making entry)					АКР	
Date Identified					30/08/2017	
Problem Detected					Sperm fraction samples were loaded straight onto Maxwell A directly after addition of Lysis Buffer/DTT instead of a 45 minuter foom temperature incubation prior to loading onto Maxwell. Maxwell cartridges were run a second time on Maxwell cardinges, were run as econd time on Maxwell cardinges, plungers and elution tubes.	

			4	Correcti	Corrective actions	Control & Stand	Control & Standard Lot numbers
Ratch ID	Date problem	Reason for failure	instrument IIsad if		campies returned /		
Batch ID	Identified	Reason for failure	nsed II	Batch audit	returned /		
			applicable		reworks	Controls	Standards
		Pro Med and Pro High is out	7000			J4 20070227	
		Pro Med and Slope out	7000	Yes	Yes	22549001 J19 MLH20070612 01	
		R2 value is out	7000	Yes	Yes	22549001 J17 MLH20070612 01	
		Slope and Pro High is out	0002	Yes	Yes	22549001 J17 MLH20070612 01	
		Slope, Pro Low and Pro Med is out	7000	Yes	Yes	22549001 J19 MLH20070612 01	
		Pro High is out	7000	Yes	Yes	22549001 J17 20070726AC 01	G10 MLH20070801 01 21927901
		Pro Med and Pro Low are out	7000	Yes	Yes	22549001 J12 20070717MA 01	G9 20070717MA 01 21927901
		Pro Low is out	7000	Yes	Yes	22549001 J12 20070717MA 01	G9 20070717MA 01 21927901
		Pro High is out	7000	Yes	Yes	22549001 J17 20070726AC 01	G10 MLH20070801 01 21927901
		Pro High out of accepatable ranges	7000	Yes	Yes	22549001 J16 20070726AC 01	G12 20070726LWC 01 21927901
		Pro High and Pro Low are out	7000	Yes	Yes	22549001 J16_20070726AC_02	G12_20070726LWC_01 21927902
		Pro High and R2 value are out	2000	Yes	Yes	22549001 J17 20070726AC 01	G10 MLH20070801 01 21927901
		Pro High value out of range	7000	Yes	Yes	23274801 L4 20070817AC 01	H5 20070816AC 01 21927902
		Pro High, Med & Low outside 3SD	7000	Yes	Yes	23274801 L1 20070817AC 01	H3 20070813AC 01 21927902
		Pro High & Med outside 3SD, low		:	;		
		between 2-3SD	7000	Yes	Yes	23274801 L4 20070817AC 01	H5 20070816AC 01 21927902
		ProHigh and Med within 2SD however					
		following quant ProHigh was outside 3SD					
		and ProMed still out	7000	Yes	Yes	23274801 L2_20070919TKM/MLH_01	H8_20070919TKM/MLH_01
		ProHigh outside 3SD and ProMed outside					
		1SD	7000	Yes	Yes	23274801 L2 20070919TKKM/MLH 01H8 20070919TKM/MLH 01	H8 20070919TKM/MLH 01
		Lamp Failure	0002	Yes	Yes	23274801 L2_20070918TKM/MLH_01	_20070918TKM/MLH_01 21927902 H7_20070919TKM/MLH_01
		ProHigh and ProMed outside 2STD and					
		following quant Prohigh and ProMed also					
		failed	7000	Yes	Yes	21280602 K12_20071204KAL/MLH_01 21927902 J3_20071204KAL/MLH_01	21927902 J3_20071204KAL/MLH_01
		ProHigh and ProMed outside 2STD and					
		following quant new standard and control					
		combination used	7000	Yes	Yes	21280602 K12 20071204KAL/MLH 01 21927902 J3 20071204KAL/MLH 01	21927902 J3 20071204KAL/MLH 01
		Thermocycler failure		>	>		
		Thermocycler failure		>	>		
		Thermocycler failure		>	>		
		Thermocycler failure		Υ	Υ		
		Positive control failure		Υ	Υ		
		Neg control with quant value		Υ	N/A		
		Neg control with quant value		Т	N/A		
		ProMed outside 3STD	0002	Yes	Yes	21927902 J7 20071204KAL/MLH 01	23274801 N1 20071204KAL/MLH 01
		ProMed outside 3STD	2000	Yes	Yes		23274801 N1_20071204KAL/MLH_01
		ProHigh Undet.	2200	Yes	Yes	21927902 l4 20080205FC/MLH 01	23274802 M2 20080205FC/MLH 01
		Positive control failed	N/A	Yes			
		ProHigh and ProMed outside 2SD	7500	Yes	Yes	21927901 J6_20080417BUA/MLH_01	23274802 M6_20080314FC/MLH_01
		Positive controls NSD	3130xlA	Yes	Yes		
		Pos Ctl Failure		Yes	N/A		
	02.06.2008	Pos Ctl Failure		Yes	N/A		

Patch audit returned / Pes					Correcti	ive actions	Control & Stanc	Control & Standard Lot numbers
1007/2008 Pos Cit Failure		Doto problem		Instrument		Samples	3 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5 5	
Pos Cil Failure Yes N/A CORTORS Pos Cil Failure Pos Cil Failure N/A M7 20080506MLH/BM 01 23274802 E Pos Cil Failure Pos Cil Failure N/A M/A M7 20080506MLH/BM 01 23274802 E Pos Cil Failure E Yes N/A N/A P Pos Cil Failure F Yes N/A P Z0080617AC 01 25090901 P Pos Cil Failure F Yes N/A N/A P Z0080617AC 01 25090901 P Pos Cil Failure Yes N/A N/A N/A P Z0080617AC 01 25090901 P Pos Cil Failure Yes N/A N/A N/A P Z0080617AC 01 25090901 P P Pos Cil Failure Yes N/A N/A N/A P Z0080617AC 01 25090901 P Z0080617AC 01 25090901 P Z0080617AC 01 25090901 P Z0080617AC 01 25090901 P Z0080617AC 01 25090901 P Z0080617AC 01 25	Batch ID	Identified	Reason for failure	used if applicable	Batch audit	returned / reworks		č
Pos Cil Fallure Yes N/A Pos Cil Fallure Pos Cil Fallure Yes N/A Pos Cil Fallure Pos Cil Fallure Yes N/A Pos Cil Fallure Yes N/A N/A Pos Cil Fallure Yes Yes N/A Pos Cil Fallure Yes N/A Pos Cil Fallure Pos Cil Fallure Yes						hordoro	Controls	Standards
Pos Cit Failure Pos Cit Failure N/A Proficity and defunded outside 3SD 7000 Yes N/A Profit plant of Profight and Profided outside 3SD 7000 Yes N/A Profit plant of Profight and Profigh		02.06.2008	Pos Ctl Failure		Yes	N/A		
Port-light and ProMed outside SSD 7000 Yes Yes W7 20080506MLHBM 013274802 IS		02.06.2008	Pos Ctl Failure		Yes	N/A		
000 Pos Ci Failure Ves N/A 000 Pos Ci Failure E Yes Yes Pos 000 Pos Ci Failure F Yes N/A Pos Pos 000 Pos Ci Failure N/A N/A Pos		90.90.90	ProHigh and ProMed outside 3SD	0002	Yes	Yes	20080506MLH/BM 01	I5 20080507MLH/BM 01 21927901
89 Pos CIT Failure Figs NA NA 008 Slope and Por High is out 7500 Yes Yes NA 008 Pos CIT Failure Yes NA PR 20080617AC 012509001 PZ 20080617AC 012 008 Pos CIT Failure Yes NA NA PR 20080617AC 012 PZ 20080617AC		17.06.2008	Pos Ctl Failure		Yes	A/N		
Residence of the control failure Free Cit Failure Vies Vies Vies PR 20080617AC 0125090901 PP 20080617AC 0125090901<		17.06.2008	Pos Ctl Failure		Yes	N/A		
Page 11 Page 11 Page 11 Page 12 Page 12 Page 12		0007,007,10	Poo Off Policie	L	25			
Stope and Pot High is out 7500 Yes N/A P2 20080617AC 01 25080617AC 01 2 20080617AC 0		25/06/2008	Pos Cti Fallure	Ц	Yes	Yes		
OB Pos CI Failure Yes N/A 0B Pos CI Failure Yes Yes No CI Failure Yes N/A Pos CI Failure Pos CI Failure Yes N/A Pos CI Failure Pos CI Failure Yes N/A Pos CI Failure Pos CI Failure Yes N/A Pos CI Failure Pos CI Failure Yes N/A Pos CI Failure Pos CI Failure Yes		10/07/2008	Slope and Pro High is out	7500	Yes	Yes	P2 20080617AC 01 25090901	P2 20080617AC 01 25206001
08 Pos CI Failure Yes N/A NA 08 Pos CI Failure Yes N/A Pos CI Failure Yes N/A 08 Pos CI Failure Yes N/A Pos CI Failure Yes N/A 09 Pos CI Failure Yes N/A N/A Pos CI Failure Yes N/A 09 Pos CI Failure Yes N/A Yes N/A Pos CI Failure Yes N/A 09 Pos CI Failure Yes N/A Yes N/A Pos CI Failure Yes N/A 09 Pos CI Failure Yes N/A Yes N/A Pos CI Failure Yes N/A Pos CI Failure Yes N/A Yes N/A Pos CI Failure Yes N/A Pos CI Failure Yes N/A Yes N/A Pos CI Failure Yes N/A Pos CI Failure Yes N/A Yes N/A Pos CI Failure Yes N/A		11.07.2008	Pos Ctl Failure		Yes	A/N		
08 Pos Cil Failure Yes N/A 08 Pos Cil Failure Yes N/A 09 ProMed outside SSD, slope > -3.3 7500 Yes N/A 09 ProMed outside SSD, slope > -3.3 7500 Yes N/A 09 ProMed outside SSD, slope > -3.3 7500 Yes N/A 00 Prox Cil Failure Yes N/A Yes N/A 00 Prox Cil Failure Yes N/A Yes N/A 00 Prox Cil Failure Yes N/A Yes N/A 00 Prox Cil Failure Yes		11.07.2008	Pos Ctl Failure		Yes	A/N		
98 Pos Cif Failure Yes N/A 98 Pos Cif Failure Yes N/A 99 Pos Cif Failure Yes N/A 90 Cot Failure Yes N/A </td <td></td> <td>11.07.2008</td> <td>Pos Ctl Failure</td> <td></td> <td>Yes</td> <td>A/N</td> <td></td> <td></td>		11.07.2008	Pos Ctl Failure		Yes	A/N		
08 Pos Cit Failure Vés N/A 08 Pos Cit Failure Vés N/A 08 Pos Cit Failure Vés N/A 09 Pos Cit Failure Vés N/A 00 Pos Cit Failure Vés N/A		11 07 2008	Pos Ctl Failure		Yes	A/N		
06 Pos Cit Failure Yes N/A 08 Pos Cit Failure Yes N/A 09 Cit Failure Yes N/A 09 Cit Failure Yes N/A 09 Cit Failure Yes N/A 09 Cit Failure Yes N/A 09 Cit Failure Yes N/A 09 Cit Failure Yes N/A 09 Cit Failure Yes N/A <td< td=""><td></td><td>11 07 2008</td><td>Pos Ctl Failure</td><td></td><td>Yes</td><td>A/N</td><td></td><td></td></td<>		11 07 2008	Pos Ctl Failure		Yes	A/N		
96 Pos CI Failure Ves N/A 08 Pos CI Failure Yes N/A 09 Pos CI Failure Yes Yes 00 Pos CI Failure Yes N/A		11.07.2000	Dos Ott Epilina		22 >			
08 Pos Cit Failure Yes N/A 08 Pos Cit Failure Yes N/A 09 Pos Cit Failure Yes N/A 09 Pos Cit Failure Yes N/A 09 Pos Cit Failure Yes Yes 09 Pos Cit Failure Yes Yes 00 Nogative control failure Yes N/A Pos Cit Failure Yes N/A Pos Cit Failure Yes N/A Pos Cit Failure Yes N/A Pos Cit Failure Yes N/A Pos Cit Failure Yes N/A Pos Cit Failure Yes N/A Pos Cit Failure Yes N/A Pos Cit Failure Yes N/A Pos Cit Failure Yes N/A		11.07.2000	Poo Off Epiling		50 /			
08 Pos CII Failure Yes N/A 08 Pos CII Failure Yes N/A 09 Pos CII Failure Yes N/A 09 Pos CII Failure Yes Yes 09 Pos CII Failure Yes Yes 09 Pos CII Failure Yes N/A 09 Pos CII Failure Yes N/A 09 CII Failure Yes N/A 09 CII Failure Yes N/A 09 CII Failure Yes N/A 09 CII Failure Yes N/A 09 CII Failure Yes N/A 09 CII Failure Yes N/A 09 CII Failure Yes N/A 09 CII Failure Yes N/A 09 CII Failure Yes N/A 09 CII Failure Yes N/A 09 CII Failure Yes N/A 09		11.07.2008	Pos Cu Fallure		Yes	N/A		
08 Pos CII Failure Yes N/A 08 Pos CII Failure Yes N/A 08 Pos CII Failure Yes N/A 09 Pro CII Failure Yes Yes 09 Pro Med outside 3SD 7000 Yes Yes 09 No Exhibit outside 3SD 7000 Yes N/A 00 No Exhibit outside 3SD Yes N/A 00 No Exhibit outside 3SD Yes N/A 00 No Exhibit outside 3SD Yes N/A 00 Yes N/A N/A 00 Yes N/A 00		11.07.2008	Pos Ctl Failure		Yes	N/A		
08 Pos CI Failure Yes N/A PRA 08 Pos CI Failure Yes Yes Yes P.2 2008 077 6MLH 01 2520 6001 O3 2008 077 6MLH 01 08 Prohigh outside 3SD 7500 Yes Yes P.2 2008 0677 AC 01 250 909 01 P.2 2008 077 AC 01 250 909 01 P.2 2008 077 AC 01 250 909 01 P.2 2008 077 AC 01 250 909 01 P.2 2008 077 AC 01 250 909 01 P.2 2008 077 AC 01 250 909 01 P.2 2008 077 AC 01 250 909 01 P.2 2008 077 AC 01 250 909 01 P.2 2008 077 AC 01 250 909 01 P.2 2008 077 AC 01 25		11.07.2008	Pos Ctl Failure		Yes	A/N		
08 Pos CH Failure Ves N/A POS 20080716MLH 01 08 ProMide outside 3SD, slope > -3.3 7500 Yes Pes PS 20080617AC_012 08 ProMed outside 3SD, slope > -3.3 7500 Yes PS 20080617AC_012509001 PS 20080617AC_012509001 08 Negative control failure Yes N/A PS 20080617AC_012509001 PS 20080617AC_012509001 09 Cut Failure Yes N/A PS 20080617AC_012509001 PS 20080617AC_012509001 Pos CH Failure Yes N/A N/A PS 20080617AC_012509001 PS 20080617AC_012509001 Pos CH Failure Yes N/A N/A PS 20080617AC_012509001 PS 20080617AC_012509001 Pos CH Failure Yes N/A N/A PS 20080617AC_012509001 PS 20080617A		11.07.2008	Pos Ctl Failure		Yes	N/A		
08 ProHigh outside 3SD, slope > -3.3 7500 Yes Yes Pos 20080716MLH 01 25206001 O3 20080716MLH 01 2500601 08 ProMade outside 3SD 7000 Yes N/A Pos 20080617AC, 01 25090901 P2 20080617AC, 01 25090901		11.07.2008	Pos Ctl Failure		Yes	N/A		
08 ProMed outside 3SD 7000 Yes PP_2_20080617AC_01250901 PP_2_20080617AC_01250901 08 Negative control failure Yes N/A PP_2_20080617AC_01250901 PP_2_20080617AC_01250901 Pos CIT Failure Yes N/A N/A PP_2_20080617AC_01250901 PP_2_20080617AC_012		05.08.2008	ProHigh outside 3SD, slope > -3.3	7500	Yes	Yes		
08 Negative control failure Yes N/A Pos CI Failure Yes <td></td> <td>11.08.2008</td> <td>ProMed outside 3SD</td> <td>2000</td> <td>Yes</td> <td>Yes</td> <td></td> <td>20080617AC 01 2</td>		11.08.2008	ProMed outside 3SD	2000	Yes	Yes		20080617AC 01 2
Pos Ct Failure Yes Pos Ct Failure Yes		15/08/2008	Negative control failure					
Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ctl Failure		Yes	A/N		
Pos Cut Failure		20.08.08	Pos Off Failure		ν ν ν	V/N		
Pos Cut Failure Pos Cut Fa		20.00.00	Dos Ott Failure		202	X < \		
Pos Ct Failure Pos		20.00.00	Doo Off Follows		50.			
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Cti Fallure		Yes	N/A		
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ct Failure		Yes	N/A		
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ctl Failure		Yes	N/A		
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ctl Failure		Yes	N/A		
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ctl Failure		Yes	N/A		
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ctl Failure		Yes	N/A		
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ctl Failure		Yes	N/A		
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ctl Failure		Yes	N/A		
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ctl Failure		Yes	N/A		
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ctl Failure		Yes	N/A		
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ctl Failure		Yes	N/A		
Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes Pos Ct Failure Yes		20.08.08	Pos Ctl Failure		Yes	N/A		
Pos Ctl Failure Yes Pos Ctl Failure Yes Pos Ctl Failure Yes Pos Ctl Failure Yes Pos Ctl Failure Yes Pos Ctl Failure Yes		20.08.08	Pos Ctl Failure		Yes	A/N		
Pos Ctl Failure Yes Pos Ctl Failure Yes Pos Ctl Failure Yes Pos Ctl Failure Yes Pos Ctl Failure Yes		25.08.08	Pos Ctl Failure		Yes	A/N		
Pos Ctl Failure Yes Pos Ctl Failure Yes Pos Ctl Failure Yes Pos Ctl Failure Yes		25.08.08	Pos Ctl Failure		Yes	A/N		
Pos Ctl Failure Yes Pos Ctl Failure Yes Pos Ctl Failure Yes		25.08.08	Pos Ctl Failure		Yes	A/N		
Pos Ct Failure Yes Pos Ct Failure Yes		25.08.08	Pos Ctl Failure		Yes	N/A		
Pos Ctl Failure Yes		28.08.08	Pos Ctl Failure		Yes	N/A		
		03.09.08	Pos Ctl Failure		Yes	N/A		

Batch ID Date problem Reason for failure Testiment Testi					Correct	ive actions	Control & Standard Lot numbers	ard Lot numbers
Pos CII - PP	Batch ID	Date problem	Reason for failure	Instrument used if	Rotch oudit	campies returned /		
Pos Ct1 - PP Yes Epi Pos Ct1 - NSD Yes Epi Pos Ct1 - PP Yes POS - PP YES POS - PP YES POS - CARRY OVER YES Epi Pos Ct1 - PP Yes Pos Ct1 - PP Yes Pos Ct1 - PP Yes Pos Ct1 - PP Yes Pos Ct1 - PP Yes Pos Ct1 - PP Yes Pos Ct1 - PP Yes Pos Ct1 - PP Yes Pos Ct1 - PP Yes Pos Ct1 - RD Yes Pos Ct1 - NSD Yes Pos Ct1 - MSD Yes Pos Ct1 - Amel + 1/18 alleles Yes Pos Ct1 - amel + 1/18 alleles & Carry over Yes Pos Ct1 - amel + 1/18 alleles & Carry over Yes Pos Ct1 - amel + 1/18 alleles & Carry over Yes Pos Ct1 - amel + 1/18 alleles & Carry over Yes Pos Ct1 - amel + 1/18 alleles & Carry over Yes Pos Ct1 - amel + 1/18 alleles & Carry over Yes Epi Pos Ct1 - carry-over of fractions Yes				applicable	Datcil addit	reworks	Controls	Standards
Epi Pos Ctl - NSD Yes Epi Pos Ctl - PP Yes POS - PP YES POS - PP YES POS - PP YES POS - CARRY OVER YES Epi Pos Ctl - PP Yes Pos Ctl - PP Yes Pos Ctl - PP Yes Carry over Fraction in E lysate Pos Yes Pos Ctl - PP Yes Pos Ctl - PP 6/18 alleles Yes Pos Ctl - RSD Yes Pos Ctl - ANSD Yes Std curve failed, ?bubbles in tubing 7500A Pos Ctl - amel + 1/18 alleles Yes Pos Ctl - amel + 1/18 alleles Yes Pos Ctl - amel + 1/18 alleles & Carry over fraction in E lysate Pos Ctl Yes Pos Ctl - amel + 1/18 alleles & Carry over fraction in E lysate Pos Ctl Yes Pos Ctl - amel + 1/18 alleles & Carry over fraction in E lysate Pos Ctl Yes Pos Ctl - amel + 1/18 alleles & Carry over fractions Yes Pos Ctl - amel + 1/18 alleles & Carry over of fractions Yes Pos Ctl - amel + 1/18 alleles & Carry over of fractions Yes Pos Ct		13/11/2009	Pos Ctl - PP		Yes	N/A		
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		21/05/2010	Pos Ctrl - NSD		Yes	N/A		

Area / Process of Concern	Examples	Potential cause(s)	Priority	Possible Solutions	Outcome of investigation	Problem closed
DNA extracts not equal to 100µL after extraction with DNA IQ on MPII		Beads blocking tips on MPII during elution stage	2	await improvements resulting from AUDIT 8227		Closed - see Audit 8227
Cofiler amps not consistent (failing on initial amp when amped at same volume as successful Profiler)		AUSLAB programming fault (for COF L and 7COF LR amp plate map refers to source as Dilution plate when there is no dilution, CAMP1C etc. work OK) Operators not noticing problem when checking plate maps	1	Fix programming in AUSLAB	SCR submitted by TEN email sent to all analytical staff and also raised in team meeting	awaiting software fix software fixed
Re-amps not consistent		Bubble in dilution plate has been noted	3	review MPII pipetting into dilution plate	PE to review pipetting Process is being audited - additionally PE expert has reviewed pipetting and made some suggestions that will be tested	
Potential Sample to Sample contamination somewhere in extraction process		?pipetting on MPII durng either lysis or elution	1	review MPII pipetting into dilution plate await improvements resulting from AUDIT 8227 - PE has made modifications to DNA IQ extraciton protocol	some pipetting modifications made to elution step	Closed - see Audit 8227
Microcons with low final volume not amping correctly		Volume too low in tube for repeateable pipetting possibly for very low volumes, after centrifuging a small bubble of sample may sit to the side of the tube	3	make volume up to 20µL with TE at end of microcon procedure Need to get PE to review pipetting ?possible to get to bottom of tube better	Initial discussions at analytical team meeting did not favour this approach - to be discussed further Improvement implemented 14/7/08 testing has shown that some liquid may remain if 20µL only in the tube (testing with pos amp control) but this did adversely affect final result, however manual amping of microcon samples with ≤20µL will be progressed from 18/8/08	to 20 (if less than 20) and amp at 20 on
		Magnet		Magnet - investigated magnabot with posts, and magnabot with flat tops New Ambion magnet tested OK - will be implemented alongside findings from Audit 8227	With magnet plate with posts - beads are not being held in place with flat top magnet, plate moves around too much	Closed - Ambion magnet utilised - see also Audit 8227
Binding and / or release may be affecting general extraction efficiency		Plate	2	Looked into Greiner 1.1mL round bottom plate Appears to not be an issue with Ambion magnet	Size not really compatible (would have to do Lysis in 300µL), with "post" magnet beads slide off magnet	Closed
		Heater adapter tile Mixing on DPC shaker	-	Insufficient mixing on DPC shaker identified as cause of decreased extraction efficiency		Closed Closed - Mixmate to be used during re- verification - see also audit 8227
DNA profiles have been obtained from unknown sources		Plastic-ware Reagents	3	DNA free spin baskets have been sourced	Axygen spin baskets and tubes sourced that are DNA free	Closed
Problems with Nuc Cleanups	see issues register	?ethanol used ?buffers present in extraction not present in cleanup	1	investigate various variables	currently underway	

1st routine automated DNA IQ extraction performed	2000	10-AON	od Loo		Mar-US	Apr-08	May-08	20-un-		000	20-00		Nov-08
פאו מכווכוו בי	29/10/2007												
BSD duet used to punch FTA saples directly into SlicPrep			4)	5/02/2008									
Start use of Off-deck Lysis													
procedure (cease use of SircPrep except for FTA samples)					19/03/2008								
Replacement of SDS with Sarcosyl in Extraction buffer (Off-deck lysis						22/04/2008							
procedure) Start traibleshooting of automated													
DAN DE extraction (use instrument								•	14/07/2008				
A for KEF only, instrument B for CW in checkerboard pattern)													
Memo sent to all DNA Analysis Staff outlining DNA IQ issues								_	15/07/2008				
Audit 8227 (audit of automated								15-28/07/2008					
UNA IQ extractions) Use of both instruments in									0000/20/60				
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for contamination) Managing Scientist apprised													
executive (through Director FSS)									29/07/2008	2	22/09/2008		
of automated DNA IQ issues DNA Analvsis departmental													
meeting held to discuss automated								.,	29/07/2008				
Managing Scientist apprised OPS											,		
of automated DNA IQ issues									31/07/2008		31/	31/10/2008	
All results derived from automated DNA IQ extractions put on hold										-	10/09/2008		
Contamination events identified													
OQ #19330 (RFIQEXT20080205_03)			pa r	batch processed 06-02-08	0QI 21	OQI 21-04-08							
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OQ #19768 (CWIQEXT20080506_01)							batch processed OQI 14-07-08 07-05-08	114-07-08					

	OQI 08-08-08	OQI 20-08-08	OQI 21-08-08	OQI 04-09-08	80-03-03-08	OQI 15-09-08	OQI 06-10-08	OQI 28-10-08	OQI 06-11-08
OQI 24-07-08	-	batch processed 26-05-08	batch processed 11-07-08		batch processed 18-06-08	batch processed 01-07-08	1	patch processed 23-06-08	processed 04-06-08
batch processed 21-04-08	batch processed 04-04-08			batch processed 16-04-08			batch processed 09-04-2008		
OQ #20231 (CWIQEXT20080417_01)	OQ #20351 (CWIQEXT20080402_01)	OQ #20422 (CWIQEXT20080506_02)	OQ #20437 (CWIQEXT20080630_01)	OQ #20615 (CWIQEXT20080409_01)	OQ #20617 (CWIQEXT20080614_01)	OQ #20690 (CWIQEXT20080628_01)	OQ #20925 & CW#21050 (CWIQEXT20080403_01)	OQ #21222 (CWIQEXT20080620_02)	OQ #21309 (CWIQEXT20080531_01)



	A					
PAC	PAC	sspu		PAC	PAC	PAC
31/1/08 - Training Portfolio Audits – are continuing, PAC to arrange training sessions for staff this year.	31/1/08 - Ongoing FACT papers for SOCOs – PAC to follow this up. QHFSS will be guided by outcome of MOU with QPS. The FACT sheets should be forwarded through SSDU, to the QPS ethics unit and for dissemination through QPS. FACT sheets to detail information pertaining to eg. blood on a rock, how to best take sample from this surface to help in receiving a profile when tested. QHFSS could gather stats on what sampling technique is best to use when collecting swabs from different surfaces such as rock, cement etc.	20/12/07 - On track: Training Module Updates – Alice currently on leave, all outstanding modules are now in draft. Please check your QIS events. 10/4/08 – There are less than 13 which are 3 months past review date.	23/11/07 – Complete: Batch functionality - Current draft does not include Questions suggestions, PAC to follow this up with Alice / Iman / Allan.	11/10/07 - On track: SSDU to reformat Issues Register. PAC will advise where it will be relocated to.	23/11/07 – Training Module for Reporting Scientists – overview of Analytical section – now with SSDU. 10/4/08 – Has been completed by JAH and PT and is with SSDU.	G:\ForBio\\Quality \text{Assurance and NATA\\Management Reports - Biology \text{Management Team\\Quality \\ Update 100408.ppt} \\ \text{10/4/08} - \text{New OQI's. #19196} - to be completed by May 2008.\\ \text{Audit findings} - #8134 \text{ Digital Image Working Party VKI has names of volunteers.}\\ \text{# 17168} - \text{PAC is to supply one more paragraph for Stats SOP (should be done by 11/04/08) then RS \\ \text{will upload.}\\ \text{Change Proposals} - \text{VKI approved the following New Change Proposals which should be ready for May Management Meeting.\\ \text{#38} - \text{GeneMapper IDX}\\ \text{#41} - \text{Batch Uploading of Profiles to NCIDD}\\ \text{#41} - 3100B \text{Upgrade to 3130xI}
additions. d. Staff	Training sign off and feedback progress including outstanding g sign offs from dan	analyses of existing staff. e. Review the	Issues Register.			Change Proposals / Project Management (Robyn)
						2.2



Change Proposals requiring Approval - #34 Kinship – WW requires this one asap. Change Proposal #33 Peak Heights – RS to advise AMc of scheduled meeting so he can attend.	AMc and M/C RS / AMc	
PREVIOUS		
 21/2/08 Complete 18695 Allan to enter an update on investigation in this OQI. On going - BRB stats - page 4 of presentation refers. Follow up - email from Robyn (22 Feb 08 12 25mm) 		
 RS to talk with Tim re non conservative ones – how long to do this fix? To alter data of affected alleles for cases 2005 onwards. 	S	1
21/2/08 - Manual calculations training session - AAP and CJA yet to undertake this.	RS	
21/2/08 - KINSHIP Change (Proposal 34) request for Stage 1 to be signed off. Please complete Impact Assessments by 2 nd April.	All	
17/1/08 - <mark>On going</mark> - Kate to propose change of phadebas paper.	Z.	
25/10/07 – Complete - Change Proposal #26 (Amended Receipt) – agreed that if the packaging is labelled on the outside then enter these details in the receipt (option 10). If not labelled then detail as eg. envelope package (option 8). Multiple items in one packaging – describe in body of statement, no amended receipt is required. VKI to summarise the 2 change proposal documents and speak with PPoint and then a date can be set to commence. Specifications for the AUSLAB table at the end of the statement are also required.		A
27/9/07 - On track - PAC to obtain information from QPS on validations of TMB, Phadebas, AP & PSA		1
27/9/07 - Needs Attn: VKI to enquire with BSAG re validation paperwork. For TMB & AP.	PAC	V
20/12/07 - ON HOLD - (to be programmed) Suggestion to have photos on screen at Shift F9 and colour to indicate specimen notes. TEN agreed to look into this.	NK VK	





	A			
Action	VKI			
3.0 NEW BUSINESS Discussion	3 year agreement certified until August 2010. If appointed to reclassified positions eg September 2008, will receive back-pay from September 2007. Information for applicants can be found on QHEPS. Links for flowcharts etc will be forwarded. 38 hour week – new arrangement. Aim for implementation by end of July 2008. It will be decided on the Unit needs not individual. Not sure at which level this decision will be made eg Director or Manager. Staff update to be arranged.	Concept Brief has been submitted and no funding will be offered. VKI will meet with all staff concerned and also meet one on one with Team Leader/Line Manager present. No CBRC submission. Questions:- How do we appoint these 16 people. Agree that whoever has a permanent position to go back to should be placed further down the list. Project people. Substantive positions. Re-sort names/dates to work out length of service. VKI will slot names into positions and Management Team to provide feedback/argue cases. Support will be given for staff to attend Win that Job training, LISS Project Manager training and effort made to find alternate campus positions.	Position to be finalised by next week (W/E – 18/04/08)	VKI and JAH will be away for three weeks commencing on 19 May 2008. CJA will be A/Managing Scientist. Travelling to Prague, London, Ottawa and Washington and visiting overseas labs for processing and refurbishment ideas etc. Architects should have been engaged and planning workshop arranged regarding the refurbishment by the time VKI and JAH return.
Topic	HP Implementation	Contracts	Major Crime Team Leader Position	Overseas Trip
ltem	3.1	3.2	3.3	3.4



is still in draft. It was suggested that a Plate Reading Workshop may be needed. PT, RS and AMc are to co-ordinate and arrange workshop.	te an te other cost Log he SOP I AMC	Feedback received has been good. Annual or half yearly event will also be good. Suggestion not to have so many 'little' awards as it diminishes the special awards. VKI happy to support anyone wishing to volunteer for SES. Requires Line Manager approval. VKI happy to support anyone wishing to apply for the NIFS secondment. The recent DNAIQ email sent out is causing doubts and concerns with DNAIQ. AMc will create an Analytical Issues Log with different tabs for certain issues. It was decided to discuss issues at the Team Meeting level to determine if issues are in fact real concerns and cannot be fixed by another means. Therefore any issues which are deemed important will be added to the Analytical Issues Log which will be located on I drive. Some plates are not being read correctly. Some do not understand that blood is evidence. The SOP is still in draft. It was suggested that a Plate Reading Workshop may be needed. PT, RS and AMC are to co-ordinate and arrange workshop. FIRMU are phoning Scientists for sub sample results when they should phone FSLU. But FSLU don't	ysis nteers nail
	they should phone FSLU. But FSLU don't rise FIRMU to phone FSLU instead of	FIRMU are phoning Scientists for sub sample results when they should phone FSLU. But FSLU dor always respond in a timely matter. It was suggested to advise FIRMU to phone FSLU instead of Scientists. VKI is reviewing communication procedures with FSLU and FIRMU so for the status quant	Sub Sample Requests
Comp plates are not being read correctly. Some of any and that blood is evidence. The SOD	b.	The recent DNAIQ email sent out is causing doubts and cor Analytical Issues Log with different tabs for certain issues. I Team Meeting level to determine if issues are in fact real co means. Therefore any issues which are deemed important which will be located on I drive.	DNAIQ Email
Analytical Issues Log with different tabs for certain issues. It was decided to discuss issues at the Team Meeting level to determine if issues are in fact real concerns and cannot be fixed by another means. Therefore any issues which are deemed important will be added to the Analytical Issues Log which will be located on I drive.	secondment.	VKI happy to support anyone wishing to apply for the NIFS s	NIFS Secondment
WKI happy to support anyone wishing to apply for the NIFS secondment. Q Email The recent DNAIQ email sent out is causing doubts and concerns with DNAIQ. AMc will create an Analytical Issues Log with different tabs for certain issues. It was decided to discuss issues at the Team Meeting level to determine if issues are in fact real concerns and cannot be fixed by another means. Therefore any issues which are deemed important will be added to the Analytical Issues Log which will be located on I drive. Reading complete any issues which are deemed important will be added to the Analytical Issues Log which will be located on I drive.	Requires Line Manager approval.	VKI happy to support anyone wishing to volunteer for SES.	Volunteers
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Communications to go out	
Action required	(inc: Officer, Due date)
4.0 BUSINESS ARISING FROM PREVIOUS MINUTES	Discussion
	Item Topic
	Item



			ogy managem	Pictor III III III III III III III III III I
	PROJECT	STATUS, RISKS, ISSUES, COMMUNICATIONS		
4.1	Mid week Mind Maps	25/3/08 - DNA Analysis to present on 23 April. Suggestions – how exhibits have been sampled / Case study / new technologies.		
		10/4/08 - DNA Analysis will be presenting a Mind Map Presentation on 23 May 2008. It was agreed to that the Yellow Team will present a specific case (Wooloowin Rapist) with other teams eg Analytical providing input as to their roles in the case. The three team's individual presentations will go for 15 mins each. CJA has volunteered to co-ordinate the presentation.		A
4.2	ANZFSS Symposium in Melbourne, Oct 08 (Cathie)	VKI to enquire re budget – funding. VKI to report to next whole team meeting. Abstracts close 6 July. August staff advised if successful with presentation / poster. A process will occur re sharing any funding. A feedback session will be held after the event. Rates for travel will be actual expenses for food, incidentals – flat rate, and staff will need to share accommodation if being funded, if not happy with this need to fund their own accommodation expense. Details to be confirmed further.	VKI	
6.3	Digital Imaging 10/3/08 (VKI)	 Current Audit was conducted. Some of the issues identified – with photographing of packages (having to photocopy also in order to read barcode information). Just photocopy if this occurs, stop the duplication. Photos being lost, due to delay in uploading to AUSLAB Annotations not being done – hand written Calibration of touch screens to be re done. Staff not complying with SOP. Need to review SOP and software. Please nominate a staff member from your team to be part of the Digital Imaging Group. Forward nominations to VKI. VKI to pick a Momt team member to chair this group. 	=	AN
			All	



Foyer Display Dates Unit Displays.doc		tors to I staff ity. and s are	ator VKI	/e have,	sthod is TEN	ter a 🛕 🗸
Foyer Display Dates for 2008 (DNA Analysis – 28 April - 6 June) what are people reporting back and what should we be reporting back to FIRMU? - what is the definition of every different type of result?" (Kylie) Reporting method for multiple items in a bag. (Justin)		Dates Unit Displays.doc Fwd Unit Display dates.vsd.rtf 21/2/08 - This is part of the communication strategy and is intended to inform our staff and vis our campus what each of the units do. It is also intended to begin to focus the attention of a on this campus on what we are here for – delivery of science and its benefits to the commun Material prepared for this could be used for Science week, therefore needs to be high quality targeted to impress politicians and the general community. Posters and/or interactive display encouraged.	Please forward nominations from each team to TEAM LDR M/C by Friday 29 February. 10/4/08 - The Foyer Display has been scheduled for end of April – May 2008. SJC is co-ordin and VKI will talk to SJC about the details.	21/2/08 - EXR Results – info in EXR should be that which is in the statement? What does FIR want in this space? CJA to speak with FIRMU (Tracey Dale) - (Results plus example of all valumnary or full details)	31/1/08 – 3 ways proposed. Due to delay in waiting for spec to be finalised, the proposed reporting monto be used – TEN to follow up with LISS re where status of spec.	Barcode (include description eg. Unknown individual) / EXR / Link field to remain blank / enter a
	Four Dienlay	Dates for 2008 (DNA Analysis – 28 April - 6 June)		"EXR results - what are people reporting back and what should we be reporting back to FIRMU? - what is the definition of every different type of result?" (Kylie)	Reporting method for multiple items in a bag. (Justin)	



4.7	Methods Page (AUSLAB)	6/12/07 - Needs Attn: Could a rep from V/C, M/C and Analytical please forward to RS a list of ALL the methods used so that these can be added to AUSLAB.	M/C & CJA	
		QIS document 17092 refers. The Methods Used in Casework page in AUSLAB is not up to date. Some methods are no longer used; some new ones are not listed eg. DNA IQ. 1. Review if required – VKI 2. Use manual process – paper rather than AUSLAB – RS	VKI	
4.8	Timeline of landmarks in For Biol eg.	11/10/07 - <mark>On track</mark> – All staff to comment and/or to write suggestions of other key landmark timeframes to include. Timeline (brown paper) is located along the corridor outside of the main lab.	VKI	
6.4	Statement Changes Case Assessment Scenarios Feedba ck 140607.doc	25/5/06— Joshua Miles is the FSS Liaison person. On going - Summary Statement Changes PhoneCall.DOC Summary Statement Changes PhoneCall.DOC Agreed that a new Statement SOP should include guidelines about appropriate phrases to include in your statements, and guidelines about what should not be written. Using the information from phone call and from the 3 example statements forwarded to DPP. A rep from M/C (Helen) and V/C (Anne) to compile this new SOP.	TEAM LDR M/C	A
4.10	DNA Certificates	24/5/07 - Need to improve linking events & OQIs. (Search QHEPS for Crisp Number?) On track - VKI to liaise with Glen Cash DPP re email.	VKI	1
	Please advise VKI if you have a request for a DNA Certificate. AUSLAB will be slightly changed.	On track - Seek advice from DPP & LALU (Need to produce a DNA Certificate for every statement OR only when requested?) Awaiting LALU advice, seeking court transcript.	VKI	18



			AN		
	RS	KDR	KDR / TEAM LDR M/C KDR	Ingrid / AMc	PAC
10/5/07 - As V/C staff will now be assisting with some High throughput cases it is important to all agree on the compilation of case files. Needs Attn: Hold a workshop in 2008 to agree on a standard method of case file compilation. Need to look at - What is the purpose Review current processes How many ways are active at present – list different ways	Impact on workload Outcome / Recommendations – Consistency of process across Biology	On track - Proposed for A/PO4 Quality to chair this Workshop. Attendees Judy, Alicia & Shannon M. (V/C) – JAH, Jacqui, Margaret, {KDR to advise of a replacement for Shannon T} Workshop Ctee to meet and make suggestions for presentation at Mgmt Team Mtg.	307/07 - On going — Some DNA Analysis staff are undertaking the Stats On-line course. To be finalised in May this year. Some DNA Analysis staff are undertaking the Stats On-line course. To be finalised in May this year. Perhaps a biology specific practical application of stats could be trained to one or two by ESR /AFP staff. On track - Need to establish what are our gaps are on all different levels and ways to address these. Both in the short term and long term. On going training would be preferred. Need to include FST also. KDR and TEAM LDR M/C putting a list of questions together (what gaps are present with current stats training — what questions have been asked) need to ensure that everything is included, including coronial. Once compiled, list sent to all Reporters to answer to find gaps.	8/2/07 - After validation is complete, the thresholds will be reviewed. Will be incorporated into 3130. Ingrid and AMc to discuss.	7/6/07 - Photography training with Murray Latter, QPS. Nomination from teams - Melissa – Blue team. Jacqui – Yellow Team Thu – Red Team
Compilation of case files.			Stats Position	Bone Threshold	Training
4.11			4.12	4.13	4.14



Biology Management Team Minutes

		6	
	ON HOLD OR AWAITING REPORT		
AS automatically re-working Al's - why? What about Al's in	Agreed to maintain status quo – reviewed when thresholds are reviewed. Sam to be included in these discussions.	1	
PP's amped at or near 20ul? (Allan)	Will explore answers to these questions.		A
Seeking feedback on	Across campus the concept of a Lab Manager position is being reviewed. This position would be responsible for and conduct the following duties (procurement, lab set up, ordering, stocks, WPHS,		
concept of Lab	lab guidelines/calibrations – does not manage staff, just the lab space). Agreed to look at the Job		
position (VKI) 10/3/08	מפאנון המווא מפעפוסף בי וווי סוופן מופמא מווע מופן מואנים המאות מופן מואנים אומים מווע מואנים אומים מווע מואנים		
Use of STAFF match Results table	23/11/07 - Currently two databases in use + an identifier matching table. Some entries are not being added to all three places. Is it necessary to have two databases?		
comparison macro &	Agreed to streamline the process. For this to occur –		
AUSLAB staff	Macro to be updated		
database. (I EN)	 Workflow – no gaps Process investigation of who is responsible. 	RS	
	Internal change process – RS to coordinate this and identify timeframes in the new year.		
	Proposed for specimen notes in reporting, input onto Case Scientist / Team communication list re "potential staff match", when this all occurs, then only the AUSLAB database will be used. UNTIL	All	1
	then using current system of 2 databases.		The second second

NEXT MEETING

The next meeting is scheduled for Thursday 8 May, 2008, Conf Room 102, 9am - 11am.



	ne: 2008 – June 20	at: WAH					s follows:	Action Communications to	7 50							
	Date and Time:	Secretariat :				SED	without amendment or amended as				many instances must we				orofiles from DNA Master /	uenced, AF to arrange for AF
PRESENT AND APOLOGIES	VKI	Conference Room 102	VKI, CJA, JAH, AJS, KDR, SM, PT, AMc, TEN, RS, PAC, EJC, WH	AAP	AF, KO, JK, CW	1.0 PREVIOUS MINUTES ENDORSED	Minutes of previous meeting held on 6 June 2008 are endorsed by VKI and WAH without amendment or amended as follows:		Discussion	2.0 STANDING AGENDA ITEMS	What Variants and Off Ladder Alleles – to include in GeneMapper ID-X (how many instances must we see these alleles in order to include them). See table below.	Table (taken from KOD a couple of years ago) Action:	To be included –	 Variants which have been published by NIFS 	 Those from DNA / AUSLAB database – expand search to include profiles from DNA Master / AUSLAB. 	 Those seen at least once / identified a fair bit (if they are not sequenced, AF to arrange for them to be sequenced on site or off site).
	Chairperson:	Venue:	Attendees:	Apologies:	Guests:				Topic		13 Update / GM IDX (Anne)					
									Item		2.1					



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100		AF		AF / TEN / AMc	AMc	₹	_			
Genescan / Sizing process. Apr June\Brown Paper - GeneScan.doc Action: Agreed to Option 4 - Current process remains the same, 3130 Operator will check Ladder, controls, and size std, then pass on to First reader for analysis.	:\GMIDX validation\Process Mapping.ppt Action:	SOPs, Training Modules, Methods of Training AF and Project Team now to finalise these.	PREVIOUS	Needs Attn - 23/5/08 - AF to forward table to TEN, ready for forwarding to AUSLAB. A macro is required in the export table to fix NSD in GeneMapper. AMc, TEN, AF to fix this.	23/5/08 - 3130 Computer can be moved, to be decided by AMc	23/5/08 - Please think about how many staff are to be trained in Genemapper initially.	<u>Presentation</u>	JAH, PT, PAC recently attended practical people management course. During 2009, 44 staff from the campus are scheduled to attend.	PAC acting Jan for the next 2 weeks.	Sue Kleidon is leaving SSDU and an EOI will be forwarded soon.
							Ē,	Training Program	b. Biology Training	Calendar
							2.2			



	A		7			1		
	EO	\K	PAC	PAC		RS	All	₹
SSDU are assisting with testing of QIS2. PREVIOUS 6/6/08 Complete - AK, RP, JW, AMc attending DPP moot court 26/27 June.	 Replacing POP STATS. EJC to liaise with PAC/ JAH/ SJC/ KDR to put together recommendations for replacement. Recommendations to include costs. GeneMapper Mixture Program Locomotion – not available as yet System used in LGC UK – can this be purchased? Cost? Build an in house program. Tack this onto the end of present Kinship project. 	O	 PAC to follow up with Paula & Justin re trainers 	 Stats course – PAC to prepare a document re the evaluation/ outcome of this course. 	G:\ForBio\\Quality Assurance and NATA\Management Reports - Biology Management Team	Change Proposal 3130B – RS to arranged for signatures AMc to add to minor change register	FTAs – Processing on 3130B will commence this afternoon. Currently approx 3000 FTAs outstanding. Overtime can be offered for staff to read plates (weeknights / weekends). Staff to at least read 1 plate during overtime. Allan to keep management team informed of need for readers to ensure deadline is met.	Change Proposal 33 Peak Height and Allelic Imbalance Thresholds AAA Analytical Section\Internal Projects\Pk Ht & AI thresholds Chiron and AMc to put together some examples which they have used to come to this decision, and
c. Training modules – review, changes, additions. d. Staff Training	sign on and feedback progress including outstandin g sign offs from gap	analyses of existing	staff. e. Review the	Register.	Change	Proposals / Project Management (Robyn)		
					2.3			



5	•		
	arrange a mtg. Mgt Team members to bring along some of their own examples and try them out with the formula. This will give assurance of the 50% for casework. Test / formula to be tested on mixtures. Papers researched for this project support 50%.	Chiron / AMc	
	Chiron to prepare a literature review report for this project, similar to the one prepared for the PSA project. Chiron to liaise with Rhys Parry.	Chiron	
	Project findings to be submitted as a technical note. VKI to take this to BSAG to put it out there, and get some review / comments / feedback from other labs.	Chiron / VKI	A
	Project has been submitted for presentation at the ANZFSS conference later this year.		1
	Quality		
	Still a few duplicate barcodes appearing. Most of these are being identified early. (FTA barcode same	All	
	Correction in Polaris of these duplicates is taking a couple of weeks. RS to advise VKI of these cases.	RS / VKI	7
	RS to forward Quality report via email.	RS	
	If your cases have BRB stats, please re do them using the new Kinship database.	All	
	RS, AMc and Analytical Staff have met to develop a plan to deal with Mixture investigations. Quants / Profiles in neg controls. These incidents will be emailed to Management Team. Meeting next week regarding mixture in ref plates.		
	 Presentations to be included in SOPs on investigation process OQI raised Reporting details – central point for investigations Filed in case file. 		
	PREVIOUS		
	6/6/08 – Stage 2 of Kinship #42 – Kinship staff to attend team mtgs, impact assessments will be due after this has occurred.		



6/6/08 – no further action at this time. Audit findings – #8134 Digital Image Working Party VKI has names of volunteers. Update 23/5/08 – RS to add note to OQI re "on hold for now, action pending".
On going - BRB stats. Follow up – email from Robyn (22 Feb 08, 12.25pm) RS to talk with Tim re non conservative ones – how long to do this fix? To alter data of affected alleles for cases 2005 onwards.
21/2/08 - Manual calculations training session - AAP and CJA yet to undertake this.
17/1/08 - <mark>On going</mark> - Kate to propose change of phadebas paper.
27/9/07 - On track - PAC to obtain information from QPS on validations of TMB, Phadebas, AP & PSA
27/9/07 - <mark>Needs Attn</mark> : VKI to enquire with BSAG re validation paperwork. For TMB & AP.
20/12/07 - ON HOLD – (to be programmed) Suggestion to have photos on screen at Shift F9 and colour to indicate specimen notes. TEN agreed to look into this.
Viewed advertisement on EZ1 Advance. Equipment for Trial will be arriving today. TEN to liaise with Victoria regarding their outcomes of using this equipment.
Project 24 sperm – I cube vs Chelex - quants. Need to establish if ProK is the problem.
Project 27 – DNA IQ recovery – new heating plate received from Promega – heats well. TEN to attend V/C and M/C meetings to talk about DNA IQ Recovery. *options around Diff Lysis
23/5/08 - <mark>Complete</mark> - Adobe Acrobat reader licences have been installed



Diology management ream minage				Please advise your teams regarding parking
y manage	TEN	TEN	JAH / CJA	
Social	23/5/08 – Needs Attn - TEN will investigate nuc /clean ups, 30% of these done are unnecessarily.	10/4/08 – Nothing further on consent form for the project, just verbal consent given for now.	23/11/07 - On track - Batch functionality was rolled out in Feb 2007 – as yet no completed training modules have been received from M/C & V/C. Update – Appts have been scheduled for M/C. V/C will be followed up in the new year. It is important for all staff undertaking Case Management to complete this training module.	<u>Timesheets</u> – new timesheet being developed, SSDU developing training to ensure correct completion of timesheets based on new timesheet. <u>Parking</u> – FSS Exec agreed to following proposals regarding illegal parking on campus. Staff rego details to be updated, initially contacted and warned, considering fines/towing for repeat offenders. <u>QIS2 Info sessions</u> – to be scheduled in July for staff to see screen shots, learn about changes Request from SSDU – if sent an appointment to review training modules, please attend or contact SSDU if you can't to reschedule. Has been people not turning up from all departments.
				Operational Group – Report (VKI)
				2.5



6			N					1	I	100	
	Action										
6	3.0 NEW BUSINESS Discussion	This will commence Monday 23 June.	Agreed on agenda for today's whole team meeting. All staff have a choice in how they react / deal with / involve themselves in the restructure / new processes from 1 July.	Perhaps a Criminal Lawyer will be engaged with respect to new changes in reporting.	Timeframes – (front end) 1 July – (new team structure) 14 July.	New Milestone wall to be established.	Need to identify new SOPs required for new structure / processes.	New team worklists have now been created in AUSLAB.	Work Experience Student – Sonya Tadrowski (Sheldon College) – 23-27 June. Sonya will be observing only, here from 9am to 3pm daily. VKI to prepare schedule for the week.	Carried forward to next meeting.	
	Topic	Accreditation Prep Plan (RS)	Restructure DNA Analysis	Issues Log						38 Hour Week	
	Item	3.1	3.2							3.3	



		4.0 BUSINESS ARISING FROM PREVIOUS MINUTES	Action required	Communications to go out
Item	Topic	Discussion	(inc: Officer, Due date)	,
	PROJECT	STATUS, RISKS, ISSUES, COMMUNICATIONS		
4.1	Plate Reading	10/4/08 - Some plates are not being read correctly. Some do not understand that blood is evidence. The SOP is still in draft. It was suggested that a Plate Reading Workshop may be needed. PT, RS and AMc are to co-ordinate and arrange workshop.	PT, RS and AMc	A
4.2	ANZFSS Symposium in Melbourne, Oct 08 (Cathie)	VKI to enquire re budget – funding. VKI to report to next whole team meeting. Abstracts close 6 July. August staff advised if successful with presentation / poster. A process will occur re sharing any funding. A feedback session will be held after the event. Rates for travel will be actual expenses for food, incidentals – flat rate, and staff will need to share accommodation if being funded, if not happy with this need to fund their own accommodation expense. Details to be confirmed further.	VKI	
4.3	Digital Imaging 10/3/08 (VKI)	 Current Audit was conducted. Some of the issues identified – with photographing of packages (having to photocopy also in order to read barcode information). Just photocopy if this occurs, stop the duplication. Photos being lost, due to delay in uploading to AUSLAB Annotations not being done – hand written Calibration of touch screens to be re done. Staff not complying with SOP. Need to review SOP and software. Need to review SOP and software. Please nominate a staff member from your team to be part of the Digital Imaging Group. Forward nominations to VKI. VKI to pick a Mgmt team member to chair this group. 		AN
4.4	"EXR results - what are people reporting back and what should	21/208 - EXR Results – info in EXR should be that which is in the statement? What does FIRMU want in this space? CJA to speak with FIRMU (Tracey Dale) - (Results plus example of all we have, a summary or full details)	CJA	



6						
37		JEN	All	M/C & CJA	VKI	VKI
		31/1/08—3 ways proposed. Due to delay in waiting for spec to be finalised, the proposed reporting method is to be used – TEN to follow up with LISS re where status of spec.	Barcode (include description eg. Unknown individual) / EXR / Link field to remain blank / enter a description. Print the screen from AUSLAB, fax through to FIRMU, and call FIRMU to confirm receival of fax, update UR note with action taken. It is very important to have documentation ie. Fax or email to confirm conversation with FIRMU, not just phone conversations.	6/12/07 - Needs Attn. Could a rep from V/C, M/C and Analytical please forward to RS a list of ALL the methods used so that these can be added to AUSLAB.	QIS document 17092 refers. The Methods Used in Casework page in AUSLAB is not up to date. Some methods are no longer used; some new ones are not listed eg. DNA IQ. 1. Review if required – VKI 2. Use manual process – paper rather than AUSLAB – RS	11/10/07 - <mark>On track</mark> – All staff to comment and/or to write suggestions of other key landmark timeframes to include. Timeline (brown paper) is located along the corridor outside of the main lab.
	we be reporting back to FIRMU? - what is the definition of every different type of result?" (Kylie)	Reporting method for multiple items in a bag. (Justin)		Methods Page (AUSLAB)		Timeline of Iandmarks in For Biol eg.
		4.5		4.6		4.7



6		N					
6	JAH / CJA / RS	D	D			60	KDR
	Johna Miles is the FSS Liaison person. Joshua Miles is the FSS Liaison person. On going - Summary Statement Changes PhoneCall.DOC Agreed that a new Statement SOP should include guidelines about appropriate phrases to include in your statements, and guidelines about what should not be written. Using the information from phone call and from the 3 example statements forwarded to DPP. A rep from M/C (Helen) and V/C JA (Anne) to compile this new SOP.	24/5/07 - Need to improve linking events & OQIs. (Search QHEPS for Crisp Number?) On track - VKI to liaise with Glen Cash DPP re email.	On track - Seek advice from DPP & LALU (Need to produce a DNA Certificate for every statement OR only when requested?) Awaiting LALU VKI advice, seeking court transcript.	10/5/07 - As V/C staff will now be assisting with some High throughput cases it is important to all agree on the compilation of case files. Needs Attn: Hold a workshop in 2008 to agree on a standard method of case file compilation.	What is the purpose Review current processes	Why Impact on workload Outcome / Recommendations – Consistency of process across Biology	On track - Proposed for A/PO4 Quality to chair this Workshop. Attendees Judy, Alicia & Shannon M. (V/C) – JAH, Jacqui, Margaret, {KDR to advise of a replacement for Shannon T} Workshop Ctee to meet and make suggestions for presentation at Mgmt Team Mtg.
	Statement Changes Case Assessment Scenarios Feedba ck 140607.doc	DNA Certificates Please advise VKI	if you have a request for a DNA Certificate. AUSLAB will be slightly changed.	Compilation of case files.			
	4. 8.	6.9		4.10			



Biology Management Team Minutes

Training	7/6/07 - Photography training with Murray Latter, QPS. Nomination from teams - Melissa – Blue team.	PAC	
	Jacqui – Yellow Team Thu – Red Team		
	ON HOLD OR AWAITING REPORT		
Use of STAFF match Results	23/11/07 - Currently two databases in use + an identifier matching table. Some entries are not being		
table	ממממנו כי מון ווויכל פונים בי וויכל		
comparison macro &	Agreed to streamline the process. For this to occur –		1
AUSLAB staff	Macro to be updated		
database. (TEN)	Workflow – no gaps		
	 Process investigation of who is responsible. 	RS	
	Internal change process – RS to coordinate this and identify timeframes in the new year.		
	Proposed for specimen notes in reporting, input onto Case Scientist / Team communication list re "potential staff match", when this all occurs, then only the AUSLAB database will be used. UNTIL	All	
	then using current system of 2 databases.		

NEXT MEETING

The next meeting is scheduled for Friday 4 July, 2008, Conf Room 102, 9am - 11am



JH-46

Report for QIS OQI as of 28/09/2022 9:41:56 PM

Report for QIS OQI -

19477 No Title Provided

OQI Details

Status	Status Closed Approved
Subject	Subject A Negative extraction control sample 346795477 was extracted on CWIQLYS20080429_01/CWIQEXT20080430_01 had been
	profiled twice confirming a partial profile in the DNA extract.
Source of OQI	Source of OQI Internal Problems (QHPSS)
Date Identified 12/05/2008	12/05/2008

OQI Creator Contact Details

Amy CHENG	Analytical		Coopers Plains
Creator	Organisational Unit/s	Service/s	Site Location/s

Investigator/Actioner Contact Details

Allan MCNEVIN	Analytical		Coopers Plains
Actioner	Organisational Unit/s	Service/s	Site Location/s

Investigation Details

Procedure/Method/Process
Root Cause Type
22/07/2008
Investigation Completed

Investigation Details

microcon results were analysed at 30RFU values. A full 9-loci profile was able to be elucidated from the concentrated sample. This was then searched against all profiles from the same extraction batch (CWIQEXT20080430_01). A match was found with Further matches were found to a mixture 333810182 and a match was found to sample 288908564 after it had undergone a During the course of the investigation two further examples of potential well-to-well contamination have been identified and taken in conjunction with two previously documented events, these events build a picture of potential systematic problems. which are all from the same case, (10.53ng/uL) had quite high quantification values and one or more were the most likely source(s) of the contamination. confirmed after re-amplification. The control was then concentrated via microcon and both the original results and the showed a single peak above 75RFU at Amelogenin after extraction. This was 8.8ng/uL) & (5.61ng/uL), (This other events have been documented as OQI's 19330, 19349, 19767, & 19768. clean up procedure with the NucleoSpin Tissue Kit, Samples 3 Quality Information System Negative extraction control samples

Preformed By

Action Details

22/07/2008

Action Complete

Action Fix Type | Changed Process

Action Description A full

reagent blanks iii) Urgent progression of audit mentioned above and investigation into findings iv) A full information review of above. A investigation report has been written and stored in I:\AAA Analytical Section\Adverse event investigations\ An extra related to platform A) ii) Processing of Casework samples on Extraction platform B in a checkerboard pattern with extraction process. This had been planned as a post implementation review but has been brought forward in view of events mentioned process audit (audit #8227 ? DNA IQ) has been commissioned to thoroughly review all facets of the automated extraction -ordinary meeting of the DNA Analysis management meeting was held 14/07/2008 and the following actions were agreed upon: i) Processing of Reference samples only on Extraction platform A (initial investigations indicated events were likely further information This OQI has been discussed in the Analytical team meeting. Staff have additionally been individually results from automated extractions with documented quality events and extractions without documented events to gain approached with questions re: concerns, possible solutions etc. by the audit team for audit #8227

Task Details

No Tasks found

Follow-up And Approval

Accepted Follow-up Status Follow-up Status Comment

8/08/2008 3:28:01 PM Amy CHENG:

Accepted as this OQI will be addressed in the Process Audit 8227

Approval/Rejection Date | 18/08/2008 Approval/Rejection Comment

Approver | Cathie ALLEN

18/08/2008 12:00:00 AM Catherine ALLEN:

Part of a larger investigation and with Audit 8227.

Associations

No Associations found

Records

No Records found

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28/09/2022

JH-47

Report for QIS Audit as of 28/09/2022 6:30:49 PM

Report for QIS Audit -

8227 DNA Extraction Process (DNA IQ)

Audit Contact Details

|--|

Audit (Lead/Internal) Contact Details

Audit Details

Date Audit Performed 15/07/2008 Audit Type Process	15/07/200 Process
Audit Status	Closed
Audit Subject	DNA Extraction Process (DNA IQ)
Audit Objective No Objective	No Objecti
Audit Scope	No Scope
Audit Criteria No Criteria	No Criteria

Audit Outcome

Audit Findings

Although some risks for mislabelling, contamination or cross contamination exist in the procedure, there are appropriate and sufficient quality control measures in place to minimise these risks. Although we observed bubbles and droplets forming at the end of disposable tips during the automated DNA IQ protocol, these were not observed to have dripped into any wells This audit was unable to determine the exact source of contamination as reported in OQI's 19477, 19768 and 19349.

28/09/2022

Audit Report

parameters within the protocol. A follow-up of samples processed in checkerboard format on batch CWIQLYS20080714_02 did not show any instances of well-to-well cross contamination, as evidenced by the absence of DNA profiles in all of the and were discarded in the tip chute. Bubble formation can be reduced and eliminated by further optimising the pipetting water blanks. We commend the department for actively engaging in a continual methods improvement process (either to improve QA/QC or increase ease-of-use and efficiency of a procedure).

The auditors have identified 28 points that may improve the automated DNA IQ extraction process, and have made the recommendations as outlined in the 18-page Audit 8227 report (Cheng, Clausen & Muharam, 2008) located in Quality Management, DNA Analysis.

Three OQI's were raised as an outcome of the audit, in order to address the 28 points of recommendation:

OQI 20367 - Automated DNA IQ process, including documentation. OQI 20368 - Enhancement of the MP II extraction platforms, including environment. OQI 20369 - Training and personnel related to the DNA IQ process.

Please refer to the associated OQI's for the specific points addressed by each OQI.

Contact Comments

Associations

Module OQI QIS Record No Title Provided QIS Record Number 20367 Association I Association Description Audit Audit QIS Record Number 9642 Association Description
--

Records

No Records found

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JH-48

Report for QIS OQI as of 28/09/2022 7:41:39 PM

Report for QIS OQI -

20351 No Title Provided

OQI Details

Status	Status Closed Approved
Subject	CWIQEXT20080402_01 was found to have a partial minor DNA profile present in the extraction positive control (
Source of OQI Audit	Audit
Date Identified 08/08/2008	08/08/2008

OQI Creator Contact Details

Kylie RIKA	Intelligence		Coopers Plains
Creator	Organisational Unit/s	Service/s	Site Location/s

Investigator/Actioner Contact Details

Allan MCNEVIN	Analytical	
Actioner	Organisational Unit/s	Service/s

Site Location/s | Coopers Plains

Investigation Details

Investigation Completed 09/12/2008	ed 09/12/2008 Root Cause Type Procedure/Method/Process Investigation Details This OQI relates to	s to
	(positive extraction control),	batch the para
	magnetic resin during the automated DNA IQ extraction process) and the stored substrates (i.e. the material originally submitted for DNA extraction processed through the initial off-deck lysis steps of the initial extraction process) for all 13	nally all 13
	samples were re-extracted. These results were analysed using GeneMapper ID-X software with a peak detection threshold of 20RFU to gain the most information. The extraction of the stored lysate for each of the 13 samples showed results consistent	hreshold of s consistent
	with that obtained from the initial extraction process. From these results, it can be concluded that the contamination of samples	ion of
	must have occurred prior to or during the	ng the
	for samples the profile form the para-magnetic less. These samples showed single source profiles consistent with one of the	ne of the
	contributors to the two-person mixtures observed after the original extraction. The other contributor was consistent with the profiles obtained from sampled	stent with the (the same
	was obtained from the original extraction, the stored lysate and re-extraction of the s	ed same
	substrate). The results show that for samples	of the
	the manual processing on	one or
	more of samples (due the 10 to 100 fold higher quantification values observed for these samples) more of samples	samples)
	substrates for samples showed no DNA profile, so no further conclusions could be drawn. The re-	The re-
	extraction of sample yielded a different DNA profile to that obtained from the original extraction and the stored	he stored
	Iysate. However when the original profile was re-analysed at 20RFU threshold, peaks consistent with the re-extracted sample were observed indicating that the re-extracted substrate has vielded the true profile and the original extraction was	cted sample
	contaminated between the same steps as sample noted above. For sample stored,	the stored
	Iysate and re-extraction of the stored substrate yielded the same DNA profile as the initial extraction. This shows that a	that a
	contamination event of this sample is unlikely and common alleles with the contaminating profile is con-incidental. The	. The
	potential steps at which contamination may have occurred are: 1. During the transfer of the lystate obtained from manual lysis into the deen-well plate via the use of the STOBstar. However this is unlikely as staff are trained to perform such	ı manual
	piesting steps with due diligence and care. This is also unlikely in the case of samples	Sacri
		proposed
	contaminating samples. Due to character space limitations, investigation is continued in Action below.	

Preformed By | Quality Information System

Action Details

During the lysis buffers and resin within a 2.2mL well) on the extraction platform DPC shaker. This had not been observed, however has been During the mixing of the deep-well plate (containing 1.5mL of protocol (fresh disposable tip for each step). If there was dripping of the lysate containing unbound DNA and this was to drip commissioned team. A number of potential contamination events were identified and each is to be investigated on batch-bystart of the automated extraction procedure (i.e. incorrect pipetting procedure), however this is unlikely as staff are trained mplemented or are under investigation as outlined in OQI's 20367, 20368 and 20369. After the cessation of the automated contamination events investigated. 4. Operator error during the manual addition of DNA IQ para-magnetic resin during the commissioned and carried out. A number of areas for improvement were identified through the audit, and these have been . 3. During the removal of the adhesive seal. It was noted during Audit 8227 that condensation on the underside of the adhesive seal was across into an adjacent well. This was possibly due to insufficient application of the adhesive seal to the interstitial barrier. result of previous OQI's raised and concerns identified around the automated DNA IQ extraction process, the extraction of via this mechanism is unlikely. Action: As a Investigation cont'd 2. Seepage of sample into an adjacent well during cold storage of the lysed material in the deep-well plate. After the cessation of processing and the carrying out of investigations, it was noted in one instance that a heavily blood-stained lysate had condensed on the underside of the adhesive seal used to seal the stored plate. This had seeped removal from the para-magentic resin to the storage plate. This procedure occurs twice during the automated extraction rom one well to another well this may account for the contamination event. A similar mechanism may occur if a bubble proposed a possible mechanism for adjacent well contamination during close scrutiny of the automated procedure. This **Action Description** not removed after centrifugation. This mechanism is considered most likely due the varied multiple-well nature of the due to the directional movement of the 8-tip arm. forms at the end of the pipette tip and bursts whilst in the vicinity of another well. This mechanism is only considered samples using the automated DNA IQ procedure was halted on the 28-7-2008. Prior to this, Audit 8227 had been DNA IQ extraction protocol, a review of all batches processed through this protocol was carried out by a specially to perform such pipetting steps with due diligence and care. This mechanism is only considered possible for the by sample by sample This mechanism is only considered likely for the contamination of sample mechanism is only considered possible for the contamination of sample 3 Action Fix Type | Changed Process by sample possible for the contamination of samples Contamination of samples contamination of sample Action Complete | 09/12/2008

Task Details

alternative to the adhesive seal and an alternative resin mixing procedure). This modified procedure is undergoing extensive

contamination events and concerns and improvements etc. that surround the automated DNA IQ extraction procedure have

been discussed at various departmental and team meetings.

verification and approval from the DNA Analysis management team must be obtained prior to re-introduction. The

extraction procedure prior to reporting will be carried out. Every DNA result obtained from these samples will be interpreted

with caution. Modifications have been made to the automated DNA IQ extraction procedure (including the use of an

batch basis. Additionally, careful review of results obtained from samples processed through the automated DNA IQ

No Tasks found

Follow-up And Approval

14/04/2009 10:42:25 AM Helen GREGG: Follow-up Status Follow-up Status Comment

Accepted

Hi Helen

I was wondering if you, or delegate, could accept the follow-up on the above OQI. Kylie Rika is away on mat leave and I need it to be closed out to print for a casefile.

Thanks

Justin Howes

Approver

14/04/2009 Approval/Rejection Date

14/04/2009 11:20:48 AM Paula TAYLOR:

Comment

Approval/Rejection

All changes to the automated process and validation of the new set-up will be detailed in a final report. Acceptance of results results, to detect whether the integrity of each sample can be verified. Retesting can be conducted on identified samples which may confirm information in the original results. Where the integrity of the results cannot be confirmed, the results will checklist, which includes, but is not limited to, the comparison of all other results from samples processed alongside these is made by reviewing the results and assessing a number of factors as per the Forensic Reporting and Intelligence Team be reported as a quality control failure.

Associations

No Associations found

Records

No Records found

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JH-49

Appendix 1

Checklist 1 - Results required for court, no time for additional work

NB: this checklist only applies for circumstances in which TIME is the critical factor, such that there is no time to undertake further quality investigations to verify the integrity of the result.

- Were the samples run though DNAIQ?
 - o No
- Release results
- o Yes
 - Go to 2
- 2. Is this the only sample for case?
 - o Yes
 - Go to 3
 - o No
 - Go to 4
- 3. Is the result first run (9PLEX) as opposed to a rework result?
 - Yes
 - Full profile, does not match any other profile on extraction batch release result. Comment in speci notes.
 - Profile matches something else on the batch from a different case. Need to investigate the circumstances of each case and document as possible contamination event. In this case it is safer to report as 'failed to pass QC'.
 - Partial profile (>12 alleles?), does not match any other profile on extraction batch – release result. Comment in speci notes.
 - If mixed DNA profile, and can either condition on known reference sample or call both major and minor, and profiles do not match any other profile on extraction batch – release result. Comment in speci notes.
 - Mixed DNA profile where the major or conditioned can be called confidently, but the minor/remaining is partial and not unique on the extraction batch

 report major, and report the remaining/minor as 'insufficient for RELIABLE interpretation'
 - Complex mixed DNA profile 'too complex for RELIABLE interpretation'
 - NSD results can be reported
 - Partial profile results can be reported if it matches to the ref sample of the owner/donor of the exhibit, or if

consistent with the other profiles obtained from the same item

 Any PP that is evidentially significant and less than 12 alleles should be reported as 'failed to pass QC'

o No

- NSD can be reported, PP insuff can be reported
- If a PP >12 alleles has been obtained on the first run and this has been reworked to give a full profile and the full profile is unique to the batch then the result is OK to report
- Anything else needs to be reported as 'did not pass QC', unless all other samples on batch have been reworked to fullest potential and has passed batch macro
- Reamps or ReGS simply to confirm OLA, Al and other similar analysis issues can be reported for 'yes' with caution
- Reamps with significant amp vol difference (ie to improve amt of information within profile) should be reported as 'did not pass QC'
- 4. Are the samples on the same extraction batch?
 - o No
- Report as for 'yes' point 3.
- o Yes
 - NSD result can be reported.
 - Any profile that matches donor/owner ref sample can be reported
 - Full profile, does not match any other profile on extraction batch release result. Comment in speci notes.
 - Partial profile (>12 alleles?), does not match any other profile on extraction batch – release result. Comment in speci notes.
 - All samples first run from same item match each other (and from staining), greater than 12 alleles, profile doesn't appear anywhere else on batch can be reported
 - Others refer to examples listed below.

Examples to illustrate 4 (yes).

Extraction batch has samples with profile matching complt. On same batch, samples from deft have been extracted, and a mix of the two is amongst the results off that batch. The mixture cannot be reported as it is not possible to tell if the mixed result is true or the result of the complt sample contaminating the deft sample.

N	
	Extraction batch contains samples from 2 defts clothing. All samples show match to complt within case. These results cannot be reported (without resampling) as it is not possible to tell if the results are a true reflection of the samples, or if the samples from one deft (true results) have contaminated the samples from the other deft.

JH-50

Appendix 1

Checklist 2 – Results for which there is time for reworking/resampling, but not time to wait for full investigations to be completed.

- 1. Were the samples run though DNAIQ?
 - o No
- Release results
- Yes
 - Go to 2
- 2. Is this the only sample for case?
 - Yes
 - Go to 3
 - o No
- Go to 4
- 3. Is the result first run (9PLEX) as opposed to a rework result?
 - Yes
 - Full profile, does not match any other profile on extraction batch – release result. Comment in speci notes.
 - Profile matches something else on the batch from a different case. Need to investigate the circumstances of each case and try to confirm profiles from spin baskets/lysates/resampling. If profiles cannot be confirmed then document as possible contamination event and report as 'failed to pass QC'.
 - Partial profile (>12 alleles?), does not match any other profile on extraction batch – release result. Comment in speci notes.
 - If mixed DNA profile, and can either condition on known reference sample or call both major and minor, and profiles do not match any other profile on extraction batch – release result. Comment in speci notes.
 - Mixed DNA profile where the major or conditioned can be called confidently, but the minor/remaining is partial and not unique on the extraction batch—rework spin baskets/lysate or consider resampling. If this does not provide additional information then report major, and report the remaining/minor as 'insufficient for RELIABLE interpretation'
 - Complex mixed DNA profile, rework spin baskets/lysate or consider resampling. – If this does not provide additional information then report 'too complex for RELIABLE interpretation'
 - NSD If inhibition indicated Nuc spin (and then m'con if PP obtained from n'spin) and confirm with spin

- baskets/lysate. Don't microcon NSD (as unlikely to confirm with spin baskets etc). If cannot confirm any profile obtained, then report as 'did not pass QC'
- Partial profile (<12 alleles?) rework sample (i.e., nuc or micro) in conjunction rework spin baskets/lysate or consider resampling. If cannot confirm the PP, then report as 'did not pass QC'
- PP insufficient can be reported as is. *

o No

- NSD can be reported, PP insuff can be reported
- If a PP >12 alleles has been obtained on the first run and this has been reworked to give a full profile and the full profile is unique to the batch then the result is OK to report
- Anything else needs to be confirmed by profiling of lysate/spin basket/resampling. If profiles are unable to be confirmed report as 'did not pass QC', unless all other samples on batch have been reworked to fullest potential and has passed batch macro
- Reamps or ReGS simply to confirm OLA, Al and other similar analysis issues can be reported for 'yes' with caution
- Reamps with significant amp vol difference (ie to improve amt of information within profile) should be reported as 'did not pass QC'

4. Are samples on same extraction batch?

No

Go to 'yes' 3

Yes

- NSD result can be reported.
- Any profile that matches donor/owner ref sample can be reported
- Full profile, does not match any other profile on extraction batch – release result. Comment in speci notes.
- Partial profile (>12 alleles?), does not match any other profile on extraction batch – release result. Comment in speci notes.
- All samples first run from same item match each other (and have been obtained from staining), greater than 12 alleles, profile doesn't appear anywhere else on batch can be reported
- If samples from different items, or are < 12 alleles, gave matching profiles go back to spin baskets/lysates or resample to confirm profile
- If samples gave different profiles that are unique to the batch then they can be reported

- If not confident in results, go to 5.
- Others refer to examples listed below.

Examples to illustrate 4 (yes)

Extraction batch has samples with profile matching complt. On same batch, samples from deft have been extracted, and a mix of the two is amongst the results off that batch. The mixture cannot be reported as it is not possible to tell if the mixed result is true or the result of the complt sample contaminating the deft sample.

Extraction batch contains samples from 2 defts clothing. All samples show match to complt within case. These results cannot be reported (without resampling) as it is not possible to tell if the results are a true reflection of the samples, or if the samples from one deft (true results) have contaminated the samples from the other deft.

Extra notes

- 1. When considering whether there is any remaining sample to submit consider remnants from sub-sampling carried out in analytical and unstained smears.
- 2. Submitting spin basket for re-extraction may provide some information to confirm the profiles obtained from the original extraction batch. For example, if a full profile was obtained, and only a partial profile is obtained after microconning from the spin basket, then this confirms the DNA in the original extract is actually the correct profile. The results can be reported.
- 3. Submitting sample from STOR plate for extraction will only exclude contamination at the elution step, not the lysis or seal removal step, it may provide more information about the plate. If the extraction was processed before March 19th (the implementation of off-deck lysis) the STOR plate will provide the confidence that results are correct. Need to check if these have been sealed post extraction

^{*} This course of action is recommended for samples where there is only time for reworking within the case. This might not be the recommended course of action where the fuller investigations of extraction batches are concerned.

JH-51

Investigation Team

Alicia
Jacqui
Shannon
Julie
Rhys
Angelina
Claire

Ingrid Amanda Kylie Emma Paula

Routine Team

Hung Rebecca Helen Kate Susan B Susan G Deb Josie Penny

JH-52

Implementation Date	Details	Project Leader	Area Affected
14/11/1991	HLA Dq alpha	SMJ (retrospectively added)	Date of first workbook entry used as implementation date
23/01/1995	D1S80, HUMTH01, FES and sex typing	SMJ (retrospectively added)	Date of first workbook entry used as implementation date
31/05/1995	Polymarker	SMJ (retrospectively added)	Date of first workbook entry used as implementation date
1/03/1996	Triplex: CSF1PO, HUMTH01, TPOX	SMJ (retrospectively added)	Date of first workbook entry used as implementation date
23/04/1996	Quad: vWA, HUMTH01, F13A, FES	SMJ (retrospectively added)	Date of first workbook entry used as implementation date
16/12/1997	Sensitivity test for 9plex (Profiler Plus)	SMJ (retrospectively added)	Date of first workbook entry used as implementation date
5/03/1998	First CS entry for 9plex (Profiler Plus)	SMJ (retrospectively added)	Date of first workbook entry used as implementation date
	Prior to routine use of NC DD, the lab used the Unknown Database in FACTS. These unknown profiles were manaually searched against the Known and Unknown databases in FACTS	CJA (retrospectively added)	
2001	The Known Offenders Database was used to store and search all ref samples. All unknown crime scene samples were manually searched against KOD as the case was completed. All unknown crime scene samples were still loaded to FACTS to search against other crime scene samples.	CJA (retrospectively added)	
9/10/2003	All profiles on the Known Offenders Database were checked by the Queensland Police Service to be included on NC DD and those profiles were bulk uploaded to NCIDD on the 9th of Oct 2003	CJA (retrospectively added)	
10/10/2003	All profiles on the datases in FACTS were bulk uploaded to NCIDD on the 10th of Oct 2003.	CJA (retrospectively added) PMT	
2nd half of 2004 (b/n Jul and Aug)	Date added to the reviewed field (LRCB1-7) for EXR and LKR testcodes	(retrospectively added) CJA	
Jun-04	Implemented use of Quantifiler for routine use for quantitation	(retrospectively added) CJA	
8/04/2005	Report issued by ESR regarding review of the use of Quantifiler Human Quantitation System at QHSS	(retrospectively added) CJA	
Jun-05	Western Australia began using NCIDD	(retrospectively added)	
15/09/2005	On Thurs the 15th September 2005, QPS RMU advised us that they no longer require Forensic Regiser case details registered on LKREXT pages and faxed to them.	Luke Ryan	Results Management Volume Crime
Oct-05	Northern Territory began using NCIDD	CJA (retrospectively added)	
9/03/2006	For cases registered 05 onwards it has been decided that to create links the case files are not required but only if the admin and tech reviews have been completed. If the admin and tech reviews have not been completed a check of the information in the case file is required therefore the case file is required to generate the link. This also means that the link sticker is not required on these case files.	Luke Ryan/Maryanne Hadfield	Results Management Volume Crime
13/06/2006	Discussions took place between VKI, MMH and CJA regarding staining of Diff Lysis slides with H&E, and the possible changeover to Christmas tree staining. It was decided that no staining of slides from this extraction type would be done, and if required, they would be stained at a later date (they will still be heat fixed).	Cathie Allen/Maryanne Hadfield/Vanessa Ientile	Analytical Section
23/06/2006	Amp volumes changed from 2ng to 1ng (as per DNA workbook). To help Case scientists with amp volumes of casework samples, it has been	Paula Taylor	All
27/06/2006	decided we will now add to the samples with multiple volumes with D1, D2 etc depending on the number of replicates so they will now appear as 12345_15ul_D1, 12345_17ul_D2 so it will be obvious which samples have multiple volumes when plate reading.	Allan McNevin	Analytical Section
28/06/2006	Change over from Quantifiler standards to Promega Male standard DNA for Quant standards and Promega Female standard DNA for Quant controls (high and low). This will take effect from run id QF#850 onwards (Amp id CW#1320 onwards). From the results of the validation work that has been undertaken this will give us a more reliable quantitation value.	Allan McNevin	Analytical Section
17/07/2006	The analytical section will submit one Quality Control spot per extraction batch for blood and cellular extractions. There is no need for individual case scientists to submit QC spots after the 17th of July 2006.	Allan McNevin	Analytical Section
17/07/2006	The operational staff will only process FTA samples on a "Link' plate singularly. Previously all samples were punched in duplicate, however since staff are dedicated to this task on a roster, the requirement for duplicates is no longer required Project No Longer Required.	Cathie Allen/Luke Ryan	Volume Crime Team
20/07/2006	From the 10th of July 2006 - the initials 'RLS' refer to Rebecca Sonter. A previous staff member Rebecca Smith also had these initials and worked in the section from the 13th of September 2004 to the 5th of August 2005. Any entries in Auslab (RLS) during those dates refer to Rebecca Smith. Any entries after the 10th of July 2006 refer to Rebecca Sonter.	(Brought to CJA's attention from RS)	Forensic Biology
24/07/2006	The analytical section will register Epithelial Lysates in Auslab. A copy entry will be done of the Sperm Fraction registration, '-E' will be added to the Client Reference field and 'from [barcode of Diff Lysis sample]' will be added to the Sample Info 1 line. The team will also be added to the 9plex page.	Cathie Allen/Allan McNevin/Mary Gardam	Analytical Section/Quality

Implementation Date	Details	Project Leader	Area Affected
25/07/2006	Change over from Quantitating samples in duplicate to singles. This will take effect from run id QF#898 onwards (Amp id CW#1388 & CW#1389 onwards). This is the second part to changes arising from validation/change management work that had previously been undertaken.	Allan McNevin	Analytical Section
26/07/2006	Re: Use of Greiner plates for FTA processing. Item removed from register as change did not actually occur.	Allan McNevin	Analytical Section
25/07/2006	Started using In-house prepared Diff Lysis controls, results to be monitored in G:\ForBio\AAA Analytical Section\Diff Lysis Controls\Diff Lysis controls 2006.xls	Allan McNevin / Justin Howes	Analytical Section
7/08/2006	PDF functionality for FBSTAT, FBSHRT, FBADD & FBAMEND is live from 8/8/06	Vanessa lentile / Sam Granato	Major Crime & Volume Crime
8/08/2006	MPREM page created in Auslab for removal of identified missing persons profile off NC DD (Refer to SOP 17152)	Kirsty Wright	Volume Crime Team
8/08/2006	EXR2 & EXR3 live in AUSLAB for use when results extend past 7 lines of data. EXR2 & EXR3 information is not transferred across the interface but must be faxed from EXR2 to QPS FIRMU.	Vanessa lentile	Major Crime & Volume Crime
31/08/2006	Stop keeping 17050s after upload for samples with and without links after page has been scanned. $ \\$	Luke Ryan	Volume Crime Team
30/08/2006	Increase of amping template from 1ng to 1 2ng. This is effect any CW from CW#1484 onwards and CoREF#32 (CoREF#33 and CoREF#34 were done using 1ng of template)	Allan McNevin / Megan Harvey	Analytical Section
19/09/2006	Addition of "-QC" suffix to Auslab barcodes of QC dots in DNAmaster	Allan McNevin	Analytical Section
6/10/2006	Moved BRB Stats v1.26 and added a password requirement to open. Made the following changes to v1.23, this version only to be routinely used from now on - Write-protected file - Protected all fields except those required for profile entry and kinship type - Renamed columns "Father", "Mother", "Child" in Paternity Trio module to "Unknown Parent", "Known Parent", "Known Child" to clarify	Tim Gardam (for Vanessa lentile)	Major Crime & Volume Crime
23/10/2006	After transferring the completed folder to AAA Results Finalised, the file must be renamed to remove the # eg. CW#1632 to CW1632. (To make the files more compatible for the new database)	Mary Gardam	All Plate Reading
26/10/2006	All statements in Auslab have been pdf'd by the admin team and VKI. There are some exceptions where difficulties have been encountered (see list below) Note that some have been accidentally pdf'd twice during this process. Please be aware if printing off any old statements. Justin/Sam are to now progress change of Stat Dec title to Statement of witness, use of new NATA logo and the inclusion of the statement appendix version 4	Sam Cave	Major Crime & Volume Crime
	PDF STATEMENTS - AUSLAB WOULD NOT VALIDATE		NO DETAILS ON REPORT PAGE SSF018929
		ı	ASUF###### entries - not done cannot be done cannot be done
30/10/2006 1/11/2006	Statement Appendix version 4 - ready for use and should be used for all statements written as of Monday 30th October All Statements are now called 'Statement of Witness' and the NATA logo has	Sam Cave Cathie Allen	Major Crime and Volume Crime
	been changed.	CJA	Ferrorio Biolo
Dec-06	Commonwealth of Australia began using NCIDD	(retrospectively added)	Forensic Biology
22/01/2007	Analytical section will now include a blank/negative control to all Nucleospin clean-up and microcon batches consisting of 100uL nanopure autoclaved water	Allan McNevin	Analytical Section

Implementation Date	Details	Project Leader	Area Affected
31/01/2007	For older cases (02/03/04) that have been sample in 2005,2006 and 2007, it has been decided that to create links, the case files are not required BUT ONLY if the admin and tech reviews have been completed. If the admin and tech reviews have not been completed a check of the information in the case file is required therefore the case file is required to generate the link. This process to change from 31/01/07. SOP to be updated.	Paula Taylor	Volume Crime Team
12/02/2007 13/02/2007	AUSLAB Batch functionality GO LIVE date Pre PCR MultiPROBE II instrument GO LIVE date	Thomas Nurthen Thomas Nurthen	All Forensic Biology teams All Forensic Biology teams
3/04/2007	For blood samples received by Property Point staff - the new test code BBCLO is ordered & filled out. The staff no longer have to add 'Blood sample' to the registration screen in the Sample Info 1 line. Biology staff trying to find out if a sample is a blood need to look for the BBCLO testcode.	Thomas Nurthen/Cathie Allen	Forensic Biology
5/04/2007	All statements are to include the ref sample source (eg buccal cell, blood, or hair) in the body of the statement to ensure that all items rec'd are documented as to what rec'd	Sharon Johnstone	Forensic Biology
5/04/2007	New lists have been configured to assist in workflow with volume crime cases that have been sampled and results are pending.9PLEXV- (Volume 9plex list) will be the list that you will go to, to assess if all the results are back for a case. If case is ready to go it will progress to the next list. If not it will just be taken off the list to wait for the last sample outstanding VRAPP- (Volume result appraisal list) This list is to be used by results management to look at samples and assess for reworks. If rework required, just taken off the list. If rework not required preferred profiles are selected CCVOL- (Case compilation list) This list you will use to find the files, print results tables, print all EPG's and track physically and electronically to case appraisal drawers.	Sharon Johnstone	Initially Voulme crime and operational officers
10/04/2007	All Quant batches to have results entered in QF QC log, with a column added to standards tab to indicate batch pass/fail. Log of failed amp bathces created, located I:\AAA Analytical Section\Test Results-AmpsTE\FailedAmpBatchLog2007.xls	Allan McNevin	Analytical Section
18/04/2007	"Items received" and "Items tested" fields on FORREC pages in AUSLAB (forensic receipts) are no longer mandatory fields. You are no longer able to edit these fields.	Vanessa lentile	Volume Crime Team/ Major Crime Team/Property Point
3/05/2007	Use of receipt barcode and item number (e.g. 123456-001) in statements is optional. May not be required if there is only one receipt issued. Exhibit barcode must be used to identify the item in the results section of a statement.	Vanessa lentile	Volume Crime Team/ Major Crime Team
6/06/2007	New Allelic Ladder incorporated into ABI Profiler Plus and Cofiler kits. See report I \AAA Analytical Section\(3100 \) Troubleshooting\(Allelic \) ladder trial May 2007\(New Allelic \) Ladder Report ver 1.doc Profiler kits from lot number 0703109 (in-house lot M received June 07) will have the new ladder	Allan McNevin	All Forensic Biology teams
4/06/2007	Use of word "neg" rather than "-" for any descriptions in any casefiles eg. TMBneg rather than TMB Can use "+" or "pos" as the "+" is harder to change than a "-".	Kylie Weller	All Forensic Biology teams
8/06/2007	Change to NCIDD introduced - NT profiles are now viewable.	Vanessa lentile	Volume Crime Team
15/06/2007	Pre-Lims completed Genotyper files were stored in I:\User3100\AAAResultsFinalised\PRE-LIMS. Due to storage restraints on I drive many of these files are now stored in J:\User3100\ResultsFinalised\PRE-LIMS.	Cathie Allen	All Forensic Biology teams
18/06/2007	Started using the Generic Interface (between Auslab and the Forensic Register)	Cathie Allen	All Forensic Biology teams
19/06/2007	Change to LKR reporting for Partial profiles. Statistical calculations will no longer be reported in an LKR.	Paula Taylor	Volume Crime Team
19/06/2007	DLYS' added to the specimen notes of SLYS and ELYS samples to allow reworking responsibility to be given to the Yellow Team Case Managers	Justin Howes, Allan McNevin	Major Crime Team (Sexual Assault Team) and Analytical Team
21/06/2007	Started using Criteria (and actions for failures) for Extraction controls as outlined in SOP24012	Allan McNevin	All Forensic Biology teams
26/06/2007	Started using Criteria (and actions for failures) for Amplification controls as outlined in SOP17130 (includes minimum average peak height for positive control ≥1200RFU at D3 and ≥600RFU at D7) based on data to be found I \AAA Analytical Section\AMP RFU monitoring\Amp RFU monitoring 2006-Jan2007.xls	Allan McNevin	All Forensic Biology teams
26/06/2007	Analytical logs (including Quantifiler logs) have been moved to I:\AAA Analytical\Analytical Logs, TG has updated the required Q'filer results macro	Allan McNevin / Tim Gardam	Analytical Section
28/06/2007	Interpol/Interstate requests will now report 9 loci matches as a match instead of "cannot be excluded as a contributor"	Paula Taylor	Volume and Major Crime Teams
29/06/2007	ALL crime scene profiles will be uploaded to NC DD. Registering of parent barcodes in Auslab into the sub-sample registration page -	Vanessa lentile	Forensic Biology
13/07/2007	end of Sample Info field. Interim fax page to be used on 9plex2 page to send to QPS FIRMU for all sub-samples used in final EXR results.	Cathie Allen	All Forensic Biology teams
TBA TBA	Networking of 9700 Thermalcyclers to PC Uploading of Thermalcycler log into AUSLAB	Thomas Nurthen Thomas Nurthen	All Forensic Biology teams All Forensic Biology teams
TBA	Linking of QIS 24486 to the 9PLEX,9FTAR,9L NK,COF L,COFILR screen masks and accessed via the SF1 function	Thomas Nurthen	All Forensic Biology teams
17/07/2007	Below threshold peaks are no longer to be interpreted during plate reading. Eg partial profile with a below threshold mixture is to be reported as a PP. The case scientist is responsible for interpretation.	Mary Gardam	All Plate Readers & Reporting Scientists
27/07/2007	Property Tag on exhibits have been changed - they now include information such as 'seized date' and 'receiving officer'. The font of other fields have been enlarged. The top of the tag now says 'Forensic Biology' instead of Exhibit Tag.	Janine King	Volume and Major Crime Teams
1/08/2007	Began matching with SA, Tas and ACT on NCIDD	Paula Taylor	All Forensic Biology teams

Implementation Date	Details	Project Leader	Area Affected
1/09/2007	Staff matches for samples will only be performed by readers and will no longer be	Magan Hansay	All Favancia Dialogy to ome
1/08/2007	checked by the analytical section when importing profiles to AUSLAB.	Megan Harvey	All Forensic Biology teams
1/08/2007	Partially inhibited samples when imported will no longer be automatically reamped. A communication will be sent to the team stating that the sample is partially inhibited and for the cs to order a rework (either reamp or n'spin cleanup).	Megan Harvey	All Forensic Biology teams
Sep-07	Victoria began using NCIDD	CJA (retrospectively added)	
5/09/2007	Volume Crime will no longer retain the print outs from NCIDD for person to person links. Only those matches involving a reference sample we are repeating for confirmation will be retained.	Paula Taylor	Volume Crime
6/09/2007	The fix for EXRs was activated in the LIVE system of Auslab. The fix ensures that only reviewed results are sent across the interface (and unreviewed results will not transfer across the interface if 'REV' has not been selected). The act of entering REV ensures that it transfers across the Generic Interface.	Cathie Allen	All Forensic Biology teams
20/09/2007	Pre-2003 Volume Crime Cases that have "No Testing Required" in their case status will remain an NTR regardless of whether or not there are UR notes to justify their status. ACTIONS: 1. Still need to enter NWQPS into the EXR page for any exhibits in NTR cases. 2.Find an empty storage box & create a storage location. 3. Store exhibits to the newly created box and transfer the entire box to FBEXR1. There is no longer any need to transfer exhibits to the NTR box. SOP WILL BE UPDATED TO REFLECT THIS CHANGE	Sharon Johnstone Kate Lee Robyn Smith	Operational Officers / Volume Crime
19/09/2007	Batch Functionality menu items changed to remove reference to DNA	Thomas Nurthen	Analytical Section
19/09/2007	Prompts added to AUSLAB for removing/returning samples in Batch functionality	Thomas Nurthen	All Forensic Biology teams
19/09/2007	Modifications to the SF7 Results History screen format in AUSLAB Columns Width increased, Specimen type now displayed	Thomas Nurthen	All Forensic Biology teams
19/09/2007	AUSLAB-SF11 Batch Allocation, Batch Creation and Batch details print out to display all storage locations	Thomas Nurthen	Operational Officers /Analytical Section
24/09/2007	Changes to the general mask GENREF mask	Thomas Nurthen	All Forensic Biology teams
24/09/2007	Changes to the general mask MICROCON (Processing Comments)	Thomas Nurthen	Operational Officers /Analytical Section
24/09/2007 24/09/2007	Changes to the general mask QUANT (Processing Comments) Changes to the general mask AMP (Processing Comments)	Thomas Nurthen Thomas Nurthen	Operational Officers /Analytical Section Operational Officers /Analytical Section
24/09/2007	Changes to the general mask NUCC (Processing Comments)	Thomas Nurthen	Operational Officers /Analytical Section
24/09/2007	Changes to the general mask NUCB (Processing Comments)	Thomas Nurthen	Operational Officers /Analytical Section
24/09/2007	Changes to the general mask NUCI (Processing Comments)	Thomas Nurthen	Operational Officers /Analytical Section
24/09/2007	Changes to the general mask NUCT (Processing Comments)	Thomas Nurthen	Operational Officers /Analytical Section
24/09/2007	Changes to the general mask HAIR (Processing Comments)	Thomas Nurthen Thomas Nurthen	Operational Officers /Analytical Section
24/09/2007	New general mask ReGenescan (REGS) Examination of items - Scientists are now requried to wipe their gloves with		Analytical Section
25/09/2007	ethonal wipes before conducting tape lifts.	Samantha Cave	All examining sceintists
27/09/2007	Print mask configured for FTAR test	Thomas Nurthen	All Forensic Biology teams
27/09/2007	Additional Export analysers configured for Amplifications batches and Amp split	Thomas Nurthen	Analytical Section
27/09/2007	macro cease to be used Removal of Stor mask from the bottom of the MPQUANT mask and quant split	Thomas Nurthen	Operational Officers /Analytical Section
27/09/2007	macro ceased to be used Autovalidation rules configured to Save preferred profile automatically if the Comment is OK (GENOT,FTAGEN & LINK masks)	Thomas Nurthen	Analytical Section
27/09/2007	Autovalidation rules configured to write the Batch D into the 2nd page of 9PLEX, 9FTAR and 9LINK test panels	Thomas Nurthen	Operational Officers /Analytical Section
27/09/2007	Microcon autovalidation mask to auto allocate to the Quant batch type if Finvol >26 Verification of new 9700 thermal cycler (9700-F) complete and used for	Thomas Nurthen	Analytical Section
3/10/2007	routine use, report located in I \AAA Analytical Section\Internal Projects\9700 Block F 2007 Routine use agreed upon by management team in meeting Thurs 27-9-07 (no comments received to date)	Allan McNevin	Analytical Section / Opertaional staff / All sections
10/10/2007	FBEB9-14 (For Biol Examination Boxes) created. These are examination boxes in the stainless steel examination area.	Justin Howes	Major Crime, Volume Crime staff
15/10/2007	Digital Imaging functionality for storage and annotation of images	Gail Hargraves	All
15/10/2007	Packages for all examined volume crime swabs, cig butts and small items photographed and destroyed after sampling. Applicable to all items received from 15/10/07	Sharon Johnstone	V/C, Operational
19/10/2007	From 10/10/07 Rebecca Sonter(RLS) has changed her name to Rebecca Gregory(RLG). Her AUSLAB login has changed from rlsf1 to rlgf1 on 18/10/07.	N/A	All
29/10/2007	First DNAIQ batch extracted on the MPII	Thomas Nurthen	All
8/11/2007	From 29/8/07 Josie Hayward (JEH) has changed her name to Josie Entwistle	N/A	All
3/12/2007	(JE). Her AUSLAB login has changed from jehf1 to jeef1. Volume Crime Low priority cases will no longer have a separate case status or EXR lines, to use started and standard EXR lines	Sharon Johnstone	Volume crime
6/12/2007	As of 5/12/2007 Kylie Weller 's new name is Kylie Rika . Admin Support, LISS, and IT, have all been informed to change my details to Kylie Rika and initials KDR.	Kylie Rika ne Weller	All
11/12/2007	FBEXS661 and FBEXS662 to be used for DLYS slides	Justin Howes/ Rose Higgins/	YT, RT, OO, AS, PPT
12/12/2007	LISS have changed my login from kdwf2 to kdrf1	Amanda Storer Kylie Rika	All
	When Copying a registration using the SF5 Copy entry function on the Registration	•	
13/12/2007	screen; The DNA priority will now copy to the new barcode.	Thomas Nurthen	All
14/12/2007	Two people required for destruction of reference samples process	Paula Taylor	V/C
7/01/2008	Started using Decapper (LifeTool Recap 96M), will be used for decapping & recapping tubes in Pre-PCR from today onwards	Allan McNevin	Analytical / All
7/01/2008	Started using thermalcycler networking software	Allan McNevin	Analytical / Operational / All
7/01/2008	From 7/01/08 Angelina Gilbert (AG) has changed her name to Angelina Keller		All
110112000	(AK). This name change is not yet effective in AUSLAB or QIS.	NA	All
14/01/2008	Crime scene profiles from Sexual Assaults only which match to complainants reference samples will not be uploaded to NCIDD. All other crime scene profiles will be uploaded to NCIDD.	SJC	Major and Volume

Implementation Date	Details	Project Leader	Area Affected
23/01/2008	FBEXA2 and FBEXA3 are pages to request when further examination of items is	Justin Howes	Major and Volume
	required to track bench, time, tests etc as per FBEXAM. Begin routine use of 3130x/A. Will begin with Reference batches then progress to		·
6/02/2008	Casework (9+1 protocol).	Allan McNevin	All
11/02/2008	Begin routine use of 7500 for quantification Change HiDi / Rox mixture from 85:1 to 85:2 (i.e. use 850µL HiDi + 20µL Rox or	Allan McNevin	All
19/02/2008	935μL HiDi + 22μL Rox). Data contained within 3130x/ upgrade project folder (9+1 protocol).	Allan McNevin	Analytical
21/02/2008	ABI7500 mask fixed, STD7 now says "0 0686ng/uL" so no need to manually fix STD7 on the ABI7500 SDS software.	Iman Muharam	Analytical
13/03/2008	Switched over from using Barnsted water purifier to new Millipore Milli-Q A10 Advantage system	Allan McNevin	Analytical / Operational / All
18/03/2008	Put "FTP" in comments field for all FTA specimens that have failed to profile.	lman Muharam	All
18/03/2008	During upload of Genotyper results into AUSLAB, manual changes are made for samples that have "FTP" in the comments field. Save the FTP profile as preferred, and change DNA profile result from "Positive" to "Negative", then validate the page.	Iman Muharam	Analytical
18/03/2008	Plate readers to source plates to be read each day from Blue "plates to be read" folder located on AS PO4's desk. Plate readers to take plates from the top working down. It is also up to the plate readers to use the batch audit function in AUSLAB to track the whereabouts of the paperwork.	Allan McNevin	All Plate Readers
18/03/2008	For specimen types CELLS, BLD, QAB, QAC initial batch type was changed from CWIQEXT to CWIQLYS. For specimen types BRE, QABR, QACR, SPOT, RCELLS the initial batch type was changed from RFIQEXT to RFIQLYS. This is to allow processing of samples through DNAIQ with off-deck lysis.	Iman Muharam	All
19/03/2008	Start Routine Processing of DNAIQ extraction using Off-Deck Lysis procedure	Allan McNevin	All
26/03/2008	Changes have been made to the Destruction Screen (DEST) in AUSLAB in preparation for enhancements to the Generic System Interface for Forensic Biology.	Vanessa lentile	Volume Crime
26/03/2008 26/03/2008	The changes include 1. Fields that are Mandatory are now displayed with an asterix "*".	Vanessa lentile Vanessa lentile	
26/03/2008	Display of the fields for Authorising Officer, Badge ID, QPS Sample Status. Please note that these fields will be populated by the GSI and so will remain blank for existing samples.	Vanessa lentile	
26/03/2008	3. Two new fields for "PROFILE REMOVED FROM AUSLAB:" and "PROF LE REMOVED FROM KINSH P:". Please note that for existing samples, the registration needs to be re-saved so that these fields are displayed correctly	Vanessa lentile	
11/04/2008	Batch types for capillary electrophoresis have changed from 3100 batches to CE batches to reflect the start of the use of 3130 instruments. e.g. batches that previously would go to 3100CWYYYYMMDD_## are now CEPCWYYYYMMDD_##. CE=capillary electrophoresis, P=Profiler, CW=casework.	Thomas Nurthen / Allan McNevin	All
22/04/2008	Started to routinely replace SDS with Sarcosyl in the DNAIQ extraction lysis protocol due to gel formation with SDS (does not occur with Sarcosyl).	Thomas Nurthen / Allan McNevin	All
28/04/2008	Resumed routine use of ABI Prism 7000 with update software version (to match the 7500) and new T/C block	Allan McNevin	Analytical / All
8/05/2008	Received the first volume crime exhibit by Australia Post	Cathie Allen	Property PointVolume Crime
	When using a profile from a previous run the comment is "USE GEN9REFXXXXXXX_XX" only. Other comments eg. OK, PP, Al cannot be included		
14/05/2008	with this comment. The preferred profile should be saved in AUSLAB (<f7> <sf6>), the profile upload form printed (<sf11>), which both readers should sign</sf11></sf6></f7>	Susan Gillespie/ Paula Taylor	Plate readers
19/05/2008	and include with the plate paperwork. ROBOTICS login increased to 20 allowable simultaneous logins (Analytical requires 13 logins, including the BSD Duet 600).	Iman Muharam	Analytical / Operational
27/05/2008	New Primary Site available called "DNA Trace Kit" with mnemonic "trace". This will be used for trace DNA tapelifts submitted directly from police. Date for	Vanessa lentile	All
21/03/2006	commencement yet to be defined. Introduced within the Forensic Register 20th May 2008 When requesting a ReGenescan of samples through L MS, a second person will be	variessa leriule	All
28/05/2008	required to check the correct 9Amp batch (and sample position number) has been correctly entered into the 9Plex processing comments, and add a specimen note indicating this has been checked (currently we are unable to use the ReGenescan processing comments section on 9plex page)	Shannon Merrick	All
12/06/2008	MIXT & MIXC will now copy the profile from the 9PLEX and put it in the Profile found fields on the mixture page	Thomas Nurthen	Major/Volume
20/06/2008	Commenced using 3130xl Platform B for routine use.	Allan McNevin	Analytical / All
	Sequence check, Pos control lot number and thermalcycler fields added to AUSLAB amp batch worksheets.		
25/06/2008	Transfer/Pool worksheet now outputs whole sample info field& processing comments (rather than truncating)	Allan McNevin	Analytical
26/06/2008	New EXR line - Mixed profile, unable to load-NC DD. Ref sample req'd is now available for use in Auslab.	Paula Taylor	Major/Volume
1/07/2008	New specimen types available in AUSLAB: QPST, EFRAC, SFRAC, DDNA, HDNA, PTISS, CPOOL, CTRAN, CSUP, MICCON, NSCON, FSS.	Thomas Nurthen	All
1/07/2008	New Primary Sites available in AUSLAB: CFAB, BFAB. This will be used for fabrics containing cells and bloods respectively, submitted directly from police.	Thomas Nurthen	All
11/07/2008	CWGENP and CWGENC prefixes added to Main module in Results Table Comparison macro to enable comparions for new samples post-01 July.	Tim Gardam / Iman Muharam	All

Implementation Date	Details	Project Leader	Area Affected
14/07/2008	For microcon samples where the final volume is less than $20\mu L$, TE will be added to make the volume up to $20\mu L$ at the end of the microcon batch. E.g. if the final volume after concentration is $7\mu L$, $13\mu L$ of TE will be added to the extract tube. This will be reflected in the results file, whereby final volume will be the volume after concentration (e.g. $7\mu L$) and the SV1 (amp volume) will always be $20\mu L$. Comments have been made to the SOP in QIS	Allan McNevin	All
14/07/2008	In order to facilitate trouble shooting, extraction platform A will be used to process reference batches only and extraction platform B will be used to process casework batches in a checkerboard configuration. This will consist of samples and extraction blanks as communicated to the Biology management, Analytical and Operational teams via email. This will be done from 14/7/08 until sufficient time as for trouble-shooting has been completed and any issues detected resolved.	Allan McNevin	All
23/07/2008	Further information has shown adverse events have occurred equally across platform A and platform B, therefore casework and reference processing will not be limited to either instrument. The checkerboard pattern mentioned above will be continued	Allan McNevin	All
28/07/2008	Missing audit trail data from January 2008 in AUSLAB cannot be restored from back up tapes as these back up tapes are not available. If required for audit trails, missing data up to the 31/12/07 and after January 2008 can be restored.	Vanessa lentile	All
28/07/2008	Outcome from extraordinary managament meeting 28/7/08 - suspend processing of samples through automated DNAIQ. Samples that are already lysed (i.e. processed through off-deck lysis) will be extracted through manual DNAIQ procedure. All extractions from 28/7 will be through chelex process until resolution of DNAIQ troubleshooting	Allan McNevin	All
7/08/2008	CSRC list enabled. FSLU to list insert if a Case Manager or Case Scientist is required due to case of high priority/media interest, or serious enough to warrant a contact point and close attention.	Justin Howes	Reporting and Intel
7/08/2008	CASEFC list enabled . This is a list to contain cases that are complete in terms of examination and results and is requiring case compilation and peer review.	Justin Howes	Reporting and Intel, Operational Staff
11/08/2008	Once profiles have been imported to AUSLAB, AS will manually list insert each barcode from the Genotyper batch (excluding Controls) onto the following lists according to the team name entered into the TEAM field on the 9PLEX page: no team - BLCM Yellow team - YELCM Blue team - BLUECM Red team - REDCM Green team - GREECM Volume team - VOLCM Orange team - BLCM This process will be in place until it can be done automatically by AUSLAB	Allan McNevin	All
15/08/2008	Any samples that have the "STAFF" flag shown by the auto-validation rules when importing profiles into AUSLAB will be noted by the person importing the results file in the specimen notes the wording used will be "Staff flag X/Z alleles". Where X/Z represents the number of matching alleles out of possible alleles	Allan McNevin	All
18/08/2008	New batch type configured "Casework manual Amp Profiler" (mnemonic CWMAMP). This batch type will be used to manually amplify sampels that have less than or equalt to 20µL of extract remaining after microcon.	Allan McNevin	Analytical / Operational teams
21/08/2008	Case managers to go through checklist as set out by working party to ensure there is no evidence of contamination at levels below what would have been detected by the extraction audit. Case manager to enter Specimen Note 'Quality Checklist Passed/Failed' for each sample that has been checked. There will be a new process in ordering reworks for Volume Crime samples only effective immediately. This means no MICROCONS or NUCLEOSPINS are to be	Justin Howes	Reporting and Intel
21/08/2008	ordered for Volume Crime samples only. Reamps can be ordered if the first 9Plex was amped at less than 20uL. Re-genescans can be ordered as normal. This means no MICROCONS or NUCLEOSP NS are to be ordered for Volume Crime samples only. Reamps can be ordered if the first 9Plex was amped at less than 20uL. Re-genescans can be ordered as normal. If 12 alleles are obtained and is loaded to NCIDD and eventually linked, a rework by any means can be ordered to get the best match probability possible for	Justin Howes	Reporting and Intel
25/08/2008	statement purposes. The following line will be added to statement preblurbs in line with NATA guidelines. 'This document is issued in accordance with NATA's accreditation requirements'.	Justin Howes	Reporting and Intel, Quality
27/08/2008	As of this date, Qld will match with NSW - thereby meaning that Qld matches with all states and territories.	Intell Team	DNA Analysis
29/08/2008	All microcon to full samples will have a final volume of 27uL. Case scientist's will only order microcon to full or microcon to half.	Megan Harvey	All
4/09/2008	FTAGEN mask - new autoval rule, looks for comment of 'REGS', adds RRREF	Thomas Nurthen	All
4/09/2008	Dilution Volumes - changes to stop 9FTAR reworks overwriting dilution volumes	Thomas Nurthen	All
4/09/2008	EXTRES mask - new autoval rule, looks for comment EL NK, returns to QUALNK batch type	Thomas Nurthen	All
8/09/2008	REQC result added to 9PLEX cummulative table, REQR added to 9FTAR cummulative table	Thomas Nurthen	All
11/09/2008	Syringes on MPII EP-A changed from 500µL to 1000µL. As of 10 September, all DNAIQ results to be put on hold. Urgent results for statements to be worked on using macro, final results for samples on batches,	Iman Muharam	Analytical
19/09/2008	autofilter function of spreadsheet - any that cannot be reported can be either reported in addendum (if committal) or quality failed (if trial or no other opportunity for investigative analysis).	Justin Howes	Reporting and Intel
22/09/2008	Syringes on MPII EP-B changed from 500μL to 1000μL.	Iman Muharam	Analytical
22/09/2008	OO staff to send information about Evidence samples to 1EVD (as opposed to 1BT,1VT,1YT,1RT,1GT) as one collection point for this information.	Justin Howes	Reporting and Intel, Operational Staff

Implementation Date 26/09/2008	Details Manufactured Phadebas paper has been sourced and is to be used within the laboratory. OO staff are no longer required to prepare Phadebas paper.	Project Leader Adrian PIPPIA	Area Affected Operational Staff, Evidence Recovery Staff, Reporting and Intel
26/09/2008	To facilitate process changes as part of GMIDX, whenever registering a Positive Extraction Control please use the following processing comments as appropriate:		
	extpc (extraction pos control cell) extpb (extraction pos control blood) extph (extraction pos control hair) extpe (extraction pos control epithelial) extps (extraction pos control semen) this allows GM- DX to recognise what control it is and checks whether the correct profile has been obtained Negative Extraction Controls (extn) stays the same	Allan McNevin / Thomas Nurthen / Anne Finch	Analytical & Operational teams
	Start using these processing comments from the very next control you register (from 26/9/08)		
1/10/2008	To facilitate process changes as part of GMIDX, whenever registering an Extraction Control start using the "Analyt" team name when registering controls in AUSLAB Start using these processing comments from the very next control registered (from	Allan McNevin / Thomas Nurthen / Anne Finch	Analytical & Operational teams
1/10/2008	1/10/08) BTEAMS and CSCI added to FBEXAM screen mask	Thomas Nurthen	All
6/10/2008	Separation of FRIT team into Routine Team (dealing with new cases and those extracted using chelex) and Investigation Team (dealing with cases processed using IQ). This separation was required in order to effectively deal with the IQ cases as these need to be investigated and interpreted with caution followed by the results either being released or failed. Process for acceptance of these results to be added to the Case Management SOP.	Emma Caunt	Reporting and Intel
8/10/2008	Interface enhancements with the Go-Live of Generic System Interface. Some minor cosmetic changes with the pages as EXRs will be phased out, and EXH pages will be the standard. New 'summary' page also.	LISS	Reporting and Intel
15/10/2008	All files with statements including statistics generated from Kinship to be reviewed. As of 15/10/2008 the data is correct. All stats prior to 15/10/2008 need to be checked. There may be discrepancies in the Asian figures.	JAH	Reporting and Intel
10/11/2008	New pre-blurb to be used for statements where QPS have done examinations/sampling. This new pre-blurb can be found in the FRIT folder in the G drive.	KDR	All reporting scientists
17/11/2008	Microcon procedure From today on, the following processing comments will apply Microcon to full - samples will be concentrated to as low a final volume as possible. This final volume will be entered into the "final volume" results field. An appropriate volume of TE buffer will be added to make the final volume of the extract to 20µL. The SV1 volume will therfore = 20 Microcon to 30 and Microcon to half - samples will be concentrated to as close to 30µL or half the initial extract volume as appropriate	АМ	All
10/12/2008	From today- when ordering Cease Work tests, also change the priority to "5". Note, cease work tests are to be ordered as a rework through the SF7 results history table.	АМ	All
	As a consequence of the first results obtained from experiments looking into the NucleoSpin cleanup efficiency this week and resultant management team discussions this morning the management team has decided to:		
11/12/2008	place the NucleoSpin cleanup procedure on hold any super-urgent samples that really need a clean-up then person requesting will need to discuss with JAH who will liaise with AM to have it processed A write-up of data (already gathered) to validate using the DNAIQ kit to perform a cleanup procedure - this won't be ready until 2009 Ongoing investigation into NucleoSpin cleanup procedure (including verification of the 1st experimental results and further testing to investigate route cause etc.)	АМ	All
	this is also going into the minor changes register (effective immediately)		
	Reagents Module now active in AUSLAB after overnight live release,		
12/12/2008	How to enter the instrument used Access the batch <f6> "Reagents" <f7> "Equipment" Type the mnemonic if you know it or use <f1> to access the look-up table <enter> then <f4> to save - this is the critical step, if you don't save, you'll just have to do it again then <sf5> to go back the main menu for the batch, make sure the reagents and other information is manually entered into the audit trail you will now be able to complete the batch</sf5></f4></enter></f1></f7></f6>	TEN / AM	Analytical and Operational Teams
	For batches such as PORTAL where there is no actual equipment used, there will be a piece of "equipment" called "software" For batches where all samples are removed (e.g. batch created in error) there will be some sort of "null" type option to choose		

Implementation Date	Details	Project Leader	Area Affected
·	Work-around for samples on an extraction batch with processing comment of "nuc after extraction" - until the cleanup investigation / processes are resolved	·	
	Analytical will, after performing the extraction		
15/12/2008	change processing comment to "hold after quant - nuc" and samples will progress to quantification (they will get quanted only) and will be removed from the amp batch allocation list (i.e won't get amped)	AM / JAH	All
	the barcodes will be e-mailed to AM (and then sent on to JAH) and JAH will make an assessment (and consult the CS where necessary) whether they a) can proceed with amp b) need cleanup but can wait c) need to be cleaned up now		
16/12/2008	There is a new process for when you want a profile removed from NCIDD: The case manager/reporter is to enter a speci note to say why sample needs to be removed from NCIDD. Print this speci note out and put in the NCIDD tray. Fill in the EXR with the removed from NCIDD EXR but DON'T validate - the NCIDD person will do this after they have removed the profile from NCIDD. Every week, there will be one day/week that the NCIDD people do removals. If you feel your removal request is urgent then please bring your speci note in person to someone in the intell team to action straight away for you.	KDR/Intell	All case managers and reporting scientists
23/12/2008	Consolidation of batch types in AUSLAB Configuration in AUSLAB so that casework samples, when registered, all go to batch types starting with "Casework XXX" (mnemonic will be CWXXX) New UR number for extraction controls for 2009:	TEN / AM	All
	FBOT404 - Neg Control		
5/01/2009	FBOT405 - Cell Control FBOT406 - Blood Control FBOT407 - Diff Lysis Control FBOT408 - Semen Control	AM	All
21/01/2009	FBOT409 - Hair Control From the next batch of cell extraction controls onwards, CJA will be the control profile (replacing VKI). Additionally, swabs will be used in place of buccal FTA punches. New SOP in draft awaiting final approvals QIS doc 25874 for the creation of extraction controls.	АМ	All
30/01/2009	Nucleospin clean-up procedure re-instated as of Monday, 2nd Feb 2009. Results from the investigation into the efficiency of the Nucleospin clean-up procedure showed that an improved recovery of DNA was evident when the B5 buffer was made fresh, at the commencement of the extraction process. Thus, this will be made as part of the Nucleospin clean-up protocol. Kandom Match Propability Wording for Statements in DNA Analysis: It is	LWC	All
5/02/2009	estimated that the probability of this DNA profile occurring, had the DNA come from someone other than, and unrelated to xxxxxxxxxxx is approximately one in yyyyyyyyy using Queensland Caucasian data. The genotype frequency, which is used in RMP reporting, is calculated based on unrelated individuals. This rewording is in response to a NATA condition. To be used in all statements as of today. Discussed at FRIT meeting 05/02/09.	JAH	FRIT
12/02/2009	From 12/02/2009 Megan Harvey (MLH) has changed her name to Megan Mathieson. This change is effective in groupwise and novell but is not effective in	NA	All
13/02/2009	AUSLAB or QIS yet . Redirect CE batches to GeneMapper batches .	CW/TEN	All
16/02/2009	Started using GeneMapper to analyse results . Implemented new thresholds . Reference: 50 RFU het, 150 RFU hom, Al 70%.	CW/TEN	All
16/02/2009 16/02/2009	Casework: 50 RFU het, 200 RFU hom, Al 50%. EPGs being attached to lab numbers in AUSLAB.	CW/TEN CW/TEN	All All
16/02/2009	The Intelligence Team will be batch uploading suitable crime scene samples from AUSLAB (however, mixtures and certain other samples will still require manual upload). With these new processes and with the implementation of Genemapper, the database upload form will still be required, however: - If the sample has been run through Genemapper and the EPG is present in AUSLAB then you do not have to attach the EPG to the database upload form. - If the sample has been run through Genotyper then the EPG will have to be	LAW	FRIT
	attached to the database upload form. The Intelligence Team will no longer be assessing each profile prior to upload to NC DD. It is the responsibility of the case scientist (and reviewer) to ensure that the correct profile has been nominated to upload to NCIDD, and that any analysis issues have been confirmed New draft SOP in plate - 23890R3		
16/02/2009	The Intelligence Team will be batch uploading all person samples through AUSLAB for all plates run through Genemapper. Outstanding plates (pre-Genemapper) will be processed through Datagate. New draft SOP in plate - 23890R3	LAW	All
23/02/2009	Z399UR3 The names of staff members in the Staff Match Macro have all been changed to be their respective FBSTF number.	IS	ALL
2/03/2009	Minor changes to the Chelex method for the extraction of Blood from Crimescene samples have been incorporated from today according to findings arising from Change Proposal#46 (main change is the use of Prot K has been incorporated).	АМ	All
3/03/2009	Analytical will analyse all negative controls using a 20 RFU labelling threshold from today. Previous negative controls are not being reanalysed.	CW	ALL

Implementation Date	Details	Project Leader	Area Affected
	GeneMapper results file fix macro now staff matches DNA Analysis staff, checks for alleles labelled with 'OL' and single allele calls at a locus. For reference batches the macro also compares profiles within the batch to each		
4/03/2009	other. DNA Analysis staff will be deactivated in AUSLAB. QPS Staff matching will continue to occur in AUSLAB. In QIS as Draft - 26045R1	CW/TEN	ALL
6/03/2009	Inactivated DNA Analysis Staff in AUSLAB database as Staff match will hold all DNA Analysis staff GeneMapper <i>ID-X</i> re-read test codes configured and available in AUSLAB for	TEN	All
10/03/2009	samples run on the 3130x/ Genetic Analyzers. This is any sample on a Genotyper batch since 10 03.08 Test codes are GMCW and GMREF.	CW	ALL
11/03/2009	Samples with EXH and FBEXAM now correctly print the Client reference on the report	TEN	All
19/03/2009	Category A-C paragraphs are to be inserted into any statements issued with DNAIQ extractions using automated platforms. These paragraphs are available in the Forensic Reporting and Intelligence team folder/Reporting Guidelines Major Crime cases will transfer across the interface as "M" - seen in Auslab as Medium Priority; Volume Crime Cases will transfer across the interface as "L" -	JAH	Reporting Scientists
6/04/2009	seen in Auslab as Low Priority; Urgent cases will transfer across the interface as "H" - seen as High Priority in Auslab. Previous cases that are registered will not be changed.	CJA	FSLU, Proprerty Point, DNA analysis
8/04/2009	For crime scene samples loaded to NC DD through batch uploading - the database upload form will be discarded and any actioning/link details will only be noted on the batch upload worksheet and therefore the database upload form will not be scanned into AUSLAB.	LAW	FRIT
	As per previous information, QPS will add 'N' to Forensic Relationship field when they no longer need that item tested. This can happen at any time through the process.		
	If pre-Started status, FSLU will close off the case.		
	If Started status or beyond, the item barcode will be sent to either personal comms (if allocated) or to the FBNLR worklist for Paula and Sharon's attention (FSLU will not add a line to the EXR). ER know about the 'N' and will not sample items with the 'N' added. Cease work (CWORK, no further and priority 5, self validate) will need to be added if it is post extraction/ pre-amp. Post-amp will continue and the profile (or no profile) will be obtained. Therefore, it is important to check personal comms regularly to ensure we are capturing the samples no longer required quickly.		
23/04/2009	If the case has some with 'N' and others without, the case is still active and all profiles will be interpreted as if they were all required. Obviously if you can stop these samples before amp, then that is what you do but if it has progressed through amp, rework as per normal case mgt (treat as a normal sample). Sometimes these cases will require a statement and it is best practice to have all samples complete to the best standard our lab can provide. Also, this practice is in the interests of full disclosure of results that the lab has obtained.	JAH	FRIT, FSLU
	A workflow is being drawn up by PMT/JAH to assist – this will include Auslab and physical locations for the casefiles. The intention eventually is to incorporate FBNLR monitoring into a roster.		
	For DNAIQ cases, the same rules apply. Remember to add a UR note that the case is closed off (case is no longer active (all 'N's') and that the investigation has not been conducted. Add to the spreadsheet that the case is complete and a comment in the appropriate column.		
	Above strategy agreed by Mgt Team 23 April, 2009.		
	As of Mon 27 April, the GREEN colour will no longer be added to cases by FSLU. This will mean BLUE, YELLOW, RED and VOL will be the only colours used by the lab. ORANGE and PURPLE will not be used in any situation.		
27/04/2009	This means FSLU will look at the 'CASE TYPE' and if it is ASSAULT (S) irrespective of the type of sexual offence, it will be given YELLOW colour. BLUE will be given to mutders and coronials. RED will be given to all other major crime cases and VOL will remain as volume. There will obviously be a lag for this to be complete, with auto-population of CM lists still occurring until all GREEN case management is complete. The PSA reporting phrase in the statement preamble has been changed to reflect the findings in a report released detailing other substances that can produce a positive result with the Biosign kit. EXR/EXH lines to change to be consistent with statements. The paragraph is:	JAH	
14/05/2009	The presence of semen on an item can be indicated by the use of a presumptive chemical test which detects a major constituent of seminal fluid (Acid Phosphatase – AP). This constituent does, however, exist in other body fluids, such as vaginal secretions. Additional presumptive chemical testing (Prostate Specific Antigen – PSA) can be undertaken and a positive reaction to both AP and PSA makes it highly likely that seminal fluid is present. The presence of semen can be confirmed via the microscopic identification of spermatozoa (sperm heads).	JAH	

Implementation Date	Details	Project Leader	Area Affected
4/06/2009	Interim examination for the presence of Seminal fluid: t has been noted that the "sensitivity" of the new PSA kits are slightly decreased. This has been verified somewhat through a small comparison with the expired PSA kit and the new PSA kit with fresh 1:4 diluted semen. Testing however, is ongoing and the supplier has been notified. As such, the use of PSA kits (719D11) has been suspended pending further investiagtion and the following exam strategies will be employed: -Micro Neg and AP pos = DLYS (examine DLYS slides of samples post DNA analysis if applicable) Micro Neg, AP Neg, no further action. EXR line will be "Presumptive seminal fluid pos. Submitted results pending" for such situations. - There is a worksheet for case managers/reporters to put the sub-sample barcode and QPrime # requiring microscopic exam of the DLYS slide (G:Forbiol\AAA Evid Recov\Diff Lysis slide exam). Further, information will be communicated as results arise.	AAP	
4/06/2009	The red line, indicating PPE is to be worn within the designated area, has been revised in the small sampling room (Rm 6115). The red line transition now commences at the end of the first set of small cupboards to the left, upon entry to the room as opposed to the end of the second set of small cupboards on which the photocopier sits, situated on the left hand side.	AAP	
10/06/2009	The PSA kits (719D11) are to be re-implemented for seminal fluid examination following recent investigations into the sensitivity of the new batch. Details of results	AAP	
16/06/2009	are available in report "PSA Kit Investigation" New Pipette calibration program in use	TEN	DNA Analysis
16/06/2009	Queensland Government introduced use of a single logo identifier for departmental activity.	Director-General	DNA Analysis
17/06/2009	As requested, the code below has been removed from the Modify 8 equation today if (CASEST = "NOTRQ" & !test_validated(CASEST)) listinsert("BNTR");	TEN	DNA Analysis
17/06/2009	An additional quality check is to be added to the first link review and second link review process. This applies for post-L MS samples only. The extraction batch for any post-L MS samples involved in a link will be checked to see if any of the linked cases appear on that extraction batch (for evidence samples the associated CRISP number/s will be used to check on any crime sample's extraction batch). If any cases are found to be involved in the same link and also processed on the same extraction batch then this link will be given to the Senior Scientist of the Intelligence Team for further investigation.	LAW	Intelligence Team
18/06/2009	To order a Person Sample sent to Kinship: - Now you DO NOT order an INCNAM in AUSLAB if you want a person sample sent to Kinship - you now order a KINSHP (note that the second "!" is missing). - You still enter an NCNAM of "default". For the page to validate you will also need to enter a Category of "KAM" or "KPM".	TG	
19/06/2009	Chelex extraction of DNA from casework samples (with the exception of Differential Lysis, Hair, & Semen extractions) will be replaced with manual DNAIQ from Monday 22/06/09.	АМ	All teams
19/06/2009	Blood, cell, QPST and FSS specimen types now allocate to CWIQEXM HP5's are able to sign as the Delegate on all Application of Leave Forms for staff	TEN	All teams
1/07/2009	from their team. [HP6 will sign the Application of Leave forms for 2 x HP5's and the Admin team] This is a retrospective communication to state that the rayon swabs have been	CJA	
2/07/2009	received from QPS as in-tube samples for approximately a month; since June 09. The 4N6 swabs are still being received.	AAP	
9/07/2009	VOLLOW worklist exists to collect samples that are from Volume Crime Low Priority cases (Case Priority 3). At the moment, samples can be manually inserted onto this list - a formula will be released in the not too distant future.	JAH	
9/07/2009	FERRO creation updated = The 'contact person' field drop down list has now been configured to show a number of departments and sub-sections that can be selected, including "DNA Analysis - Evidence Recovery". When creating a FERRO relating to an exhibit non-conformance please complete this field. This will allow QPS to contact the appropriate person with enquiries about a FERRO received.	AAP	
13/07/2009	As of today Abigail HOULDING will go under the in -tray FBSI36. I have confirmed with Tom N that we are fine to use this in-tray as of today and LISS will re-configure.	AAP	
16/07/2009	As of the 20-07-09 sub-samples from whole exhibits (excludes QPS tubes received) will be submitted in the 2ml or 1 5ml tubes to Analytical for processing and not placed within a 5ml tube for transfer. The 2ml or 1.5ml tubes will be barcoded (which is current process) and cleansed prior to sending to Analytical. SOPs will be amended in due course if applicable.	AAP	
16/07/2009	The Methods Page in Auslab will no longer be used. If already filled out, this page can be validated as is'. If not filled out, the testcode can be deleted.	JAH	
17/07/2009	As of 17-07-2009 all of the data within DNA Datagate has been transferred into the DNA Analysis Database (DAD). Datagate data will now be accessed through DADI (DNA Analysis Database Interface) and DNA Datagate will no longer be used.	LAW	
24/07/2009	As of the 27-07-09 all volume crime tubes and whole item sub-samples received prior to and including 16-08-09 will be processed as Priority 4 and have the 9plex substituted with POLD. All volume crime in-tubes and whole item sub-samples received on the 17-08-09 and onwards will be registered as Priority 3 and will have a 9plex requested. All Major crime in-tubes and whole item sub-samples will be registered as Priority 2 have a 9plex requested. This process is no longer current. All samples registered with a 9plex.	AAP	
24/07/2009	As of 24-07-2009 all negative controls processed within the laboratory will be interpreted at CE Quality Checking stage with a 16 RFU labelling threshold in GeneMapper D-X v1.1.1.	CCW	All teams
28/07/2009	Formulae functional in Auslab to direct VOLUME cases of Priority 1 and 2 to VOLCM worklist, and Priority 3 cases to VOLLOW.	JAH	
28/07/2009	Paperless casefile receipts can be tracked to FBP1 -10 (Paperless 1 to 10) located in the write-up area. These storage racks can allow 500 entries each.	JAH	

Implementation Date	Details BLUEREV has had its name changed to VCREV (Volume Crime Review) to	Project Leader	Area Affected
31/07/2009	assist with paperless case mgt.	JAH	
6/08/2009	A new batch type has been created for all samples prior to 17-08-09 CW HOLD .	MLM	All teams
6/08/2009	Any samples that need to have a priority changed that is on the CW HOLD batch type please list insert sample onto list GRM , so analytical staff can asign the sample to the next batch type. The wording for the priorities has now been changed Priority 1 High priority has changed to 1- Urgent Priority Priority 2 Medium priority has changed to 2 - High Priority	MLM	All teams
6/08/2009	The mnemonics for these has not changed as LISS indicates that it may effect certain lists. This does not effect Property Point as the priority is automatic from QPS.	PT	
6/08/2009	Please note the following changes to Evidence Recovery protocols as of Monday the 10-08-09: - casefiles will no longer accompany the examining scientist during sampling - examination strategy will be available in the UR notes for each case, resolution/amendments and notification of completion of examination (by way of	AAP	
6/08/2009	initials and date of examiner) will be completed in the UR notes in AUSLAB - The "Project detsruction box" used to retain samples not processed for DNA Analysis will be renamed to " tem Retention Box ##"	AAP	
11/08/2009	The mnemonic's for CWIQLYS and CWODL have been switched. Now CWIQLYS is for Casework IQ Off Deck Lysis and CWODL is for DNAIQ Lysis CW.	MLM	Analytical / all
11/08/2009	New reference DNAIQ Extraction (IQEXTREF) and Off Deck Lysis (IQLYSREF) batches have been created to process reference samples on the automated platforms.	MLM	Analytical / all
14/08/2009	FBSI30 intray name changed from KCO intray to Paperless Intray Extraction platform B has been implemented for the automated DNAIQ™ extractions of volume crime casework samples. These samples are being	JAH	
20/08/2009	extracted in a checkerboard format of samples and negative controls. Samples and controls on an extraction batch will be processed together through analytical. The first batch of samples has been extracted on the instrument today.	CCW/MLM	Analytical / all
21/08/2009	Old ForBiol Structure folders burnt to 3 DVDs (3 copies) located in CD rack on shelf in Admin area.	JAH	
28/08/2009	As of Monday the 31-08-09, all tubes which are to be manipulated are to have an FBX with the examination bench and equipment box (if applicable) entered. The time taken to perform the examination is not applicable.	AAP	
28/08/2009	As of Monday the 31-08-09, bleach is to be used as part of the decontamination procedure of utensils used during examination. First step is to place utensils into a vial of bleach, then manually wipe with toweling; second step is to place in ethanol and then finally burn excess ethanol off over bunsen burner.	AAP	
7/09/2009	New casefile review worklist established- STATREV. Cases with statements for review are to be list inserted and tracked to STAREV location (physically in main write-up area). Slips to go on front of file with boxes to check depending on 'grading' of case.	JAH/AJR	
11/09/2009	Boxes of files at PRSENT stored in decon/drying room have been relocated to the exhibit room shelves: FBEXS43 to 70 and 85-98.	JAH	
24/09/2009	From 24/09/2009 Claire Perrin is changing her name over to Claire Gallagher . This change is effective in AUSLAB, Groupwise, Novell and QIS2 as of today. AUSLAB: cpf2 is now cgf1. (Still waiting on AUSLAB lists)	LAW	All
24/09/2009	Effective immediately, a new process is to be implemented when reading slides. For sperm counts <1, two vernier readings are to be recorded aswell as being observed, signed and dated by a second sampling scientist. In addition, the two screws at the back of the microscope platform are to be tightened prior to using the microscope and again prior to recording the vernier reading. For any questionable sperm, a senior scientist is to be called in to make the final call.	MJC/JAH/PT	
1/10/2009	FTALO is allocated to FTAOSD, FTALR is allocated to FTARUN, FTA RPT is allocated to FTARPT, FTA LNK is allocated to FTARPT. This has been changed from the Automated DNAIQ extraction allocation.	CCW/TEN	
6/10/2009	Negative control UR changed from FBOT404 to FBOT501 as the upper limit of numbers of samples that can be ordered has been reached	TEN	All
6/10/2009	FBPR45-55 have been created for casefile storage.	JAH	
12/10/2009 14/10/2009	FBCM23-26 have been created for casefile storage In-tube boxes containing registered but unreviewed or unmanipulated tubes that require storage overnight will now be stored and tracked to the relevant day's shelf	MJC AKL/AM/PT	Evidence Recovery / Analytical
21/10/2009	in the Exhibit Room and not the freezers in Analytical. New manual receipts are being utilised at Property Point. Carbon copy type receipts are no longer being used and superseeded by QIS form "Forensic Receipt", 26584V2.	AAP	
9/11/2009	Photocopiers are no longer to be used as part of examination of items. The use of photographs uploaded to AUSLAB has superseded this form of recording.	AAP	
25/11/2009	A new Auslab worklist, MIXEXR , has been created for paperless cases. Samples with mixture results will be inserted on this list by the case manager for review. The MIXPEER list sorts on date received so that results can be validated in order of receipt.	KL	Intelligence Team
30/11/2009	When PCR product (Post L MS) is discarded a batch audit entry will be entered against batch.	MLM	
30/11/2009	New team structure within FRIT commenced. Reporting 1 (AJR senior), Reporting 2 (EJC Senior) and Reporting 3 (SMH/ M Seniors). No change to Intel. Some workflow changes using lists and drawers to better manage reworks implemented.	JAH	

Implementation Date	Details	Project Leader	Area Affected
1/12/2009	LISS have made some slight changes to our CM lists to help out with prioritisation. The lab no. are now sorted on a new column which records when the 9Plex was first ordered. The DNA priority column has been deleted as all samples on these lists are P2. The AMP2CW and AMP3CW have been replaced with RRCW and GMCW as these are more frequently used. The sample will still populate the list if an AMP2CW or AMP3CW are ordered, it is just that they won't display in a column (the AMP1CW will always be displayed anyway). These apply to BLUECM, VOLCM, REDCM, YELCM and BLCM.	JAH	
8/12/2009	The small sampling room (Room 6115) will no longer be used for the examination of items. It is now a semi-clean yellow line only room where gloves are the only form of PPE required. t will be used for the photography of packaging for in-tube items.	RGM	
8/12/2009	The in-tube process has been amended so that each tube is wiped with bleach and ethanol when it is removed from the CSSE.	RGM	
9/12/2009	Evidence samples will no longer be added to the 1WPP list. They will be added to the 1EVD list or the communications list of the case scientist if applicable.	RGM	
10/12/2009	Interim lines to be added when ordering reworks. Final result will be updated not incorrect lines. Linked no. field not required to be filled in. Lines to be reviewed by a second scientist.	JAH	
14/12/2009	During the link process those person samples that have been run through Genemapper (REFGMP batches) will have their JPEGs uploaded to AUSLAB rather than being printed and scanned.	KL	
24/12/2009	Forensic Receipt = AUSLAB has now been configured to only print one copy of the receipt. As such no receipts will be transferred from Property Point. If a receipt is required for the case file then simply print one.	AAP	
4/01/2010	New UR numbers for extraction controls were made for 2010. Negative control FBOT 516, Positive Control Blood FBOT 517, Positive Hair FBOT 518, Positive Dif Control FBOT 519, Positive Semen Control FBOT 520, Positive Cells Control FBOT 521.	CCW	All
19/01/2010	Extraction platform A has been implemented for the automated DNAIQ™ extractions of casework and reference samples. The first batch will be extracted today.	CCW/ARM	All
19/01/2010	The plot setting in GMIDX for printing profiles has been changed from "Scale to Maximum Y" to "Scale individually". The settings have been changed for both plot views "Print Profile" and "Print Profile Size". The appearance of printed profiles and PDFs should slightly change to zoom closer for profiles.	CCW	All
21/01/2010	automated DNAIQ extraction procedure: the STORE plate (containing supernatant retained after binding with magnetic beads has occurred) will no longer be retained for one month after extraction, it will be discarded immediately after extraction.	ARM	Analytical / all
1/02/2010	Batch filler macro now being used to combine two off deck lysis batches	TEN	All
2/02/2010	Automated DNAIQ extraction batches now have 2x pos (FBOT517), 2 x neg	TEN	All
4/02/2010	(FBOT516), 10x blank (FBOT524) samples, max 76 samples batch New Processing comment configured - ENVM Environmental Monitoring to be used for QPS and DNA Analysis Environmental samples Automated casework extractions, processed as locked batches will progress to	TEN	All
11/02/2010	GeneMapper batches with the batch mnemonic of GMPCW (1st batch ID GMPCW20100211_01). All manually extracted batches and any re-works will be processed through GeneMapper with the old batch mnemonic of CWGMP.	ARM	all
11/02/2010	EB checking commenced on GMPCW batches processed using locked batches. EB checking performed by FRIT members. modified rack 403508484, changed description from "Evid Swabs To Be Sam" to	JAH	
15/02/2010	"Evid Sample - Manual" , started to store evidence swabs in this rack as they arrive	TEN	Operational/Evidence recovery
22/02/2010	instead of the fridge New HR forms - Application for Leave, new RAF ~ now called AVAC form Communication with Bob HOSKIN (chief GMO) has brought around following changes to SAIK collection:	WH	All
24/02/2010	- slides not to be prepared from swabs - corner of swab sheath will not be cut This information has not official become part of the process, as currently under review, and will be via communication to GMOs and FNEs. This change will be	AAP	
24/02/2010	gradual so expect some SA Ks to follow the above changes and others not to. Processing comments added for RRREF & RRCW rework test codes	TEN	All
24/02/2010	Reactivation of CSCI on the 9PLEX,9FTAR,COF L & COFILR test panels	TEN	ALL
24/02/2010 26/02/2010	Changes to FBSTAT - code changes to make statement more efficient Addition of new comms list to highlight Evidence Samples that have to be manually processed by a scientist, eg, swabs, hair, etc. The new list name and Mnemonic is "Evid Samp - Man Process"; "ESMP". OO staff will now use this list instead of 1AAP. This change will coincide with the new storage location ("Evid Sample – Manual" – 403508484, located on FBEXS384) for these types of Evid samples.	TEN AAP	All
3/03/2010	The re-genescan (RRCW) processing comment can now be edited on the 9PLEX2 page. This is to be used (replacing use of specimen notes). Enter the processing comment in the following format: batch id, position eg. CW9AMP20100303_01 Pos 14	ARM	All
18/03/2010	As of today standard size trace DNA kits will not require manipulation (cutting the tape) due to Analytical extraction process changes. Any tape that is received larger than the standard trace DNA kit will however be manipulated and the appropriate EXH line completed (SRMI)	AAP	
23/03/2010	As of 22/03/2010 intell letters are being emailed to G Simpfendorfer and K Gee Kee for interim link results for major crime and other post Sept 2009 samples that are not priority 3. This is a temporary measure until the QAPEND (interim) and QAFINAL test codes are available. Workflow discussed in intell meeting and flowchart given to each intell team member.	KDR	Intelligence Team
24/03/2010	Fix to a fault with GSI, 9FTAR test code will no longer be ordered by the GSI to prevent AUSLAB registering samples that have not been received. Fix also prevents AUSLAB registering a sample with a pathology lab number	TEN	All

Implementation Date	Details	Project Leader	Area Affected
24/03/2010	Fix to the Ammended statement fault- samples with something in this panel can	TEN	ALL
30/03/2010	now be resaved. Insp David Neville, Quality Management Section of QPS advised, in an email, that the QPS Executive had approved the process of posting of swabs and tapelifts	CJA	DNA Analysis, Property Point, SSLU
16/04/2010	from Major Crime offences to commence. As of Monday the 19th of April the registration of tube samples will change with the addition of a new specimen type to distinguish between tube samples that required manipulation, "QPSTM", and those that do not "QPST". Additionally extra primary sites have been configured in order to aid in collating information from AUSLAB when required. Furthermore, FBX is to be added to tube samples regardless of whether a	AAP	
	manipulation is required or not.		
16/04/2010	Part of the documentation of a sample received by the laboratory is the recording of the labelling and sealing method via photography. In the past if a CSSE (crime scene sample envelope) did not have any labelling or sealing information on one side of the packaging a photograph was not routinely taken. With the advent of new smaller CSSEs bearing a template for additional information to be entered by QPS, the majority of samples incurred two photographs one from each side of the envelope. However on the occasion where no additional information is added to one or the sides, no photograph is to be taken. If this instance a specimen note is added (on each individual sample) to state that "photography was not conducted on the reverse side of the envelope due to no additional information"	AAP	
20/04/2010	Fault fixed in Batch functionality. Undetermined Quantitation results were displaying as 0.00, these results will now display as Undetermined. (AUSLAB Q1 release notes)	TEN	All
20/04/2010	Storage location within the exhibit room (DNA Analysis Unit) for tube items, whole items and items "On hold" has changed. Exhibits currently are located on shelves FBEXS22 to 35 and will change to FBEXS696 to 700 and 702 effective immediately. Please note, all shelves have been named to show the location for each sample type (tube Vs whole exhibit, etc) for deliveries Monday to Friday.	AAP	
21/04/2010	Creation of MCREV worklist to contain Priority 1 and 2 samples (major crime and upgraded Volume crime) that are paperless and require review.	JAH	
29/04/2010	Comment for occasion where no photograph of CSSE back is required (see minor change 16/04/2010) has been amended so that it will fit into 1 hot key. New wording is: "No additional information on rear side of envelope - no photograph required"	RGM	
04.05 2010	BOX 2 Extracted DNA Samples DNA# 2488-4919 BOX 3 Extracted DNA Samples DNA# 2488-4919 BOX 3 Extracted DNA Samples DNA# 4920-7345 BOX 4 Extracted DNA Samples DNA# 4920-7345 BOX 5 Extracted DNA Samples DNA# 9776-12242 BOX 6 Extracted DNA Samples DNA# 15013-18012 BOX 7 Extracted DNA Samples DNA# 18013-18012 BOX 7 Extracted DNA Samples DNA# 18013-21012 BOX 9 Extracted DNA Samples DNA# 18013-2612 BOX 10 Extracted DNA Samples DNA# 23613-2612 BOX 10 Extracted DNA Samples DNA# 26213-28812 BOX 11 Extracted DNA Samples DNA# 28813-31408 BOX 12 Extracted DNA Samples DNA# 34109-34808 BOX 13 Extracted DNA Samples DNA# 34109-36808 BOX 15 Extracted DNA Samples DNA# 346809-39407 BOX 16 Extracted DNA Samples DNA# 39408-42107 BOX 17 Extracted DNA Samples DNA# 45200 BOX 18 Extracted DNA Samples DNA# 45201-48200 BOX 19 Extracted DNA Samples DNA# 48201-51400 BOX 20 Extracted DNA Samples DNA# 48201-51400 BOX 20 Extracted DNA Samples DNA# 51404-54600 Validation Project BOX? Unlabelled -Spinbaskets Another BOX? Unlabelled -Spinbaskets BOX 21 Tubes 57601-5900 BOX 22 Tubes 57601-5900 BOX 25 Tubes 66601-69400 BOX 26 Tubes 66901-72600 BOX 27 Tubes 72601-75600 BOX 27 Tubes 72601-75600 BOX 28 Tubes 75601-78600 BOX 29 Tubes 75601-78600 BOX 29 Tubes 76801-81600	Quality	All
4/05/2010	If the case is paperless, the FBAR page will be N/A for all fields except the RHS top (profiles uploaded and reviewed). The LHS refers to the casefile and as there is no casefile, the fields don't apply (therefore N/A). The paperwork is checked for alterations/additions/corrections as part of the presumptive EXH review. If the case is paper with no statement, the FBAR page will be filled out on the RHS by FRIT (this should already be happening). The file will be sent to peer and a FBPR location. Admin will complete the page numbering, initialling etc. and complete the FBAR page on the LHS. They will write 'yes' for page # on pages, page # on cover, N/A for corrections, N/A for exam notes (these are both tasks completed as part of pres review) and 'no' for results table. If a statement is eventually required, a new FBAR and FBTR will be required. For cases with statements, the reviewer adds 'no' to pg# fields and yes for the rest, track to admin to issue statement. Admin go back to FBAR to change 'no' to 'yes' after they have written the page numbers, initials etc on every page. The audit trail records who changed what fields. The files will therefore not need to return to the peer reviewer – make sure the	JAH	Reporting Scientists
4/05/2010	what helds. The liles will interestore not need to return to the peer reviewer – make sure the green paper slip has to direct to FBCFF1 not back to the reviewer. Paperless case management training complete for FRIT members and this form of case mgt is now incorporated into FRIT rosters.	JAH	FRIT, Admin, ER

Implementation Date	Details	Project Leader	Area Affected
6/05/2010	MCMIX and VCMIX worklists created to capture mixtures requiring review for paperless P1/2 and P3 samples respectively. MIXT & MIXC can be ordered on the following specimen types QAB - Quality Blood	JAH	FRIT, case managers
6/05/2010	QABR - Quality Blood ref QAC - Quality Cells QACR - Quality Cells Ref QAEL - Quality Epi Lys QFTA - Quality FTA QAH - Quality Hair QASL - Quality Sp lys QAS - Quality Sperm	TEN	ALL
14/05/2010	A new ERROR file has been created for each forensic department. The new code for DNA Analysis is ERRDNA0000001. This replaces the previous code of ERRF000001.	JAH	All corrections officers
25/05/2010	As of 26/05/2010 controls (ProHigh, ProMed and ProLow) will no longer be used on quantification batches.	MLM	ALL
31/05/2010	As of 31/05/2010 the intell team is now using a new link workflow involving the use of the LKR (link tracking) worklist. This is to help prevent links being missed/lost and to aid in reducing link TATs. The SOP will be updated but the new workflow can be located at G:\ForBiol\AAA Forensic Reporting & Intel\Intel team.	KDR/TG/Intell	Intelligence Team
31/05/2010	As of 31/05/2010 the Analytical team will be discarding any of the following substrates start after extraction. Chewing gum, cigarette butts, fingernails, toothbrush, straws, extraction positive controls, CTS samples, stamps, envelopes, environmental samples in-house and QPS. Specimen notes will be added against each sample when the substrate is discarded.	MLM	All
15/06/2010	New team structure within FRIT commenced - Reporting 1-3 with defined tasks. No change to Intel. Details in G:\ForBio\nDNA Analysis Team Meetings\Forensic Reporting and Intelligence Team\2010\Apr-Jun.	JAH	FRIT- Reporting teams
18/06/2010	An additional line added to preambles (excluding paternity preamble) of : 'As a representative of the laboratory, I am only able to comment on the processes performed within DNA Analysis.' This is to simply show what we can comment on.	JAH	Reporting Scientists
21/06/2010	Slight change to QFLAG process. Profiles of at least 3 contributors are too complex for QFLAG checking unless significant major profile obtained. If not through QFLAG process, note to specimen and Batch Audit notes that it did not go through the process. SOP updated and in review.	JAH	QFLAG checkers and Case Managers.
23/06/2010	As of today a new version of the Genemapper macro will be in use. Homozygote peaks and low threshold peaks will no longer need to be edited and PDF's will have LPH on profiles that are partial.	MLM	ALL
25/06/2010	The second 7500 instrument for quantification has been put into routine use as well as the new Limit of Detection (LOD) of 0.0021ng/uL and the new Limit of Reporting (LOR) of 0.0063ng/uL.	KML	ALL
2/07/2010	Photography of item packaging by the operational staff has been changed from Rm 6115 to 6110. The change will take effect as of Monday the 05-07-10.	AAP	
6/07/2010	ERT-AS TRANSFER BOXES (1-20) to replace "off deck" and "manipulation" in tube boxes for in-tube processing from 07-07-2010	KDS/AAP	
12/07/2010	Slight changes to complex DNA profile interpretation including establishing criteria to report 'cannot eliminate the possibility' and to state the assumptions of the number of contributors to major/minor profiles.	JAH	Case managers and Reporting Scientists
14/07/2010	Blood, cell, QPST and FSS specimen types now allocate to CWIQEXM instead of rework portal and samples on FTAEXT batches will go to RFIQEXT.	MLM	ALL
19/07/2010	Small change to processing of in-tube samples: sample will now be transferred to the ERT-AS box and the packaging location added to a destruction box after sample registration by Operational staff.	RGM	ALL
22/07/2010	Area Warden for Block 3 Level is Matthew Hunt, Wardens are Biljana Micic and Tyson Kleidon. Controlled Documents are being made to reflect the new positions.	VLC	ALL
29/07/2010	Sign-off by management team for the commencement of routine manual processing in block 3, using room number 3188 for reagent preparation, room 3189 for manual extraction, room 3194 (sorting area) for manual quantification and amplification reaction setup and room 3196 for PCR cycling and CE fragment analysis (Thermalcycler's 9700 A, 9700 B, RT-PCR 7500 A and 3130xl A).	ARM	All
2/08/2010	As of today the Evidence Recovery Team will log in AUSLAB all reagents used when conducting presumptive screening tests. Tackle boxes associated to the Stainless Steel benches in the exam room	RGM	ER, FRIT
4/08/2010	will be stored on top of the benches instead of below the hard drive near the floor. In their current locations, the tackle boxes are accidentally kicked whilst completing computer work and as such a potential for DNA transfer. The change is also a OH&S initiative.	AAP	
4/08/2010	Update to ABI 7500 export mask to remove positive controls	TEN	Analytical
4/08/2010	Sign-off by management team for the use of Thermalcyclers 9700 C and 9700 D for use in room 3196 (new CE/PCR lab in block 3) Quantification Mask updated fit an extra 3 samples, update the DNAIQ off-deck lysis batches to accommodate one extra sample and auto DNAIQ extraction batches extra two more samples.	ARM	All
5/08/2010	·	ARM	Analytical / OO
	Rework PORTAL re-started, slightly modified rules All priority 1 - send to manual DNAIQ All priority 2 - send to manual DNAIQ priority 3 & 4 - send tape lifts to manual IQ, everything else to off-deck lysis.		
6/08/2010	New freezer in exhibit room instead of cold room, new allocations have been assigned.	JSM	
9/08/2010	NCIDD worklist to be used for paper and paperless cases.	KDR/KSL	FRIT

Implementation Date	Details	Project Leader	Area Affected
10/08/2010	Re-work portal - return to standard rules All priority 1 samples & all tape-lift (Trace DNA kit) samples - send to manual DNAIQ allocation list, all other samples send to Off-Deck Lysis allocation list	ARM	Analytical
16/08/2010	Sign-off by management team for the commencement of routine automated processing in block 3, using room number 3188 for reagent preparation, room 3189 for off-deck lysis, room 3191 and extraction MPII A for automated extraction, room 3194 and pre-PCR MPII A for quantification and amplification setup and room 3196 for PCR cycling and CE fragment analysis (Thermalcycler's 9700 E, RT-PCR 7500 B and 3130xl B).	ARM	All
23/08/2010	As of today all consumables used in the manufacture of SAIKs has been transferred to the storage units under Block 3.	AAP	
16/09/2010	Stutter below thresholds outlined in SOP 17137 are to be removed in mixture samples	MLM	All
17/09/2010	Sign-off from the management team for the routine processing utilising extraction MPII B in room 3191, pre-PCR MPII B in room 3194, and thermalcycler 9700 F in room 3196	ARM	All
22/09/2010	New test panel released to LIVE CRREF Cofil ref rerun New batch analysers released to LIVE	TEN	All
22/09/2010	3130FTA - for FTA plates specifically 3130GMCR - for Cofil Reference specifically Updates to batch analyser masks released to LIVE - added Negative control Analysis module automatically for Neg ctls and Environmental samples, Added UR	TEN	All
22/09/2010	number to mask 3130GMP 3130GMC 3130GMR	TEN	All
22/09/2010	New test panel released to LIVE CRRCW Cofil CW rerun Modify 8 equation updated	TEN	All
22/09/2010	SNC DD & FNCIDD should now revalidate after loading Uploaded to NCIDD in Status. If the Comment OK is in the Link Comment, the equation will copy completed and the date into the 9LINK panel	TEN	All
24/09/2010	Ladders and Controls will now be converted to JPEG. And UR details will now be displayed on the PDF and JPEG.	MLM	All
30/09/2010	The test code BBCLO is now orderable on specimen type QABR - Quality Blood Ref.	TEN	All
4/10/2010	FTA boxes have been re-configured so that they can be tracked from shelf to shelf and Property Point to DNA Analysis Unit to enable full tracking of FTA samples	TEN	All
6/10/2010	New result for the BTEAMS result field - mnemonic = ERT & Description = Evid Recov New list created, mnemoninc = ERTL Addition to the modify 8 equation - this code list inserts a sample if they meet the volume list flag 1 or 2 and have ERT as the team onto the Evidence Recovery List	TEN	All
6/10/2010	RFCALC worklist created to replace IAM worklist - Reference Calc Check worklist	PT	
11/10/2010	EVDCM worklist created for evidence samples that require rework - replaces the TEN worklist.	KL	
20/10/2010	FBEXA2 and FBEXA3 areable to be ordered on the QABR specimen type New racks for FTA processing 41327 3019 - FTA TRANSFER 1	TEN	ALL
21/10/2010	41327 3028 - FTA TRANSFER 2 41327 3005 - FTA BENCH 1 41327 2995 - FTA BENCH 2 Work is to completed in the exhibit room by workmen associated with ABI group on the 1st, 2nd and 3rd of November. Alternative arrangements have	TEN	All
29/10/2010	been made for exhibits to remaining in Property Point until required at which time a DNA Analysis staff member will transfer the item and associated paperwork. The workmen will be accompanied by a security person at all times within the exhibit room.	AAP	
3/11/2010	New UR numbers for negative and blank controls have been created. FBOT548 = negative extraction control and FBOT549 = blank extraction control.	MLM	All
8/11/2010	AUSLAB issues hindering photo upload and slowing processing. A back up plan has been devised for times where AUSLAB upload is extremely slow >5mins and is as follows - take photo and upload to folder within I (I \text{VERT exhibit photography}) - add specimen note "AUSLAB upload issues, photo to be entered at later date - photos with then be uploaded when AUSLAB running faster and corresponding package transferred to destruction box labelled "photos not uploaded" (these packages not to be destroyed until photos are uploaded and reviewed	AAP	
10/11/2010	- these photos will be uploaded when AUSLAB is running normally/faster and associated pakages transferred to a destruction box Note = ERT scientists will have to persist with items that require freezer storage, SAIK etc, as it becomes a lot more complicated Fault in batch fucntionality resolved batch audits affected 04/11/10 to 10/11/10 inclusive. For further details regarding this fault, refer to SIR 3805 (Batch Functionality - Unable to enter audit trail entry)	TEN	All
11/11/2010	(LISS Call 2435620) Orford freezer moved from 3190 to 3192, upright freezers moved from 3192 to	TEN	All
	3190		

Implementation Date	Details	Project Leader	Area Affected
12/11/2010	Scheduled electrical outage to Block 6, Level 1 on 13th and 14th of November. Following temporary storage as follows Exhibits stored to freezer locations FBDFS1-4 and return location 447778685 in freezer located in Exhibit Rm transferred to Property Point (FBEXFL)	AAP	
	Exhibits located to Analytical fridge, FBAFL1, in Exhibit Rm transferred to secure, refriferated storage location underneath Block 3 Reagents and controls stored in fridge located in exam rm, 6110, transferred to secure, refrigerated location underneath Block 3 Scheduled electrical outage to Block 6, Level 1 on ZUTT and ZIST of November. Following temporary storage as follows Exhibits stored to freezer locations FBDFS1-4 and return location 447778685		
18/11/2010	in freezer located in Exhibit Rm transferred to Property Point (FBEXFL) Exhibits located to Analytical fridge, FBAFL1, in Exhibit Rm transferred to secure, refrigerated storage location underneath Block 3 Reagents and controls stored in fridge located in exam rm, 6110, transferred	AAP	
22/11/2010	to secure, refrigerated location underneath Block 3 As of today the automated decappers will be in regular use for the Pre-PCR and automated extraction MPII's.	MLM	Analytical
25/11/2010	AUSLAB Corrections List AUSCRL created for corrections eg image deletion & moves	VLC	All
1/12/2010	Evidence Recovery Laboratory clean and environmental sampling has been cancelled due to impending laboratory relocation. Additional lines added to F1 look-up menu for LKR's (LKR1-3)	AAP	
1/12/2010	Mixed Profile, partial Major component. MPPMA Mixed Profile, partial minor component. MPPMI Partial DNA Profile PDNA	PMT	Intelligence Team
9/12/2010	Partial DNA Profile - possible sub-threshold peaks PDPTP 9 loci DNA Profile - possible sub-threshold peaks DPPTP MPQUANT~SS mask and QUANT worksheets have been updated in AUSLAB.	MLM	Analytical
5/01/2011	Samples will now be in positions 18,19 and 20 instead of controls. Faulty printer HP3005 (FBLASER3) replaced by HP 2420n (formally FBLASER8)	TEN	All
10/01/2011	& FBLASER9), reconfigured as FBLASER3 Only use Axygen tubes for extractions, ie. Do not use Simport tubes	CI	
10/01/2011	Component replacement log. An electronic log (I \AAA Analytical Section\Analytical Logs\Component replacement log 2011.xls) has been created for the recording of any components that are replaced on any piece of equipment within the DNA suite of DNA Analysis	ARM	Analytical & Operational teams
12/01/2011	The new Evidence Recovery lab is in use from 12pm 12th Jan 2011. Storage location DAER1 now refers to Rooms 6123 and 6124.	AKL	All
17/01/2011	New Evidence Recovery lab bench and tackle box name changes - examining areas will be labelled Examination Area 1 to Examination Area 15 (rooms 6123 and 6124). The associated tackle boxes will also be labelled the same as the bench it is assigned to eg, Examination Area 1, etc Examination forms will be amended and in the mean time the examining scientist need only write down the examination bench labelling as this will refer to the tackle box as well.	AAP	
21/01/2011	Between the period 12th of Jan 2011 and 20th of Jan 2011 the abbreviation for examination benches/boxes used on exam records may have been EA1 to 15 (denotes Examination Area 1 to 15). This abbreviation has been amended to EX1 to 15 to reflect AUSLAB Mnemonic. Also configurations for the "Examination trolley" filed on the FBX page was not configured commencment of new lab use with the new examination bench results (Examination Area 1 to 15) and as such the old exam box IDs (FBEX13) have been used. Configuration requests for additional results ("Examination area 1" to "15") to "examination trolley" field for FBX2 has also been ordered. Major audit trail incident in AUSLAB, audit trails are lost on some samples or partially available, initial testing indicates that audit trails may reappear after	AAP TEN	ALL
24/01/2011	resaving registration, LISS still investigating for a cause. AK1 - Adam Kaity and BUAF1 - Belinda Andersen to the F1 lookup for test code	1214	/ NEC
28/01/2011	FBSCI.	TEN	All
3/02/2011	Statements are to adopt the following way of writing dates: Date Format After seeking advice, the date format will follow the guidelines from the QH Corporate Correspondence and Brief Guide and Quick Reference Document. Section 7.4 Dates: Correct use is 7 April 2009 not 7th April 2009 or 7/4/09. DPP has also indicated that use of date format such as dd/mm/yy (7/4/09) leads to the possibility of American date format being applied. Time Format A 24 military format of mm:hh will be applied to reports similar to the current AUSLAB time display.	JAH	
7/02/2011	All DWASH~ss lot numbers incorrectly recalled in Consumable inventory in AUSLAB, in use lots re-added with -1 suffix, Consumable audits updated with details	TEN	ALL
16/02/2011	Admin alerted to the report issued page for Evid Certificates . This will now be completed when forwarding Evid Cert. via fax/post. (implemented by FRIT back in Oct 2010)	WAH	
23/02/2011 23/02/2011	New specimen type available in AUSLAB: STAFF New Justices Act 6(C) automatically adds to statements.	TEN JAH	ALL
24/02/2011	Implemented the use of the new BSD Punching machine for routine use New UR prefix - FBUNK - Forensic Biology unknown created in AUSLAB for	TEN	All
24/02/2011	unkown profiles	TEN	All
24/02/2011 28/02/2011 28/03/2011	Unknown FBSTF profiles renamed with FBUNK numbers Generic email address for Intel : FSS DNAIntel Generic comment "for processing" to be used for sample removal hotkeys	TEN JAH MLG	

Implementation Date	Details	Project Leader	Area Affected
4/04/2011	As of today, LKRs will have one review only performed by a competent 2nd reviewer. Intel will also introduce QCPend and QCFinal for reporting links as soon	LAW/KL	Intelligence Team
WC W2011	as they are actioned whilst awaiting the confirmation of person profiles (this eliminates the need to write interim link reports). The CCR 78820 has been released to LIVE 9LINK panel has had the FTALND	22	gooo . ou
6/04/2011	removed so it always populates. Also the LNKCM Lab list rules (Add and Del) have been removed for 9L NK in LIVE.	TEN	
6/04/2011	Samples for cease work will now be given a priority 6. Exhibit brown cardboard boxes to be slowly replaced with plastic, lidded boxes. 16 plastic boxes were transferred to Property Point to commence	MLM	All
12/04/2011	usage. These plastic boxes are to be bleach and ethanol cleansed at the Evidence Sorting room, 6117, stage and then again when transferred to main exam room 6124/6123.	AAP	
14/04/2011	Start to use UNKM or UNKF for QPS environmental samples that do not match QPS staff, DNA Analysis staff or exhibits. Due to limited supply of strip septa used for the reservoir of the 3130xl instruments, the following protocol will be in place until further notice. Rather than the standard procedure of changing these each day, the reservoir septa will be replaced with fresh septa on a Monday and Wednesday only. On Tuesday, Thursday and Fridays, the following protocol will apply	TEN	
27/04/2011	Septa are to be removed, washed thoroughly with nanopure water, then dried with a kim-wipe and returned to the same reservoir. Removing, washing and replacing one at a time is the best procedure to ensure no mixups. AB technical experts had no specific recommendations around the washing	ARM	CE / Analytical
	(as it's not recommended) but agreed that washing with nanopure water, returning to the same reservoir, and ensuring they are completely dry would		
6/05/2011	Samples from environmental sampling in the laboratory are now priority 2 not 1	PMT	
17/05/2011	ER team is registering the tube lot numbers used for samples and subsamples	JSM	ER, FRIT
18/05/2011	PSA kits have run out. New procedure fluid from tube to be tested with P-30 - transferred to new tube with same barcode. Must be a minimum of 8 drops. Tube retained in specially marked box in laboratory freezer. Number added to Excel speadsheet G/ForBiol/AAA Evidence Recovery/supernatant testing-P30. Specimen note added. Put original tube in for DLYS.	JSM	ER, FRIT, Analytical
26/05/2011	In sexual assault cases if micro neg and AP pos, add 2 EXH lines. 1st line: Micro neg for sperm. 2nd line: Presump. AP test positive, submitted-results pending. This will continue until the P 30 test is validated. Printing profiles from GMIDX will revert to Adobe PDF instead of CutePDF	JSM	ER, FRIT
31/05/2011	writer, due to the larger size of CutePDF which takes more time to convert and upload into Auslab. This will continue until Auslab is able to provide quicker uploading of files from CutePDF.	PMT	ALL
20/06/2011	Cease using the rationing protocol for reservoir septa for 3130xl instruments as normal supply has resumed	ARM	CE / Analytical
20/06/2011	Configured FTAEXT prep worksheet in LIVE system	TEN	Operational
20/06/2011	FRIT structural change from 3 Reporting Teams to 2 (Reporting 1 and 2). No change to tasks required of staff. Document saved in G:\ForBiol\AAA Forensic	JAH	
28/06/2011	Reporting & Intel\Team Structures. FTAEXT mask released to LIVE.	TEN	Operational
4/07/2011	Effective immediately, QPS environmental samples will be reworked only once, unless a searchable profile is obtained.	PMT	Quality and projects
11/07/2011	Commence reporting Volume Priority 3 samples with undetermined quant values to QPS using new EXH NDNAD - "No DNA detected". Worklist VOLUND created for this process.	PMT	ALL
11/07/2011	Internet access granted to all staff. Not available on group logins ie BIOLOGY	JAH	
14/07/2011	and ROBOTICS p30 kits have now been validated and are ready for use to replace the PSA kit.	AAP	ERT, FRIT
26/07/2011	FBSTR - Storage required list altered in AUSLAB, the Storage position column has been widened and case number & case scientist been removed.	TEN	Operational
22/08/2011	Commencing routine use of the Maxwell-16 MDX for the extraction of DNA from blood swabs Volume Crime Priority 3	ARM	ALL
29/08/2011	No longer flaming of differential lysis extraction slides - slides to be wiped with 100% ehanol before use and heat fixed on a hot block after slide is made in line with ERT procedures - comment to be made in Chelex extraction SOP Semen examination and subsequent sample submission When large items (eg, AP fabrics) are required to be separated into multiple sections each section will undergo complete semen testing (micro and PSA). Sub-	ARM	Analytical
8/09/2011	samples will be determined from the scientific results and case history (ie, semen detected = DLYS; semen not detected = cells if touch DNA will be beneficial to addressing allegations. Exam of internal vaginal swabs (SA K) = if HVS and Vulval are micro pos (≤ 1+ sperm) but LVS is micro Neg, submit all swabs for DLYS. Seminal fluid testing not rerquired on LVS prior to submission.	AAP	
14/09/2011	ERT have implemented electronic storage of slides prepared in-house. The slide storage boxes are titled "ERT Slides 1, 2, etc". The slide storage boxes will be kept in lab room 6124 until full upon which they will be transferred to the exhibit room. The slides will be tracked via the parent barcode (EXH/FBX) attahced to the underside of the slide; eg, HVS slide will be stored via the SA K barcode. Comments to be made against Spermatozoa SOP.	ААР	
19/09/2011	Implementation of tapelifts and priority 1 swabs on the Maxwells. The original QP127 will be handed back to the QPS with the receipt after it has	CI	
20/09/2011	been scanned and checked in Auslab. This will commence from 26 Sept. SA K paperwork to be included with SAIK. If Auslab is down, the QP127 will be photocopied and scanned in later.	JAH	

Implementation Date	Details	Project Leader	Area Affected
22/09/2011	Links are to be sent to QPS if only one person had been previously reported as	SMJ	Intelligence Team
26/09/2011	involved and has been since destroyed Maxwell can be used for fabrics, scrapings, Cigarette butts, paper, FTAs, and	AM	All
16/11/2011	Chewing gum. New print mask for LKR released to LIVE system	TEN	ALL
17/10/2011	Name Change: From 10/10/11 Kate LEE is now Kate P PPIA in Groupwise. From 13/10/11 Kate LEE has changed from kslf1 in AUSLAB to kspf1. List names and mnemonics in AUSLAB are yet to be changed.	KSP	All
19/10/2011	Name change: From today Kate's corrections login has changed from crksl1 to	KSP	All
20/10/2011	crksp1. New UR for negative blank extraction controls FBOT1051.	MLM	All
31/10/2011	New batch CE 400HD (CEHD) for samples to be rerun using the size standard 400HD and new batch CE Investigation (CE NVES) for samples to be rerun with 2uL of PCR product.	MLM	All
21/11/2011	When carryover is seen in DLYS controls, and the profiles are expected (given they are known), the message 'see CWD F ' will no longer be added to the speci notes. Appropriate notes will be made in the batch audit.	JAH	
2/12/2011	Nem print mask for FERRO released to the LIVE system	TEN	ALL
7/12/2011	Relocation of Year 07 case files from front compactus to exhibit room compactus.	WAH	
9/12/2011	Intel Reports (excluding Quality Intel Reports) need to be emailed to QPS DRMU in .pdf and.doc formats. Word .doc to be password protected for modification with 'FRIT'.	JAH	
9/12/2011	GREECM resurrected to be used for cold case management Name change: AUSLAB list names and mnemonics for Kate have now all been	JAH	
15/12/2011	changed (ksp, 1KSP)	KSP	All
6/01/2012	Casefiles without Statement of Witness will be counted (and checked) with the total no. on the front cover, but will not have page numbers written on the pages. No change to files with statements.	JAH	
6/01/2012	Statements and Evid Certs will be scanned and uploaded directly into QPRIME by SSLU. These reports will only be faxed if urgent, or need to go to other clients (eg. DPP). New casefile slips for Admin assistance to be used. Reports need to be with Admin by 2pm to make mail and QPRIME upload that day.	JAH	
16/01/2012	Fingernail and hair substrates can now be processed using the Maxwell	ARM	
	instruments. These substrates will be discarded. Alternative CE processing. Temporary measure to conteract apparent CE		
24/01/2012	carry-over being observed. Amplification batches to be split into to two CE batches such that a folder of HiDi/Rox is run prior to each folder of samples. The batches are to be re-combined for a single GeneMapper batch. This will be in place from this date until notified as being ceased.	ARM	Analytical / all
2/02/2012	Name Change: From 02/02/12 Lisa WESTON is now Lisa BENSTEAD in Groupwise. From 08/03/12 Lisa WESTON has changed from lawf6 in AUSLAB to labf1 and crlaw6 to crlab1. List names and mnemonics in AUSLAB are yet to be changed.	LAB	
3/02/2012	Alternate CE processing noted in 24/01/2012 ceased. Instead, all samples will proceed through CE as per normal. All amplified samples will then have repeat CE process done using the RRCW (or RRCW2 if needed) test. The repeate CE plate will be processed in a 180 degree roatation of the amp plate orientation (i.e. sample that was in postion 1/A01 on the original CE run will re-run on repeat CE in position 96/H12, sample in postion 2/B01 will be done is position 95/G12 and so forth. All samples will be analysed at case management stage for congruence between repeate CE runs.	ARM	All
11/02/2012	Plate readers to include zoomed pdf images of each sample profile read on transposed casework plates to aid with interpretation. Ladders and controls on casework plates and all samples on reference plates do not require zoomed images at this time. Effective as of 11/02/12 until deemed unnecessary.	PAF	
16/02/2012	A new storage box has been configured for individual tube samples being tracked from the Evidence sorting room to the ERT lab: 511900897 - "ERT Lab Storage Box".	AAP	
20/02/2012	Advice from WH&S is that disposable pipette tips do not require to be disposed of in an approved sharps container. Pipette tip waste to be disposed of into bins lined with biohazard bags, then bags cable tied and discarded into biohazard wheelie bins.	MLG	Analytical
27/02/2012	New statement testcode FBSOW to be used. This includes new preamble and new Appendix (v5). New extraction procedure for the Differential Lysis of Epithelial and Sperm	JAH/TEN	
27/02/2012	DNA using DNAIQ chemistry and Maxwell 16 MDX instruments - batch ID Casework Diff Maxwell batch code CWDMAX. Cessation of epis for P2 Female SAIKs.	ARM/JAH	All
28/02/2012	Alternate CE processing noted in 03/02/2012 ceased and a return to normal / routine processing (including the return to routinely using Axygen branded plates and plate septa)	ARM	all
29/02/2012	New testcodes for amended and addendum statements : FBAMEN and FBADDE . New App v5 and preamble to be used. Plate reading process noted on 11/02/12 to include zoomed images of	JAH/TEN	
1/03/2012	samples on casework plates to cease as normal/routine CE processing of plates has commenced 29/02/12. Zooming of images to apply to transposed casework plates within this period only, zooming of images does not apply	PAF	
1/03/2012	to plates processed routinely from 29/02/12. Current contacts at QPS DRMU for incorrect EXH/LKR, Intel letters, advice: Gerard Simpfendorfer, Ruben Collopen, Emma Bellamy, Kathryn Lukowski Name Change, ALSI AB user page for Paula Taylor previously pmff1 changed to	JAH	
8/03/2012	Name change: AUSLAB user name for Paula Taylor previously pmtf1 changed to Paula Brisotto pmbf1. Email address changed from paula_taylor@health qld.gov au to paula_brisotto@health.qld gov.au	PMT	
12/03/2012	New offline template for Evidentiary Certificates is saved to G:\ForBiol\AAA Forensic Reporting & Intel\AAA_Audits-Evidentiary Certificate. This includes Appendix v5.	JAH	

Implementation Date	Details	Project Leader	Area Affected
13/03/2012	FBINTL testcode available in live to be used for Intelligence Reports. A scan fo the signed copy, and unsigned pdf (from Shift Insert in AUSLAB) to be sent to DRMU.	JAH/TEN	
26/03/2012	Batch filler macro being used to create maxwell, off deck lysis and microcon batches in AUSLAB as a workaround until AUSLAB has been fixed.	MLM	Analytical
10/04/2012	Batch filler macro no longer being used to create Maxwell, off deck lysis and microcon batches.	MLM	Analytical
16/04/2012	PDF and JPG 's have been deleted from P:\ for Casework samples from 2009 and 2010.	MLM	All
21/04/2012	INTELL work list used to associate CRISP NO's to FTA Elimination samples. ERT - comment added against SOP 17185 "Detection of Azoospermic Semen in	MLG	Operational
22/05/2012	Casework Samples' mentioning that p30 and PSA are interchangeable terms referring to the protein targetted during the p30/PSA testing for the possible presence of seminal fluid.	AAP	ERT, FRIT
26/05/2012	NCIDD version 6.0.0 released allowing loci other than Profiler Plus to be loaded	SMJ	Intelligence Team
14/05/2012	Flagging OS samples for case-managers. OS (MPH) samples can be "flagged" as possible sources of CE carry-over for subsequent samples processed n in the same capillary for the next 12 runs.	ARM	Case managers and Reporting Scientists
28/05/2012	QPS have introduced a new FTA envelope for use in the collection of FTA reference samples. This envelope does not contain the name or DOB of the person. We have advised QPS that two identifiers are needed on the FTA envelope, which DNA Analysis uses as part of their quality check. QPS will use the SPI number from QPRIME, which transfers to AUSLAB as the UR/Case no. and the barcode number as the two identifiers. The QPDNA number will be hand written on the envelope's by the QPS, signed and dated. The Operational team will check the barcode and QPDNA number against this information in AUSLAB. If the numbers do not match, it will be returned to QPS SMU as per standard process. The SOP will be updated, and the EV DS association worklist changed.	РМВ	OO, Ali
4/06/2012	FBLABEL2 and FBLABEL 11 have been swapped, FBLABEL2 is now located at the GeneMapper computers and FBLABEL11 inside evidence recovery lab	TEN	ALL
7/06/2012	Intel Reports (excluding Quality Intel Reports) and other DRMU comms need to be emailed to A/Snr/Sgt COLLOPEN and also to Emma BELLAMY and Kathryn LUKOWSKI. Reports to be addressed to A/Snr/Sgt COLLOPEN.	JAH	Case managers and Reporting Scientists
12/06/2012	Maxwell C and D have been signed off by management and can now be used for routine extractions	ARM	Analytical
13/06/2012	Quality paragraph to be added to statements that have samples amplified with kits containing reagent contamination.Paragraph located: G:\ForBio\AAA Forensic Reporting & Intel\Quality System Events\PPlus contamination_2012	JAH	
1/07/2012	OQI#32000 - all microcons on hold (except urgents) until resolution of Profiler Plus issues.	PMB	ALL
27/06/2012	Commenced amplifications using new batch on Profiler Plus with lot #1205077 of Primer (including microcons on hold) (after using 'affected lot numbers' since early May that has 'extraneous peaks')	PMB	ALL
28/06/2012	From July 2012, the number of QHFSS environmental samples will be reduced for each of the lab areas to no more than 10 samples per month - targeting high risk areas. This is to be monitored by the Senior Scientists of each team. If there is a rise in the number of profiles seen in the ENVM samples, the number of samples taken the following month can be increased for investigation.	РМВ	Analytical, Evidence Recovery and Quality.
5/07/2012	For a coronial case - if multiple reference samples are received for the same person (determined by DNA number/Name/DOB); only one sample will be routinely processed. O her samples are to be stored and only profiled if required. FTA cards are to be profiled in preference to blood samples	PMB/TEN	ALL
18/07/2012	CE daily maintenance Septa are to be changed on Mondays & Wednesday, on Saturdays as well if work is performed over the weekend. On Tuesday, Thursday and Fridays septas will be rinsde trhroughly with Nanop water, air dried and returned to use in the reservoir from which they came.	ARM	
19/07/2012	QPS Environmental samples will be processed as priority 3, and follow the same process as Volume Crime Priority 3 samples for undetermined quant values - report as "No DNA Detected" and not progress to amplification.	РМВ	Evidence Recovery / Analytical
19/07/2012	Current contacts at QPS DRMU for incorrect EXH/LKR, Intel letters, advice: Ruben Collopen (A/Sr/Sgt), Emma Bellamy, Megan Lamsam	JAH	FRIT
19/07/2012	Name change in Auslab: Tara Neylan is now Tara Prowse and tlnf1 is now tlpf1	CJA	ALL
20/07/2012	Name change in Auslab: Alanna Speirs is now Alanna Darmanin and aksf1 is now akdf1	AKD	ALL
24/07/2012	As of today plate readers will no longer print hard copies of EPG's and no longer need to list out the sample barcodes of the samples that require quality flags in the	MLM	ALL
24/07/2012	batch audit trail. This will be done by the QFLAG checker. Name change: in Novell: Alanna Speirs is now Alanna Darmanin with the email	AKD	ALL
1/08/2012	address Alanna_Darmanin@health.qld gov.au As of today Operational staff will be converting profiles to PDF and JPG.	MLM	ALL
9/08/2012	FBEVC testcode available in AUSLAB for DNA Evid Certs. As of today only single person samples involved in a link that were profiled prior to	JAH/TEN	Reporting Scientists
10/08/2012	Feb 2005 will be repeated. Single person samples involved in interstate links will not to repeated unless specifically requested.	SMJ	Intel team
17/08/2012	Missing person sample and unknown deceased sample matches on NC DD will be reported to QPS DRMU through an LKR. An LKREXT page will be included to provide information regarding the NC DD category. An email will be sent to DRMU identifying the LKR as a missing person or Unk Deceased match.	РМВ	Intel

Implementation Date	Details	Project Leader	Area Affected
4/09/2012	As of today all information related to GM DX is stored on P:\(\). Only 10 users can access P:\(\) which has been restricted to certain computers within the DNA Analysis Unit. O:\(\) has been created for use within the DNA Analysis Unit and this stores DDS upload and Profile PDFs, no other information is to be stored to this drive. Profile PDFs will be regularly archived to P:\(\).	MLM	ALL
7/09/2012	If you need the FBADDE to be a stand alone statement, order the APPVER at the same time, and you are able to edit the Appendix field.	PMB	Reporting Scientists
14/09/2012	Reference samples being repeated for link confirmation will no longer be given priority 1, very few samples are now requiring repeat and therefore making small batches to accommodate is not very time or cost efficent. The time delay impact is minimal.	SMJ	Intel team, OO's and Analytical
26/09/2012	Powerplex 21 for Reference samples validation report signed off by Management. Reference samples (Intel and Evidence samples) commenced processing using PowerPlex 21.	PMB	All
26/09/2012	PowerPlex 21 for Reference samples will be reported in LKRs as "Full DNA profile (PP21)" and "Partial DNA profile (PP21)". Profiler Plus reporting will remain unchanged.	PMB	Intel team
26/09/2012	New AUSLAB batches for FTAs. R21EXT (Reference PP21 Ext Prep), R21GM (Reference PP21 GeneMapper) R21OSD (Reference PP21 OSD), R21RPT(Reference PP21 RPT) R21RUN (Reference PP21 RUN) R21FTA (Reference PP21 FTA),	TEN	All
26/09/2012	New preamble for ref samples profiled with PP21 saved in G:\ForBiol\AAA Forensic Reporting & Intel\AAA. Reporting guidelines\Preambles. For samples that have the 20/20.1 issue at D3, plate readers will be adding the comment "ReGS OLA@D3[20.1]" when plate reading. From Project#54, it was	PMB,EJC	Reporting Scientists
28/09/2012	established that the 20.1 was running just off bin, and should be a 20 designation. The 400HD Rox was the recommended rework strategy for these samples. When case managing these results, we need to order a ReCE using the 400 HD Rox. There is no test code available for this, so please email Allan requesting this rework strategy. This process will be implemented for Profiler Plus. PP21 does not appear to have this issue at D3.	РМВ	Case managers and Reporting Scientists
13/11/2012	Update to DRMU contacts to go to: - collopen RubenB@police.qld gov.au - Lamsam megan@police.qld gov.au - McIntyre.OliviaM@police.qld gov.au - Bellamy.EmmaJ@police.qld.gov.au	JAH	Case managers and Reporting Scientists
19/11/2012	Plate readers to accept as complete on first run partial DNA profiles for FTA intel samples only with 34 alleles in PP21 (including full P+ loci).	PMB	Plate readers, Intel team and case managers
23/11/2012	Plate readers to accept as complete on first run DNA profiles containing one Allelic imbalance for FTA intel samples only on the condition that there is no indication within the DNA profile of more than one contributor. On the second run, a profile with more than one Al may be accepted in consultation with a Senior Scientist.	РМВ	Plate readers, Intel team and case managers
3/12/2012	ER ordering PP21 amps. STRmix for interp of PP21 profiles.	TEN/MM	CD and reporters
10/12/2012	Manual H&E staining of slides commenced today From now on, the Quality flag checker will be responsible for reviewing the sample	JSM	ER and reporters
11/12/2012	on the ENVM list.	PMB	Quality flag checkers, Quality
11/12/2012	New Panel, Bin and Stutter files PowerPlex_21_IDX_V1.1 uploaded to GM DX. New Analysis Methods and Panels created for casework (PowerPlex21_casework and PowerPlex_21_IDX_v1.1.1), reference FTA plates (PowerPlex21_reference and PowerPlex_21_IDX_v1.2) and reference amp plates (PowerPlex21_ref_amp and PowerPlex21_DX_v1.3)	MLM	Analytical
19/12/2012	0.01 ng/µL lowered to 0.008 ng/µL for auto-MIC for Priority 1 and 2 samples	JAH	CMs
19/12/2012	Appendix v6 LIVE in AUSLAB. This is for PP21 and STRmix statements (includes FBEVC, FBSOW and FBADDE).	JAH	Reporting Scientists
20/12/2012	From this date forwards, all Profiler plus CE re-run batches will prepared using Gel company ROX labelled size standard rather than the AB ROX labelled size standard.	ARM	Analytical, all case-managers & reporters
17/12/2012	From this date lookup lines for PP21 in LKR's are available for use however only lines for ref samples Full DNA profile PP21, Partial DNA profile PP21 and Interstate DNA profile are to be used.	SMJ	Intel team
11/01/2013	A complete un-install and re-install of GeneMapper software version 1.1.1 was performed.	MLM	All
14/01/2013	Email group at DRMU until further notice: collopen RubenB@police.qld gov.au - Lamsam megan@police.qld gov.au - McIntyre.OliviaM@police.qld gov.au - Bellamy.EmmaJ@police.qld.gov.au Simpfendorfer.GerardM@police.qld gov.au	JAH	Case managers and Reporting Scientists
4/02/2013	Cease half-vol amp profiling. Report SS, Complex unsuit, No DNA and DNA insufficient profiles. Full-volume reactions to be assessed.	JAH	
6/02/2013	From this date forward, Lai-Wan Chen (LWC) has changed her name to Lai-Wan Le (LWL) and her AUSLAB username is now lwlf1. Novell, Groupwise and QIS2 have all been updated to reflect this change.	LWL	All
12/02/2013	Profiled' added to the comment field on profile page to ensure pages remain validated and do not 'unvalidate' after the data is acknowledged by QPS. New batch ID's created for full volume PP21. Samples manually extracted will have	JAH/TG	
13/02/2013	the following batch ID's CW21FVA (amp), CW21FCE (CE) and FCW21GM (GM DX). Samples auto extracted will have the following batch ID's FVA21CW (amp), FCE21CW (CE) and FGM21CW (GMIDX)	MLM	All
19/02/2013	New batch ID's created for PP21 Re-run CE batches for both full and half volume, CWFVRCE (full volume ReCE) and CW21RCE (half volume ReCE).	MLM	All
22/02/2013 13/03/2013	Amplifications at full-vol PP21 started for routine analysis. Reporting full-volume PP21 amps for 2 and 3 person mixtures commenced.	JAH JAH	

Implementation Date	Details	Project Leader	Area Affected
22/03/2013	As of 2 30pm, no longer doing EB checks routinely. They can be performed for FGM batches if need be.	JAH	
28/03/2013	CFLIT Tray relocated from Admin Area to Write Up Area in Evid Recovery	WAH	All & Property Point Staff
8/04/2013	OS log no longer required to be kept by Analytical Section. If staff need to access information regarding instrument and batches, can refer to I:\AAA Electronic Workflow Diary.	JAH	
30/04/2013	QP identifier no longer required to be written/stamped on each page within a casefile as per NATA communication. Current DRMU contact list:Simpfendorfer.GerardM[OSC] - Simpfendorfer.GerardM@police qld.gov au Hartel.JensO[OSC] - Hartel.JensO@police.qld.gov au	TEN	all
30/04/2013	Linton.NicoleS[OSC] - Hart-eniso@police.qlu.gov au Linton.NicoleS[OSC] - Linton.NicoleS@police qld.gov au Bellamy.EmmaJ[OSC] - Bellamy.EmmaJ@police.qld.gov.au McIntyre.OliviaM[OSC] - McIntyre.OliviaM@police.qld gov.au Nauschutz.MarneyS[OSC] - Nauschutz.MarneyS@police.qld gov.au Lamsam.Megan[OSC] - Lamsam Megan@police qld.gov au	JAH	
6/05/2013	Volume Crime samples will be processed in Profiler Plus from this date forward.	CJA	All staff
27/05/2013	Case Mgt streamlining strategies (Proposal #126 - includes 4 proposals to QPS) implemented into result release. Minor change doc in #126.	JAH	
4/06/2013	Insp Tony Carstensen advised by phone that any item requiring saliva testing needs to be authorised by either: Superintendent of Forensic Services Branch, Inspector of Scientific (HQ) or Inspector of DNA before it can be received by Property Point. This change is effective immediately. Property Point have been advised.	CJA	All & Property Point Staff
7/06/2013	QP127s are no longer required. If presented, they will be scanned and returned to delivery officer. If any instructions are required for examination, they will come across into AUSLAB in the clinical notes.	JAH	All staff
14/06/2013	Consulted with Gerard Simpfendorfer, samples involved in a link that were initially tested in Profiler Plus and have been since tested using PP21 are not required to have that information updated in a link unless: 1) The link is no longer valid or 2) the sample status changes for a CW sample e g single source to 2 person mixt or 2 person mixt to 3 person mixt	SMJ	intel
19/06/2013	Re-implementation for the use of 3130xl A for CE of Casework and amplified reference samples following laser change - no changes to GM DX and STRmix parameters required	ARM	all teams
20/06/2013	Use Australian datasets, applicable Theta value and 1-tail 99% CI for Paternities in Kinship until STRmix v2 is released. Two mutations to equal exclusion until formal direction from BSAG is received. Minor Process chage doc in #105.	JAH	
21/06/2013	Kinship locus sort order has been fixed so that loci sort in the proper PP21 order.	TJG	Case managers and Reporting Scientists
4/07/2013	Project #91 (Maxwell pre-lysis) is implemented as of today. Lysates extracted using the Maxwell pre-lysis method may be kept at room temperature for up to 120 hours prior to processing on the Maxwell instruments.	KML	Analytical
5/07/2013 8/07/2013	In GMIDX - stutter thresholds for panels (1 2 and 1.3) have been updated to two decimal places (they are currently only at one decimal place). Additional Wash Module (Super 3 wash module) implemented for use with both	KML ARM	all all
11/07/2013	3130xl instruments (refer management team minutes dated 05-07-2013) The Sample Information field for subsamples that have been copied from the registration of EXHs have always transferred to QPS, but the visibility has just been turned on. This should prevent phonecalls to SSLU for sub-sample description. For Case Mgt, this means when creating a barcode for NCIDD upload from STRmix mixtures, the sample info field goes to QPS. If the result is from a sub-sample (eg. HVS from SAIK), please add more information to the registration of the upload to inform QPS what the sub-sample relates to.	CJA	
11/07/2013	Eg. SA K with upload from HVS:	007	
	Sample info field: UKM1 – HVS – 987654321 (SA K barcode)		
	NB: For EXHs that we create, we still need to call DRMU with the item information (Eg. PM samples, Paternity Results).		
	Current DRMU contact list		
29/07/2013		JAH	
5/08/2013	Following variance and baseline checks on 3130xl B using a 5 second injection time (see OQI 34817), 3130xl B has been approved by the management team for routine processing of casework samples amplified with the PP21 amplifica ion kit. Maxwell 16 instruments can now be used for the extraction of tissue with the	ARM	analytical (all)
26/08/2013	exception of paraffin embeded tissue which is not validated for use with the Maxwell instrument	ARM	all
18/10/2013	Interim CM workflow using STRmix assistance implemented. 1DA list used for paperless and FBSI17 used to track casefiles	KDR	All case managers and reporting scientists and STRmix data entry role
1/11/2013	Emails regarding Intel vs Evidence FTA samples are to go to FSS Biology Quality for sending to S/Sgt McLaren and/or Insp Carstensen.	JAH	Case managers and Reporting Scientists
7/11/2013	Change to the sample submission process for semen positive SAIKs. See Section 6.1 of QIS document 32106 Examination of Sexual Cases	LBR	ERT, case managers and reporting scientists

Implementation Date	Details minor changes to STRMix Report Macro: - rounding when less than LR=10 to 1 sig	Project Leader	Area Affected
8/11/2013	figure (also applies to Hd) - removed the LR total line so that we only have the HPD LR Total available (which is the only one we use anyway) added words to explain the figure - added string of **** to make it clear when favouring non-contribution.	TJG	Case managers and Reporting Scientists
14/11/2013	Interim CM workflow using STRmix assistance updated on 7 Nov 2013 and again on 14 Nov 2013 - changes relate to Assessor deciding if they STRmix or send it to STRmixer and who removes samples from coloured CM lists for both paper and paperless processes	KDR	All case managers and reporting scientists and STRmix data entry role
21/11/2013	Re-implementation for the use of 3130xl B for CE of PP21 Casework samples following laser change - no changes to GM DX and STRmix parameters required	BUA	all teams
26/11/2013	Change to sample submission process for AP positive fabrics (i e. QPS AP screening). If the item is tested and is semen negative (i.e. micro and P30 negative) we will not submit any samples for DNA (previously samples were sumbitted for cells). The screening test results will be reported to QPS. QPS may request samples be submitted for cells for semen negative AP positive fabrics, and this will be listed in the Clinical Notes. QPS may subsequently request further testing of a semen negative AP positive fabric.	LBR	ER, FRIT
4/12/2013	Stutter macro has been verified for use and is now in QIS. Plate readers to commence using.	KML/JAH	All
9/12/2013	Only Profiler Plus (Volume priority 3 samples) to be processed on automated extraction at this time. This change is for operational convenience	ARM	Analytical
9/12/2013	Timesheets Up to the certification of EB7 29th of Feb 2009, all FSS employees worked under the Variable Working Hours Arrangements for Corporate Office Employees, which mandated sign on and sign off at 15 minute intervals. There is no such mandate in the current arrangements for both Public Service and District Health employees (HP, AO and OO). Therefore from the 9th of Dec 2013, all staff are able to sign on and off when they actually start and finish work (ie 8.07am until 5.22pm). This also means that there is no rounding as previously was used (ie started work at 8.07am therefore rounded to 8am - this discontinues from this date as well).	CJA	All staff
11/12/2013	From 13/12/13, all phone calls to 3000 9629 regarding unplanned absences should be made after 8am (rather than after 7 30am).	CJA	All staff
13/12/2013	All casework plates (PP21 and P+) are to be read using the following core comments: NSD, SS, MIX, COMPLEX. This is to direct samples to the correct Case Management Batch Functionality Lists. Current list of DRMU contacts until further notice	KML/JAH	Plate readers, Analytical, FRIT
9/01/2014	Simpfendorfer.GerardM@police.qld.gov.au Linton.NicoleS@police.qld.gov.au Collopen.RubenB@police.qld.gov.au Bellamy.EmmaJ@police.qld.gov.au Lamsam.Megan@police.qld.gov.au	JAH	
20/02/2014	Palmer.CarolynL@police.qld.qov.au Thermalcycler F returned to full use after return from service and calibration from	LBR	FRIT, Analytical, ER, Case Managers
21/02/2014	AB and BTS 3130xl A returned to full use (i e. PP21 CW in addition to P+ and PP21 Ref) "New" yellow team cases are now paperless, with FMO and examination notes to	LBR	FRIT, Analytical, ER, Case Managers
6/03/2014	be scanned into AUSLAB on completion of SAIK presumptives. "Old" pink or blue FMO notes have a casefile made up as done previously. Scanning to occur on SAIK barcode.	JSM/ JAH	FRIT, ER, Case Managers
10/03/2014	Plate Readers to convert all P1 samples to PDF at plate reading stage to enable timely CM.	PAF/JAH	Plate Readers, FRIT
13/03/2014	At plate reading, readers can accept partial Amelogenin for Intel samples effectively immediately (as NCIDD does not currently match on Amel). Approved at management team meeting	KDS	Plate readers, FRIT
18/03/2014	Maxwell B has returned to routine use after replacement and subsequent verification of the rod assembly	BUA	Analytical
21/03/2014	Plate reading computers moved to Block 3, in the old Operational Officer hot desk area. Laboratory Assistants moved to Block 6, to the Quality & Plate Reading area	KDS & CJA	Operational Officers & Plate Readers
24/03/2014	$jpgs\ will\ cease$ for PP21 profiles. Zooms for P+ will commence. Pdfs and $jpgs\ will$ continue for P+ and reference samples.	JAH	Case managers, Operational Officers
31/03/2014	New EXH now available for use EXREV This exhibit had extra information on the back of the crime scene sample envelope. Where writing or barcodes are present on the back of a CSSE, an extra photograph is taken which must be reviewed during examination and prior to final results being released. These extra reviews increase the processing time for samples and slow down the release of final results. Please ensure all information is limited to one side of the envelope.	АН	ERT / All reporters
1/04/2014	QPS staff matching for profiler plates moved from AUSLAB matching to smatch matching (5 previous profiler plates with QFLAGS rerun to test MACRO - rerun accepted)	KDS	FRIT, Analytical, ER, Case Managers
1/05/2014	Introduction of electronic visitors log (to replace hardcopy sign-in sheet) File is stored in: G:\ForBio\AAA Administration\Visitors Log	KDS	Forensic DNA Analysis
5/05/2014	Management team have approved GII batch status upload changes: All batches, except for FTA batches, will no longer have "Completed" batch status loaded to AUSLAB using the GII. All batches will have a "Started" status loaded, and the next status update will be either "Passed, Failed, See batch, <3 alleles in neg ctls".	MLG	FRIT, Analytical, Operational, Case Managers

Implementation Date	Details	Project Leader	Area Affected
7/05/2014	Advised by Rhys Skinner and Morgan Pascoe that all emails from QH travel via a secure pathway to other government clients (namely the QPS, Justice and the Coroner). Therefore all emails containing case related information is able to be sent from a @health.qld.gov.au account to a @police.qld gov.au securely. [Detail: QH email travels via CITEC before proceeding to QPS, the pathway from QH to CITEC and CITEC to QPS is all secure and private].	CJA & Linda Morley	Forensic DNA Analysis, SSLU, Forensic Property Point
	Given the large amount of Priority 1 cases we have been receiving of late, we sought and received approval from Insp Carstensen to introduce some interim processes to streamline the reporting of the P1 samples to ensure results are reported in the agreed timeframes. This is an interim measure to enable the critical results to be reported to the QPS for all of these urgent cases during this peak time.		
20/05/2014	- If we receive a number of P1 samples for the case, and the results are all indicative of the same unknown profile, we will select the most suitable and probative profile for interpretation and loading to NCIDD, and any matches will be reported on this sample within the urgent timeframe We would liaise with the QPS to determine if these remaining results can be downgraded to Priority 2 samples. This will enable the reporting scientists to allocate their time to interpreting and reporting other urgent P1 samples. The allocated scientist will ensure the results for all downgraded samples are reported in a timely manner.	РМВ	FRIT
	- A reference sample from the complainant, for example from a sexual assault, as well as ownership of the item is critical for the interpretation of any DNA results obtained. Without these, interpretation of the resulting DNA profiles is limited and may not provide information that can be loaded to NC DD. If P1 samples are all		
26/05/2014	Use of Item ownership list (mnemonic "ELF") for investigating if items can be attributed to certain person. Samples list inserted to the ELF list will be sent by SSLU to QPS questioning if ownership of the item the sample has been taken from can be determined, or if the owner is unknown. This is to assist with samples where conditioning is a reasonable approach to mixture interpretation. The ERT will be list inserting samples at registration if the sample is from an item of clothing or has been taken from the body (e.g. body swab) and there is no item ownership in AUSLAB or on the packaging / paperwork and if there are reference samples collected for the case. FRIT will be able to use the list at case management when item ownership information is required. SSLU intend on sending the list of samples to QPS once a day, responses will be added to the UR notes and e-mails scanned into AUSLAB as per current processes.	ARM	ERT / FRIT
28/05/2014	Sample barcodes no longer scanned into Maxwells as of Analytical Team Meeting on 14/04/2014 (see meeting minutes for further info).	LBR	Analytical/CMs
30/05/2014	LKREXT pages no longer need to be created if there is an evidence sample related to multiple cases involved in a link. Only one Case number is to be chosen with to put into the LKR line. QPS gave the OK	SMJ	Intel/QPS
11/06/2014	NAD reference samples require a rework to confirm the profile. This rework is to be ordered by the plate reader when reading ReGs plates	KDS	All
11/06/2014	As per agreement from all members of management team (raised at 5th of June management team meeting) - removed the requirement for ERT staff to initial and date tubes containing samples submitted to AT that are labelled with a printed AUSLAB barcode / number. Refer e-mail stored in G:\ForBio\NAAA Evidence Recovery\Planning and Equipment\Planning and Equipment Archive	ARM	ERT
23/06/2014	As of 23/06/2014 Coronial blood FTA submissions will be submitted directly for EREF processing (REF21 and R21RUN/R21OSD not required prior to EREF).	KDS	All
1/07/2014	STRmix v2.0.1 to be used for any new case received today. New macro for the report to be used with v2. Also, 32 allele cut-off for SS profiles implemented - no need for LR calcs on these. Any LRs calculated with STRmix V2.0.1 to have LRs calculated using all 3 datasets and the most conservative LR to be quoted. Factor of N LR to be quoted.	EJC, RGM, RJP, TG, JAH	Case Mgt
8/07/2014	As per project #136, AP reagent is now to be stored frozen in 2ml and 50ml aliquots with an expiry date of six months.	АН	ERT
9/07/2014	For FNCIDD and all SNC DD batches - the NCIDD status and date uploaded can now be updated using the results file function in the batch. The NC DD status on the SNC DD page will now also copy back through to the COMIX page for mixture samples (once the registration page of the COMIX is resaved).	LAB	FRIT
18/07/2014	Appendix v7 live in AUSLAB. To be used in conjunction with STRmix v2 casefiles.	JAH	Reporting Scientists
21/07/2014	LKRK (tem No) field on LKR page modified to 16 characters from 14 and DUML (Result/Status) field length modified to 56 characters from 58. Thise samples where the result/status would be ### due to shorter length were printed and scanned prior to modification. Change to the process for environmental cleaning of the ERT lab. Same cleaning protocols in pace. The change involves more focus on cleaning of non-work areas such as windows / window sills etc. Those work surfaces and utensils that are used	SMJ	Intel
6/08/2014	on regular (daily or close to daily) basis, that are cleaned prior to and after use (e.g. exam benches and exam box contents) will not be cleaned as part of the environmental clean. Other areas that have previously been included in the clean (e.g. outer surface of fridges, non-examination benches and cupboards etc.) will continue to be cleaned. FRIT staff no longer required to assist with the clean. Increased environmental sampling will be conducted for a short period to monitor this change.	ARM	ERT / all
25/08/2014	Pre PCR MPIIs not able to be verified at 1uL. Move to manual amps for all amps requiring addition of 1uL volumes.	LBR	Analytical

Implementation Date	Details Unit D-A settings export 0 alleles at ally one locition for allo 12 alleles lot FF2 I (as	Project Leader	Area Affected
	on ID-A settings export a linetes at any offerior for the ID-A linetes for FZ-1 (as this includes stutter). The allow for any potential QFLAGs for profiles with more than this number of alleles at any of the loci, the following process is to be implemented: When case managing a P+ case - if your profile is complex unsuitable for comparison, then nothing further needs to be done. This would be unsuitable for quality flag checking.	•	
26/08/2014	If your profile is able to be interpreted (complex cannot exclude/ complex unable to load, or a major/minor), and at any loci has 7 or more alleles, then an assessment needs to be made – if the major or "large" peaks are captured within the first 6 alleles for each loci, then you will know those peaks have undergone quality flag checking. If any peaks within the major or 'large' peaks fall outside of the first 6 alleles at more than one loci (as the staff match macro allows for 1 mismatch) then a quality flag check can be undertaken for the major/large peaks by the Quality and Projects Senior Scientist. This sample can be inserted onto the BQUAL list with a speci note "for Quality flag checking". Kirsten or delegate will then perform the quality search on the larger peaks within the profile and add a speci note, inserting back onto the case managers comms list.	РМВ	Quality flag checkers, case managers
29/08/2014	For quality flag checking, this will really be only a minor impact as well. For those Email received from Paula Campbell - Office of State Corner (via Lisa Farrelly) confirming all tissues including DNA samples can be destroyed under the Form 6. Refer to scanned emails in G:\ForBiol\QPS Communications\Comms from the Coroner Email received from Gerard Simpfendorfer has confirmed that QPS no longer	KDS	Reporting/Intel/Operational
29/08/2014	require the case information to inform them when a reference sample is an evidence sample, in an LRK page. The link state is still required to be QP or QL but the case number is not required. Reference sample specimen notes for warm links	SMJ	Intel
29/08/2014	involved in LKR's are also no longer required. 7500 A returned to service. See Change Management #156 Change in process for Sampling of FTA cards by ERT team. No requirement for	LBR	All
3/09/2014	creating of examination notes. FBEXAM will be sufficient to record who sampled the card, what time and when. See records located in G:\ForBiol\AAA Evidence Recovery\Planning and Equipment\2014 - FTA card sampling process change. Note: for all other reference samples (e.g. swab, hair etc.) examination notes will still be made as per current process.	ARM	ERT / reporting
3/09/2014	Change in process for sampling of reference samples by ERT. (discussion carried out during meeting held on 02/09/2014 regarding BSD instrument). From now onwards, Exam bench 15 will be dedicated to the processing of any reference sample designated as a reference sample at the time of examination by ERT. All reference samples (regardless of type) will processed on this exam bench.	ARM	ERT / reporting
10/09/2014	Model maker results for 3130B for STRmix v2 have been approved by Mgt Team. No change to the values generated pre 3130B filter change.	LBR,JAH	Case mgt, Analytical
11/09/2014	Successfully verified Pre-PCR MPII A & B today at 1ul, 5uL, 10uL and 20uL. We have created an adjusted performance file which was used for the verification. As such we will be returning both Pre-PCR MPIIs to use for 1uL - 20uL pipetting. Any volumes less than 1uL will be sent to a manual pipetting batch.	LBR	Analytical
30/09/2014	The Priority look-up table for samples has been changed. We now have 7 priorities, starting with 0 – Urgent, 1 – Priority, 2 – High Priority, 3 – Medium Priority etc through to 6 - Cease. Priority "0" will only be used for QPS requested 3 day TAT samples (and associated controls). Priority "1" will be used for all other controls and urgent rework samples. Samples will sort in the lists according to this priority "0" being at the top, then 1 through to 6. Blanks by default will continue to sort at the very top of the list. Batch paperwork with "0" priority samples will be stamped "Urgent". Batches with "1" priority samples will be stamped with "Priority" (this will be handwritten in the first instance, until we get a 'Priority' stamp). This will clearly differentiate them from any other routine batches.	РМВ	All
1/10/2014	Implement new Workflow Diary to consolidate all pre-existing instrument workflow diaries.	LBR	Analytical
9/10/2014	External swabs to be submitted for cells when SAIK determined to be semen neg. The is an update to the findings from project #143.	JAH	ERT, case managers and reporting scientists
13/10/2014	Implement use of STRMIX (STRMIX decon) and STRMIXCM (STRMIX case management) worklists in place of 10HC and 1DA respectively.	JAH	FRIT
28/10/2014	Commence use of Semen Quality Control Donor#4 for DLYS batches. Lab# 603294808 and FBOT#1179. It is PP21 EXTPS 2 in GMIDX.	MLM	Analytical
11/11/2014	Model maker results for 3130A for STRmix have been approved by Mgt Team. No change to the values.	LBR,JAH	Case Mgt, Analytical
13/11/2014	The Priority look-up table for samples has been reverted back to 1 – Urgent 2 – High Priority, 3 – Medium Priority etc through to 6 - Cease. Priority "0" has been renamed DNU (for Do Not Use) and has been deactivated. To manage the QPS versus internal P1 samples, the Analytical Team propose the following: - Include all Analytical HP4s as a CC on the Urgent authorisation emails – mark QPS authorised P1s as 3 Day TAT - Maria (in her HP4 role as the Urgent Monitor) will monitor and communicate to team members so they know which samples are QPS approved P1s. The other HP4s and LBR will be a redundancy for this. - All P1s are visible on the Batch Workflow Table in AUSLAB so sample progress will be monitored on this table. The Analytical_Reporting interface spreadsheet will not longer be used (as this was implemented during CE downtime). The GREECM and GTEAM worklist implemented to monitor the P0 samples will no longer be used for routine processing.	РМВ	All
27/11/2014	processing. Start using the RFBSDIQ batch type for locked EREF batches. This is used with BRE specimen type for controls	KDS	Analytical and Operational Teams

Implementation Date	Details	Project Leader	Area Affected
17/12/2014	Refer to OQI 38198 (STRmix miscoding). Data gathered on possible affected samples on 16/12/14. Any samples not on data grab that are not affected (as seen from STRmix 'green screen') do not require a note that unaffected. Any samples affected will need to be on hold until implementation of STRmix v2.0.6	JAH	Case managers
5/01/2015	New EXH lines available to use. See G:\ForBiol\AAA Forensic Reporting & Intel\EXH spreadsheets for QPS (versioned)\2014\EXH proposals for QPS_102014v3_approved for QPS testing.	JAH	Case managers
9/01/2015	AUSLAB missing storage audit fault identified. Fault is retrospective to 10th December - reported under problem number 9549. Refer to Adverse event 33 in the Adverse events log for additional details (I:\Adverse Events DNA Analysis\adverse Events log)	KDS	All
19/01/2015	New case management workflow changes implemented - no longer use Batch Functionality to case manage. See I:\AUSLAB\AUSLAB Calls\2014\New CM 2014	PAF, TEN	All
	Update to DRMU contacts. Comms (eg 'incorrects') to go to: - Simpfendorfer.GerardM@police.qld gov.au		
29/01/2015		JAH	FRIT, Mgt
30/01/2015	STRmix v2 0 6 installed today.V1.05 deconvolution - run LR in V2.0.6 using caucasian dataset only (factor of N will not apply) V2.0.1 deconvolution - run LR in V2.0.6 using all 3 datasets (factor of N will be reported) V2.0.6 deconvolution - run LR in V2.0.6 using all 3 datasets (factor of N will be reported)	RGM, EJC, JAH	Case managers
10/02/2015	FTA Intel samples with a profile of NSD on an FTA plate will now be given a comment of 'NSD' and will be reworked.	MLM	Plate readers
12/02/2015	New version of the Forensic DNA ANalysis Workplace Health and Safety SOP QIS 23945 - "A minimum of two staff members must be present within a laboratory area when performing the following tasks: using scalpels and other sharps, naked flame, liquid nitrogen, gas, chloroform and instruments with large moveable parts (e.g. MPII). It is sufficient for another staff member to be present within the adjoining office area when someone in the laboratory is performing other tasks, including but not limited to photography, labelling tubes, Maxwell extraction, manual quantification or amplification, microscopy, phadebas and in tube samples. If any staff member is mopping or entering the cold or freezer room alone, they must ensure that another staff member is aware of their location and approximate time of task." Update to DRMU contacts. Comms (eg 'incorrects') to go to: - Simpfendorfer.GerardM@police.qld gov.au	AKD	Laboratory areas.
13/02/2015		SMJ	FRIT, Mgt
16/02/2015	3500xL in use for direct amp FTA PP21, and config for FTA punching has changed to 3500xL platemaps. Update: Due to 3500 POP4 polymer supply issues, implementation of 3500xL for direct amp referenace samples did not occur. FTA batch configuration has been returned to 3130 plate configuration. Implementation to occur at a later date upon receival of POP4. Update: POP4 received. Commence use of 3500xL as of 04/03/2015. FTA Batch configuration changed to 3500xL.	LBR/BM	All areas
27/03/2015	Based on change to 3000c. Based on change Mgt #149, a guideline for determining number of contributors is implemented from today.	JAH/EJC/RGM	Case managers
27/03/2015	Destruction of reference samples to include crossing out of the paper version of EPG if a paper case file exists	SMJ	Intel team & Operational team
9/04/2015	No casefiles to be made up for SA Ks. SAlKs with blue or pink sheets that cannot be scanned will be tracked to exam filing and then to paperless folders.	JAH/JSM	ER, FRIT, Admin and Operational teams

Implementation Date	Details latest DRMU email list:	Project Leader	Area Affected
	(please still send to me even		
14/04/2015	though I am away)	JAH	all teams
15/05/2015	FBUNK26 and FBUNK27 are complex mixtures. Upon discussion between EJC and KDS it was decided that these should be removed from the staff database and both staff match macros as they are not suitable for searching against.	EJC	All teams
15/05/2015	Created i:/Projects folder to be used as a centralised source of all Forensic DNA	AKD	All teams
23/06/2015 29/06/2015	Analysis project data, mostly via shortcuts. Guidelines for Number of Contributors final report signed off. New server for GM DX setup and GMIDX v1.4 installed.	EJC, RGM MLM	case managers All teams
7/07/2015	3500xl A implemented for analysis of Extracted Reference samples from today	LBR	All Teams
31/08/2015	STRmix V2 0.1 has been removed from laptop with asset number 10367658. STRmix V2 3 06 has now been installed on this laptop. Forensic DNA Analysis no longer has a copy of STRmix V2 0.1	EJC	Case Managers
14/09/2015	FTA cards to be sealed after punching. Person sealing the envelope to be	KDS	OO Team
21/09/2015	identified. Date of sealing to be included on the sealing label All reference blood samples to be processed in Evidence Recovery and not	KDS	OO and ER team
12/10/2015	processed on the BSD machine Thermal cycling block replaced on Real Time PCR system 7500A. 7500A back in	HKP	All teams
27/10/2015	use 15/10/2015. Carolyn Hoffman (QPS) called and advised that Forensic Analysis does not need to fax evidence association sheets to QPS DNA Unit anymore. Effective immediately	KDS	OO Team
29/10/2015	Admin will scan examination paperwork into the relevant whole item barcode in AUSLAB. Same process as per SA K examination paperwork. Effective immediately.	JAH, ARM, WAH	ER, Admin,case managers
4/11/2015	Quantifiler Trio DNA Quantification Kit is now in routine use for casework and reference samples.	PA/MLM	All teams
11/11/2015	Implementation of Mantis for dispensing water and mastermix for reference plate processing	KDS/MAM	All teams
17/12/2015	Change 96 well plates for DNA Amplification and CE setup from Axygen to SSI Ultraflux 96 well plate. See associated Minor Process Change.	LBR/MLM	Analytical
23/12/2015	Mantis placed out of routine use - for FTA processing (Refer to minor change form)	KDS/MAM	All teams
6/01/2016	New Y-Intercept ranges have now been calculated and set for SAT, LAT and Y-Target (calculated from all quant standard curve results since Nov 2015). I:\Change Management\Proposal#152 - Validation of Quantifiler Trio and Y-Filer Plus\Quant Trio\Implementation\Y-Intercept Monitor New email address for DRMU group: DNA Management@police qld.gov au. The	PA	Analytical
14/01/2016	members of this group list will be managed by QPS. No change to the sending of Quality letters to Inspector Carstensen.	JAH	All teams
22/01/2016	dd/mm/yyyy date format adopted as per ISO8601 for all teams in Forensic DNA Analysis	LBR	All Teams
22/01/2016	3500xL B verification signed off. Instrument currently being used for validations and as a back up instrument to 3500xL A.	MLM	All teams
11/02/2016	NIST-STRBASE 1036 combined data set added to Kinship population data, required for CTS.	TJG	FRIT
15/02/2016	Quant Trio v1.4 program to be used on the MPII's.	MLM	Analytical
5/04/2016	Auto Extraction MPII A is no longer in use. All extractions will be performed by Maxwell and manual processes. GeneMapper Data server changed over. All data relating to GeneMapper D-X, CE	MLM	All teams
8/04/2016	and PDF profiles have been moved from I: O:J:\ and are all now located on GM DX P:\.	MLM	All teams
14/04/2016	Intuitive exclusions process stored at G:\ForBiol\AAA Forensic Reporting & Inte\AAA_Reporting guidelines\PP21 and STRmix case mgt\Intuitive Exclusions can be used as of today. A comment has also been added to the CM SOP.	KDR	
10/06/2016	As a result of risk assessment QIS#23352 - gloves are now to be worn by staff when handling yellow biohazard bins	KDS	OO team (and all as applicable)
15/07/2016	The process of P+ CW epgs being jpeg'd and uploaded to AUSLAB is no longer in use. Staff need to use pdf finder instead. Comments will be added to the appropriate SOPs.	KDR	All teams
21/07/2016	Profiler Plus and Powerplex 21 casework amplifications which require a dilution to be processed by manual amp and not on the MPII	LBR	Analytical, Quality, FRIT
19/07/2016	Hannah Pattison comms:1HKP and storage: FBSI77 (HKPF1 intray)	KDR	all teams
22/07/2016	Cassandra James comms: 1CJF5 and storage: FBSI78 (CJF5 intray) WEN size standard is now implemented. All settings are now set for 3130 & 3500,	JAH	all teams
1/08/2016	Genemeapper and all docs associated are in QIS. MPII taken offline for crime scene samples. Reference samples still processed on	HKP	All teams
3/08/2016	MPII. P1 and P2 samples processed manually. P3 samples on hold (after extraction).	PMB	All teams

Implementation Date	Details	Project Leader	Area Affected
8/08/2016	Effective from this date: All items where semen examination is required will be submitted for Differential Lysis regardless of microscopy result during Evidence Recovery processing. All samples with a negative microscopy result at the time of examination will automatically have the slide made from differential lysis extraction examined as a matter of routine. For samples with an initial Micro Neg result will simply have a "Submitted - results pending" EXH line, and after examination of the diff slide, a further EXH line (e.g. "micro neg for sperm") will be added depending on the results of the microscopy result and any AP / p30 testing conducted during examination	ARM	ERT / AT / Reporting
12/08/2016	A re-assessment of pipettes has been completed - they are now determined to be non-critical pieces of equipment as several checks (including positive and negative controls, original NATA calibration certificates and internal 3 monthly checks with traceable ARTEL equipment and reagents) are in place to ensure pipettes are within range.	KML/KDS	All teams
29/08/2016	MPII recommenced processing of P3 samples. Manual dilutions for P3 samples (off the MPII) will continue. Reference samples still processed on MPII. P1 and P2 samples remain with manual processing.	PMB	all teams
12/09/2016	MPII recommenced processing all samples. Dilutions recommenced on the MPII for all samples.	PMB	all teams
30/09/2016	GM DX "Zoom PP21 Print Profile" print setting scan range changed from 65bp to 60bp to make the range the same as PP21 Print Profile setting. Added new Batch type for Diff Lysis Retain Supernatant - CW Retain Sup. Diff Maxwell (CWRSDMAX~SS).	KML	all teams
25/10/2016	Specimen types for this batch are: Sperm Lysate (SLYS~SS) Epithelial Lysate (ELYS~SS)	LBR	all teams
25/10/2016	Upgrade MVS software - Nil changes to function of software, change to operating	KML/KDS	Analytical/Operational
4/11/2016 21/11/2016	system compatibility only As of today - plates readers to PDF urgent samples and do the zoom as well QIAsymphony implemented today following final approval of Project# 168	KML LBR	all teams all teams
22/11/2016	Single point checks on thermometers to be conducted at 70 degress (no longer ice point) from this date. FSS purchase of Fluke oil bath has enabled this change. Temperatures can now be checked at a range closer to "standard use"	KDS	Analytical/ER/Operational
25/11/2016	Project files (that reach signed project proposal stage) are loaded to FR as a backup for network files. Refer to change management SOP	KDS	All
29/11/2016	QlAsymphony removed from use following liquid residue on exterior of drip catchers	LBR	All
13/12/2016	QIAsymphony - new SP extraction protocol received with modifications to waste disposal received from QIAGEN. Modification to protocol leaves some waste liquid in the used cartridges (previous protocols removed all liquid from the cartridge at end of use and dispensed into waste reservoir - QIAGEN suspected this was causing bubbles on the end of the tips). Blank run was processed with new protocol and no residue on exterior of drip catchers observed. Two test runs using casework samples were processed on 13/12 and 14/12 during the day so that these runs could be observed and no residue on exterior of drip catchers was observed. QIAsymphony returned to full use (including overnight runs)	LBR	All
16/12/2016 10/01/2017	Temporary method of SAIK blood storage and processing (from 15/07/2016 until Dec 2017 - final date TBC). When blood sample is removed from SAIK, property point add a receipt barcode, track the blood by receipt barcode pending QPS registration of the blood sample. QPS will either register the sample or request its return or destruction. No processing can be completed until the blood sample has been registered by QPS. UR notes are added to the case by property point to indicate the sample is received and stored in the fridge (due to no registered barcode for blood being available). Property Point will also send an FR task to QPS to request registraion 3130xl B laser changed and baseline calculations performed. LOD 16 RFU and LOR 40 RFU retained. Seel:\AAA Analytical\Audits and Reviews\AAA 3130-3500\Baseline Reviews\Review Baseline 3130xl B post laser change January 2017 for relevant files and calculations.	KDS	Reporting and OO team
16/01/2017	Decision Point: Post laser change on 3130xl B, Model Maker was run and the pre- laser change values were retained.	Mgt/EJC/RJP/AAP	case managers
17/01/2017	STARlet A implemented for routine use today. Second email requesting unknowns to be compared to mixtures that might be	LBR	All teams
3/02/2017	obtained from P1 items is no longer going to be received from QPS. All Uks in cases where P1 items are requested, are to be compared to mixtures (in P1 items only).	JAH	case managers
9/02/2017	Minor Process Change: troubleshooting added to Section 7 of QIS 19978 for situation where CE spectral calibration is failing due to sub threshold dye peaks: The amount of 5C Dye added to the dye preparation is increased by 1.5 x $$	LBR	All teams
24/02/2017	Quantifiler Trio Y-Intecept calculcation method and thresholds modified for all targets. See associated Minor Change form. Comment added to SOP	LBR	all teams
24/03/2017	QlAsymphony has been verified and returned to use following software upgraded to HID v1.0 Minor Process Change form has been completed. For P2, PP21 samples: Unallocated casework (working from MIXCM, SSCM, COMPCM) - Do not add CS initials if ordering rework - Add CS initials if no further rework required, ready to CM sample	LBR	all teams
10/04/2017	Allocated casework/operations - Continue to add CS initials when ordering reworks When ordering reworks to CS in the CS in th	МОН	case managers

When ordering rework, use XPLEX page 'comments' to detail reasons $% \left\{ \left(1\right) \right\} =\left\{

nplementation Date	Details Test Amp Criteria for Profiler Plus and PP21 modified. See Minor Process Change	Project Leader	Area Affected
11/04/2017	and comemnts against QIS 19994	LBR	All Teams
5/06/2017	FBCFF1 and FBPR1 will no longer have physical locations in Block 3 Reporting. The only physical locations will be in Admin Block 6.	JAH	case managers, Admir
15/06/2017	Implementation of Forensic Register for receipt of new cases See Minor Change Form: Comparing the use of Sigma Proteinase K and QlAamp	KDS	all teams
30/06/2017	DNA Investigator Proteinase K for use in QIAsymphony Pre-Lysis. Sigma Pro K can now be used as a substiture for QIAamp DNA Investigator Kit Pro K. Comment added to SOP 33758	LBR	all teams
10/07/2017	Reference sample collections removed from SAIK form (QIS31281) as per QPS advice	KDS	ER
10/07/2017	QIAsymphony AS module put into use today (2uL and 18uL verifications have passed)	LBR	All teams
11/07/2017	Oral swabs taken as reference samples (as per GMO notes) and present with SAIKs to be noted in examination, and returned to SAIK. Nil action required as a reference sample - as per QPS advice	KDS	All teams
24/07/2017	STARlet B implemented for Pre-PCR	LBR	all teams
14/08/2017	Test Quant criteria and program changed to be in line with Test Amp Criteria, as per Minor process change. Comments added against QIS 19994.	MLM	All Teams
19/09/2017	See Minor Change Form: Comparing the time between reagent preparation and addition to samples. Reagents can now be prepared first thing in the morning for use throughout the day. Comments added to QIA 34044 and 34132.	MLM	All Teams
2/11/2017	Current arrangements in DNA Management Section: A/Insp Ewen Taylor, A/Sen Sgt Libby Harris; S/Sgt Ruben Collopen (Quality)	JAH	all teams
7/11/2017	Pre-Lysis storage period extended from 4 days to 8 days - see relevant Minor Change Proposal	LBR	All Teams
7/11/2017	FPP will no longer continue to check exhibits in Auslab. This means all new sample submissions will be processed in FR. AUSLAB reworks will continue to be	PMB	All Teams
27/11/2017	processed in AUSLAB until further notice. AUSLAB Evidence association of FTA Evidence samples (FR submitted) no longer	KDS	All Teams
20/44/2047	required BSD software upgraded from version 4 0.4 to version 4 0 5 0 and in use (on	KDO	00.7
29/11/2017 22/01/2018	Windows 7 PC) Profiler Plus reagents have been exhausted (AB have ceased manufacturing more	KDS LBR	OO Team All Teams
22/01/2016	reagents). All P3 samples now to be processed in PP21 DNA Insufficient for Further Processing EXH line to be used for P1-3 samples in the	LDK	All Teams
12/02/2018	range from LOD <x<0 0088ng="" td="" ul.<=""><td>JAH</td><td>All Teams</td></x<0>	JAH	All Teams
20/02/2018	Emails for 'incorrects' are no longer required to be sent from Seniors to DNA Management for FR samples. AUSLAB samples still require an email.	JAH	case managers
23/02/2018	Eyewash station removed from Rm 6110 - no longer required	KDS	All Teams
3/04/2018 5/04/2018	Saftey shower removed from Rm 6110 - no longer required Cease analytical processing in AUSLAB. Further reworks to be done in FR	KDS PMB/JAH	All Teams All Teams
13/04/2018	Project 192 approved and Bone extraction on QIAsymphony implemented. PCI bone extraction ceased.	F IND/JAIT	All Teallis
26/04/2018	HP2 Forensic Technicians to be trained in the manual sampling of Reference Blood FTA samples. See minutes of management team meeting (today's date)	ARM	Evidence Recovery
6/04/2018	Interim workflow for P3 Single source profiles. If profile appears affected by amp issues, copy down designations without STRmix and load to NC DD thosees profiles where there is one peak for a locus (allele, 0). NTER1 result line to be used.	JAH	case managers
27/04/2018	Unchecking the RPT boxes and QAF boxes for link lines not to be reported because they were warm links are now going to be reported as for normal cold	SMJ	Intel
23/04/2018	links. QlAsymphony B implemented following verification approval (Project 194)	LBR	All teams
30/04/2018	All fridges and freezers in Forensic DNA Analysis deemed non-critical (for temperature monitoring) as per Project #195. Calibration on Fridges and Freezers	KDS/KML	All teams
4/05/0040	not required Archiving of PP21smatch as at 12/04/2018. Archiving Staffdatabase key as at	KDO	A II 4
4/05/2018	04/05/2018. No longer required as all data secure in FR (access to Quality and Managing Scientist only) Admin will use Unallocated Statement worklist in FR to see what casefiles require	KDS	All teams
9/05/2018	creation. A note will be added to the statement request when casefile is created. Admin can be emailed for non-statement cases that are requested to have casefiles created.	JAH	Reporting Scientists
11/05/2018	As at 11/05/2018 any reference samples with Category=Reference that do not require processing (eg. Coronial blood tubes), can be finalised with an appropriate result line eg. IPNE - tems prioritised not examined at this time. This will enable to sample to complete and remove from the 28-Audit reference list	KDS/KML	all teams
11/05/2018	Please note that SSLU will now only use the FR for court notifications; all comms and notices will be located in the one spot.	JAH/Andrea	Reporting Scientists
28/05/2018	Admin Generic Email Account has changed from FSS_DNA_Analysis_Admin@health.qld.gov.au to	Wendy	all teams
13/06/2018	FSS FDNA.Admin@health qld.gov au as of today Project #188 approved - Retain Supernatant on Maxwell 16 for Differential Lysis and samples which do not require differential lysis extraction (routine retain supernatant) has been validated and implemented.		
14/06/2018	As at 14/06/2018 eye wash stations to be flushed monthly (no longer weekly) as per risk assessment 26268	SAB/KDS	all teams
24/07/2018	See relevant Minor Process Change - Quant transition auto dilution threshold has been reduced from >10ng/uL for P2 and >20ng/uL for P3 samples to greater than or equal to 5ng/uL for P1, P2 and P3 samples.VSTS job 974 has been completed to update this threshold in Quant Transition.	LBR	all teams
19/07/2018	Case File Identifier on each page of case file - whether case file has a statement or not. Standard 4.13.2.1 refers System to uniquely identify, or link all records in or pertaining to the case record	Wendy	all teams

Implementation Date	Details Email from S/Sgt Simpfendorfer, QPS DRMU: Hi Kirsten,	Project Leader	Area Affected
	Can I request that Intel letters be sent to our business account in the future so that either the Inspector, Snr Sgt Collopen or myself can action them.		
	The business account email is		
	If you have any questions please feel free to contact me.		
31/08/2018	Kind regards	PMB	All
	Gerard		
	Gerard Simpfendorfer Senior Sergeant 4009415 Officer in Charge, DNA Results and Sample Management Units DNA Management Section, Forensic Services Group Operations Support Command, Queensland Police Service		
Aug-18	Insp Neville advised Forensic Officers to only submit 1 trace DNA sample in Volume Crime Cases Email from Insp David Keatinge, QPS Quality Management Section, advising change in process for QPS labelling of items, as follows: Good morning Cathie	CJA (added retrospectively)	
4/09/2018	FSG has changed policy with respect to the labelling of DNA subsamples in crime scene envelopes. The inclusion of any details in the 'Exhibit Description' field is now optional. For your information.	PMB/CJA	ED Poporting
4/09/2016	Regards Dave	PINIB/CJA	ER, Reporting
	QPS direction regarding change in procedure for exhibit descriptions acknowledged by CJA 04/09/2018.		
5/09/2018	NC DD-IFA (N FA/Bonaparte) is now live for all jurisdictions. Implementation to follow. NWQPS samples: FRIT to validate finalised and PDA worklist samples only. Lab	JAH, SMJ	FRIT
12/09/2018	staff are to validate all those still in active analytical processing. In-tube samples not yet extracted will be repackaged and returned to QPS (by HP2 staff). Other samples (non-intube) not yet extracted will be stored to "No Further Work" boxes. Once processing has been intiated samples will be stored to permanent storage boxes	KDS/ARM/LBR	All
19/09/2018	NC DD delete samples are now able to be ordered on barcodes that are not identical. E.g <9digit ref samples reworked for confirmation can be deleted by ordering the NCIDD delete in the exhibit testing of the new confirmed barcode	SMJ	FRIT & Quality
24/09/2018	CEQ checkers will be leaving -1 repeat stutter peaks on casework positive controls. PDFs will now show the stutter peaks on the positive controls	MLM	All
24/10/2018 3/01/2019	FR Downtime occurred 24/10/2018. LR's no longer reported for intel links	KDS SMJ	All Intel
16/01/2019	STRmix v2 6 0 implemented in this month, no earlier than 16 Jan.	EJC, ARM, JAH	Case managers
4/02/2019	Sen.Scientist Quality and Projects to meet with project teams regularly (Refer to minor change document)	KDS	All
4/02/2019	QlAsymphony A and B re-implemented today following approval of Project #201	LBR	All Teams
15/02/2019	QS5 implemented today (7500 removed from use)	LBR	All Teams
	I have chased up with QPS on current contact points:		
	- Contacting for ownership information in the absence of a Forensic Officer: Sgt Ken Gee Kee		
28/02/2019	 Contacting for information on likelihood of obtaining reference samples: Sgt Carolyn Hoffman Contacting for information on statement timeframes, or further items being received, notification of DPP-initiated statements: Sgt Ken Gee Kee Urgent release of results so a statement can be released: S/Sgt Ruben Collopen. 	JAH	FRIT
1/04/2019	FR Workflow Diary for Analytical and Operational staff implemented.	MLM	Analytical and Operational Teams
10/04/2019	The D12[16 3] allele has been included as a virtual bin in the PP21 allelic ladder. See relevant Minor Change form.	LMF/LBR	All
20/05/2019	STARlet C implemented for CE prep today	LBR	All
22/05/2019	Stratification of Paternity, paired kinship and missing person calculations implemented. Use of QIS 35100 calculator to accompany Kinship calcs. As at 30/05/2019 for casework and reference sample RE-CE results as a first/only	JAH	Paternity Reporting staff
30/05/2019	result can be accepted without rework. This change has been made with the implementation of the STARlet in CE; As plates are now scanned by the instrument (to confirm correct batch/plate selection using the barcode label), and the pipetting positions are file determined (no manual selection which can be susceptible to human error).	KDS	All
6/06/2019	See Minor Process Change "Updated CE STARlet Whole Plate Transfer Program" regarding change to order of CE plates being prepared on the STARlet C.	LBR	All
21/06/2019	Start STRmix v2.6.2 upgrades - HP2 PCs upgraded, routine use of STRmix v2.6.2 by HP2 staff start 24/06/2019	ARM / JAH	All staff with STRmix v2.6.0 installed on PC
1/07/2019	See Minor Change document "Additional virtual bins added to ladder at D6 [18.3,20 2], D21 [29.2] and D12[16.3] for reference samples" regarding addition of virtual bins as per document title.	LMF/LBR	All Staff

Implementation Date	Details Change in bone processing equipment cleaning protocol:	Project Leader	Area Affected
5/07/2019	Cleaning of the bone crushing equipment using the dishwasher as per Proposal #148; Use bleach and / or Trigene followed by 70% ethanol (as appropriate) to clean the remaining equipment in line with other Evidence Recovery and Analytical laboratory equipment protocols	ARM	ER
11/07/2019	Finalisation of STRmix v2 6 2 upgrades - all required PCs now have had this	ARM / JAH	All staff with STRmix v2.6.0 installed on PC
24/07/2019	version installed and challenge plates run with results checked. First use of destruction functionality in FR live	KDS	All teams
30/07/2019	See Minor Change form regarding addition of Penta E [26] virtual bin for casework and reference	LBR /LMF	All teams
19/08/2019	Implement QIAGEN Pro K for routine maxwell and differential lysis batches.	SCN/LBR	Analytical
23/08/2019	First Intelligence Report using NIFA released to QPS Default setting for deconvolutions is 2x iterations: 200 000burn in, 100000 post burn	SMJ	Reporting
29/08/2019	in.	JAH/HKP	All staff with STRmix v2.6.2 installed on PC
13/01/2020 16/01/2020	GM v1 6 implemented from today Cotton swabs now used for positive blood and diff controls	LBR JSM	all areas ER/ Analytical
3/02/2020	Baseline reviewed on 3500xL A following laser change. Previously implemented LOD and LOR thresholds maintained. See report I:\AAA Analytical\Audits and Reviews\AAA 3130-3500\Baseline Reviews\Review 3500A baseline post laser	LBR	All Teams
10/02/2020	change 20191105\Final Report STRmix v2.7 implementated on some PCs, to be rolled out from this date. Some deconvolutions from this date will be in v2.7.	JAH	All staff with STRmix installed on PC
8/05/2020	Chewing gum samples to be processed on Maxwells from today given. Background: some chewing gum brands partially dissolve during the pre-lysis step of extraction, producing a viscous lysate. The QIAsymphony has difficulty pipetting these lysates. When processed on the Maxwell, these lysates are added manually by the operator to the cartridge and therefore this is less of an issue.	LBR	ERQ
19/06/2020	Maxwell FSC implemented	LBR	All Areas
26/06/2020	Equipment list (I Drive) archived, no longer required all equipment within FR Refer to Minor Process Change document - Use of Automatic Baseline Function	KDS	ERQ
14/07/2020	when Analysing Quantification Results on the QuantStudio®5. Baseline function	LBR	All areas
11/08/2020	changed to automatic from Monday 20/07/2020 Any new samples for cases previously processed with STRmix v1 05, v2.0.6 and v2.5.11 are to be run with STRmix v2.7. If references are received for these old cases, strategy to be discussed with Senior Sceintist. I:\Change Management\Minor	JAH, KDR, SMJ	Case Managers
25/08/2020	Change Forms - completed\STRmix versions and retention To contact DNA Mgt by phone, to use 3364 6916 as the best number. Contact via email continue to use generic DNA Mgt email address.	JAH	Case managers
25/08/2020	The incoming mask for QPSFREG (QPS Forensic Register) GSI Interface has been updated to prevent EXH (Exhibit Reports) requests being registered in AUSLAB when received via QPRIME interface	PMB/TEN	All areas
9/09/2020	The incoming mask for POLARIS GSI interface has been updated to prevent registration of new FTA samples, destructions and withdrawals continue to be registered in AUSLAB (change request LC-1730)	PMB/TEN	all areas
20/10/2020	Teeth Extraction using QIAsymphony approved and implemented.	LBR/MJC	All Teams
16/11/2020	Replacement of BSD 600 Series II with BSD Ascent instrument for FTA processing	KDS/CKS	All Teams
30/11/2020	Implementation of new approach to examination for semen / spermatozoa based on outcomes of Project $\#181$	ARM	All teams
6/01/2021	Implementation of the INT (tem has been examined/subsampled) to be used for all parent items that will not have a final result line issued (eg SA K parent barcode).	CJA	All Teams
11/02/2021	Minor change completed: Al and homozygous thresholds for controls and ENVM recorded (see minor change form) for application on CW 3500xL implementation on 15/02/2021	LBR	All Teams
11/02/2021	Separate authorisation from the state coroner is not required if QPS request testing on tissue wax blocks. See G:\ForBiol\QPS Communications\Comms from the Coroner	JAH	ER and reporters
15/02/2021	3130x/ removed from use. 3500xL implemented for all casework PP21 processing	LBR	all teams
4/03/2021	Missing person module of N FA released today. This function allows for restricted searching of pedigree or individual against only Unknown deceased samples	SMJ	Reporting
16/03/2021	Removal of "Hair examination" from NATA scope of accrediation as at 16/03/2021 refer to (I:\Quality & Projects\NATA\Reduction in scope 2021) Communication to staff commenced as a result of removal of NATA hair	KDS	All teams
14/04/2021	examination accreditation. Refer to I:\Change Management\Communications.	KDS/JAH	All teams
4/05/2021	Implementation as of 19 April. Implemented Minor Change - Mastermix Position change from today	LBR/AKD	Analytical
13/05/2021	Implemented STRmix v2.8 for casework from this date. At this stage by exception, routine will follow when suite of licences are received, installed and tested.	JAH, ADA, ARM, CLJ, EJC	
21/06/2021	Implemented Data Collection Software v4 on 3500xL A	LBR	All Teams
14/07/2021	Action from OQI 54954 - Kinship program no longer has the dataset STRBASE ALL (1036) visible for selection. The only datasets visible for selection are three PP21 datasets, and three STRBASE datasets for paternity CTS cases. At the same time as making this minor change, the default datasets in Kinship are now marked to be the three PP21 datasets, with the three QLD datasets no longer visible for selection. If these datasets are required at some stage, a user with a higher level of access (eg Team Leader) can reinstate from the Administration tab.	JAH	Reporting
19/07/2021	SS with high stutter guidelines distributed today. Comment added to SOP 17117. Email sent to all Cmers. See also G:\ForBiol\AAA Forensic Reporting & Intel\AAA Reporting guidelines\Proposed SS guidelines.	KDR	Case Managers
28/07/2021	HP2 Forensic Technicians to sample blood cloth samples in line with exisiting competency for the sampling of FTA cards and Guthrie cards	ARM	Evidence Recovery

Implementation Date	Details	Project Leader	Area Affected
20/09/2021	Admin team to no longer create casefiles for Category 1 and 2 cases with statement requests unless they are Sexual Assaults. Comments added to SOPs 17117 and 34248. Email to be sent to all Reporting Scientists and to Admin.	AKL	Reporting Scientists, Admin
19/10/2021	Minor Process Change: Promega change of PP21 QC detection from 3130 series to 3500 series instruments.	LBR	Analytical
12/11/2021	Minor change to process - Any manual reference samples that are received e.g. hair and other non-FTA samples should be assessed by Evidence Recovery and the category type checked. If the category type is anything other than "reference" it should be referred to the Quality team to have the category changed to "reference". A notation will be added to the sample to state the reason for changing the category type, for example "Category manually changed from "Hair" to "Reference" by Quality team to ensure correct reference processing".	Quality	Evidence Recovery
10/01/2022	Implementation of AB Proflex thermal cyclers to replace AB 9700s- Project# 199	LBR/MLM	Analytical
2/03/2022	Effective immediately blood positive controls will now be labelled orange, to differentitate them from semen controls that will continue to be labelled white POVA checks and STARlet checks now done at 3 volumes (nominally 10%, 50%).	SMCK/KDS	ER&Q
25/03/2022	and 100% of POVA volume). This is the addition of the 50% volume, previously only 2 volumes 10% and 100%	KDS	ER&Q
13/04/2022	Data Collection Software (DCS) version 4 implemented on 3500xL B	LBR	All
20/04/2022	Custom protocol added to QIA-AS A and B instruments to verify at 10uL. For all results provided to clients via email - will need to provide the results as a PDF in addition to any other formats and this statement must be included in the	LBR	All
21/04/2022	email: "The pdf document in this email is the report issued by Forensic and Scientific Services. Any other attachments or information provided is not considered to be the issued report."	Quality	All
3/05/2022	New version of Foresic Register (FR) has gone live at this date	PMB/KDS	All
17/05/2022	New Model Maker settings applied from 17 May to PCs with STRmix. Settings based on findings from Proposal #199	JAH	Case managers
19/05/2022	Additional line added to Appendix: The signed Statement is the report issued by Forensic and Scientific Services. Any other attachments or information provided is not considered to be the issued report. To be used for any new SoW from this date.	JAH	Reporting Scientists
19/05/2022	QS5-A was overloaded with .eds files stored on the instrument and ceased processing new batches. These files are the quantification experiment files that are used to re-open a previously processed batch. They are initially saved to the instrument computer D drive and are then manually transferred to I drive each month. Emma Campbell (ThermoFisher) deleted all eds files from the years 2018 - 2020 from the instrument and it is now functioning again.	AKD	Analytical
6/06/2022	As per Premier's request we have commenced amplifying all samples in the previous DNA Insufficient range 0 001 – 0 0088 ng/ μ L.	LBR	All
23/06/2022	National Policy - Cross Jurisdictional Familial DNA searching for the Investigation of Crime in Australia 2022 approved by ANZPAA Board for release.	JAH	NIFA staff
27/06/2022	Cig butt standard wording - description changed from -"Approximately 5 x 5 mm of filter paper was sampled for further analysis" to "A portion of filter paper was sampled for further analysis"	JSM	ER and Reporting Scientists
13/07/2022	Change to the Quant trio SOP implemented on 12/07/2022: "Quant Trio user guide recommends centrifuging at 3000 rpm to remove bubbles from the quant plate. Current SOP lists speed as 2000 rpm. Update to 3000 rpm as this is more effective to remove bubbles." The PP21 User guide and this SOP is silent on centrifuge speed to remove bubbles. Previously 2000 rpm was used for PP21 for full volume/EREF and FTA. 3000 rpm is used for consistency for Quant and all PP21 amp types.	LBR	Analytical - Laboratory Assistants.
5/08/2022	A/DG Memorandum received requiring an immediate change to wording in Statements of Witness, effective from the next statement in draft. Wording to be added to body of statements and for results reported as D FP - DNA Insufficient for Further Processing result line. Low levels of DNA were detected in this sample and it was not submitted for further DNA profiling. The sample may have insufficient DNA to result in a DNA profile suitable for interpretation. It is possible that further testing may result in an interpretable DNA profile in some cases.	A/DG David Rosengren via A/EDFSS Helen Gregg	Reporting Scientists
12/08/2022	Pre-PCR STARlet computers upgraded to WIN10 with Venus 4 software. STARlet B verified and implemented on 15/08/2022. STARlet A verified and implemented on 18/08/2022.	AKD/LBR	Analytical Scientists
19/08/2022	As per A/DG Memo 19/08/2022 - all P1 and P2 samples in quant range 0.001-0.0088ng/LL will undergo an automatic microcon to 35uL, and then an amplification. If the scientist considers it potentially beneficial to perform a second amplification post-microcon, this will need QPS approval due to the exhaustion of the DNA extract. Follows that any sample that might be exhausted (eg consideration of microcon to full) will need QPS aproval.	A/DG David Rosengren via A/EDFSS Helen Gregg	Analytical and Reporting Scientists
12/09/2022	As per crown law advice (Email CJA 12 09.2022) Statements signed and produced from the 9th of September 2022 onwards should refer to Rex, rather than Regina.	CJA	Admin and reporting scientists
14/09/2022	3500 xl B has a new computer connected. All PP21 protocols have been transferred from 3500 xl A to B. Plate ran on 14/09/2022 and passed. 3500 xL B	SCN/BUA/PMB	Analytical Scientists
23/09/2022	back in use. CE STARlet computer upgraded to WIN10 with Venus 4 software.	AKD	Analytical Scientists

Details of communications that have occurred (to who, when and Other notes for consideration how)			T 0 T 0	All reporters wa microsont Teams on C2/02/2021 EXEMPLAR ONLY
Actions	Removed artefact from FR GeneMapper table.	Adds contributor to otherwise single Annotated eggs and re-loaded to Forensic source assumed known contributor. Register	Notations added to case in Forensic Register.	Added to odd profile register
Assessment of Issue		Adds contributor to otherwise single source assumed known contributor, beint to fortefoot and sometimes tutth	another contribution dropping out.	no experialitare of money, unie of resources required.
Teams Affected				Reporting
Who can made the decision on what needs to happen?				
Has it been seen before? (if Yes - Where?)			>	(in Case XXXXXXXXX) John Smith
Define Issue		Apparent artefact at D18S51. Artefact shifts between labelling as a 17.1 or 17.2 variant	allele. No stutter is observed for this artefact.	Only observed in samples from pen-anal, rectal or penile areas
Item added by				/01/2021 John Smith
Date Raised				1/01/2021

JH-53

EB Check - overview and workflow

- Macro includes profiles greater than 6 alleles.
- Macro has separate tabs for single source and mixtures.
- Macro displays matching profiles in groups. The profile highlighted in white is the profile that the profiles underneath have matched to.
- Macro displays loci of matching profiles in different colours: green is for at least 2 alleles matching, yellow is for 1 mismatch, light orange is for 2 mismatches, bright orange is for a mismatch at Amelogenin.
- EB check to be performed at desk, single profile to be displayed on screen or printed and checked against matching profiles flagged by the macro.

Samples run through analytical

1

Batch paperwork placed in in-tray near plate reading computers

 \downarrow

Plate is read by Plate reader

Once plate is read, profiles are PDF'd and JPEG'd by reader

Plate reader enters batch ID onto whiteboard on side of filing cabinet at the end of the plate reading bay

QPS/Quality Flag check

- batch audit note entered
- if Quality Flag checking completed before EB check, transfer paperwork to EB checker (enter batch audit note)

1

EB check (2 people per day based on roster system)

- From AUSLAB main menu: 5, 2, 2, enter batch ID, CTRL F11, 'y', save in I:\auto 'space' ext 'space' ur\'Batch ID'.txt
- Run Macro from I:\Macros\EB Macro, enable macros, click on 'Extraction Batch Check', enter initials, double click on Batch ID
- assess matching profiles (using # of mis-matches, peak heights etc)
- complete 'EB check sample detail investigation' form for closely matching samples, have Quality Flag checker review, scan and place file with Batch within P:\ drive as
 - GMPCWyyyymmdd nn EBsample, retain with Batch paperwork
- Quality Flag checker and EB checker will need to check Evidence Recovery details in order to investigate possible contamination at that stage eg. Bench details, time/date of examinations, operator
- complex mixtures to be brought to attention of Quality Manager, specimen note "profile too complex for EB check (note batch ID)" to be entered
- any suspected contamination will result in the batch being quarantined until investigation complete
- enter batch audit note to say EB completed and if passed
- file EB check paperwork with batch paperwork in analytical

Once comments from both Quality Flag checker and EB checker have been entered and no remaining flags are to be investigated:

- last person to enter batch audit note completes batch (F7in AUSLAB on batch), returns paperwork to analytical (note entered)
- case managers free to manage results from batch (if both Quality Flags and EB check have passed)

Case managers to manage rework results (>6 alleles) and perform EB check

- access EB Macro, click on 'Show update & Check Macro (for reworks)
- enter details into fields (sample ID, profile obtained, reader comments)
- click on 'Update and check'
- find the original CWGMP 9PLEX batch to write over
- assess updated profile against matches
- if sample passed, enter specimen note 'EB check for rework performed passed'
- file rework EB check paperwork in blue folder next to STAREV intray

November 14, 2008

Background

Following a request from the Director, Mr Greg Shaw, a review of procedures was conducted by Drs Sloots and Whiley (the reviewers) at the Forensic and Scientific Services laboratory, Clinical and State-wide Services, Coopers Plains, pertaining to the extraction of nucleic acids from samples submitted for analysis.

The reason for this review related to a previous episode in the laboratory which resulted in anomalous results and which appeared to be linked to the operation of robotic instrumentation utilised in the nucleic acid extraction process.

During their visit, the reviewers were made aware of the operations applied in the general laboratory from receipt of specimens to issuing of results, and then examined in detail the bench process relating to the pre-digestion of specimens and the extraction of nucleic acids using the Perkin Elmer MultiPROBE II PLUS HT EX with Gripper Integration Platform.

All aspects of these operations were scrutinised including staff input and instrument operation.

Findings

It was obvious to the reviewers that extensive measures were applied by all staff to prevent the misidentification or cross contamination of samples. There was appropriate use of personal protection equipment and other protective measures to prevent contamination of the work environment with extraneous nucleic acid.

The procedures currently in place for the Off-Deck Lysis and MPII extraction appeared to be adequate and specifically designed to prevent cross contamination of test samples.

We agree with the Forensic Services Management team that the previous issue of possible cross-contamination of samples most likely related to the use of adhesive film in sealing the deep-well plates used in the Off-Deck lysis procedure. The type of plate used, and the period of storage at reduced temperatures have in our experience caused similar problems in molecular diagnostics. The subsequent decision to change this procedure to the use of capped tubes has clearly solved this problem.

The use of robotic equipment for the extraction of nucleic acids has some considerable benefits for a busy laboratory, and prevents human error introduced as a result of repetitive actions. However, the efficient use of such instruments requires the proper maintenance and calibration be performed at the requisite time intervals. These appeared to be adequately performed at the time of review.

It may appear that the original issue concerning the cross-contamination of samples in the deep-well plates could have been prevented if this change in procedure had been fully validated against existing protocol when the new method was introduced. Although most

laboratories would have considered this change to be minor and therefore accepted without validation, it clearly demonstrates that all changes in procedure, no matter how minor, need to be validated according to a standardised protocol before their introduction as standard operating procedure.

Items for Further Consideration

During the review process some items were identified which may require further consideration by the management staff of the Forensic and Scientific Services laboratory.

These are:

- Develop a standard validation protocol for each procedure based on the guidelines described by J Butler (<u>www.promega.com</u>; September 2006). Incorporate these into the Standard Operating Procedures for the laboratory.
- 2. We advise that the number of negative controls included in each batch of extractions be increased to comprise at least 10% of the total number of specimens tested. These controls should ideally be distributed randomly over the plate. Currently one negative control is included with 47 samples.
- Quality assessment might be increased by testing a control plate once every 3-4
 weeks on each of the MultiPROBE II PLUS platforms. We would suggest alternating
 between the soccer ball, zebra and checkerboard formats.
- 4. It was noted that the magnetic particles used for the nucleic acid extraction had a tendency to settle quickly, thereby blocking the filter tip and potentially producing a false-negative result. At the time of review this was not a problem as the attending operator was diligent in observing all stages of this process. We would like to reiterate however, that constant observation by the operator of all processes leading up to and including the addition of magnetic particle is necessary to ensure that failure of the robotic system does not occur.
- 5. Finally, it was noted that the laboratory design allowed traffic from the amplification/post-amplification area into the lysis/extraction areas. Presently this carries moderate contamination risk, as the amplification protocol is limited to 28-32 cycles. However, if this protocol is changed in the future to detect low copy nucleic acid (greater than 32 cycles) the risk of carrying post-PCR product into the extraction area would be high, and work-flow dynamics must then be carefully examined to minimise that risk. Likewise, sample cross-contamination during specimen handling and extraction processes will assume greater relevance when contemplating detection of low copy nucleic acid, and would necessitate stringent validation of all steps.

Theo P Sloots, PhD, Grad Cert Management.

David M Whiley, PhD,LLB, G/ad Cert Law

JH-55

June 2009

Department of Justice and Attorney-General Office of the Director of Public Prosecutions State Law Building 50 Ann Street BRISBANE Q 4001

To Whom It May Concern:

In December 2008, the A/Managing Scientist of the DNA Analysis Unit and the Senior Director of Queensland Health Forensic and Scientific Services advised the Director of Public Prosecutions, Executive Director and two Principal Crown Prosecutors of DNA testing that had been conducted in a period where some results were the subject of an adverse event.

Written advice has been received from the Solicitor-General and Crown Law regarding disclosure of these events within DNA Analysis evidentiary Statements. The adverse event has been identified as falling within three categories. The advice was that an insertion of a paragraph into statements, detailing information about the particular category, was an appropriate course of action.

The addendum statement is to replace any previously issued evidentiary Statement of Witness and the recommendation was that it would be issued directly to the DPP.

Hardcopy records of any investigation into the adverse event and subsequent results have been included within the casefile, which will be retained with the DNA Analysis Unit indefinitely.

A copy of the casefile is available, upon request with adequate time frames, to either Prosecution or Defence at anytime.

Yours sincerely,

Cathie Allen A/Managing Scientist DNA Analysis Unit



Your ref:

Our ref.

LT8/HEA027/7206/HUR

Rob Hutchings

Department of Justice and Attorney-General

19 December 2008

Mr Greg Shaw A/Senior Director Pathology Queensland Queensland Health

Attention:

Cathie Allen

Acting Managing Scientist

DNA Analysis

Dear Mr Shaw

DNA test - Prosecution Disclosure

I have received the joint opinion from the Solicitor-General, Walter Sofronoff QC and Mr Peter Davis SC, as requested. A copy is attached for your records.

You will see that the opinion substantially reflects what was discussed in conference with Mr Davis. I would draw your attention to paragraphs 23 to 28, on pages 7 to 9.

Counsel have identified the three categories of result to which you directed our attention, namely:

Category A – Tests that have been conducted in the relevant periods, but there is no evidence or suggestion of evidence of an adverse result;

cc:



DNA test - Prosecution Disclosure

Category B – Tests that have been conducted in the relevant periods, and there has been an adverse result but there is no sample available to perform a second test; and

Category C – Tests that have been conducted in the relevant periods and there has been an adverse result, there is a sample available to perform a second test, and the second test has been performed.

With respect to Category B, Counsel consider that it is appropriate that there be no reporting. I understand this is the decision you have already taken.

In respect of Category A, your report should disclose:

- (a) That the test was conducted in a period where some tests were the subject of adverse results;
- (b) That the test was not the subject of any adverse result;
- (c) Explain how the conclusion was reached that the test was not subject to any adverse result:
- (d) If there was re-testing, disclose the results of that re-testing; and
- (e) The case file should be preserved and the DPP advised of the existence of it with a covering letter.

With respect to Category C circumstances, your report should disclose:

- (a) That the test was conducted in the period during which there were adverse results;
- (b) That the test was the subject of an adverse result;
- (c) The cause of the adverse result;
- (d) The fact that the test was re-done;
- (e) What steps were taken to eliminate adverse results in the re-testing; and
- (f) The case file should be preserved and again, an accompanying letter should be sent to the DPP advising of the existence of it.

As Counsel have mentioned, both they and my office would be happy to help you settle on the final words of any particular report you might like to send to us.

Thank you for your instructions. If I can be of any further assistance, please do not hesitate to contact me on 3239 3375.

Yours faithfully

Rob Hutchings
Assistant Crown Solicitor
for Crown Solicitor

encl



Solicitor-General Queensland

MEMORANDUM OF ADVICE:

RE: QUEENSLAND HEALTH – DNA TESTING; DISCLOSURE OF ADVERSE RESULTS

Introduction

- We are briefed to advise as to what disclosure ought to be made in forensic scientists' expert reports where irregularities or possible irregularities have occurred in forensic deoxyribonucleic acid ("DNA") tests conducted by Queensland Health in certain periods in 2006 and 2007.
- 2. DNA testing has become an important forensic tool especially in criminal cases. The current testing examines ten regions of DNA. One of these gives an indication as to the gender of the donor. Characteristics are identified from each of the other nine sites. Statistical data then identify the percentage of the population which has those particular characteristics at that particular site. That process is repeated in relation to each of the nine sites, thus narrowing enormously the statistical chance of two persons within the area of the crime scene having the same DNA profile.
- 3. There are numerous recorded cases of convictions being obtained in reliance upon DNA evidence where, without that evidence, there would not have even have been a case for a trial judge to leave for a jury's consideration.
- Of course, the reliability of the test results is dependant upon the integrity of the testing process.

- 5. The National Association of Testing Authorities ("NATA") is Australia's national laboratory accreditation authority. Queensland Health's laboratories comply with NATA requirements.
- 6. The Evidence Act 1977 now contains various provisions to facilitate the admission of DNA evidence. Importantly, s 95A of the Evidence Act provides:

95A DNA evidentiary certificate

- (1) This section applies to a criminal proceeding.
- (2) A certificate, in the approved form, purporting to be signed by a DNA analyst and stating any of the following matters is evidence of the matter—
 - (a) that a stated thing was received at a stated laboratory on a stated day;
 - (b) that the thing was tested at the laboratory on a stated day or between stated days;
 - (c) that a stated DNA profile has been obtained from the thing;
 - (d) that the DNA analyst—
 - examined the laboratory's records relating to the receipt, storage and testing of the thing, including any test process that was done by someone other than the DNA analyst; and
 - (ii) confirms that the records indicate that all quality assurance procedures for the receipt, storage and testing of the thing that were in place in the laboratory at the time of the test were complied with.
- (3) If a party intends to rely on the certificate, the party must—
 - (a) at least 10 business days before the hearing day, give a copy of the certificate to each other party; and
 - (b) at the hearing, call the DNA analyst to give evidence.
- (4) If the chief executive receives a written request from a party for a copy of the laboratory's records relating to the receipt, storage and

Sections 95A and 133A

- testing of the thing, the chief executive must give the party a copy of the records within 7 business days after receiving the request.
- (5) If a party intends to challenge a matter stated in the certificate, the party must, at least 3 business days before the hearing day, give the chief executive and each other party notice, in the approved form, of the matter to be challenged.
- (6) A party challenging a matter stated in the certificate may, with the leave of the court, require the party relying on the certificate to call any person involved in the receipt, storage or testing of the thing to give evidence at the hearing.
- (7) The court may give leave only if the court is satisfied that—
 - (a) an irregularity may exist in relation to the receipt, storage or testing of the thing about which the person to be called is able to give evidence; or
 - (b) it is in the interests of justice that the person be called to give evidence.
- (8) Any equipment used in testing the thing at the laboratory is to be taken to have given accurate results in the absence of evidence to the contrary.
- chief executive means the chief executive of the department within which the Health Services Act 1991 is administered.

 DNA analyst means a person who holds an appointment as a DNA analyst under section 133A.

 DNA profile means the result from DNA analysis.

 hearing day means the day fixed for the start of the hearing of the proceeding.

 party means the prosecution or a person charged in the proceeding.
- 7. Certificates pursuant to s 95A of the *Evidence Act* are frequently tendered in criminal proceedings. While the certificates are not conclusive evidence of the things certified, they are often accepted by the defence without question. This is no doubt because confidence has been established in the testing regime.
- 8. Sometimes tests can be redone. This depends upon the quality and nature of the biological sample available. In the common case of vaginal swabs for instance, the DNA testing consumes all biological material. In other examples, such as large blood stains, it is often he case that not all the material is used so a second test can be performed.

The Facts Here

- 9. Over the past decade in particular, techniques for the extraction and analysis of DNA have developed and become more sophisticated. Queensland Health purchased four automated platforms which perform the functions of extraction of DNA from the relevant samples, quantification of the DNA in the samples, and amplification of the DNA to aid analysis.
- 10. Over the periods between February and July 2006 and January to October 2007, deficiencies in the processes were discovered when six batches of tests were found to be contaminated. Auditing and retesting was conducted, which identified a further ten batches as being questionable.
- 11. The questionable tests were the subject of "adverse results". There are numerous adverse results which, in one way or another, damage the integrity of the testing process. Most of the tests within the 16 batches have been retested. All except two of the retests have confirmed the original results. In one of those cases, a report had been issued. It has been, or will soon be, the subject of a supplementary report.
- 12. The Director of Public Prosecutions ("the DPP") was alerted to the problem and advice was sought from him as to how disclosure should be made in the DNA testing reports of the difficulties that were encountered. The DPP has declined to provide advice on the wording of the disclosure of adverse results. The DPP says that it is not for the prosecuting authority to draft reports which will then be tendered in cases which the DPP is presenting to the Court. This is a little odd as there is nothing inappropriate in lawyers settling expert reports provided, of course, that the settled report truthfully represents the opinion of the expert.
- 13. A general advice was received from the DPP.² That advice was:

Each matter that is reported upon that was tested in the relevant period should disclose that fact [the fact of the difficulties with the machines over that period]. In my view you should disclose that it was tested during the period when adverse results occurred and whether an adverse result occurred with respect to that particular matter.

Email Todd Fuller, Assistant Director of Public Prosecutions to Greg Shaw of Queensland Health, 11 December 2008

If there was no adverse result you will then need to explain why you exclude that an adverse result occurred with respect to it. If there was an adverse result, or a real possibility of an adverse result, you will then need to explain what that result was, the cause of it and how that cause was eliminated when the matter was retested. The more detail you provide the less time your scientists will have to spend answering questions about the procedure.

The Question for Us

14. Queensland Health wishes to make proper disclosure of the irregularities in the testing, but does not, if possible, wish to use the word "contamination" for fear that this might damage public confidence in DNA analysis in Queensland. Advice is sought as to how the disclosure should be framed.

The Duty of Disclosure

- 15. There is a duty of disclosure imposed upon the prosecution. In Queensland, that duty primarily rests with the DPP. There are both statutory and common law duties of disclosure.
- 16. Section 590AB of the *Criminal Code* provides:

590AB Disclosure obligation

- (1) This chapter division acknowledges that it is a fundamental obligation of the prosecution to ensure criminal proceedings are conducted fairly with the single aim of determining and establishing truth.
- (2) Without limiting the scope of the obligation, in relation to disclosure in a relevant proceeding, the obligation includes an ongoing obligation for the prosecution to give an accused person full and early disclosure of—
 - (a) all evidence the prosecution proposes to rely on in the proceeding; and
 - (b) all things in the possession of the prosecution, other than things the disclosure of which would be unlawful or contrary to public interest, that would tend to help the case for the accused person.
- 17. The term "possession of the prosecution" is defined in s 590AE of the Code:

590AE Meaning of possession of the prosecution

- (1) For a relevant proceeding, a thing is in the possession of the prosecution only if the thing is in the possession of the prosecution under subsection (2) or (3).
- (2) A thing is in the possession of the prosecution if it is in the possession of the arresting officer or a person appearing for the prosecution.
- (3) A thing is also in the possession of the prosecution if—
 - (a) the thing is in the possession of—
 - (i) for a prosecution conducted by the director of public prosecutions—the director; or
 - (ii) for a prosecution conducted by the police service—the police service; and
 - (b) the arresting officer or a person appearing for the prosecution—
 - (i) is aware of the existence of the thing; and
 - (ii) is, or would be, able to locate the thing without unreasonable effort.
- 18. Various sections of the *Code* deal with disclosure that must always be made,³ mandatory disclosure,⁴ disclosure on request⁵ and how disclosure is made.⁶
- 19. It can be seen that s 590AB:
 - (a) Does not limit the common law duty of disclosure;
 - (b) Places the obligation of disclosure upon "the prosecution";
 - (c) Defines the object of the duty of disclosure as "things in the possession of the prosecution".
- 20. By s 590AE, things are in the possession of the prosecution, relevantly, if they are:
 - (a) In the possession of the DPP;

Section 590AH

Section 590AI

Section 590AJ

Sections 590AK and 590AK

- (b) In the possession or control of the arresting officer.
- 21. The *Code* provisions provide similar, if not identical, disclosure obligations to those imposed by the common law.⁷
- 22. Queensland Health strictly does not have a duty of disclosure. However, Queensland Health ought to make proper disclosure in reports of any matter that is relevant to the opinion expressed in the reports and should make all relevant material available to the DPP so that he may make proper disclosure. These steps are necessary for a number of reasons, namely:
 - (a) It is honest and proper to do so;
 - (b) The discovery of relevant material after conviction can, even when there was no duty to disclose the material, lead to the quashing of a conviction;⁸
 - (c) If proper disclosure is not made, then Queensland Health scientists, when giving expert evidence, are open to suggestions that they have failed to make proper disclosure and are biased. This of course undermines public confidence in the system.

The Way Forward

- 23. The focus so far, especially by the DPP, has been upon disclosure in the scientific reports. However, there are documents contained on case files which either are or may be relevant. These documents will evidence the testing, the peer reviews and the compliance with other NATA requirements and will document adverse results and the retesting. Those documents are clearly subject to both statutory and common law duties of disclosure and we have no doubt that the DPP would make those documents available to the defence upon request.
- 24. There seem to be three (3) categories of circumstances which arise here which, of course, require different responses. These are:

Grey v The Queen (2001) 75 ALJR 1708; Mallard v The Queen (2005) 224 CLR 215.

Rattan v The Queen (1974) 131 CLR 510, Mickelberg v The Queen (1989) 167 CLR 259, and for a detailed analysis of the distinction between "fresh" and "new" evidence, see the judgment of Jerrard JA in R v Katsidis, ex parte: Attorney General of Queensland [2005] QCA 229

- (a) Category A tests that have been conducted in the relevant periods but there is no evidence or suggestion of evidence of an adverse result;
- (b) Category B tests that have been conducted in the relevant periods and there has been an adverse result and there is no sample available to perform a second test;
- (c) Category C tests that have been conducted in the relevant periods and there has been an adverse result, there is a sample available to perform a second test and the second test has been performed.
- 25. We understand that, with Category B circumstances, a decision has been made that the results will not be relied upon. That is appropriate.
- 26. In Category A circumstances, the report should disclose:
 - (a) That the test was conducted in a period where some tests were the subject of adverse results. Obviously, the words should be drawn in such a way as to fairly explain what that means;
 - (b) That the test was not the subject of any adverse results;
 - (c) An explanation should be given as to how the conclusion was reached that the test was not subject to any adverse result;
 - (d) If there was retesting, then the results in relation to both tests should be disclosed.
 - (e) Further:
 - (i) The case file should be preserved;
 - (ii) When the report is delivered to the DPP, there should be an accompanying letter advising of the existence of the case file and describing in broad terms what documents are contained in it.
- 27. With Category C circumstances, the report should disclose:

- (a) That the test was conducted in the period over which there were adverse results;
- (b) That the test was the subject of an adverse result;
- (c) The cause of the adverse result;
- (d) The fact that the test was redone;
- (e) What steps were taken to eliminate the adverse result in the retesting;
- (f) Further:
 - The case file should be preserved;
 - (ii) When the report is delivered to the DPP, there should be an accompanying letter advising of the existence of the case file and describing in broad terms what documents are contained in it.
- 28. It is not possible for us to draw the actual wording as this involves scientific input. There is some concern about use of the word "contamination". That word need not be used if the circumstances can be otherwise accurately described. Instructing solicitors can certainly assist in the drawing of the wording and we can, if required, settle that wording.

We advise accordingly.

With compliments

WALTER SOFRONOFF OC Solicitor-General

PETER J DAVIS SC

Chambers

19 December 2008

JH-57

PETER J DAVIS S.C. Barrister-at-Law ABN 29 578 313 997



Mr G R Cooper Crown Solicitor 11th Floor, State Law Building 50 Ann Street BRISBANE QLD 4000

Your Ref: Mr Rob Hutchings

MEMORANDUM OF ADVICE

RE: QUEENSLAND HEALTH - DNA TESTING: DISCLSOURE ADVERSE RESULTS

In December of 2008 the Solicitor General and I advised¹ as to issues arising consequent upon deficiencies being discovered in DNA² tests conducted by Queensland Health in periods between February to July 2006 and January to October 2007. The Solicitor General and I advised as to the appropriate approach to be taken by officers of Queensland Health to the disclosure in DNA testing reports of the deficiencies which have been identified. I have now been provided with draft paragraphs which are proposed to be included in reports.

It is unnecessary to further analyse the facts or the issues as these matters have been canvassed in the earlier advice of the Solicitor General and me.

Suffice it to say there are three categories of circumstances which have arisen. These are:

1. Category A – tests that have been conducted in the relevant periods but there is no evidence or suggestion of evidence of an adverse result;

By written advice

Deoxyribonucleic acid

- 2. Category B tests that have been conducted in the relevant periods and there has been an adverse result and there is no sample available to perform a second test;
- 3. Category C tests that have been conducted in the relevant periods and there has been an adverse result, but there is a sample available to perform a second test and the second test has been performed.

A decision has been made, appropriately I think, that results in Category B circumstances will not be relied upon. However, an approach as to Category B circumstances must be settled upon as in some cases there will be many samples and the report will deal with Category B circumstances as well as Categories A or C.

I have been provided with a draft report which contains disclosures in relation to each of Categories A, B and C. The draft disclosures are as follows:

Category A

"Testing for this case has been conducted in a period where some results were the subject of an adverse event. An adverse event includes a deficient operation or function identified to have occurred within the automated extraction process utilised in testing during the period October 2007 to July 2008. Testing for this case was not the subject of any adverse result. An adverse result is a result which has been affected by an adverse event, whose integrity cannot be verified. This conclusion has been reached by conducting a review of the results and assessing a number of factors, including, but not limited to, the comparison of all other results from samples processed alongside this result, to detect whether the integrity of each sample can be verified. Retesting has been conducted on identified samples which have confirmed information in the original results³."

Category B

"Testing for this case has been conducted in a period where some results where the subject of adverse events. An adverse event includes a deficient operation or function identified to have occurred within the automated extraction process utilised in testing during the period October 2007 and July 2008. Within this case, the adverse event is demonstrated to have affected a result or results and the integrity cannot be verified. There was no remaining sample for retesting to be conducted. These results have therefore been reported as follows 'these samples did not pass our Quality System requirements at the DNA analysis stage and therefore the DNA profiling results relating to these samples cannot be reported'."

The last sentence will only appear where there has been retesting but as I understand it, that will be done as a matter of course.

Category C

"Testing for this case has been conducted in a period where some results were the subject of adverse events. An adverse event includes a deficient operation or function identified to have occurred within the automated extraction process utilised in testing during the period of October 2007 and July 2008. Testing for some samples within this case has been the subject of an adverse event. The cause of the adverse event was identified to have occurred within the automated extraction process. Portions of the sample remained available for further testing. Retesting has been conducted, using an alternative manual extraction method and all quality assurance checks were satisfactory. These samples have been reported as they have been assessed as no adverse event having been detected and the results have passed all quality assurance checks."

Subject to the qualifications below, I consider that the draft disclosures in relation to each of Categories A, B and C are adequate and appropriate. The qualifications though are as follows:

1. The draft report is broken into two sections. The first is the statement of the scientist. This part of the document identifies the scientist, explains his/her position within Queensland Health and his/her qualifications and then proceeds to identify the sample before recording the results of the testing. There is also, in this part of the report, some general explanation of DNA profiling and other associated issues.

The second part of the report is "Appendix 1" headed "Procedural Overview for the Forensic Biology Laboratory of Queensland Health Pathology and Scientific Services (QHPSS)" and "Appendix 2" headed "Technical Information Relating to DNA Profiling at the Forensic Biology Laboratory of Queensland Health Pathology and Scientific Services (QHPSS)". Appendix 1 and Appendix 2 together deal with procedural and technical issues concerning DNA profiling generally. The two appendices do not concern the particular case the subject of the report.

The disclosure is contained within Appendix 1. While I can see the logic behind including the disclosure in Appendix 1, I think that as a matter of prudence, the disclosure should appear in the body of the "statement of witness" itself. Whether the case falls within Category A, B or C, depends on the particular circumstances of the testing in that particular case. Therefore it appears to me that it ought to appear in the "statement of witness" section of the report. The inclusion of the disclosure in Appendix 1 opens the possibility of a suggestion being made to Queensland Health scientists that the disclosure has been "buried".

2. The disclosures in relation to all categories refer to periods of testing in "October 2007 and July 2008". As I understood the instructions given to the Solicitor General and me, the

WIT.0016.0188.0601

-4-

periods in questions were "February and July 2006" and "January to October 2007".

Obviously, the disclosures have to nominate the correct periods.

3. When a report contains tests performed in either Category A or Category C circumstances,

the report should be delivered to the Director of Public Prosecutions ("the DPP") with an

accompanying letter advising of the existence of the case file and describing in broad terms

what documents are contained in it. This will enable the DPP to make whatever disclosure

of documents he considers appropriate. It will also enable the DPP to properly respond to

requests for documents made by defence legal representatives.

In practice, I expect that these issues will most likely arise initially, at least, in the cross-

examination of scientists at committal proceedings. It will be important that any scientist who is

called to give evidence after the delivery of any of these reports which make a disclosure is in a

position to give detailed evidence as to the issues which arose in the periods in question.

Yours faithfully

Peter J Davis S.C.

Chambers

JH-58

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MPII Enhancements Update

26 March 2009 Chiron Weber



Outline

- OQI investigations
- Automated DNA IQ enhancements
- Testing results 1st method
- Testing results 2nd method
- Re-implementation proposal

OQI Investigation

- 15 OQI in total have been raised
- Both EP-A and EP-B
- Dripping was noted
- Condensation adhering to seal was noted
- IQ Off-Line June 2008
- Not possible to find exact cause of contamination
- No clear pattern of events

OQI Investigation

- Investigation
- Contaminated samples found in store plate
- Contamination occurred prior to removal of lysate to store plate
- Mainly off-deck processes prior to lysate removal to Store
- Liquid STORstar
- Storage in fridge with sticky seal
- Addition of Resin
- Very few steps on platform no "drips" seen at these
- Availability of DNA in Store plate indicated low binding efficiency

Automated DNA IQ enhancements

- 1. Change syringes from 500µL to 1000µL
- Change in off-deck lysis volume from 500µL to 300µL
- 3. Removal of liquid STORstar
- 4. Change of deck layout
- 5. Automated addition of DNA IQ Resin
- 6. Off-board mixing of DNA IQ Resin
- 7. Magnet changed from PKI to ABI
- Electronic platemap changed for volumes & new steps
- Risks considered with regards to droplets on side of tips
- 10. Changes reviewed

Change syringes

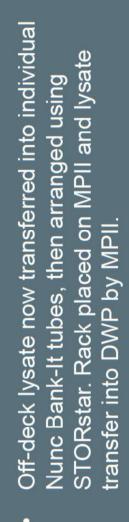
- Now using 1000µL syringes.
- Reduced number of draws and therefore longer lifetime.

Change in off-deck lysis volume

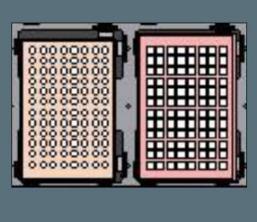
- Volume of extract in well/tube is reduced to minimise contamination risk during shaking/incubating.
- Translates also to a reduction in reagent usage.
- Minimal difference in yield (as found from QPS Tapelift Trial).

Removal of liquid STORstar

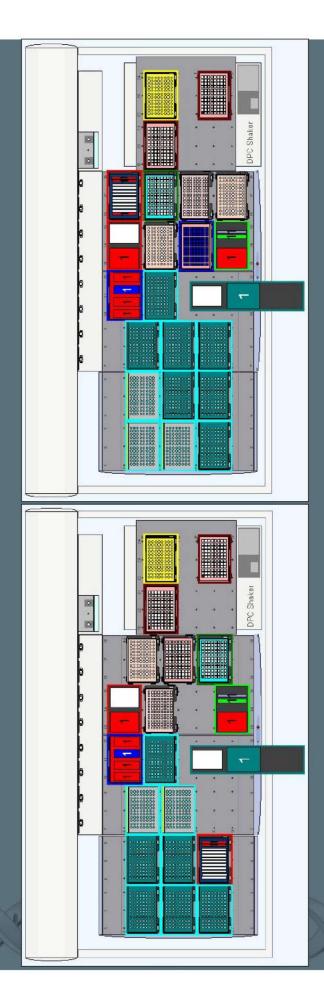
- Off-deck lysate was transferred into 96-DWP with aide of STORstar. DWP was sealed with adhesive film and stored.
- Removal of adhesive film was a contamination risk
- Condensation adhering to seal, did not centrifuge down



Lysates can be stored in fridge or freezer for extended periods.



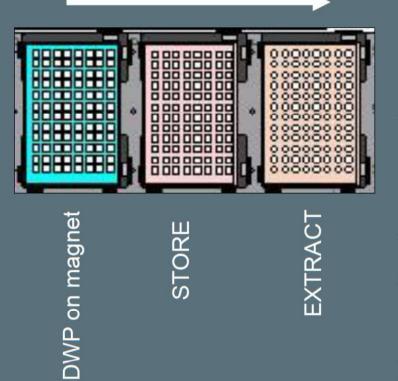
Change in deck layout



V6.2

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DWP

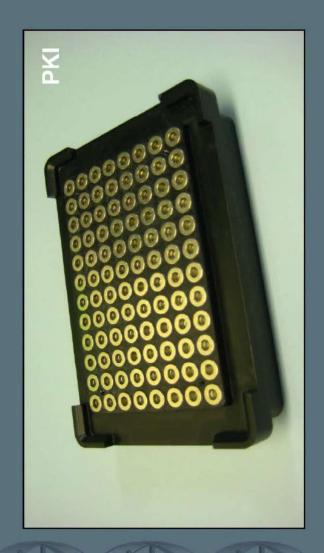
The STORE plate is removed off the deck prior to addition of elute to the EXTRACT tubes.

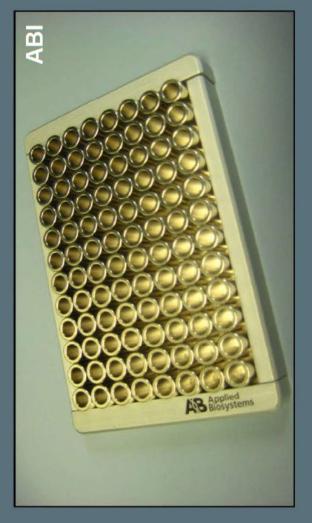
Automated addition/mixing of resin

- Resin was added manually and then mixed by MPII ~1hr.
- Resin now added by MPII, followed by addition of lysis buffer (no mixing on deck) ~15min.
- New tips are used for every addition of resin.
- User intervention required: seal plate using septa and shake on MixMate at 1100rpm for 5mins followed by centrifugation at 3000rpm for 2mins to bring down any drops.
- Septa is removed and DWP returned to MPII.
- This mixing is KEY to improving the DNA recovery.

Magnet changed

- PKI magnet had corners that required the user to "click in" the plate. If the plate is not straight, resin loss can occur.
- ABI magnet has no corners and the magnets are raised, so the plates sit better and no resin loss occurs.
- Some impact on yield. Resin loss = DNA loss.





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Platemap changed

- Electronic platemap for MPII has changed to reflect new pipetting volumes.
- The addition of the Nunc "Iysate" rack from off-deck lysis meant the addition of 1 new column of information in the platemap.
- Has been configured and used for validation plates.

Risks considered

- Static attraction of the tip causes some reagent types (e.g. lysis buffer) to adhere to the tip side.
- These are only reagents and the risk of sample contamination is very low.
- Pre-step, post-step and transport air gaps were optimised to make the pipetting cleaner.
- Droplets and bubbles have not been observed by operators with **V6.2**

Changes reviewed

PerkinElmer National Liquid Handling Specialist was asked to review the automated program and provide comments for improvement.



While observing the test, the problems were noted and then the modifications done and the DNA Analysis Team was advised to run the test again with the modifications. Actual extraction protocol liquids were used to completely mimic a "real" extraction run. modifications, the DNA IQ extraction protocol is a sound, neat protocol.

"

"

Result

25 Plates extracted

- Intermittent contamination observed
- Average once every 2 plates
- Multiple events on each plate
- No clear pattern observed
- A single source profile in blank observed
- Strong full profile
- Unknown

Verification II

Septa Mat

- Previously swabbed septa mat after mixing
- No quant values or profiles deemed fit for purpose

Sodium lodide test

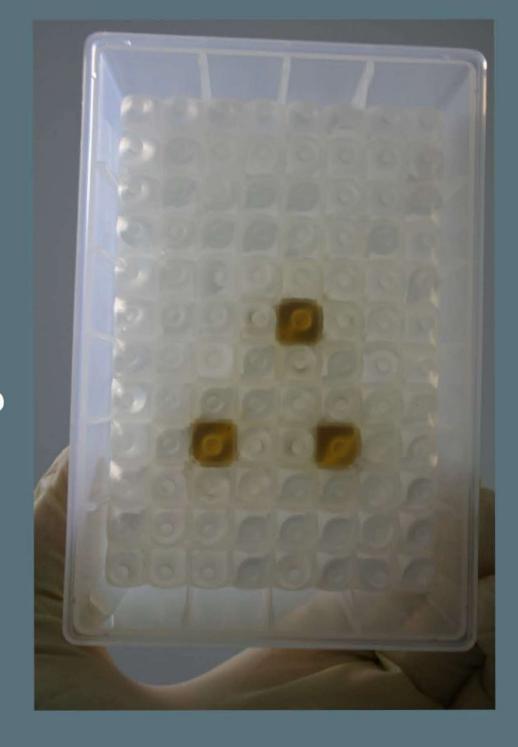
- Highly sensitive 1 in 10 000 dilution of 3M Nal will cause a reaction in contact with bleach and cardboard
- Cardboard turns a black purple colour
- No False positives

Nal - Before mixing



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Nal – after mixing



Nal – after centrifugation



Nal - Top of plate after Septa removal





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Alternate seals

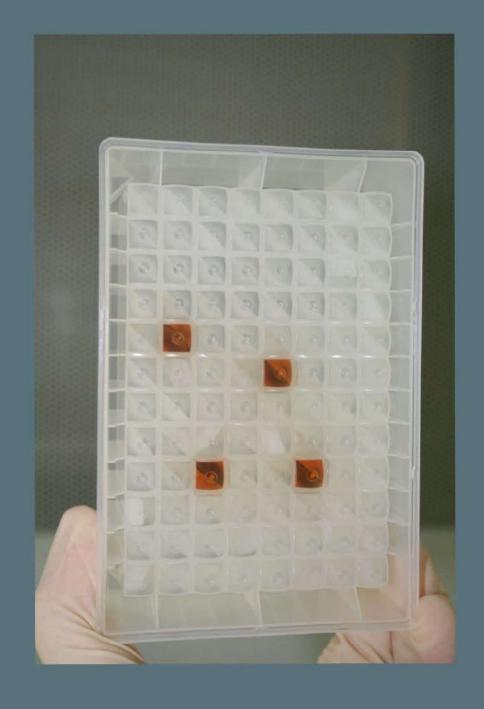
- Adhesive plastic seal
- Adhesive aluminium seal
- Heat seals being investigated

Aluminium seal

- No cross contamination after mixing and centrifuging
- Seal can be pierced
- Well to well transfer observed after plate was sealed and held upside down for 2 minutes



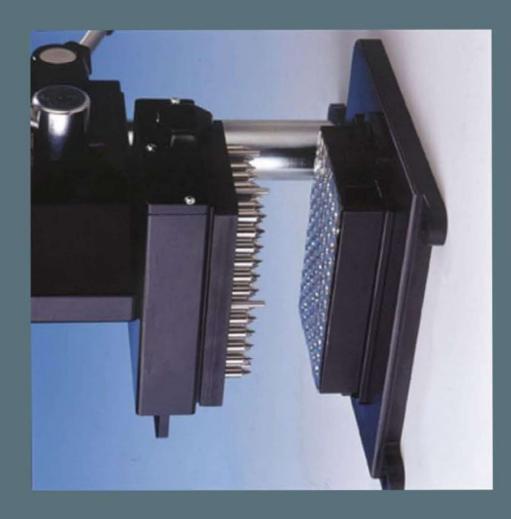
Nal - Automated steps



Sample access after sealing

- Verification plates with seal piercing
- Suspected issues with adhesive seal removal
- Verification plates pierced using PCR plate
- Disposable
- Does not contact sample
- Perfect size pierce
- Cost effective
- Nal tests showed no cross contamination using this method
- No evidence of sample on PCR plate

Manual piercer - Abgene



Proposed method

- Drill press
- Custom plate holders
- PCR plate
- Pierce plate??
- Custom made piercing plate??

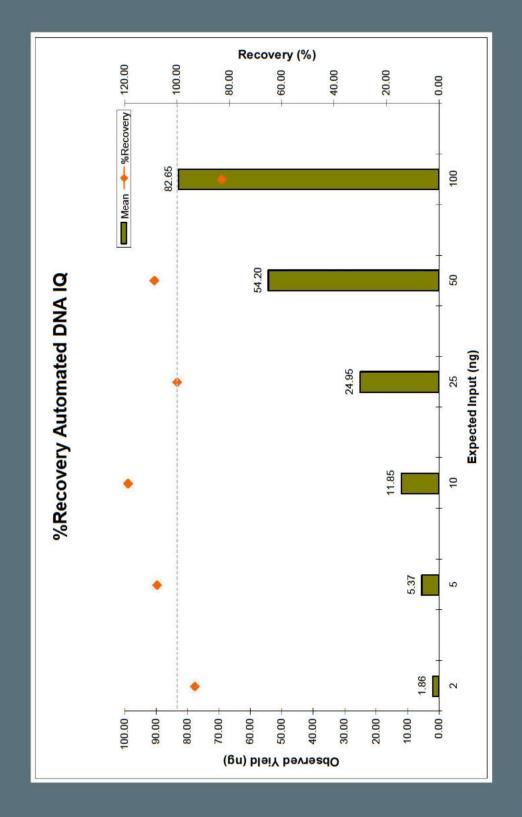
Pierced plate



Anti-contamination plates

- 1 efficiency plate
- 3 soccerball plates V6.2
- 3 checkerboard plates V6.2
- 3 zebra stripe plates V6.2
- Extracted on EP-B and EP-A
- All results obtained for EP-B
- EP-A results at quant and amp
- Male/Female samples at 1200ng
- Blanks analysed individually
- Any fluorescence above background was noted and repeated.

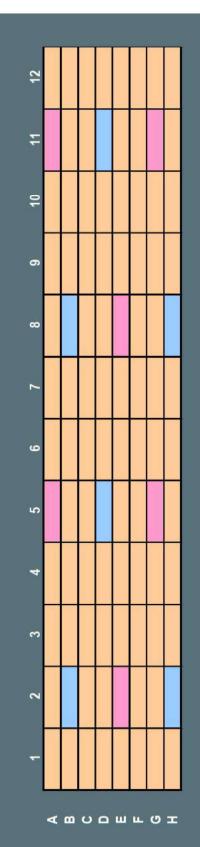
Efficiency



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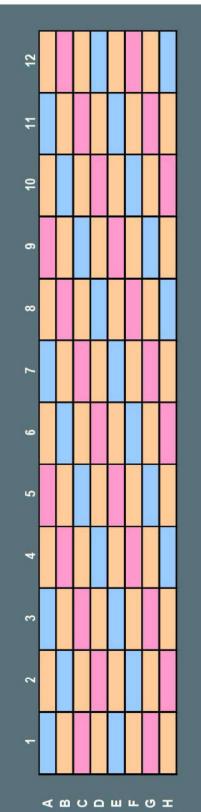
Soccerball

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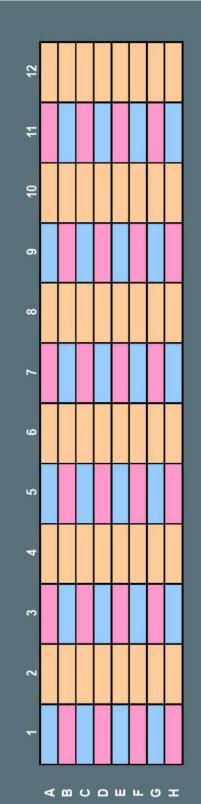
Blank Male Female

Checkerboard



Blank Male Female

Zebra Stripe

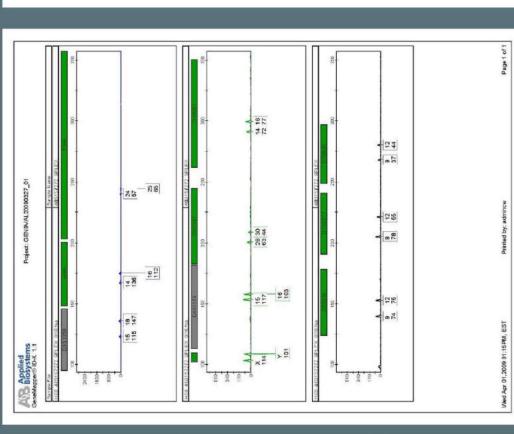


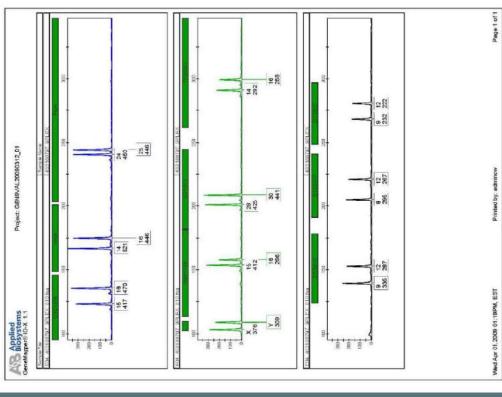
Blank Male Female

Profiles in blanks observed

- 3 Observations of spurious profiles
- » Single source
- » No match to staff
- » No matches to previously obtained profiles
- » Same profile occurred twice 3 weeks apart
- Further investigation to be conducted
- » Labware?

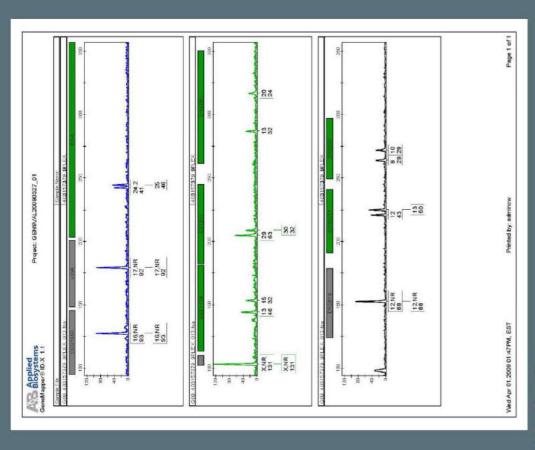
Profile in Blank





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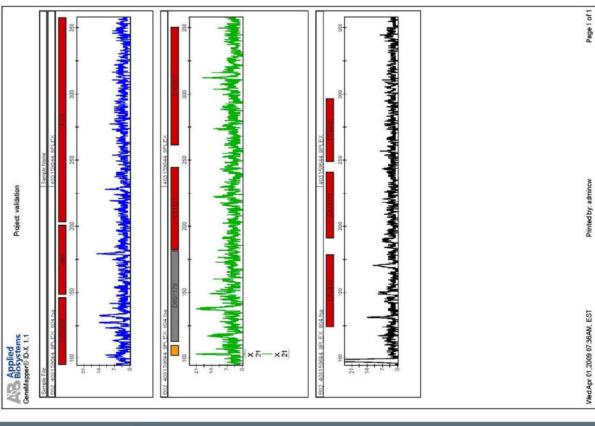
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Blanks

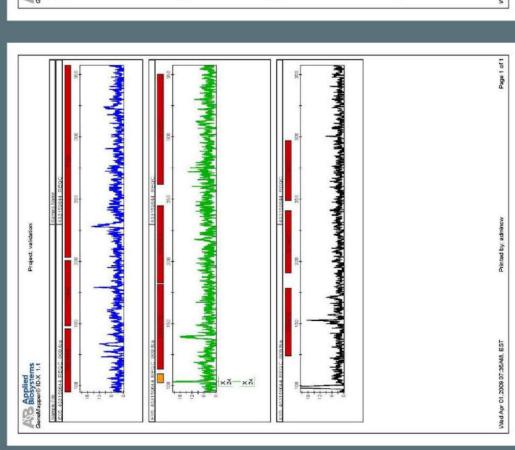
- 1 other blank with quant value
- 0.000692
- REQC 0.005240
- All other blanks Undet, profiles NSD

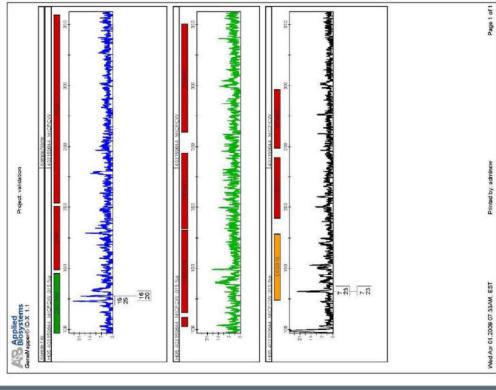


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009

Profile in Blank??





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External audit

- Assoc. Prof Theo Sloots and David Whiley
- They will write a summary report of their observations.
- The adhesive seal that we had used on the off-deck lysis plate was a problem. This process is no longer in the current version of DNA IQ.
- The MP II and current version of the automated DNA IQ is OK.
- Commended us on our environmental monitoring and monthly cleaning regimes.
- Commended us on our thorough approach to identifying and diagnosing the problems.
- We're "ahead of the pack" and learning lessons that nobody else has come across yet. I

Conclusion

- Enhancements have been made to the MP II. These changes have been tested and approved by a PerkinElmer LHS.
- The changes have caused an increase in efficiency and recovery of DNA.
- Changes have been made to the process surrounding the automated extraction.
- Off-deck mixing increases efficiency
- Seal piercing is required
- Anti-contamination checks have been run and results are as expected. **
- Feedback from external auditors were very positive.

Proposal

- Move to checkerboard extractions of reference samples
- Re-direct all RPT and LINK samples to EFTA
- Continue with off-deck mixing to maintain high efficiency
- Use the seal piercing method
- Samples read as per normal by readers
- Blanks and samples monitored by CCW
- Below threshold peaks in blanks

Below threshold mixtures in samples

 All profiles to be confirmed with previous alleles obtained – as per plate reading guidelines

Proposal

- Management has approved the proposal
- Considered low risk
- Every sample will be monitored by CCW
- If an adverse event is observed proposal will be reassessed
- No set time frame for reference sample extractions

Further information

- Further information will be emailed as fact sheets
- I will be in CR-611 from 11-12am tomorrow (02.04.09).
- Answer questions/concerns
- Run through results

JH-59

Order	Project	Details	Scientist	Due dates	Audience	SOPS, TMS?	SOPS, TMS? Project Discussed?	Absentees
-	Basic Probability	Laws Probability vs Odds Normal distribution, SD, HWE, LE LR vs Gentotype Freq vs Match Prob vs prob exclusion. Product Rule	MOH/CG	14-Jun	All Case Managers		JAH 20/04/12	AJR, JMW, SLB, AK, KDR, DRN, TJG, LAB, CJA
2	Data Sets	No of profiles Significant sizes Generation of frequencies Donors Sampling uncertainty HPD Truncation Stratification	SJWDRN	21-Jun	Reporting Scientists		JAH 20/04/12	AJR AK KDR TJG AAP MOH
8	Theta	How it will be reported	AJR	25-Jul	Reporting Scientists		JAH 23/04/12	AK KDR CJA AAP RJP SJM AAQ SMJ
4	Bayes Theorem	Frequentist vs non-frequentist What it is and how it relates to Forensics, and to DNA profiling	PAF	28-Jun	All Case Managers		JAH 20/04/12	CJA, AJR, SLB, HMW, KDR, AK, TJG, LAB, KHR, MB, VLC, ARM, MOH
5	Thresholds	History in QLD Derivation/Validation LOD vs LOR	JMW/PJT	5-Jul	Reporting Scientists		JAH 20/04/12	ARM,CJA,AK,ADA,SJM,MOH,AJR
9	Dropout	Background Zp model Probability of dropout Binary vs Continuous Drop models	M/JEE	12-Jul	All Case Managers		JAH 20/04/12	AJR, EJC, ADA, TJG, RJP.MOH,LAB,KHR.TVN.AK
7	Maths	Prior/Posterior Probs MCMC, Metropolis Hastings Algorithm Stutter Biological modelling	RJP	19-Jul	Reporting Scientists		JAH 20/04/12	JMW, AK, DRN, KDR
∞	Interpretation methods	Composite Consensus RAMNE RAMNE DNASenCE LCN and Omagh Bombing case Low template DNA Mixtures- 3 appearing as 2 etc	RGM/OHC	26-Jul	All Case Managers		JAH 20/04/12	CJA,KDR,AK,AAQ,SJM,SLB,VLO,EJC
6	STRmix program	Program itself Background - kit sensitivity, sampling Relatives	EJC	ТВС	All Case Managers	SOP/TM	yes	
10	Court	Boundaries	EJC/JAH	твс	Reporting Scientists		yes	
11	Linkage	background DNA in general Kits Affect on stats Incest	ADA	21-Jun	Reporting Scientists		EJC April	AJR, AK, KDR, TJG, AAP, MOH
خ	Case Management	New Workflow	AAQ/TG/JAH/EJC	٠.	All Case Managers	SOP/TM	JAH 20/04/12	

Minimum requirements Presen Rep Refere

Exercises, demonstrations

Presentation Report References Comments to SOPs?

JH-60

Erin Shearer

From: Justin Howes

Sent: Thursday, 30 November 2017 12:50 PM

To: Allan McNevin; Amanda Reeves; Cathie Allen; Kirsten Scott; Kylie Rika; Luke Ryan; Paula Brisotto;

Sharon Johnstone; Wendy Harmer

Subject: Project #184 for review

Attachments: Report_Evaluation of the efficacy of Microcons_v1.doc

Hi all

Please find attached a report for Project #184 – Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon® Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

This has a due date of <u>Wednesday 20 December</u> for feedback. Please be mindful of this due-date and schedule time to review.

Thanks Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Forensic & Scientific Services, Health Support Queensland, **Department of Health**

HSQ's vision | Delivering the best health support services and solutions for a safer and healthier Queensland.

Queensland Health acknowledges the Traditional Owners of the land, and pays respect to Elders past, present and future.



Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon[®] Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

November 2017
Justin Howes and Cathie Allen



Great state. Great opportunity.

Project Proposal #184 Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon® Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

Published by the State of Queensland (Queensland Health), November 2017



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For more information contact: Forensic DNA Analysis, Forensic and Scientific Services, Department of Health, GPO Box 48, Brisbane QLD 4001.

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Project Proposal #184 – Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon® Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

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Document Details

Contact for enquiries and proposed changes

If you have any questions regarding this document or if you have a suggestion for improvements, please contact:

Contact officer: Justin Howes

Title: <u>Team Leader – Forensic Reporting</u> and Intelligence Team

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Version history

Version	Date	Changed by	Description
1.0	30/11/2017	Justin Howes	Document Created.

Document sign off

This document has been approved by:

Name	Position	Signature	Date
Cathie Allen	Managing Scientist		

The following officers have endorsed this document

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Justin Howes	Team Leader FRIT		

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Project Proposal #184 – Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon® Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

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Name	Position	Signature	Date
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1. Abstract

All samples that underwent a Microcon® process were evaluated and categorised into whether there was meaningful information obtained or not. This evaluation focussed primarily on samples processed in 2016 that underwent an 'auto-microcon' process. Arguably minimal value in proceeding with this automatic processing step was found. Given this, further workflow streamlining processes could be implemented that would provide significant processing efficiencies, and cost and time savings such that these efforts could be better placed in processing higher DNA-yielding samples.

2. Introduction

Microcon® Centrifugal Filter Devices desalt and concentrate macromolecular solutions such as DNA-containing solutions. They employ Amicon's low binding, anisotropic, hydrophilic regenerated cellulose membrane [1].

The use of Microcon® filters to concentrate extract has been a standard post-extraction process within Forensic DNA Analysis to reduce the volume of extract from approximately 100uL to $\leq 20\mu L$ for amplification with AmpF ℓ STR® Profiler Plus®, and to $\leq 35\mu L$ for amplification with PowerPlex® 21 system (PP21).

Since the implementation of PP21 amplification kit within Forensic DNA Analysis for casework samples in December 2012, extracts with low Quantification values were recommended to be concentrated. Templates of <0.132ng were found to exhibit marked stochastic effects after amplification ^[2]. Consequently, a workflow that directed extracts automatically to a concentration step based on Quantification value was implemented ('auto-microcon' process).

Anecdotally, the suitability to provide the Queensland Police Service (QPS) with DNA profile Intelligence from extracts that have been concentrated has been noted to be limited. Furthermore, extracts that are of low quant value that have been automatically concentrated have been observed to rarely yield DNA information for QPS.

NB. Project #163 – Assessment of results obtained from 'automatic-microcon' samples [3] was conducted to evaluate the results of samples that were processed with the 'auto-microcon' process. A recommendation of this project was to re-evaluate after the introduction of the Forensic Register in conjunction with the use of Quantifiler® Trio DNA Quantification Kit.

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This recommendation was based on the perceived ease of retrieving data from the FR as opposed to AUSLAB, and with the thought that the FR would soon be implemented. For the purposes of this project, it is not considered essential to have the FR implemented if the data can be retrieved from AUSLAB. However, it is considered important that the data be spanning a sufficient period of processing, and be based on the same Quantification system namely the Quantifiler® Trio DNA Quantification Kit.

The purpose of this project is to evaluate the suitability for interpretation of DNA profiles that may be obtained after the post-extraction concentration step using the Microcon® centrifugal filter devices. This evaluation includes an assessment of those samples that underwent the 'auto-microcon' process. This evaluation is based on a data mine of extracts in the year 2016 that were concentrated with Microcon® centrifugal filter devices, and assesses the 'suitability' of PP21 profile outcomes as a function of quant values obtained from using the Quantifiler® Trio DNA Quantification Kit.

This evaluation looks at two data sets as a function of the Quantification value:

- PP21 DNA profile outcomes from extracts that were processed through the 'auto-microcon' process;
- 2. PP21 DNA profile outcomes from all extracts that were concentrated with the Microcon® filter devices.

3. Resources

The following resources were required for this validation/project:

Forensic DNA Analysis staff and computer time to retrieve data from AUSLAB and to use Microsoft Excel.

4. Methods

4.1. Data retrieval from AUSLAB (LIMS)

Data was retrieved from AUSLAB using Extended Enquiries. Data was searched for samples that had a testcode of 'XPLEX' and 'MCONC1' ordered in the year 2016 in Forensic DNA Analysis. Samples with the XPLEX testcode were High Priority (P2) samples.

The data was output with the corresponding Quantification value and the reported DNA profile interpretation (Exhibit Report Line in the Exhibit Report

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(EXH)) for that particular barcode. If the barcode was a sub-sample, the corresponding EXH line for the sub-sample was output.

For ease of data interrogation, the RAW data (I:\Change Management\Proposal#184 - Evaluation of the efficacy of Microcons\Data\RAW Data from AUSLAB) had a column added to describe whether the sample underwent the 'auto-microcon' process ('AUTO' = 0.001ng/ μ L<Quant <0.0088ng/ μ L) or not ('MANUAL' = Quant >0.0088ng/ μ L). Another column was added to describe whether there was a Quantification value returned in the data collation ('TRUE' = Quant value obtained), or not ('FALSE' = no Quant value obtained (ie. 0 ng/ μ L).

The data excluded samples that had not returned a DNA profile result, Quality samples (including environmental monitoring samples), have no quant value in the data export, or have quality issues noted.

4.2. Data interrogation

The data was interrogated by assessing the DNA profile outcome results reported as Exhibit Report lines as a function of the Quantification value.

The Exhibit lines were interrogated and grouped into two interpretation outcomes as follows:

- 1. 'Fail': DNA profile interpretation outcomes of 'Complex unsuitable for interpretation', 'No DNA profile', 'Partial unsuitable for interpretation', 'No DNA Detected';
- 'Success': All other DNA profile outcomes.

5. Experimental Design

5.1. Experiment 1: Assessment of 'auto-microcon' results

Intent

Evaluate the 'success' or 'fail' outcomes for PP21 samples that were processed in 2016 through the 'auto-microcon' workflow.

Data Analysis

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The samples applicable to this experiment had Quantification values in the range $0.001 ng/\mu L$ to $0.0088 ng/\mu L$, and a total number of samples that were processed this way was determined. This total number excluded environmental samples, samples without Quantification values, samples not requested for further work, samples where quality flags were raised, and samples that had not returned results at the time of data collection.

DNA profile interpretation outcomes were grouped into either 'success' or 'fail' as a function of the Quantification value. A percentage of samples that fell into these categories was determined.

The 'auto-microcon' data could be expressed as a function of Quantification value.

Of the DNA profile interpretation outcomes of 'success', the data was broken down further to determine the percentage of samples that were reworked prior to the DNA profile outcome of 'success'.

The percentage of samples that had an 'auto-microcon' process and led to an NCIDD upload was obtained. This data could be filtered further into the outcome from the NCIDD load, at the time of data collection.

5.2. Experiment 2: Assessment of all DNA profile results from extracts that have had a concentration step.

Intent

Evaluate the 'success' or 'fail' outcomes for PP21 samples that were processed in 2016 and underwent a post-extraction concentration step using Microcon® centrifugal filter devices.

Data Analysis

The samples that were applicable to this experiment had Quantification values above $0.001 ng/\mu L$, and underwent the Microcon® process. This included the 'auto-microcon' samples, and those that had a Microcon® rework performed (termed 'manual'). This combination of data was termed 'combined data'.

A total number of samples that were processed this way was determined. This total number excluded environmental samples, samples without Quantification values, samples not requested for further work, samples where quality flags were raised, and samples that had not returned results at the time of data collection.

DNA profile interpretation outcomes were grouped into either 'success' or 'fail' as a function of the Quantification value.

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The percentage of samples that fell into these categories ('manual' and 'combined') was determined. 'Manual' referred to the samples beyond the 'automicrocon' range that were reworked with the Microcon® process, and 'combined' referred to all samples ('auto-microcon' and 'manual').

There was a point where the number of 'success' samples was approximately the same as the number of 'fail' samples when the Microcon® process was performed. This appeared to be approximately Quant = 0.02ng/uL. Therefore, the data was interrogated further at a Quantification value lower than this mark to determine what percentage of samples in certain ranges led to DNA profile interpretation outcomes of 'success'.

From this data, a sub-section of samples was interrogated further to evaluate the effect on DNA Intelligence that was obtained. A range of samples with Quantification range up to 0.015ng/uL was chosen and a total number of samples was determined. This Quantification value was chosen as it was the approximate value where all samples below this value that underwent a Microcon® process, led to an approximate, round figure of 85% 'fallure'.

With this Quantification value chosen, the data was interrogated further. The percentage of samples in this range that were determined to be a 'success' and were reworked further was determined.

The percentage of samples that were in this Quantification range and led to an NCIDD upload was determined. This data could be filtered further into the outcome from the NCIDD load. This data could then be used to evaluate the potential for samples to not provide meaningful DNA Intelligence to QPS if the Microcon® process was re-defined in some way.

5.3. Experiment 3: Datamine of the difference in pre- and post-Microcon[®] Quantification values

Intent

Evaluate the difference between the values obtained from the Quantification process in samples that have had a Microcon® concentration step applied.

As this is purely a datamining experiment, only the samples that have yielded a result of 'success' was examined.

Data Analysis

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The samples applicable to this experiment had Quantification values above $0.001 ng/\mu L$ where the final result was 'success'.

The range was further refined as per Section 5.2, such that samples that had Quantification values between $0.001 ng/\mu L$ and $0.015 ng/\mu L$ were examined.

This range was considered by the author to be able to provide a sufficient demonstration of the trend of the data.

6. Results and Discussion

6.1 Assessment of 'auto-microcon' results

For samples in the 'auto-microcon' Quantification range, the total number of samples that were processed this way (excluding certain samples as per Section 5.1) was N= 1449 samples.

The percentage of samples that resulted in a determination of 'fail' was 89.4% (Fig 1). As expected, the number of 'fails' increased when the Quantification decreased and approached the Limit of Detection of Quantification ie. 0.001ng/uL (Fig 2). This was considered to be due to there being less DNA detected in the extract, and therefore less DNA to concentrate.

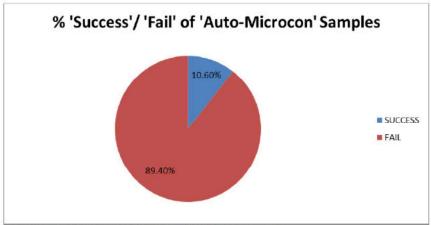


Figure 1: Percentage 'Success'/ 'Fail' of 'Auto-Microcon' samples.

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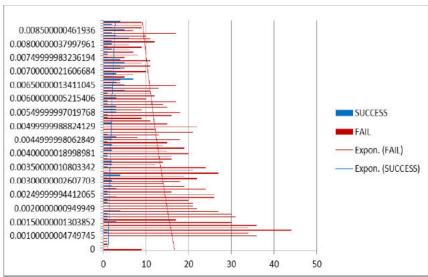


Figure 2: Spread of data and categorised as 'Success'/ 'Fail' for 'Auto-Microcon' samples.

In order to reach a DNA profile interpretation outcome of 'success', it was found that 74.7% of samples had an additional rework to the Microcon® process (Fig 3).

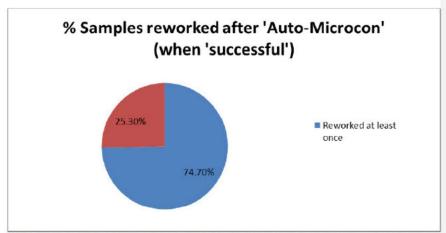


Figure 3: Percentage of 'Auto-Microcon' Samples that were reworked at least once and led to a 'successful' DNA profile outcome.

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In putting the data behind Figures 2 and 3 together, if an 'auto-microcon' process was not conducted and was subsequently requested by the client for samples in this Quantification range, there would be approximately a 10% chance of obtaining a 'successful' DNA profile interpretation. Furthermore, in order to achieve that outcome, approximately 75% of these 'successful' samples would have needed a further rework. This means, for these samples, there would be a turnaround time factor for the client to consider, and in a potential fee-for-service model with requesting clients, being prepared to have increased processing costs associated with these low-quant samples would be a client consideration.

If samples were not processed through the 'auto-microcon' process, what DNA Intelligence would the client miss out on? To evaluate this, the 'success' data was drilled down to the samples that had some NCIDD interaction and in particular, where they were the only samples in the case that were NCIDD-suitable for that particular profile (Fig 4). This represented 1.86% of all 'auto-microcon' samples. In looking at samples that provide *new* Intelligence, that is DNA information available for future linking, or has provided a cold-link, this equated to 1.45% of all 'auto-microcon' samples.

This 1.45% of samples would be the pertinent value for the client to consider if the 'auto-microcon' process was not performed. In considering this, it would be important to evaluate the time and cost for processing, and the opportunity to concentrate efforts on other higher yielding samples. In saying this, with the ease of communication through the Forensic Register, these samples could process if the client has no other forensic Intelligence assisting the matter, or if the item is considered to be of critical priority.

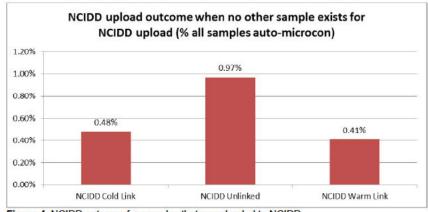


Figure 4: NCIDD outcome for samples that were loaded to NCIDD

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Ultimately, this data means that for approximately 90% of samples that underwent an 'auto-microcon' process, there is arguably negligible DNA profile Intelligence for the client. If the 'auto-microcon' was not applied, there would be the following advantages, including but not limited to:

- -the potential to make available at least 1449 processing positions for other samples including further available positions that would have been used for reworks,
- -the lack of a need for the considerable efforts required to prepare and process Microcon® (and further rework) batches for this number of samples,
- -consumable and labour savings in the end-to-end processing of these samples, and
- -time and effort could be redirected in the laboratory workflow to other activities including service extensions like Y-STR profiling.

6.2 Assessment of all DNA profile results from extracts that have had a concentration step.

All samples from 2016 that had a Microcon® process were determined. The total number of samples was N= 2201 samples, excluding certain samples as per Section 5.1.

The percentage of samples that resulted in a determination of 'fail' was 78.5% (see Fig 5). As expected, in looking at the spread of the 'combined' data, the number of 'successes' increased when the Quantification increased (Fig 6).

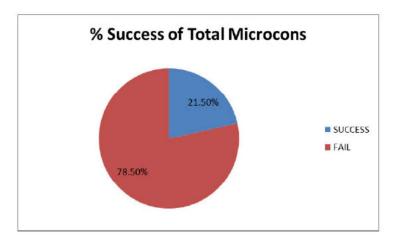


Figure 5: Percentage 'Success'/ 'Fail' of all Microcon® samples ('combined' data).

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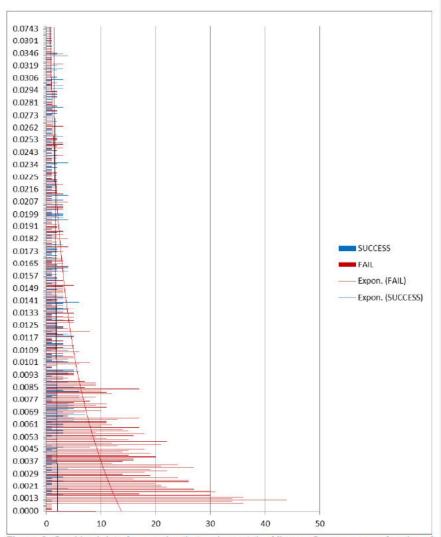


Figure 6: Combined data for samples that underwent the Microcon® process as a function of Quantification value.

As mentioned in Section 5.2, the Quantification value where there was roughly the same number of 'success' and 'fail' samples was approximately 0.02ng/uL. It must be noted that this is a rough estimate at this particular Quantification value, and it is based on limited samples that returned that Quantification value. It can be argued that taking a range of Quantification values to look at the overall success/fail percentages could provide the client with approximate likelihoods of obtaining meaningful DNA Intelligence.

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A number of ranges were looked at to determine the percentage 'success' of samples with Quantification values in various ranges (Fig 7). The ranges were established up to the highest Quantification value of 0.02ng/uL. As expected, the percentage 'success' increased as the Quantification increased due to the higher amount of DNA in the extract available to be concentrated.

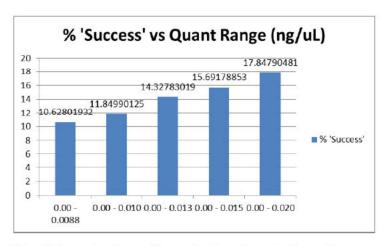


Figure 7: Percentage 'success' for samples that underwent a Microcon® process

In viewing the data in Fig 7, a limitation is that all samples that fell in the 'automicrocon' range, had a Microcon® process performed, whereas there are samples that are in higher Quantification ranges that might not have required a Microcon® concentration rework step to yield useful DNA profiles. These samples were not evaluated.

A lower Quantification value to where the number of 'successes' roughly equalled the 'failures' was chosen to be the upper end of data ranges that were evaluated further. The value chosen was 0.015ng/uL. Table 1 and Figure 8 describe the risk to NCIDD upload for samples in these ranges if Microcon® concentration steps were not performed.

Table 1: NCIDD outcome for samples that were loaded to NCIDD in various Quant ranges

	% No other samples to Upload in Quantification ranges (Q)				
	Q 0.00ng/uL to 0.01ng/uL (total samples in range 1519)	Q 0.00ng/uL to 0.0133ng/uL (total samples in range 1696)	Q 0.00ng/uL to 0.015ng/uL (total samples in range 1778)		
NCIDD Cold link	0.92	0.88	1.01		
NCIDD Unlinked	0.53	0.77	1.24		
NCIDD Warm Link	0.46	0.83	0.90		

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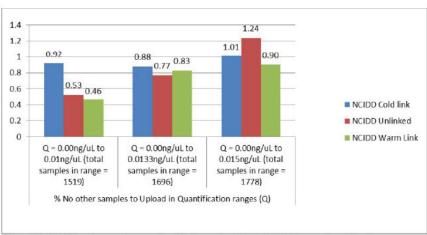


Figure 8: NCIDD outcome for samples that were loaded to NCIDD in various Quant ranges

Approximately 1.45% of samples in the Quantification range up to 0.01 ng/uL resulted in 'new' DNA Intelligence. This percentage is the same as that found in the 'auto-microcon' range. This percentage increased to 1.65% and 2.25% for the Quantification ranges up to 0.0133 ng/uL and 0.015 ng/uL respectively.

The number of further reworks required to obtain 'success' outcomes decreased as the Quantification increased. This is not unexpected given higher DNA yields detected would not necessarily require as many reworks in order to yield DNA profiles.

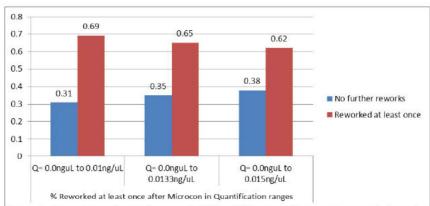


Figure 9: Percentage of samples reworked (in addition to a Microcon® process) in various Quantification ranges.

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6.3 Datamine of the difference in pre- and post- Microcon® Quantification values

The samples applicable to this experiment had Quantification values above $0.001 ng/\mu L$ where the final result was 'success'. The range was further refined as per Section 5.2, such that samples that had Quantification values between $0.001 ng/\mu L$ and $0.015 ng/\mu L$ were examined.

As the Microcon® process concentrates the DNA extract from approximately 100uL to approximately 35uL, in theory it would be a reasonable expectation to obtain approximately two to three-fold increases in DNA Quantification after concentration. Figure 10 shows the plot of the differences found for samples that resulted in 'success'.

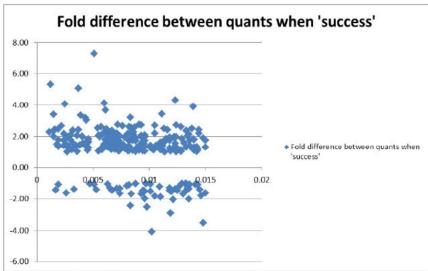


Figure 10: Quantification differences pre and post concentration

The findings are not unexpected as the scatter focusses mostly around two-fold increases in Quantification. It was also not unexpected to observe the variable results. Anecdotally, variability in success rates is found at profile management stage when assessing results of samples that have had this concentration step.

DNA can be lost in the process as seen in Fig 10 where the Quantification values decreased after concentration. Variability in results could be attributed to a number of things, including but not limited to the slight differences between operators and instrumentation, the differences in substrate type and level of degradation, and the variability in Quantification result.

Project Proposal #184 – Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon® Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

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7. Conclusion and Recommendations

The data analysis demonstrated that there was arguably minimal value in performing the 'auto-microcon' concentration step. This opinion was formed by analysing the data from 2016 where it was found that for all samples that underwent the 'auto-microcon' step, 89% did not yield meaningful results.

It was found that in considering all samples that underwent a Microcon® step at some stage in 2016, 78.5% did not yield meaningful results. As expected, when the Quantification value increased, the percentage of meaningful results increased. However, it was also demonstrated in the data analysis that the Quantification values did not always improve after Microcon®, but where they did, the magnitude of change was roughly equivalent to the change in volume (from neat to concentrated sample).

Based on the data analysis, the following recommendations are offered:

- 1. Cease 'auto-microcon' processing with the following exceptions:
 - a. Priority 1 samples (Critical Priority); and
 - b. Coronial/DVI samples where profiles are mostly single-source and quite often incomplete profiles may be enough to provide Intelligence on possible identity.
- Cease processing all Priority 3 samples up to the Quantification value of 0.0133ng/uL (template of 200ng).
- 3. For samples in the range described in Recommendation 2, automatically send result information via the Forensic Register to QPS at Quantification stage. This result information is recommended to be the exhibit result line of 'DNA Insufficient for Further Processing'. This recommendation is an extension to the current 'No DNA Detected' process, which looks at Priority 2 samples yielding Quantification results of less than the Limit of Detection.
- Re-analyse Priority 2 samples in the range 0.0088ng/uL to 0.0133ng/uL after a six month period of processing to evaluate whether Recommendation 2 can be extended to Priority 2 samples.
- 5. Communicate the change in process to QPS and ensure that QPS are aware that for samples in the ranges mentioned in Recommendations 1 and 2, that they could be requested for Microcon® concentration steps at any point in time. This request can be made via the Forensic Register after they have received the 'DNA insufficient...' result line.

Project Proposal #184 – Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon® Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

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7.8. References

Formatted: Bullets and Numbering

- [1] QIS 19544v11 Concentration of DNA Extracts Using Microcon Centrifugal Filter Devices
- [2] PowerPlex® 21– Amplification of Extracted DNA Validation. Megan Mathieson, Thomas Nurthen, Cathie Allen. December 2012. Forensic DNA Analysis.
- [3] Project #163 Assessment of results obtained from 'automatic-microcon' samples. Josie Entwistle, Allison Lloyd, Kylie Rika, Thomas Nurthen, Cathie Allen. August 2015. Forensic DNA Analysis.

Project Proposal #184 – Evaluation of the Efficacy of a Post-Extraction Concentration Step Using the Microcon® Centrifugal Filter Devices in Yielding DNA Profile Intelligence.

Procedure for Resolving DNA Profile Interpretation Differences of Opinion

1 Purpose

The purpose of this procedure is to describe the approach to resolving differences in scientific opinions associated to DNA profile interpretations.

2 Scope

This procedure shall apply to situations where there is a difference in scientific opinion related to DNA profile interpretations. This could be at item-reporting stage (initial DNA profile interpretation) or at statement stage (collating results for a case into a court statement).

3 Definitions

Nil

4 Actions

The workflows in the appendices are for situations where the difference in scientific opinion is at the Profile Data Analysis (PDA) stage (Appendix 2) and at statement stage (Appendix 3). The guidelines (Appendix 1) are to be read in conjunction with Appendices 2 and 3.

The Line Manager Checklist (Appendix 4) is to be used by the Line Manager to guide them in providing assistance to the resolution of the particular difference of opinion.

The templates in the appendices are to record the Interpretation summary by the independently selected scientists (Appendix 5), and the outcome summary by the Team Leader (Appendix 6).

5 Records

Appendices 5 and 6 are templates for recording independent interpretations and Team Leader summaries. These should be added to the Forensic Register (FR) as sample notations against the barcode for the DNA profile under discussion.



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6 Associated Documentation

QIS: <u>17117</u> – Procedure for Case Management

QIS: 33773 – Procedure for Profile Data Analysis using the Forensic Register

QIS: <u>34006</u> – Procedure for the Release of Results Using the Forensic Register

QIS: 34322 - Technical and Administrative Review of Records Created in the Forensic Register

7 References

ISO/IEC 17025 Application Document: Legal (including Forensic Science) – Appendix. Effective February 2020. Section 7.7

Forensic Science Regular Guidance, Cognitive Bias Effects Relevant to Forensic Science Examinations FSR-G-217



8 Amendment History

Version	Date	Updated By	Amendments
1	Aug 2021	J Howes	First version

9 Appendices

- 1 Guideline to approaching differences of scientific opinion in DNA Profile Interpretation
- Workflow for Dealing with Differences of Scientific Opinion in DNA Profile Interpretation: PDA Stage
- Workflow for Dealing with Differences of Scientific Opinion in DNA Profile Interpretation: Statement Stage
- 4 Line Manager Checklist
- 5 Independent Profile Interpretation Instructions and Findings
- 6 Team Leader Summary



9.1 Guideline to approaching differences of scientific opinion in DNA Profile Interpretation

Date	19 August 2021
References	This information is to be read in conjunction with the QHFSS Forensic DNA Analysis Approach to Dealing with Differences of Profile Interpretation Workflow diagram
	 QIS <u>34322</u>: Technical and Administrative Review of Records Created in the Forensic Register, Forensic and Scientific Services ISO/IEC 17025 Application Document: Legal (including Forensic Science) – Appendix. Effective February 2020. Section 7.7 Forensic Science Regular Guidance, Cognitive Bias Effects Relevant to Forensic Science Examinations FSR-G-217
Background	In the end-to-end Forensic DNA Interpretation and reporting process, there are a number of points where scientists may have differing interpretations of the DNA Profiles obtained. These points create junctures in the process. How these differences are handled are not specifically covered in the current Standard Operating Procedure. (34322V2). To date, these junctures had been handled inconsistently.
Purpose	To provide a consistent approach to working through the differences of scientific opinion for the interpretation and review of DNA Profiles
Applicable to	All Forensic DNA Analysis staff who are required to perform interpretation and review of DNA Profiles.
Requirements	A scientist performing the:
	 interpretation of the DNA profile must be competent in this task, including the use of relevant statistical software technical review must be competent to perform the process that they are reviewing and to interpret mixtures using the relevant statistical software.
Time considerations	The end-to-end process of obtaining and reporting a DNA profile should be completed within 10 working days.
	All staff are required to perform their sections of the process within the recommended time frames to ensure a 10-day turnaround.

Role title	Function
Profile Data Analysis (PDA) Entry	The individual who performs the interpretation of the DNA profile and statistical analysis for result entry to the FR.
PDA Review	The individual who independently interprets the results from PDA Entry. Performs an administration and technical review of the results and validates the result for release to the client. This is a quality step undertaken prior to release of the results to the client.
Line Manager	Assist in the resolution of differences of opinion, utilising the check list to ensure a standardised approach is taken. Note: This role is a management role, to guide the process forward. It is not an interpretation role.
Team Leaders	Enters the process if the PDA Entry and Reviewer's interpretations do not align and the Line Managers are not able to resolve it at level. The Team Leaders role is: • For differences of interpretation: to authorise the 3 independent interpretations



- For Unusual Profiles: to set up a Round Table discussion for unusual profiles to decide on a process for interpretation,
- For Finalised Unusual Profiles: to decide how to share knowledge and if any SOPs, guidelines or workflows need updating.

Process overview	Stages	Comments
	DNA Recovery	The Evidence Recovery Team conducts the DNA Recovery Process
	DNA Profile Creation	The Analytical Team creates the DNA Profile
	DNA Interpretation	Competent staff within the Forensic DNA Analysis Team undertake DNA profile interpretation and review. Conducted in a two-stage verification process:
		Initial Assessment, called PDA Entry
	·	 Validation step, called PDA Review
	FR Profile Finalisation	The profile is finalised in FR and distributed according to standard distribution processes
	DNA Statement Writing	The Reporting Team conducts a two-stage verification process:
		Statement writing
	_	Statement review
	Court Reporting	The Reporting Scientist may be called to provide expert testimony.

Process by Stages and Steps

Process	Steps	Comments
DNA Recovery		
DNA Passyon/	Receive evidence	
DNA Recovery	2. Processes evidence	
DNA Profile Generation		
	3. Generation of a DNA Profile/s	
DNA Profile Creation	 DNA profile is made available to Case Managers 	
DNA Interpretation		
PDA Entry	The initial scientist conducts the PDA Entry (Initial assessment)	If the profile is <i>new</i> , something not previously seen, raise it with your Line Manager to be considered as approach as new and emerging practice.
	6. The reviewing scientist conducts the PDA Review (Validation Step)	
PDA Review	IF the interpretations:	The PDA Review is required to communicate face to face that they have a different
	6.1 align – profile is validated, and it may go to Statement Writing process	interpretation within 24 hours of conducting their PDA Review.
	6.2 don't align - the PDA Reviewer informs the PDA Entry of their view as a first/initial step.	

Differences of Opinior	Steps Steps	Comments
Communicate the difference of opinion	7. PDA Entry and Reviewer meet to discuss the different interpretations	If the differences are:
		straight forward: initial feedback can be done via email; or,
		unusual: this communication should progress to in person/phone /MS Teams.
Capture the different scientific interpretations	8. PDA Entry and Reviewer use the FR Free Text field to <i>capture</i> their comments on their different interpretations.	



Seek guidance from the Line Manager(s) 9. PDA Entry and Reviewer discuss their interpretations with the PDA Entry's Line Manager. They should reference their captured differences of opinion.

The LM role is to Guide the Scientists through the process (not to do an interpretation) to ensure that each aspect has been captured.

A check list is developed to create consistency at this stage.

Request an Independent Review Process

10. IF a resolution path is not visible, THEN the Independent Review Process is to be requested of the other Team Leader (Not the Team Leader of the PDA Entry person)

Authorise an Independent Review Process

11. Team Leader organises the Independent Review Process during from a roaster system including deidentifying the Profile

Conduct an Independent Review Process

12. Three scientists conduct their own interpretations and provide them to the Team Leader.

OUTCOME: Three independent interpretations provided to the nominated Team Leader

Summaries Independent Review

- 13. Team Leader writes a summary report and distributes it to the:
- PDA Entry
- PDA Reviewer
- PDA Entry's Line Manager

Note: The Summary Report must go to those listed at the same time.

Distribute the findings

14. Team Leader, Line Manager and PDA Entry and Reviewer read the summary report.

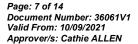
Acting on the findings

15. Based on the report findings;

IF the independent interpretations:

15.1 align - THEN validate the result and it may go to Statement Writing

15.2 align however differ to either the PDA Entry or PDA Reviewer either person can request their role is reassigned.





15.3 don't align THEN how to move forward is reviewed on a case-by-case bases. Most likely establishing a round table to decide how to move forward.

Considerations

Next actions will be based on a case-by-case basis.

Statement Writing Process

THE END

Additional considerations and guidelines

Differences of opinions

Differences of opinion are to be expected. It is also expected that the different views are identified and recorded throughout the review process.

This is important because it:

- is a requirement through NATA
- shows consideration of various possibilities
- shows process transparency

When can a PDA Entry or PDA Review Scientist withdraw from the process?

Each individual should be able to withdraw from the process if they disagree with the interpretation or approach undertaken. There are two likely stages that an individual may want to withdraw from the process:

- Discussion with the Line Managers when exploring how to move forward.
- After the Team Leader Summary Report has been shared back to the PDA Entry and Reviewing scientists.

It is highly recommended that the latter is the most appropriate point to withdraw as the independent review process will indicate the individual/s who will be able to be assigned to the DNA profile interpretation.

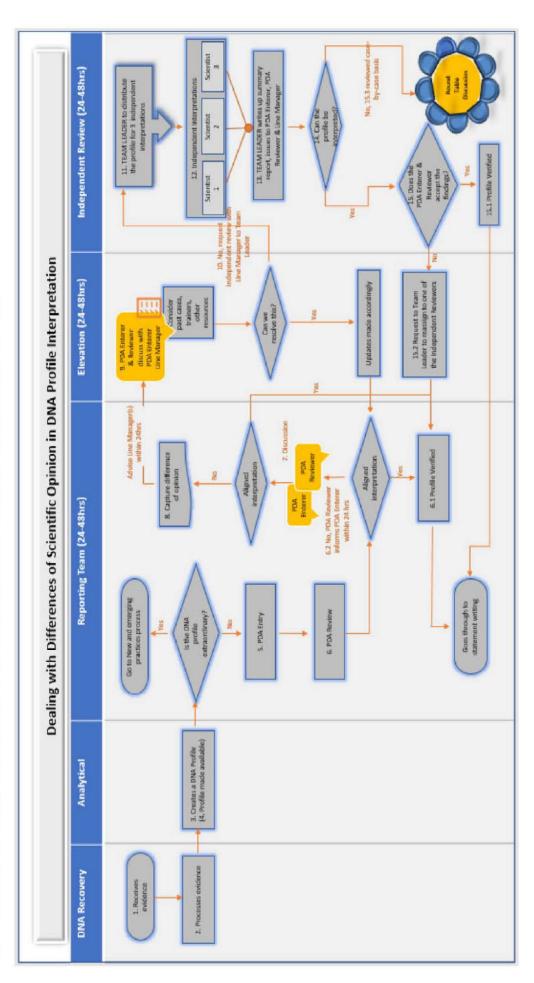
Key Elements

The process is intentionally designed to:

- Provide consistency in how we approach our differences
- Create space at different points in the process for scientific discussion to exist
- Create efficiencies in the process
- Promote discussion between PDA Entry and Reviewing scientists prior to engaging Line Managers, or other internal and external individuals.
- Provide time guidelines to ensure the end-to-end processes are completed within the recommended turnaround times.
- Reduce the number of people who are engaged at the PDA Entry and Review stage if there is a
 difference of interpretation. This allows for the availability of three independent reviewers later in
 the processes if required
 - NB. A technical review must not be performed such that it 'shifts the perceived responsibility of the scientific findings from the examiner to the reviewer'.

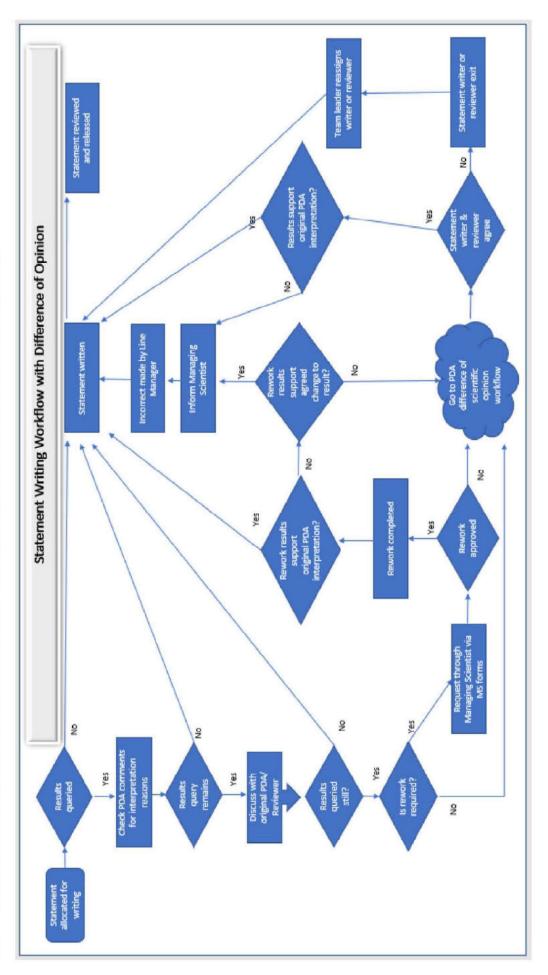


Workflow for Dealing with Differences of Scientific Opinion in DNA Profile Interpretation: PDA Stage 9.2





Workflow for Dealing with Differences of Scientific Opinion in DNA Profile Interpretation: Statement Stage 9.3





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9.4: Line Manager Checklist

Role

The role of the line manager is not to look at the profile to give their opinion. The role of the manager is to facilitate the discussion between the two parties to ensure that each one understands each other's reasoning and considers the alternate opinion.

The aim is to talk both parties through the opinions of each other to help them assess the most likely explanation of the evidence and to come to an agreement. Such a discussion may require the line manager to ask each to reconsider new information and to reconvene. Alternatively, the line manager may also try to facilitate a view to compromise given the noted risks.

Checklist to assist discussion

- Have you discussed the result and the reason behind your chosen interpretation?
- Do you understand the reasoning behind each other's point of view?
- Have you documented the reasons behind your interpretation?
- What are the differences between the interpretations?
- What are the risks associated with reporting in each way?
- Is it covered in the SOP? If so, does it support one interpretation over the other?
- Is there any other supporting literature that should be read and considered in the interpretation?
- Which interpretation would be more consistent with the remainder of the case?
- Has this result been reported to the QPS? If so, does the result need to be corrected? Will
 the client need to be informed?

9.5: Independent Profile Interpretation – Instructions and Findings

Details

Independent Reviewer's Name: Ima Person A

Date Required: DD / MM / YYYY

Date completed: DD / MM / YYYY

Instructions

You have been provided with a Crime Scene profile for review. This includes copies of all amplifications and may include Reference Samples and STRmix outputs if appropriate. Inferences should not be made from the number of amplifications and presence or absence of a STRmix analysis.

You are requested to perform an independent interpretation of this DNA profile. You are instructed to do this without discussion with colleagues, and without viewing any information within the FR.

If you require any information that has not been provided, you are to e-mail the Team Leader (or delegate) who has requested you perform this review.

Return your findings to the requesting Team Leader (or delegate).

Profile details

FR number	QP number	Exhibit barcode
3	- E	

Category	Description and Located / Owner
Swab Blood	Of something from somewhere

Quant value (ng/uL)	Amp 1 volumes	Amp 2 volumes	Amp 3 volumes
0.XXXX	SV1 TV1	SV1 TV1	SV1 TV1
	SV2 TV2	SV2 TV2	SV2 TV2
	Y-quant:	Y-quant:	Y-quant:
	Deg Index:	Deg Index:	Deg Index:

Other relevant profile information (e.g. if diluted, microcon, SFRAC etc.):



Independent Findings

Please answer the following questions:

Is the profile interpretable? (Y/N)

Reason(s) for profile not interpretable (if applicable)

Number of Contributors assessment (1-4)

Is conditioning appropriate? (Y/N)

Reasons for not conditioning (if applicable)

For each reference barcode, please state whether it should be an assumed contributor (AC), requires comparison (LR) or can be intuitively excluded (Excl)

Are there any risks associated with any particular interpretations (e.g. false exclusion? Incorrect NCIDD upload), if so why? Can these be mitigated against? (explain)

Further comments:



9.6 Team Leader Summary

Details

Report prepared by: Person A - Team Leader, Team X

Date: DD / MM / YYYY

Profile details

FR number	QP number	Exhibit barcode
-		-

Independent Findings

Independent Reviewer 1

[Copy in Independent Reviewer's assessments and findings]

Independent Reviewer 2

[Copy in Independent Reviewer's assessments and findings]

Independent Reviewer 3

[Copy in Independent Reviewer's assessments and findings]

Summary

Do all three reviewers agree on an interpretation? (If so, what is that interpretation. If not, on what points do they agree or disagree?)

Have all risks been considered and appropriately mitigated against?



JH-62

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	Statement	N/A	10	0	5	8												16	15
Cases Finalised F	Reviewed	N/A	2	4	6	16												12	8
2	Evidentiary Certificates Issued	N/A	0	0	0	0				-	1	0						0	0
Interpretations	Entry	N/A	285	403	529	425	+	+	+	+	+	+	+	+	+	+	+	352	311
	чемем	N/A	345	313	335	324	+	1	1	+	1	+	+	+	+	4	+	2/1	407

FR Week Number		19 2/05/2022	20 9/05/2022	21 16/05/2022	22 23/05/2022	23 30/05/2022	24 6/06/2022 1	3/06/2022 2	20/06/2022 27	27 27 106/2022	28 4/07/2022 11	29 1/07/2022 18	30 25	31 25/07/2022 1/0	32 '08/2022 8/04	33 8/08/2022 15/	34	35 22/08/2022 3	36 29/08/2022	37 5/09/2022	38 12/09/2022
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	Evidentiary Certificates Issued	0	0	0	0	0	0	0	0	0	0	0	0		0	0	0				
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	Entry	146	178	281	467	382	418	999	398	438	455	382	639	+	+	319	554	0	0	0	0
Interpretations	Review	262	382	445	458	438	295	418	486	453	344	316	454	505		265	382	0	0	0	0
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- Inke	Poloscod	61	73	157	130	135	205	133	12.1	2000	118	126	154	ł	+	ł	137	0	0		0
	Scene	69	122	142	143	91	244	147	118	276	125	143	123	H	H	H	116	0	0	0	0
NOIDU Opioads	Person	120	28	347	184	578	374	227	230	219	261	173	547			302	339	0	0	0	0
	Statement	12	16	21	24	6	6	11	12	12	14	10	17				10	0	0	0	0
Cases Finalised	Reviewed	18	16	13	20	20	8	10	14	8	13	10	15				21	0	0	0	0
	Evidentiary Certificates Issued	0	0	0	0 5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Interpretations	Entry	762	8/1	300	460/	382	418	000	398	438	455	382	639	+	1	319	200		0		0
	A CARROLL	707	700	200	25	200	207	101		8	5	200	100	┨	$\frac{1}{1}$	202	700	,		,	,

FR Week Number		39	40	41	42		44	45	46	47		49	20	51	52
		19/09/2022	26/09/2022	3/10/2022	10/10/2022 1	17/10/2022	24/10/2022	31/10/2022	7/11/2022	14/11/2022	21/11/2022	28/11/2022	5/12/2022	12/12/2022	19/12/2022
REPORTING 1															
FTE Variance	Week														
	Statement														
Cases Finalised	Reviewed														
	Evidentiary Certificates Issued														
PDA	Interp Result Entry														
	Interp Result Review														
REPORTING 2															
FTE Variance	Week														
	Statement														
Cases Finalised	Reviewed														
	Evidentiary Certificates Issued														
PDA	Interp Result Entry														
	Interp Result Review														
TOTAL REPORTING				ľ	İ						ľ		ĺ		
				İ	t	İ	t	Ī	ĺ	I	Ī	ĺ	Ī	Ī	
				l	İ			ĺ							
FTE Variance	Week	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Statement	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Cases Finalised	Reviewed	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Evidentiary Certificates Issued	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Interpretations	Entry	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Review	0	0	0	0	0	0	0	0	0	0	0	0	0	0
INTELLIGENCE															
Links	Released														
	Source														
NCIDD Uploads	Person														
FRIT		39	40	41	42	43	44	45	46	47	48	49	20	51	52
		19/09/2022	26/09/2022	3/10/2022	10/10/2022 1	17/10/2022	24/10/2022	31/10/2022	7/11/2022	14/11/2022	21/11/2022	28/11/2022	5/12/2022	12/12/2022	19/12/2022
FTE Variance	Week	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Links	Released	0	0	0	0	0	0	0	0	0	0	0	0	0	0
NCIDD Holoads	Scene	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Person	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Statement	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Cases Finalised	Reviewed	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Evidentiary Certificates Issued	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Interpretations	Entry	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Review	0	0	0	0	0	0	0	0	0	0	0	0	0	0

				AVERAG
VEADI	Y DATA			ES per
ILAKL	IDAIA			week
		COUNT	ERAGES 2	2022
Links R	eleased	4257	119.88	137.00
CIDD Uploa	Scene	4711	129.53	178.64
DDD Opioa	Person	7754	232.37	174.36
	Statement	393	14.29	10.36
ses Finalis	Reviewed	411	14.78	11.36
	Evid Cert	0	0.06	0.00
terpretation	Entry	15395	408.9	537.82
terpretation	Review	12471	384.49	347.91

From 1 Nov 2021

RT1 (400.15hrs

Total

Name	Av Hrs/ week
Sharon	38
Adrian	38
Angela	27.75
Rhys	30.4
Alicia	38
Anne/Mega	22.8

		Interp	retations E	ntered	Int	erps reviev	ved			State	ments		
	From 03/01/202 2	Total		Average per hour	Total	Weekly Average	Average per hour	Total Written	Weekly Average Written	Average per hour	Total Reviewed	Weekly Average Reviewed	Average per hour
RT1	(400.15hrs	8704	264	0.659	7053	214	0.534	187	6	0.014	236	7	0.018
RT	2 (352hrs/	6696	203	0.576	5139	156	0.442	206	6	0.018	175	5	0.015
Int	el (38hrs/v	#REF!	#REF!	#REF!	#REF!	#REF!	#REF!	#REF!	#REF!	#REF!	#REF!	#REF!	#REF!
		·											

Average

per hour

Interps reviewed

Weekly

Average

#DIV/0! #DIV/0! 0 0.000

Total

Statements

Average

per hour

#DIV/0! #DIV/0! #DIV/0! #DIV/0!

Total

Weekly

Average Written

Total

Written

Weekly Average Reviewed

Average

per hour

0.020 0.015

Cassie	38
Emma	38
Jacqui	38
Kerry-Anne	38
Josie	38
Kylie	38
Matthew	38
Angelina	38
Claire	25.2
Ingrid	38
Thomas	38
Penny	38
Deb	22.8
Allan	38

Tegan 38

Justin 38

Susan

0

736.95 TOTAL (no Total Repo 736.95

R1 R2 Intel 384.95 352

 Percent Reporting

 R1
 52.23557

 R2
 47.76443

Percent FRIT R1 52.23557 R2 47.76443

Note; From 10/05/2021 Statement is recording the number of statements prepared. Prior it was the number of statements issued

Interpretations Entered

Weekly

Average

#DIV/0! #DIV/0! 0 0.000

Average

per hour

From 01/11/2021

Sharon Adrian Alicia 38 38 38 27.75 38 38 38 38 38 Angela Anne/Mega Cassie Emma Jacqui Josie Kerry-Anne 30.4

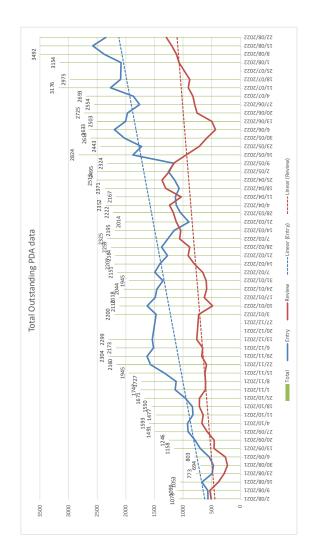
Kylie Allan 38 38 25 22.8 38 38 38 38 Angelina Claire Deb Ingrid Matthew Penny Tegan Thomas

> R1 R2 Total 400.15 351.8 751.95

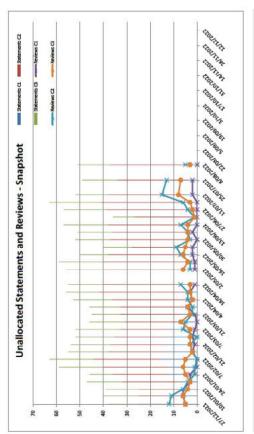
IT (not including JAH) R1 53.21497 R2 46.78503

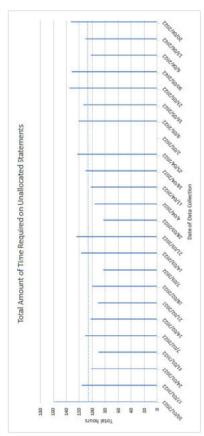
FRIT	FR Week No.	
FTE Variance	Week	
Links	Released	
	Scene	
NOIDE Opioads	Person	
	Statement	
Cases Finalised	Reviewed	
	Evidentiary Certificates Issued	
anoitatoranotal	Entry	
ווופואובושוחוא	Review	

NB. Only counting what is available (ie. not including 'grey-ed out samples') Worse than week prior	Better (or the same) than week prior																												JAH absent week starting 25/07/2022 - data line removed																						
Review		728	483	629	700	282	000	802	883	1060	1041	1417	1117	472E	1031	1303	1367	1241	1166	949	719	589	434	499	755	796	827	910	884	1070	1115	1198	1290	0	0	0 0			0	0		0	0	0		0 0	0 0	0	> c		0
Entry	!	1472	1627	1489	1407	1350	1495	1405	1376	1376	1154	#011 408	1049	1777	1136	1068	1146	1254	1158	1875	1724	2024	2199	2004	1970	1758	1866	2266	2091	2084	2377	2577	2351	0	0	0 0			0 0			0	0	0		0 0	0 0	0	> 0		_
	Complex	0 !	10	0	-	- c		0 4	c		0 0		t C			ď		0 0	0 0	0 0	0	0	11	0	0	11	16	3	9	9	13	4	9													†	1	†	†	‡	-
P3	H	247	79	113	77	7	9 4	13	90	20	02	98 8	170	240	115	271	321	253	282	230	159	114	41	23	150	152	108	87	31	220	189	214	219								+	1			1	+	1	+	+	+	_
PDA Review	SS	+	0	9 7	- 0			14 0		24		2 2	14		7 60	4 0	2		- 6	, -	-	3	5	4	1 35	1 7	5 12	25 15	8 5	3 1	33 4		3 8										1		1	+	$\frac{1}{1}$	+	<u> </u>	<u> </u>	_
P2	Š	461 4	362 1	487	876	550		1 1	Ì	000		724 154	1000	 	840 0	ľ	977	. «	+	- -	551	449	ŀ	ŀ	508 2	562 21	578 2	659 2	3 999	629	659 3	722 2	785 23				1	1			1			1	1	+	$\frac{1}{1}$	+	+	+	_
	SS	1	-	ł	+	+	+	38	ļ	Ŧ	ļ	16	ļ	-	+	ļ	ļ	ŀ	+	+	ŀ	ŀ	ŀ	H	ŀ	43 6					217 (225	249 7													1	+	+	+	1	_
	Complex	32	36	09	1 00	108	100	121	14.4	4 6	88	200	50	60	20	71	82	105	95	145	143	162	194	203	196	164	180	202	189	138	146	162	173													\dagger	\dagger	\dagger	\dagger	+	-
P3	XIW	009	654	614	700	866	000	649 605	663	611	570	755	433 708	2000	937	587	572	625 625	610	1003	824	906	976	932	843	992	856	988	962	838	1019	1076	1029													T		1	1	†	_
Entry	SS	14	52	47	43	40 65	71	13	3.4	36	20	22 11	14	09	00	110	113	135	130	260	200	236	254	249	184	169	191	222	230	222	289	304	282																		
PDA Entry	Complex	29	62	52	7/	0/	ò	44	QV V	73	2 2	24,0	31	30	30	27	37	502	90	44	37	54	9	46	58	42	35	51	- 29	64	62	47	72																	\int	
P2	XIW	208	746	633	929	520	010	253	185	460	412	282	302	000	200	214	285	280	202	331	477	483	492	378	515	479	456	583	482	682	269	296	629				\downarrow		\downarrow								1	_	_	_	
	SS							14 4											ı	ı	ı	ı		196			2 148		169				136	12	21	210	310	31.	710	310	710	210	2	210	210	210	77	710	210	310	?
	Date	4/01/2022	10/01/202	17/01/2022	24/01/202	31/01/2022	1102/202	24/02/2022	2/03/2022	2/03/202	14/03/2022	21/03/202	29/03/2022	4/04/2024	11/04/2022	18/04/202	26/04/2022	4/05/202	9/05/202	16/05/202	23/02/202	30/05/2022	6/06/202	13/06/2022	20/06/202	27/06/2022	4/07/202;	11/07/202	18/07/2022	1/08/202;	8/08/2022	15/08/202	22/08/2022	29/08/202	5/09/202.	12/09/2022	19/09/202	20/09/202	3/10/2022	10/10/2022	1//10/202	24/10/2022	31/10/2022	2202/11//	14/11/2022	707/11/12	28/11/2022	5/12/2022	12/12/12/2	19/12/2022	26/12/2017



	Mondays	Data collected at cern on Mondays Entry Review Total	Review	Total		
	2/08/2021	564	509	- 1		
	3/08/2021	508	533	- 1		3500
	23/08/2021	401	202	-		
	30/08/2021	469	225	1		
	6/09/2021	543	260	ı		3000
	13/09/2021	669	459			
	20/09/2021	790	456	- 1		
	27/09/2021	911	280	- 1		2500
	4/10/2021	922	671	- 1		
	11/10/2021	82/	020	- 1		
	26/10/2021	020	710	1530		000
	474479034	930	200	1		2000
	9/11/2021	1130	609	1707		
	15/11/2021	1308	647	1		
	22/11/2021	1560	202	1		1500
	20/11/2021	1630	129	1		
	6112021	1818	1000	1		
	13/13/2021	1515	260	1		
,	3/04/2022	1337	700	-		1000
7	3/01/2022	14/2	87/	- 1		
n -	10/01/2022	1627	483	- 1		
4	17/01/2022	1489	629	2118		200
2	24/01/2022	1457	587	- 1		i
9	31/01/2022	1350	595	- 1		
7	7/02/2022	1493	658			
8	14/02/2022	1405	802			
6	21/02/2022	1271	913	ı		
10	28/02/2022	1376	883	ı		
1	2/03/2022	1265	1060	1		
- \$	14/03/2022	1454	7000	1		
2 5	14/03/2022	1134	104	- 1		
13	21/03/2022	/68	111/	- 1		
14	29/03/2022	1049	1173	- 1		
15	4/04/2022	1117	1235	2352		
16	11/04/2022	1136	1031	2167		
17	18/04/2022	1068	1303	2371		
18	25/04/2022	1146	1367	2513		
19	2/05/2022	1254	1241	2495		
000	0/05/2022	1458	1166	2324		
24	18/05/2022	1875	0/0	2824		
7 6	10/03/2022	10/0	240	2024		
3 2	23/03/2022	1724	2 0	2443		
53	30/05/2022	2024	689	2613		
24	6/06/2022	2199	434	2633		
25	13/06/2022	2004	499	2503		
26	20/06/2022	1970	755	2725		
27	27/06/2022	1758	962	2554		
28	4/07/2022	1866	827	2603		
200	11/07/2022	2266	040	3176		
67	1907/2022	2200	2 6	2000		
30	18/07/2022	2091	884	2975		
			JAH absent	week startir	ng 25/07/2022 - dai	 data line removed
32	1/08/2022	2084	1070	3154	3154	
33	8/08/2022	2377	1115	3492		
34	15/08/2022	2577	1198	3775		
35	22/08/2022	2351	1290	ı		
36	29/08/2022		-	1		
37	5/00/2022	0 0	0 0	0		
5	4200/2022					
8	2703/2027	0 0	0 0	0 0		
39	19/09/2022	0	0	0		
40	26/09/2022	0	0	0		
41	3/10/2022	0	0	0		
42	10/10/2022	0	0	0		
43	17/10/2022	0	0	0		
44	24/10/2022	c	c	c		
	24/40/2022		0			
2 4	7141,0000	0	0			
ţ.	444410000					
4	14/11/2022	0	0			
48	21/11/2022	0	0	0		
49	28/11/2022	0	0	0		
20	5/12/2022	0	0	0		
51	12/12/2022	0	0	0		
25	0000000					
	13/12/2022	0	0	0		





	Total hours of work required on unallocated statements (hrs)	NB. This uses the estimated times for C1-3 as found in Statement Time worksheet	158.13	116.07	101.82	70'DR	00.111	102.84	27.18	83.01	147.01	10. 10.	27.47	OR 18	402.73	110.05	122.77	0	120.89	113.72	135.27	131.52	102.13	110.35	53.44	402.47	129.82	195.67	176.73	0	105.53	0	0 0	-	000	0	0	0	0	0	0	0	0	0	0	0	0	-	0
	Total Unallocated statements	æ	40	39	47	48	BC	03	CC	£ 02	20	70	45	787	53	28	55	0	99	59	90	40	52	21	20	90	97	52	49	0	51	0	0	0		0	0	0	0	0	0	0	0	0	0	0.0	0.0	0	
	8	KO .	9	4	0	c c	0	00	40	2 6	0 0	2		2		1 60	0 67		9	4	8	5	4	4	4.	- 0	3 6	0 00	7		3		1	1				l	Н			1				1	1	t	t
Reviews	8	12	=	8	4	-	- (0	70	200	9 9	0 4	o c	76	7		7		3	3	7	8	89	4		-	* «	45	13		5			Ī													İ	1	Ī
	2	ĸ	8	9	6	e (0	9	7	- 6	,	- 0	-	¥	+ 0	4 67	-		1	1	2	2	0	2	0	0	0	6	6		0																		
200	ខ	13	15	14	15	10	BL	18	11	17		8	CI	43	47	14	45		17	19	12	12	15	15	BL	0.0	22	14	15		14					×													
Statements	g	20	18	17	24	23	87	26	27	22	32	3 12	27	20	32	37	33		34	35	33	25	34	33	3 5	25	30	33	30		33														I	I	I	I	
	5	9	8	00	8	13	71	20	200	2 5	2 *		3	- 10	0 4	2	7		2	5	5	3	3	4	4- 0	•	+ 6	2 10	4		4															Ī	Ī	Ī	T
Ī	Week starting (date) NB. bata collected the Monday following the week.	27/12/2021	4/01/2022	10/01/2022	17/01/2022	24/01/2022	31/01/2022	A MANAGED A	2402/2022	28/12/2012	TANAMAN	44103/2002	24/03/2022	2002000	A/MA/2012	11/04/2022	18/04/2022	25/04/2022	2/05/2022	9/05/2022	16/05/2022	23/05/2022	30/05/2022	6/06/2022	13/06/2022	ZUNDO/ZUZZ	4/07/2022	11/07/2022	25/07/2022	1/08/2022	8/08/2022	15/08/2022	22/08/2022	ZWUSYZUZZ EJMOJOWYS	12/DB/2022	19/09/2022	26/09/2022	3/10/2022	10/10/2022	17/10/2022	24/10/2022	31/10/2022	7/11/2022	14/11/2022	21/11/2022	28/11/2022	5/12/2022	12/12/2022	26/12/2022
	Data collection date	4/01/2022	10/01/2022	17/01/2022	24/01/2022	31/01/2022	1102/2022	14/02/2022	2022000	7102/2022	44/02/00/00	14/03/2022	21/03/2022	AINAPORT	11/14/2022	18/04/2022	25/04/2022	2/05/2022	9/05/2022	16/05/2022	23/05/2022	30/05/2022	6/06/2022	13/06/2022	ZUNDO/ZUZZ	ZINDIONIZA ZINDIONIZA	44/07/2022	18/07/2022	1/08/2022	8/08/2022	15/08/2022	22/08/2022	29/08/2022	3/US/2022	19/19/2022	26/09/2022	3/10/2022	10/10/2022	17/10/2022	24/10/2022	31/10/2022	7/11/2022	14/11/2022	21/11/2022	28/11/2022	5/12/2022	12/12/2022	19/12/2022	2/01/2023
Week Number			1	2	es .	4	0	9		0 0	40	200		43	44	15	16	17	18	19	20	21	22	23	24	20	27	28	30	31	32	33	34	30	37	38	39	40	41	42	43	44	45	46	47	48	43	000	52

_																																					
423	441	302	355	448	508	236	281	477	431	521	457	526	302	649	501	452	591	490	550	009	506	442	492	573	573	575	437	512	259	271	443	326	427	200	515	631	430
734	622	617	526	379	504	544	484	296	831	842	758	0	0	0	#REF!	278	504	663	376	626	767	761	574	372	652	715	720	909	253	503	464	444	298	476	417	534	468
612	518	515	438	316	420	453	404	497	692	701	632	0	0	0	#REF!	232	420	553	314	522	639	634	479	310	543	969	009	504	461	419	387	370	248	397	347	445	390
691	989	584	421	099	604	923	662	923	986	842					608	099	757	418	969	852	845	889	413	724	767	008	672	614	699	516	493	331	679	463	593	520	511
37	38	39	40	41	42	43	4	45	46	47	48	49	20	51	2	3	4	2	9	7	8	6	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
13/09/2021	20/09/2021	27/09/2021	4/10/2021	11/10/2021	18/10/2021	25/10/2021	1/11/2021	8/11/2021	15/11/2021	22/11/2021	29/11/2021	6/12/2021	13/12/2021	20/12/2021	3/01/2022	10/01/2022	17/01/2022	24/01/2022	31/01/2022	7/02/2022	14/02/2022	21/02/2022	28/02/2022	7/03/2022	14/03/2022	21/03/2022	28/03/2022	4/04/2022	11/04/2022	18/04/2022	25/04/2022	2/05/2022	9/05/2022	16/05/2022	23/05/2022	30/05/2022	6/06/2022

460	487	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
383	406	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
541																												
25	26		28		30			33		35	36		38		40	41	42	43	44	45	46	47	48	49	20		52	
13/06/2022	20/06/2022	27/06/2022	4/07/2022	11/07/2022	18/07/2022	25/07/2022	1/08/2022	8/08/2022	15/08/2022	22/08/2022	29/08/2022	5/09/2022	12/09/2022	19/09/2022	26/09/2022	3/10/2022	10/10/2022	17/10/2022	24/10/2022	31/10/2022	7/11/2022	14/11/2022	21/11/2022	28/11/2022	5/12/2022	12/12/2022	19/12/2022	26/12/2022

INSTRUCTIONS FOR USE

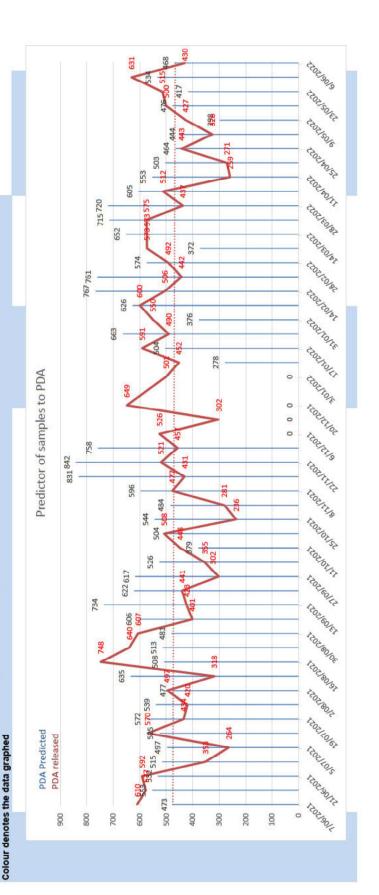
On Thursday afternoon of the week starting (as per Column A), access the Management Review Analytical Team data in the FR (NB. This data is collected per week number). Add together the values for the following rows: Diff Lysis Retain Supernatant, Differential Lysis DNA IQ, Maxwell 16 DNA IQ, Nucleospin Tissue, QIASymphony, QIASymphony - Integrated, Retain Supernatant DNA IQ. This becomes the total Extractions for the week.

After adding this value to Column D, a value should automatically populate in Column G. This value factors in the average number of extractions that end up with an interpretation of No DNA Detected or DNA Insufficient Multiply the Total Extractions by 1.2. This is to account for the data being collected on Thursday and assumes equal number of daily extractions. Add this total value to column D.

When a value automatically populates in Column G, another value automatically populates in Column H. This value factors in an approximate rework rate of 1.2.
The final value in Column H is the predicted amount of samples on the PDA lists for the coming week based on the values obtained from the Analytical processing in the previous week.

Information only. NB. Column C collection discontinued data collection from 19 October, 2020: used to determine the average of 25% for NDNAD and DIFP used for estimator

* Data collected on Thurs PM to enable plan for coming week. Figure is multiplied by 1.2 to account for collecting the data on a Thurs (ie. 4/5 of the week, assuming equal no. exts per day)
Data collected on Monday morning following the week starting [date]



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19	2/05/2022	601.12	483 30	18	146	262		8.33		-180 23	0,00	42.10			26	2324
8	25/04/2022	601.12	468 09	15	311	407		5 64		-4 96	00 808	194 02			22	2495
17	18/04/2022	601.12	445.43	12	352	271		3 09		51.33	00	08.33			22	2513
16	11/04/2022	601.12	438.76	2	294	236		-1.78		-2.16	10 00	30.37			26	2371
15	4/04/2022	752.15	551.78	2	512	478		-6 04		139 55	7000	220 94			23	2167
4	28/03/2022	752.15	686.61	7	637	420		-6.73		173.54	7	66.701			46	2352
13	21/03/2022	752.15	740.19	18	601	514		3 20		101 37	20 11	17 / 71			45	2222
12	14/03/2022	752.15	714.09	15	624	426		0.72		141.99	90	101.09			46	2014
=	7/03/2022	752.15	726 50	17	262	333		2.47		104 61		2.44			52	2195
9	28/02/2022	752.15	632.71	8	520	371		-4.65		92.92	0.00	83.12			25	2325
6	21/02/2022	752.15	673.02	11	578	232		-2.46		123.71	74.00	-/4.23			54	2259
80	14/02/2022	752.15	705.22	14	609	352		-0.10		132.98	0.80	31.13			22	2184
7	7/02/2022	752.15	701.68	11	719	367		-3 03		245.37	11	47.74			63	2207
9	31/01/2022	752.15	631.43	10	540	406		-2.63		113.78	0,1	118.70			26	2151
ĸ	17/01/2022 24/01/2022 31/01/2022	752.15	529.06	10	374	368		-0.58		1688	407.00	121.28			46	1945
4		752.15	229 60	16	425	324		481		47.27		09.38			47	2044
က	10/01/2022	752.15	624 28	6	529	335		-3.49		107 61	r C	50.95			39	2118
2	3/01/2022	601.12	368 07	4	403	313		-3.36		154 56	7	145 53			40	2110
FR Week	Week	Expected Hours	Observed Hours	Statements reviewed	Interp Result Entry	Interp Result Review	Statements reviewed variance (0.020 per Obs	hour)	Interp Result Entry variance (0.675 per Obs	hour)	Interp Result Review variance (0.455 per Obs	nour)	Outstanding Unallocated	Statements after week	starting [date]	Outstanding PDA after week starting [date]

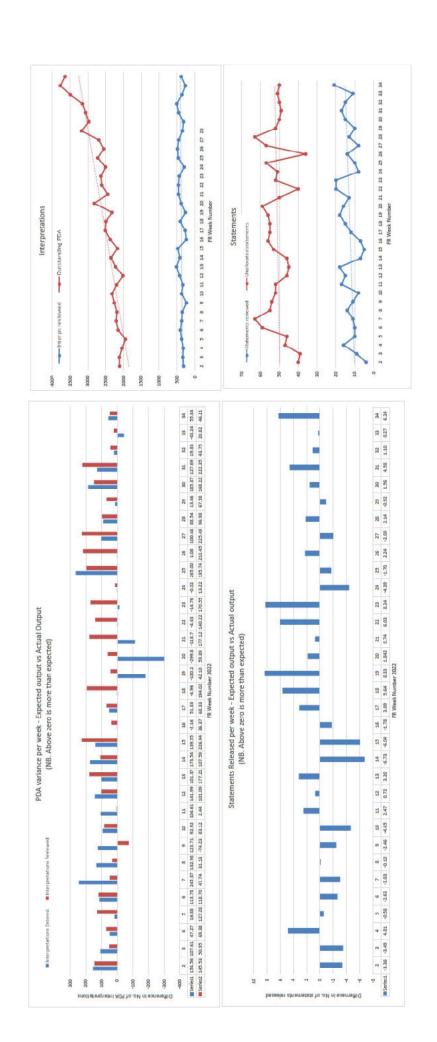
FR Week	20 L	73	22	23	24	22	56	27	28	59	30	33	32	33	34	32	36	37
Week	9/05/2022	16/05/2022	23/05/2022	23/05/2022 30/05/2022	6/06/2022	13/06/2022	20/06/2022	27/06/2022	4/07/2022	11/07/2022	18/07/2022	25/07/2022	1/08/2022	8/08/2022	15/08/2022	22/08/2022	29/08/2022	5/09/2022
Expected Hours	752.15	736.95	736.95	736 95	736 95	736.95	752.15	752.15	752.15	752.15	752.15	752.15	752.15	752.15	752.15	752.15	752.15	752.15
Observed Hours	707.92	612.92	698.41	587.79	619 29	585.19	588 03	500 03	542.90	545 99	672.05	62120	90:269	536.66	738 01	00 0	0.00	00.0
Statements reviewed	16	13	20	20	8	10	14	8	13	10	15	17	15	11	21	0	0	0
Interp Result Entry	178	300	467	382	418	099	398	438	455	382	639	547	489	319	554	0	0	0
Interp Result Review	382	456	458	438	295	462	486	453	344	316	454	505	360	265	382	0	0	0
Statements reviewed																		
variance (0.020 per Obs																		
hour)	1.842	0.74	6 03	8 24	-4.39	-1.70	2 2 4	-2.00	2.14	-0 92	1 56	4.58	1.10	0.27	6.24	000	0.00	00.0
Interp Result Entry variance (0.675 per Obs																		
hour)	-299.85	-113.72	-4.43	-14.76	-0.02	265.00	1 08	100.48	88 54	13.46	185.37	127 69	1983	-43 24	55.84	00 0	0.00	00.0
Interp Result Review variance (0.455 per Obs																		
hour)	59.89	177.12	140.22	170 55	13.22	195.74	218.45	225.49	96 98	67.58	148.22	222 35	43.75	20 82	46.21	0000	0.00	0.00
:																		
Outstanding Unallocated	_																	
Statements after week																		
starting [date]	26	20	40	52	51	22	36	22	63	52	20	49	20	51	20	0	0	0
Outstanding PDA after																		
week starting [date]	2824	2443	2613	2633	2503	2725	2554	2693	3176	2975	3064	3154	3492	3775	3641	0		

FR Week	38	39	40	4	42	43	4	45	46	47	48	49	20	51	52
Week	12/09/2022	19/09/2022	26/09/2022	3/10/2022	10/10/2022	17/10/2022	24/10/2022	31/10/2022	7/11/2022	14/11/2022	21/11/2022	28/11/2022	5/12/2022	12/12/2022	19/12/2022
Expected Hours															
Observed Hours	0.00	000	00'0	0.00	0.00	00:0	00 0	00 0	00.0	00 0	000	00.0	0.00	00 0	00:00
Statements reviewed	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Interp Result Entry	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Interp Result Review	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Statements reviewed															
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hour)	00.0	000	0.00	0.00	00:0	00:00	000	000	0.00	00 0	00 0	0.00	0.00	00 0	0.00
Interp Result Entry															
variance (0.675 per Obs															
hour)	0.00	000	0.00	00:0	0.00	0.00	000	000	00.0	000	000	0.00	00.00	00 0	0.00
Interp Result Review															
variance (0.455 per Obs															
hour)	00.0	000	0.00	0.00	00:00	00:00	00 0	00 0	0.00	00 0	000	0.00	0.00	00 0	0.00
Outstanding Unallocated															
Statements after week															
starting [date]	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Outstanding PDA after															
week starting [date]															

ediately prior and after obtained. Data added is average of RED TEXT

DATA Information

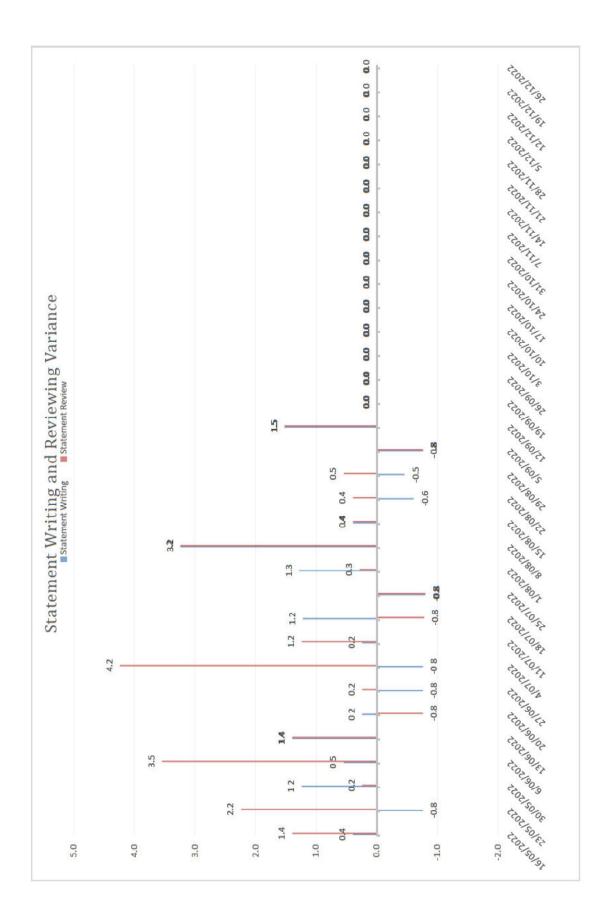
1. Especial fours are the hours expected as per contract. If there is one public holiday (eg, four-day week), the expected hours are only if
2. Observed hours are the actual hours a person worked in the week. For example, if a If I staff member was SICX for one day in the
2. Observed hours are the actual hours a person worked in the week. For example, if a If I staff member was SICX for one day in the
3. The data is based on 6 months of collation in 2021 for the two Reporting Teams as they were at that time. This source data excluded
4. Statement review and writing data is assumed to be equal is time allocated per week (factor of 0.000 per hour). This factor is used in



8	•	Г																Г	77 74	1 11								
iew	Variance	1.4	2.2	0.2	3.5	1.4	8.0-	0.2	4.2	1.2	8.0-	-0.8	0.3	3.2	0.4	0.4	9.0	8.0-	1.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Statement Review	Observed	2	3	1	4	2	0	1	2	2	0	0	1	4	1	1	1	0	2				9	o e				
Sta	Expected	9.0	0.8	0.8	0.5	9.0	0.8	0.8	0.8	0.8	0.8	0.8	0.7	0.8	9.0	9.0	0.5	0.8	0.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
ing	Variance	0.4	-0.8	1.2	0.5	1.4	0.2	8.0-	-0.8	0.2	1.2	8.0-	1.3	3.2	0.4	9.0-	-0.5	-0.8	1.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Statement Writing	Observed	1	0	2	1	2	1	0	0	- 1	2	0	2	4	1	0	0	0	2	8 18				Y.				
Stal	Expected	9.0	0.8	0.8	0.5	9.0	8.0	0.8	0.8	0.8	0.8	0.8	7.0	0.8	9.0	9.0	0.5	0.8	0.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	Variance	-3.8	7.7	13.7	34.6	10.2	-7.3	-15.3	35.7	23.7	4.7	-3.2	2.6	38.7	7.2	10.2	2.6	-2.3	5.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
PDA review	Observed	10	25	31	45	24	10	2	53	41	13	15	19	99	21	24	13	15	16									
	Expected	13.8	17.3	17.3	10.4	13.8	17.3	17.3	17.3	17.3	17.7	18.2	16.4	17.3	13.8	13.8	10.4	17.3	10.9	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	Variance	8.5	-5.7	4.4	-3.4	-18.5	30.4	26.4	-23.7	-2.7	-16.3	14.0	2.7	7.0-	1.5	-7.5	22.6	12.4	17.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
PDA Entry	Observed	29	20	30	12	2	99	52	2	23	10	41	30	25	22	13	38	38	34	E ()			9) 0					
	Expected	20.5	25.7	25.7	15.4	20.5	25.7	25.7	25.7	25.7	26.3	27.0	24.3	25.7	20.5	20.5	15.4	25.7	16.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
urs	Observed	30.4	38.0	38.0	22.8	30.4	38.0	38.0	38.0	38.0	39.0	40.0	36.0	38.0	30.4	30.4	22.8	38.0	24.0									
Hours	Expected	38.0	38.0	38.0	38.0	38.0	38.0	38.0	0.88	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0	38.0
Week starting		16/05/2022	23/05/2022	30/05/2022	6/06/2022	13/06/2022	20/06/2022	27/06/2022	4/07/2022	11/07/2022	18/07/2022	25/07/2022	1/08/2022	8/08/2022	15/08/2022	22/08/2022	29/08/2022	5/09/2022	12/09/2022	19/09/2022	26/09/2022	3/10/2022	10/10/2022	17/10/2022	24/10/2022	31/10/2022	7/11/2022	14/11/2022

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						3	View Val					17.8	12.4	5,1	-2.3				2000
0.0	0.0	0.0	0.0	0.0	0.0	3	Entry and Review Variance				22.6		10.2	2.6		-7.5			402015
0.0	0.0	0.0	0.0	0.0	0.0		PDA Entry	38.7						7.2	-0.7				404 404 404 404 404 404 404 404 404 404
							Id						14.0	5.7	-3.2		5.3		10001
0.0	0.0	0.0	0.0	0.0	0.0				35.7		23.7				-2.7		-16,3	-23.7	402
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38.0	38.0	38.0	38.0	38.0	38.0				34.6				10.2		-3.4		-18.5		401/80/K
21/11/2022	28/11/2022	5/12/2022	12/12/2022	19/12/2022	26/12/2022	50.0		40.0			30.0	20.0		10.0 0.3 7.7	-3.8	-10.0	-20.0		404 404 404 606



Erin Shearer

From: Kylie Rika

Sent: Wednesday, 25 May 2022 10:23 AM **To:** Justin Howes; Sharon Johnstone

Subject: RE: Predictor Phase 2

Thanks Justin

Kylie

From: Justin Howes

Sent: Tuesday, 24 May 2022 11:58 AM

To: Kylie Rika ; Sharon Johnstone

Subject: RE: Predictor Phase 2

Yes, I wanted this to digest with you before I speak more on it and I want to get one going for an individual, in addition to this team-based one. I will work on that this week and look to set a meeting next week or two on this.

justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services

Prevention Division, Queensland Health



Please note that I may be working from a different location during the COVID-19 Pandemic. The best contact method is via email.

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From: Kylie Rika

Sent: Tuesday, 24 May 2022 10:39 AM

To: Justin Howes ; Sharon Johnstone

Subject: RE: Predictor Phase 2

Hi Justin

Would it be possible to get together and have you demonstrate/walk through - I will understand better this way.

Thanks

Kylie

From: Justin Howes

Sent: Wednesday, 27 April 2022 3:29 PM

To: Sharon Johnstone Kylie Rika

Subject: Predictor Phase 2

Hi

Yesterday I had a look at the data from the two Reporting Teams for 2022 and did a quick evaluation of how it looks compared to the KPI Predictor data that I spoke to you about in your CSPs.

Recall:

- The KPI Predictor (attached) used the evidence gathered for 6 months from the KPI data in 2021. This data was not including Allison, Allan or Luke so it was a look just at the KPI data for RT1 and RT2. This data was then averaged per week and compared to the observed hours. Formulae then could be generated based on observed output per hour. It could then be used to estimate the expected output for team and individuals based on the expected hours in the coming week. For example, PERSON is expected to be at work 38 hours next week so the data driving the formulae shows they can be expected to output x PDA entry, PDA Review, statement draft and review. Similarly, the team is expected to have 400 hours next week, so can expect to see in KPI data y PDA Entry, review, Statements etc..
- The best cell to use is C57 in the Predictor spreadsheet because this includes all data from all members of RT1 and RT2 (irrespective of the sub-team they work in). You can add the 38 hours here to get an idea of individual, or you can add your 352 or 400 per team if that is your expected value for the coming week.
- After the week, you can then use the same formulae to see how the observed data went compared to the
 observed hours. So you might have been expecting 400 hours, but only observed 300, so you can add those
 hours in to the cell and see how your total data for the week went.
- This is data that is the product of staff adding the KPI data weekly for 6 months. It therefore is the final outcome of PDA and Statement work after all other work (eg. mandatory training, project work, plate reading etc) is conducted. It can't really be used for staff not performing review, or statement work at this stage ie. Megan or Tegan. However, you can simply average their own data and generate targets that way. You might want to then take their hours out of the rest of the team targets.

The next step for me was to see how 2022 has gone with the Predictor to see what differences we have between expected and actual/observed. This way we could see if the team data is showing better (or not) data than expected. Before looking at the data, I had thought we would see our PDA data better than expected because Allison, Allan and Luke's data wasn't included in the Predictor, so in effect, it is very much a minimum average...if that makes sense... Attached is the DRAFT look at this, and please go to Input vs Output worksheet for the graphs. I simply used the factor within the Predictor eg. 0.675 PDA Entry, to multiply with the observed hours to generate Expected data, and subtract that from the Observed data. This is then graphed below.

This data shows RT1 and RT2 data combined is performing better than the Expected data from the Predictor for PDA Entry/Review. This can now be used in so many ways going forward! You can see that the Statement data is not as high as Expected, and that is demonstrating the focus on PDA work. The outstanding data in a separate worksheet is overall quite stable this year showing we are releasing what we are receiving; we are still trying to catch up with the increase in number of items we had in late 2021. One way we can integrate the PDA Predictor is that, for example, we can estimate the number of samples to come to PDA eg. 400 and check that against the KPI Predictor to see that we have 700 hours of staff expected next week. With 700 hours, we are looking at 472 Entry and then after the week, we can evaluate how the team has gone with that goal of at least 400 results to enter. We would hope to see above the line for PDA Entry given the expected PDA entry output of 472. It can also be used at the individual level by looking at PERSON and knowing that they have a pretty average looking week ahead, so your expectation is that their output is comparable to the expected data from these spreadsheets.

Overall, I think this is really getting somewhere in meeting the expectations from staff and the organisation in using an evidence-based approach to setting realistic goals, and then evaluating the data to measure performance and ensure accountability. The next step is for this to digest with you both a bit, then to meet some time in the next couple of weeks to refine and discuss implementation points.

Happy reading!

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services Prevention Division, Queensland Health



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Wash your hands regularly to stop the spread of germs.

Erin Shearer

From: Kylie Rika

Sent: Tuesday, 5 April 2022 8:23 AM

To: Justin Howes

Subject: RE: SS High stutter guidelines_Final

Thanks Justin

I know these are guidelines but am I correct in thinking that unless there is a valid reason to deviate from them, then this is the preferred approach of the lab?

Thanks Kylie

From: Justin Howes

Sent: Monday, 4 April 2022 1:45 PM

To: Kylie Rika

Subject: RE: SS High stutter guidelines_Final

Hi Kylie

I will try to catchup with Angela on this. There is a comment against 17117 and for the next version, it would be a good time to consult case managers again and see what opinions can be listened to.

Thanks

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services Prevention Division, Queensland Health



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From: Kylie Rika

Sent: Monday, 4 April 2022 12:51 PM

To: Justin Howes

Subject: FW: SS High stutter guidelines_Final

Hi Justin

Do you know if Angela was able to do this and which SOP makes reference to it? Just had a staff member query around these guidelines.

Thanks Kylie

From: Justin Howes

Sent: Friday, 6 August 2021 3:05 PM

To: Angela Adamson

Cc: Kylie Rika ; Sharon Johnstone ; Allison

Lloyd

Subject: SS High stutter guidelines_Final

Hi Angela,

I have been speaking to Paula about this document and we think it should be put into the format in QIS 31548v6 which is the Minor Process Change Form. This way it ensures we are going through a change mgt process (albeit a minor change) and will be pdfd so it is not able to be edited (ie. for version control). It also documents the areas affected eg. notification, SOP to update etc.

As lead author, could you please find some time to add this to the template?

I have CCd the seniors as I know this was a body of work that was previously thought to be complete until this request.

Thanks Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services Prevention Division, Queensland Health



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Erin Shearer

From: Justin Howes

Sent: Monday, 22 August 2022 4:37 PMTo: Kylie Rika; Sharon JohnstoneSubject: RE: PIM agenda meeting and actions

Attachments: Pullup and ignore locus_16122021.xlsx

Hi,

I had asked BSAG and kept the survey in G: drive normal location. I don't recall anything from literature.

Re SS, I had asked if you both feel any more discussion is needed on this. I am sure staff would want to have a scientific discussion on many things and this could be one. I am not sure what staff would want so could be worth more discussion? The SOP has the guidelines and perhaps there are further considerations that could be discussed at the PIM that might improve the SOP? If there is an action item from a meeting already held that shows appetite for a discussion, then I think that would be good for staff to continue discussing as a group.

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services

Prevention Division, Queensland Health

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From: Kylie Rika

Sent: Monday, 22 August 2022 2:58 PM

To: Justin Howes >; Sharon Johnstone

Subject: RE: PIM agenda meeting and actions

Thanks Justin

Did you follow up on:

7. I will dig around. I had asked Angela for some lit searching and I know nothing came through. I think I asked BSAG as a survey item and sent finding through, but will double check.

Also, Justin, are you OK with the SS guidelines being the default position, unless really good reason to deviate?

Thanks Kylie

From: Justin Howes

Sent: Monday, 22 August 2022 2:40 PM

To: Kylie Rika ; Sharon Johnstone

Subject: RE: PIM agenda meeting and actions

Hi, I think you can both get together and move forward on the PIM. I would interested to hear how it goes.

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services Prevention Division, Queensland Health



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From: Kylie Rika

Sent: Monday, 22 August 2022 1:46 PM

To: Sharon Johnstone ; Justin Howes

Subject: RE: PIM agenda meeting and actions

Hi both

Just touching base on this. Justin, did you have any final thoughts before we start moving? I have just had a training meeting with Tegan (nearly finished her mix rev training). She let me know that she is encountering different ways that people are doing things and I let her know that we are trying to get some of the interpretation issues resolved.

Thanks Kylie

From: Sharon Johnstone

Sent: Tuesday, 31 May 2022 1:34 PM

To: Kylie Rika Justin Howes

Subject: RE: PIM agenda meeting and actions

None from me



Sharon Johnstone

Senior Scientist – Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Prevention Division, Queensland Health

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From: Kylie Rika

Sent: Tuesday, 31 May 2022 1:12 PM

To: Justin Howes **Cc:** Sharon Johnstone

Subject: RE: PIM agenda meeting and actions

Thanks Justin

- 3. I don't think plate reading is affected here. In thinking on this point again, another option would be to consider modelling -2 rpt stutter in STRmix. This would remove all ambiguity. We would need to re-do model maker, but maybe this option could be the best?
- 8. I will chat with you Justin on this point

Any final thoughts before we start actioning?

Thanks Kylie From: Justin Howes

Sent: Tuesday, 31 May 2022 12:59 PM

To: Kylie Rika

Cc: Sharon Johnstone

Subject: RE: PIM agenda meeting and actions

Ηi

I have some general points re below to consider:

- 3. Is there something here that needs to be communicated with plate readers? I don't think so as the points mostly relate to leaving the peak labelled. Just checking that there is no impact on plate readers.
- 4. I would think that stoch effects for 4p profiles would be a reasonable expectation given amped at 0.5ng or less, and split between at least 4 contributors in various ways. Would a reamp really assist low level ones like this as dropin values would come into effect a bit more too?
- 5. This is an interesting point for CMers discussion and could be good for one/two to put guidelines together on. Essentially, it is a reasonable assumption based on info so we could potentially condition on more that we currently do. I have had this point on my whiteboard for some time and would be interesting what comes out of it.
- 6. Another good discussion point. This could be a BSAG survey point.
- 7. I will dig around. I had asked Angela for some lit searching and I know nothing came through. I think I asked BSAG as a survey item and sent finding through, but will double check.
- 8. Does this need more discussion at a PIM? It is a comment against 17117 and I don't think it is particularly controversial (in my opinion), so as a guideline to assist opinions, do you think it needs any more fleshing out?
- 9. This could be a minor change request and would need some consultation with QPS if the default changes. It would be an interesting discussion point, esp alongside point 4 which considers a poss benefit in reamping samples. A Mic to full presents only one shot at it without knowing how many conts could be in the sample and pushes the case manager to make a decision not too dissimilar to approaching a P3 sample. I know some staff are amping without MIC at all for some of these low level samples so it would be interesting how the discussion goes.

Overall, excellent and looks like a pretty full agenda.

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services

Prevention Division, Queensland Health

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From: Kylie Rika

Sent: Tuesday, 31 May 2022 8:23 AM

To: Justin Howes **Cc:** Sharon Johnstone

Subject: PIM agenda meeting and actions

Hi Justin

Here are the meeting minutes (and action items) from a meeting Sharon and I had last week. Let me know if you are OK with us starting on the action items and/or if you would like to discuss.

Kylie and Sharon met to discuss the email from Emma, Angela and Cassie [Inconsistencies with interpretations, 01 April 2022]

1. Unresolved D8

We are seeing higher peak heights in our amps at the moment which means that the D8 issue is being seen again. This issue is where D8 amps higher than the rest of the profile and STRmix is unable to resolve the major peaks. There seems to be some inconsistency in the way people are handling this. Our previous advice was to try to resolve the issue where possible. The method for this is to re-decon at double accepts and if this doesn't work to amp down to try to reduce the peaks heights at D8. Not all people are doing this. Additionally we do not have an agreed solution if amping down doesn't help.

How would you like to progress this?

KDR to check if staff have been informed of the pathway of double iterations and amping down to resolve over-amped D8. If yes, KDR to send an email from FRIT seniors as a reminder. KDR to ask STRmix trainers to keep an eye out for times when this pathway doesn't resolve so that a discussion can occur with line manager, reporter and STRmix trainer

2. Saturation point

Since we are seeing larger peak heights, sometimes we reach saturation (30,000rfu). This is being missed because people are not used to seeing it. A reminder needs to be sent out. Additionally the question is asked whether peak heights >30,000rfu are ok for reference samples – the answer to this is yes because STRmix doesn't use the peak heights of the ref.

KDR to send an email from FRIT seniors as a reminder.

3. -2 rpt stutter

There are inconsistencies with how people approach potential -2 rpt stutter peaks that sit in a +1 rpt stutter position. Some people remove them, some people leave them labelled. Our advice is as follows:

- If the peak is below the +1 rpt stutter threshold leave it labelled
- If the peak is above the combined +1 and -2 rpt stutter threshold leave it labelled

• If the peak is above the +1 rpt threshold but below the -2 rpt threshold run STRmix and see if it is modelled as stutter some of the time. If it won't falsely exclude then leave it labelled. This requires some discussion about % weighting of the peak being designated as allelic.

How would you like to progress this?

> KDR to send an email from FRIT seniors on recommending the first two points. The third point can also be included in the email with recommendation that if anyone has this scenario occur, let line manager know so a discussion can occur with line manager, reporter and STRmix trainer.

4. 4p mixtures an the use of ratios in determining NoC

Low level 4p mixtures can be difficult to assign NoC due to AI and inconsistent ratios. Should we be amping twice to assist with identifying stochastic effects versus true peak heights of the contributors?

> SMJ to get more information from STRmix trainers

5. <u>Inconsistent conditioning</u>

This is still causing issues and needs some guidelines.

Discussion point at PIM to get views. Then perhaps task someone with putting some guidelines together.

6. Mutations

There has been some discussion around whether our guidelines for dropping the locus when a mutation is present is appropriate/too strict. We had implemented the current process based on advice from Duncan Taylor many years ago. It is not known whether this advice still stands or whether it has been reconsidered.

How would you like to progress this?

Discussion point at PIM to get views. If large split in views then seek advice from statspwg or bsag

7. Pull-up in stutter position

In October last year I put together a workflow for dealing with pull-up in stutter position. This workflow still has not been finalised and the issue continues to occur.

- > JAH, KDR and SMJ to search emails, diaries to find where this was last left. Is workflow ready to go to case managers for feedback?
- 8. Use of s/s quidelines & inclusion in the SOP so that everyone is interpreting these profiles in the same way
 - > KDR to speak with JAH re guidelines being default position, unless really good reason to deviate
- 9. <u>Can we change the DIFP process so instead of mic to 30ul, they are m</u>ic to full?

- > JAH checking with Steve Foxover if QPS are mass ordering further processing on a set of cases.
- SMJ to send an email from FRIT seniors asking if case managers want mic to 30ul or mic to full as default
- > KDR to write up PIM agenda and send appointment etc..

Thanks
Kylie and Sharon



Kylie Rika

Senior Scientist, Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services Prevention Division, Queensland Health

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Handle the first intended the order of the control	
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Dighter than in a suspected mutation/triallele situation, do you have criteria that describe when a locus may and falls on the shoulder of the faller peak in extreme cases, this may lead to the false exclusion of a minor resolved however this usually only deficite the student cannot be shoulder of the faller peak in extreme cases, this may lead to the false exclusion of a minor resolved however this usually only deficite its stutter variance parameter. Provided that all other diagnostic identifying possible evidence peak issues (i.e., mising stutter peak)s prior to interpretation. Issually the peak ifegrated, Regards, Clint Cochrane H. Justin, Response below from one of our reporting FS When F as a seasigned as a single 19 peak) or if there is a reproducible artefact (generally if following comparison there is an unintuitive LR (either exclusion or elevated non-exclusion) we at trace levels, stutter peaks that are a single 19 peak) or if there is a reproducible artefact (generally if following comparison there is an unintuitive LR (either exclusion or elevated non-exclusion) we a	ps affected by underlying pull-up, Given that profile analysis and interpretation are undertaken by different staff in different work units, it is likely that we would not identify this unless unintuitive results were ress the issue hologically (e.g., re-amplification with a smaller amount of target DNA). If this still failed to correct the issue, we would consider ignoring the locus during STRmix interpretation.
The most common reason for ignoring a locus is where there is a suspected 1 base pair resolution issue (ir and fails on the shoulder of the allen peek. In externe cases, this may lead to the false peek exclusion of a minor resolved however this usually only affects the stutter variance parameter. Provided that all other diagnostic resolved however this usually only affects the stutter variance parameter. Provided that all other diagnostic identifying possible evidence peak issues (i.e., missing stutter peaks) prior to interpretation. Usually the pee STRmix results at the affected locus and consider re-interpretation with the locus ignored if unintuitive result to pee this helps. Get back to me if you have any follow up questions. Regards, Clint Cochrane Hi Justin, Response below from one of our reporting FS 1.Bo you have criteria for when a peak is removed from STRmix consideration? For example, if a peak (stutt affecting the deconvolution and then consider removing the peak? **** Pis note, this is after a rework is performed affecting the deconvolution and then consider removing the peak? ***** Pis note, this is after a rework is performed affecting the deconvolution and then consider removing the peak? **** Pis note, this is after a rework is performed affecting the deconvolution and then consider removing the peak? **** This is a supported mutation/triallele situation, do you have criteria that describe when a locus may contend contemporated. However, we do not have strict guidelines as to the maximum number of loci or molecular weil interpreted. However, we do not have strict guidelines as to the maximum number of loci or molecular weil maximic.	s there a maximum number of loci that may be ignored and is this number affected by the MW location of the loci that are bo
FASS	, closely sized peaks that differ in size by 1 bp). While such peaks are typically able to be resolved if their peak heights are similar, they may fail to be resolved where one peak is substantially shorter than the other clone. This is readily identifiable by assessment of the primary and secondary diagnostics in STRmix (an exclusion at a single locus but inclusionary LRs at all/most other loca). Stutter peaks may also fail to be so we entuitive/closeptable, we would not ignore the locus for such a result (we would be comfortable to explain why the stutter variance was elevated). Newer versions of STRmix can assist with this issue by the norther of the peak. If this peak appears to be fairly high in height, we would generally ignore the locus in the initial deconvolution. Otherwise, we would closely assess the Its were produced.
FASS	
FASS	
WA	
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WA	affected by puli-up, do you remove from the data prior to deconvolution, or do you run with STRmix (and not remove the que
WA	comes difficult to assess whether there is a true peak presentigiven the masking. This is also the case where a microvariant is Iy we would re-amp to try and resolve the problem biologically before we consider it statistically.
WA	nitially increasing the DNA input where possible) to determine whether the issue can be replicated. Generally, unintuitive LR ally and we can justify our decision making scientifically, we will (gnore the locus.
WA	s there a maximum number of loci that may be ignored and is this number affected by the MW location of the loci that are bs
WA	hat cannot be accurately assigned, we don't ignore multiple loci. If there is a requirement to ignore multiple loci, I would sug nored. If there is clear justification to ignore a locus (that can be supported scientifically), I would consider potentially ignori
WA	
WA	

Justin Howes

From: Justin Howes

Sent: Monday, 24 August 2020 11:45 AM

To: Kylie Rika

Subject:Summary of meeting 18 August, 2020Attachments:RE: Summary of meeting 18 August, 2020

Hi Kylie,

Thankyou for your feedback on the first attempt at recalling our meeting last week. I have attached the thread to this email.

Thanks for meeting on Tuesday at 2pm to hear your ideas relating to LOD; I have tried to summarise the meeting below. Please let me know at your earliest convenience if I have not recalled accurately, or inadvertently omitted key points. If no reply or amendments, I will take it that this is an accurate reflection of the meeting.

Justin started the meeting by talking about a range of other items not including LOD. This took up 25min of the half hour slot. Then Justin said he was sorry we only had 5mins left as he knew Kylie wanted to leave early. Kylie replied to say it's OK she can stay as long as needed.

- Kylie mentioned that she wanted to come to the meeting to share her idea with Justin before taking to the management team as she felt that having Justin's support for the idea would go some way in the mgt team forum.
- Kylie mentioned that she wrote a paper ('Review: ...') and sent to Reporting Scientists and took on feedback through that process. Then she thought that we could make the threshold lower but then you would always have the situation of what to do with peaks below LOD. Then Kylie wrote a separate email with further thinking on the topic that was the idea of relaxing the rigid threshold of our LOD.
- Some key parts of Kylie's thinking related to:
 - o observations in samples of peaks that were below LOD that upon rework, these peaks went above LOD and in one situation (of the three examples presented) the peak went above LOR.
 - Trying to minimise 'incorrects' as she felt that if these examples went to statement stage, they would be reworked and then lead to amended results later.
 - Using all of the information in the profile and drawing on experience. This could be like observing a clean baseline and peaks in bins below LOD with good morphology, and using this as information for no. contributors, or rework given the examples where reworks assisted the profile interpretations.
- Kylie felt that while some feedback was to wait for Verifiler Plus, she felt that the idea couldn't wait as she wanted to help with some of the amendments that are occurring now. Feels that need to use this information to either rework, use in no. contributors or for exclusionary purposes.
- Justin mentioned at this stage that the intention of the meeting from his point of view was to listen to understand the idea, and not to provide opinions on the points for now, rather to assist in the process of seeing how the idea could be developed.
- Justin mentioned that he would be happy to support the progression through the formal change management process and through that, the scope of the idea could be developed.
- Justin said that what he was hearing was that there could be profiles with clean baselines and sub-LOD signals and these observations could promote a rework, and that could assist the profile interpretation.
- Kylie indicated that wanted my support for the idea before going to the Mgt Team at this stage. Justin suggested that if Kylie wanted to discuss the idea with the management team in a way other than coming to a meeting and discussing, she could meet with the relevant people and go through the idea. Kylie didn't reply to this.
- Kylie mentioned that she is working with Sharon on a task that Justin set and one point was on labelling subthreshold peaks but felt that was not a good idea in her opinion to be labelling these for STRmix. Justin asked if Kylie had spoken to Sharon on that point Kylie said not as yet. Justin recommended talking to Sharon as he read it completely differently in that the peaks in the LOD-LOR range could be labelled as the

Analysis Method is used for CEQ checking, and could help reduce amendments by labelling the range. Possibly with GMIDx v1.6 and pdfing of batch that we could have these available for case management early to help – either saved in the sample, or the batch- but not for creating a file for STRmix. Yet to be explored more with Kerry-Anne but that was how Justin read it. Kylie acknowledged that that sounded different and could be what Sharon meant.

- Kylie spoke about the paper she wrote and that it specifically mentioned that management team were not included at that stage as she wanted to hear feedback from staff on the discussion of the idea in the paper before going to Mgt Team. Justin mentioned that at some point, the idea would go to the decision making group being the mgt team and explore it and we would all accept whatever outcome is reached.
- Kylie mentioned that she knows Sharon does not support the idea and after the phone call from Justin and from previous conversations, felt that Justin didn't support it. Justin asked when this was Kylie said she remembered from years ago where Justin had said not to look below the LOD, and was aware that Justin had mentioned that recently to a staff member. Justin didn't reply to this.
- Kylie mentioned that she understands that Sharon felt disrespected and that Justin was, and would she do things differently now? She said she probably would but said she didn't understand why it appears to be so controversial.
- Kylie said that there would always be the need to consider less than LOD and Kylie raised FaSTR DNA can assess the baseline dynamically and relaxing the LOD seemed to be a good lead into that. Justin mentioned not sure if that is the case as FaSTR DNA hasn't been looked into thoroughly and that initial looks by Allan are that it may not be do what we thought it might. It might be that a threshold still needs to be added.
- Journal article provided by Kylie which had the conclusion copied into her report. Kylie read the conclusion. Justin agreed with the information and confirmed that written in 2015 and that the AT for the authors is the LOR here, and the progression of the thinking of the authors is such that the Artificial Intelligence work occurred around and after that time, and perhaps this is where FaSTR DNA came from. Justin mentioned that still not sure about what is in the product as yet. Justin said it is good that other labs are interested in FaSTR DNA.
- Justin spoke about other labs and that is difficult to draw parallels as most are on 3500, but do know about NT where they have a LOR of 175RFU and drop-in level of around 90RFU and that they don't look below even if peak-like observations below.
- Justin said that in hearing the information it seems that in developing the idea, you would need to find the decision points eg. clean baseline and sub-LOD signal decision point: option of rework, or report; sub LOD signal and rework and it disappears and nothing else appears: decision point.
- Kylie said she didn't know what was so wrong with the idea in the report. Justin said time for reflection and to trust in the workshops that we have on communications coming up; to trust in the process. Kylie said she didreflect often and hoped others did too.
- Justin recommended that Kylie think of what practical outcome to focus on, and spoke to some key words outcome, intention, consequence and risks associated, and to focus on the positive.
- Some discussion on a separate matter of Change Mgt process for Intelligence vs Evidence reporting pathways and what that could deliver for the lab, including ability to focus on other things that lab could do. The idea involved much FR development and education, but could have some benefit. We then also talked about RT2's brainstorm on a new results reporting structure and Justin said he will revive that project as it does provide ways to reduce result amendments.
- At the end, Justin recommended Kylie to speak with Sharon on labelling peaks between LOD and LOR and to see if that is something that could be practically implemented to help staff. Justin recommended Kylie read the FaSTR DNA manual to see if it might do what seemed to be advertised in that it could calculate thresholds per sample/injection. Justin reiterated that happy to support the initiation of a change management request if Kylie wanted to do that, and through that formal process the idea can be scoped out. Justin said maybe the better way of doing the change request was to discuss what things need to be looked at or considered for RW pathways etc... Kylie said she didn't really understand because in order for her to devise guidelines on what to consider might be suitable to RW, first she needs permission to let staff know that they are able to actually look at peaks below LOD
- Kylie spent the last ~10 minutes of the meeting explaining to Justin how she feels that no matter what she says or does, she feels that she is always seeming to be the bad guy. Kylie said she doesn't know why she is treated this way because no one will actually sit down and tell her why or what they think of her. Justin's response was to trust the process with Tess. Kylie asked Justin to tell her what the opposing argument was to her idea about LOD, because if there is some massive dangerous aspect she has missed, please let her

know, because all she can see is risk in NOT looking at peaks below LOD – Justin didn't respond. Kylie mentioned how the sample that Allan and her worked on took 5 months to resolve just because Allan decided to dig his heels in and he had Justin's support. Kylie mentioned that at the end of the day, her view on the sample was correct (>3p) and Allan hasn't even bothered to acknowledge that. Kylie said that she wasn't sure why Allan's opinion carried more weight than hers with this sample given she has 20 years' experience interpreting profiles. Justin said that these profiles are tricky and subjectivity was the reason.

- Meeting ended approx. 4:40pm.

Regards Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

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Erin Shearer

From: Justin Howes

Sent: Tuesday, 19 July 2022 10:59 AM

To: Sharon Johnstone

Subject: FW: Initial request for new project **Attachments:** Analytical threshold test proposal.docm

Hi, can you please do this? I had sent an email to you and Kylie on our focus on the key strategic projects for now. We can potentially approve the idea, but not able to afford time to this at the moment esp during the CoI and while we have strat projects in progress.

Thanks

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services Prevention Division, Queensland Health

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From: Kirsten Scott

Sent: Tuesday, 19 July 2022 10:56 AM

To: Justin Howes **Cc:** Paula Brisotto

Subject: FW: Initial request for new project

Justin,

We need to have this decision written down and the document finalised please. Attached again for your reference.

When complete, please forward to Quality so they can store the document and communicate the outcome to the initiators and relevant management team.

Kirsten

From: Justin Howes

Sent: Monday, 20 June 2022 4:08 PM

To: Kylie Rika

Cc: Sharon Johnstone Kirsten Scott

Subject: RE: Initial request for new project

Hi, I can sort this out manually tomorrow.

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services Prevention Division, Queensland Health



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Wash your hands regularly to stop the spread of germs.

From: Kylie Rika

Sent: Monday, 20 June 2022 3:34 PM

To: Justin Howes
Cc: Sharon Johnstone

; Kirsten Scott

Subject: FW: Initial request for new project

Hi Justin

Given that the proposal is from both Adrian (R1) and Tom (R2), are you able to do this for Kirsten as there is not enough room for both Sharon and my signatures on the proposal.

Thanks

Kylie

From: Kirsten Scott

Sent: Monday, 20 June 2022 3:07 PM

To: Kylie Rika Thomas

Nurthen < Th >; Justin Howes

Cc: Adrian Pippia ; Paula Brisotto ; Chelsea

Savage ; Chelsea

Subject: RE: Initial request for new project

Kylie,

Thanks for the notification.

Can I have those notes added against the proposal, and have it E-signed and sent to me to finalise? We need to complete the paperwork with notes and a signature so it is acknowledged as an idea. We can reopen at any date in the future, or we can pre-set a review date if you wish.

If it is unsigned and on-hold there is no record of the decision or acknowledgment of the outcome of the process.

Thanks Kirsten

From: Kylie Rika

Sent: Monday, 20 June 2022 2:34 PM

To: Kirsten Scott
Thomas Nurthen
Cc: Adrian Pippia
Savage
Subject: RE: Initial request for new project

Thomas Ruman Summan

Hi all

Justin, Sharon and I have chatted about this.

Unfortunately at this time we are unable to allow staff time on a new project such as this one proposed. If we can get our PDA/rev total numbers down to say the levels of Sept 2021, then we could have another look at this project. I realise this is disappointing and we hate to have to slow down possible improvement initiatives, but as a team, we can only do so much. We have critical strategic projects and MM work as a priority over other projects currently.

Can we please put this project on hold for now – to be re-visited at a later date.

Many thanks
Kylie and Sharon

From: Kirsten Scott

Sent: Monday, 20 June 2022 1:54 PM

To: Sharon Johnstone

; Kylie Rika

; Kylie Rika

; Paula Brisotto

; Chelsea

Savage

Subject: RE: Initial request for new project

Afternoon All,

I have not seen this back in quality yet.

Has it been signed with notes and description as to the outcome of this proposal?

We can not leave it "pending"

Thanks Kirsten

From: Kirsten Scott

Sent: Friday, 10 June 2022 12:45 PM

To: Sharon Johnstone >; Thomas Nurthen ; Kylie Rika ; Justin Howes

Cc: Adrian Pippia >; Paula Brisotto

Subject: RE: Initial request for new project

Tom and Adrian,

Thanks for raising this.

We will need a clear decision about what is happening with this idea from the FRIT seniors, and to document that.

If it is going on hold pending Model maker/Proflex changes, and/or Verifiler, and/or time within the workunit to do this work, can it be signed off for now as on hold pending some of these reasons, and state the reasons on the form. We could add a review date, to revisit this project idea when we have the capacity to look at it.

Kylie, Sharon or Justin can you decide who is the most appropriate person so sign of and finalise the decision? When signed please forward to Quality to PDF, store and finalise

Kirsten

From: Sharon Johnstone

Sent: Wednesday, 8 June 2022 11:01 AM

To: Thomas Nurthen ; Kirsten Scott

Rika

Cc: Adrian Pippia

Subject: RE: Initial request for new project

Hi Tom,

I've had a look at the proposal. As a proof of concept I am happy to support this. Given that Model Maker work is still being finalised for the proflex, I would expect that it would be best to select data post implementation of those parameters. It would be interesting to know how much people are looking below threshold now with the 3500 as I was under the impression that information there is not considered as rigorously as for the 3130. Such work would also need to be balanced with BAU so the amount of time anticipated to complete the proposal would be a critical element to the decision to go ahead.

Thanks for putting ideas forward in pursuit of steam lining our work.

Regards, Sharon



Sharon Johnstone

Senior Scientist – Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Prevention Division, Queensland Health

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From: Thomas Nurthen

Sent: Thursday, 2 June 2022 3:09 PM

To: Kirsten Scott ; Kylie Rika >; Sharon Johnstone

Cc: Adrian Pippia

Subject: Initial request for new project

Hi Please find attached an Initial request for a new project put forward by Adrian and myself.

Thanks



Thomas Nurthen (He/Him)

Reporting Scientist - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Prevention Division, Queensland Health

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Queensland Health

Forensic and Scientific Services

Initial Request

Stage 1

		Proposal #:		
Proposed by :	Thomas Nurthen	Date:	02/06/2022	
	Adrian Pippia	Due Date:		
Title of Proposal:	Performance of Analytical threshold (AT) vs Limit of Detection /Limit of Reporting (LOD/LOR) PowerPlex 21 samples.			
Project type	☐ Administration ☐ IT/LIMS ☐ Laboratory ☐ Data mining/analysis ☐ External Project ☐ Other			
Brief Outline of Proposed Change				
The aim is to assess the performance of an Analytical threshold (AT) vs LOD/LOR for PowerPlex 21 samples.				
Consistent with the observations in Project#186, consistent high level pull up is regularly observed in casework PP21 samples. The presence of artefacts interferes with the determination of the number of contributors, requires more reworking, and can falsely increase the peak/stutter heights leading to inaccurate STRmix modelling and subsequent LR inaccuracies.				
We propose a proof of concept where the PP21 baseline (Analytical threshold is a single threshold, where we assess on the same information that STRmix can see) is calculated from the last 3500 model maker data using the weighted regression method of baseline determination.				
Dye specific analytical thresholds (AT) will be calculated and tested against the samples used for project #231 verification of STRmix 2.8, amplification positive controls and extraction positive controls.				
The amount of pull up removed/allele count/stutter count/ stutter heights and effect on the number of contributors will be assessed.				
No additional amplifications or capillary electrophoresis re-runs will be required.				
Line Manager :		Recommendatio	n:	
Signature:		Proceed to minor change Proceed to full project plan Place on hold or abandon Reason:		
Proposal restarted by:		Date:		
Approved By: Signature: Date:		Reason:		

Please convert to PDF, e-sign and lock document on completion.

Advise quality FSS BiologyQuality@health.qld.gov.au when finalised.



JH-68

Erin Shearer

From: Justin Howes

Sent: Wednesday, 18 May 2022 11:30 AM **To:** Kylie Rika; Sharon Johnstone

Subject: RE: rep/rev pairings

HI

I think if you both focus on what the enhancement could contain, then please add it to the system. It could have many parameters – users, case and profile type. We recently added the KPI information enhancement, so I think this is a good time to add any other data to help monitor team work and allocations. The hope is that data enhancements could be grouped and worked on by bna after v2 implementation settles.

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services Prevention Division, Queensland Health



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From: Kylie Rika <Kylie.Rika@health.qld.gov.au> Sent: Wednesday, 18 May 2022 11:11 AM

To: Sharon Johnstone ; Justin Howes

Subject: RE: rep/rev pairings

Thanks Sharon

As I mentioned, I don't have an issue with people self-allocating at all. My concern (not alarm) is around the possibility that the self-allocating is repeatedly happening with pairings involving the same 3 people.

As Justin mentioned, the only way to prove that this is definitely happening is to be able to look at trends by putting in an enhancement for stats looking at repeated rep/rev pairings.

Going into the P2 rev list regularly has shown me a possible repeated pattern of self-allocating within the trio, but there is no hard data to back up what I am suspecting anecdotally.

If you and Justin don't have the same concern, no problem – we can just wait for an enhancement and assess then.

Thanks Kylie

From: Sharon Johnstone

Sent: Wednesday, 18 May 2022 10:57 AM

To: Kylie Rika ; Justin Howes

Subject: RE: rep/rev pairings

Hi,

I've just looked into these cases and I can't see any reason for concern. To have a consistent PDAer and reviewer for bigger cases or ones that may have complications is quite normal. What exactly caused you concern about these ones? I don't understand the alarm

Regards, Sharon



Sharon Johnstone

Senior Scientist – Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Prevention Division, Queensland Health

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From: Kylie Rika

Sent: Tuesday, 17 May 2022 2:17 PM

To: Sharon Johnstone ; Justin Howes

Subject: RE: rep/rev pairings

The following are examples currently sitting on the P2 rev list that have been self-allocated:





My concern here is the potential formation of an "interp silo" along with bias in review process.

From: Kylie Rika

Sent: Tuesday, 17 May 2022 2:07 PM

To: Sharon Johnstone ; Justin Howes

Subject: rep/rev pairings

Hi both

As I have been going through the P2 rev list this week reviewing, I've noticed a few self-allocated cases involving rep/rev pairings of Jacqui, Adrian and Tom.

TBH this isn't the first time I've noticed self-allocated cases amongst just this trio.

I don't have a problem with staff self-allocating cases, more so, the selective self-pairing of rep/rev amongst these three.

How do you think we should address this?

Thanks

Kylie



Kylie Rika

Senior Scientist, Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services Prevention Division, Queensland Health



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JH-69

NOTE 1:

When wording your statements it is important to remember that the comparison is being performed by STRmix and therefore the conclusions are based on statistical interpretation. Intuitive checking is performed only to ensure that STRmix is giving an appropriate interpretation. Therefore statements such as 'Mr X cannot be excluded as having contributed to this profile and therefore I have considered the following propositions' are not appropriate under this model. Your statement should refer only to your assumptions and the statistical interpretation.

NOTE 2:

A link between the profile obtained and the assumption of number of contributors is recommended.

This could be written for mixtures in the following ways:

 The mixed DNA profile(s) obtained from this sample indicates the presence of DNA from three contributors. As such, an assumption of three contributors has been made for statistical analysis.

<u>Or</u>

 A mixed DNA profile has been obtained from this sample. Based on the information within this DNA profile, an assumption of three contributors has been made for statistical analysis.

This could be written for single source in the following ways:

- The DNA profile(s) obtained from this sample indicates the presence of DNA from a single contributor. As such, this DNA profile matches the reference DNA profile of XY.

<u>Or</u>

- The DNA profile(s) obtained from this sample matches the DNA profile of XY.

Example wording

Unknowns

123456789 Swab (A), near rear door

123456789 Swab (D), floor in foyer near charge counter

The DNA profiles obtained from these samples [match each other and] do not match the reference DNA profiles associated with this matter. Each of these DNA profiles indicated male gender.

Single Source

123456789 Swab (E), floor in charge area

The DNA profile(s) obtained from this sample indicates the presence of DNA from a single contributor. As such, this DNA profile matches the reference DNA profile of XY.

Based on statistical analysis, it is estimated that the DNA profile obtained is greater than 100 billion times more likely to have occurred if the DNA originated from Mr X, rather than if the DNA originated from someone other than and unrelated to Mr X.

OR

The DNA profile obtained from this sample matches the DNA profile of Mr X.

Based on statistical analysis, it is estimated that the DNA profile obtained is greater than 100 billion times more likely to have occurred if Mr X had contributed DNA rather than if he had not.

123456789 Swab (B), floor near cells

Nonconditioned Mixture

The mixed DNA profile(s) obtained from this sample indicates the presence of DNA from three contributors. As such, an assumption of three contributors has been made for statistical analysis.

The reference DNA profiles of John, Sam and Carol have been compared with this mixed DNA profile separately, in order to assess whether or not any of them may have contributed DNA. Based on statistical analyses, the results are as follows:

In favour of contribution:

John - It is estimated that the mixed DNA profile obtained is approximately 4 times more likely to have occurred if he <u>has contributed</u> DNA rather than if he has not.

In favour of non-contribution:

Carol – It is estimated that the mixed DNA profile obtained is approximately 100,000 times more likely to have occurred if she <u>has not contributed</u> DNA rather than if she has contributed DNA.

Inconclusive:

Sam – It is estimated that the mixed DNA profile obtained is <u>equally likely</u> if he has contributed DNA rather than if he has not.

Conditioned Mixture

Conditioned Mixture

The mixed DNA profile(s) obtained from this sample indicates the presence of DNA from X contributors, one of whom could be Carol. Since this sample is said to have been collected from Carol, it would not be unexpected to find DNA which could have come from her. In order to interpret this mixed DNA profile an assumption of DNA from X contributors, one of whom is Carol, has been made.

The reference DNA profile of John has been compared to this mixed DNA profile, to assess whether or not he may have contributed DNA along with Carol.

Based on statistical analysis it is estimated that:

In favour of contribution:

John - It is estimated that the mixed DNA profile obtained is approximately 4 times more likely to have occurred if he <u>has contributed</u> DNA [along with Carol] rather than if he has not.

In favour of non-contribution:

John - It is estimated that the mixed DNA profile obtained is approximately 4 times more likely to have occurred if he <u>has not contributed</u> DNA rather than if he has contributed DNA.

Inconclusive:

John - It is estimated that the mixed DNA profile obtained is <u>equally likely</u> to have occurred if he has contributed DNA rather than if he has not.

Excluded:

Based on the assumption of X contributors and the presence of DNA from Carol, the following reference samples are excluded as potential contributors to the mixed DNA profile obtained: John et al

Not unexpected findings

Rectal swab Anterior lower gum swab

The DNA profiles obtained from these samples [match each other and also] match the reference DNA profile of Carol. As these samples are said to have been taken from Carol, the finding of DNA which could have come from her is not unexpected, and therefore no statistical analysis has been performed.

Insufficient DNA 123456789 Graph 21; swab; pop bottle

This sample contained insufficient DNA to be suitable for analysis and was not tested further.

No DNA Detected 123456789 Graph 9; swab; cot 123456789 Graph 2; swab; flyscreen

DNA was not detected in these samples and therefore they were not tested further.

Complex – no STRmix 123456789 Graph 11; swab; right thong 123456789 Item 6; tapelift; back of hand

The complex mixed DNA profiles obtained from these samples indicate the presence of DNA from more than three contributors and are therefore unsuitable for statistical analysis.

Complex – unsuitable

123456789 Graph 5

Due to the complex nature of this DNA profile, including uncertainty as to the number of contributors, in my opinion this DNA profile is not suitable for meaningful interpretation.

JH-70

Erin Shearer

From: Justin Howes

Sent: Wednesday, 8 July 2020 11:29 AM

To: Emma Caunt

Subject: RE: taking into account combined stutter

Hi

Sure, I will let the seniors know that there is information in the manual to be considered. More/expanded information will be in the future interp SOP.

Thanks Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

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From: Emma Caunt

Sent: Wednesday, 8 July 2020 7:52 AM

To: Justin Howes

Subject: RE: taking into account combined stutter

Ok, will do.

Kylie's email mentions that some staff are not following this approach, will this be addressed to get everybody doing the same thing?

From: Justin Howes

Sent: Tuesday, 7 July 2020 4:23 PM

To: Emma Caunt

Subject: RE: taking into account combined stutter

Hi, ok it is mentioned to some degree as a consideration. Perhaps focus this in more detail in the future 'interpretation' SOP.

Thanks Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

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From: Emma Caunt

Sent: Tuesday, 7 July 2020 2:54 PM

To: Justin Howes
Cc: Kylie Rika

Subject: RE: taking into account combined stutter

Hi

I have checked the SOPs and have found the following:

QIS 17117 doesn't mention anything about combining stutters

QIS 33773 Section 23.5 states:

"A combination of stutter thresholds can be considered when determining the number of contributors for loci where +1 / -1 / -2 repeat stutter peaks of different alleles overlap."

This isn't very detailed, but does make the point.

The number of contributors guidelines (in the new Basics of Interpretation SOP) does talk about the removal and combining of stutters, but I feel that the information is quite outdated now that we are using new and improved stutter models with v2.7 (the guidelines were written when we were using v2.0.6). In it's current form it is quite confusing.

What would you like me to do? How would I go about updating the number of contributors guidelines?

Thanks

Emma

From: Justin Howes

Sent: Tuesday, 7 July 2020 11:05 AM

To: Emma Caunt
Cc: Kylie Rika

Subject: RE: taking into account combined stutter

Hi Emma

I see no need to consult the PWG then.

Please review the STRmix and interpretations SOPs and training material to ensure this information is available and documented. I think the information below is pretty clear and logical/intuitive.

Thankyou

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

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From: Emma Caunt

Sent: Monday, 6 July 2020 4:08 PM

To: Justin Howes **Cc:** Kylie Rika

Subject: RE: taking into account combined stutter

Hi

I would be happy to take this point to the statspwg, but I don't think that there is any point. The information is readily available in the STRmix User's Manual, additionally all other labs are modelling post stutter in STRmix and therefore don't have this issue.

Hopefully the below information will help.

STRmix has always modelled cumulative peaks. With the first version of STRmix this was only peaks consisting of allele and stutter, however they were modelled as a purely additive effect. This is the model that we are currently using.

The most recent versions of STRmix model all stutter types and it still considers them as additive.

From page 40 of the user's manual:

Where T_9^1 is the total allelic product generated from the mass parameters and genotype set. The proportion of allelic component in E_0^1 is:

$$\mathbf{A}_{9}^{1} = \frac{E_{9A}^{1}}{SR_{10}^{1}O_{10}^{1} + E_{9A}^{1} + FSR_{8}^{1}O_{8}^{1}}$$

And the proportion of stutter component is:

$$S_9^1 = \frac{SR_{10}^1 O_{10}^1}{SR_{10}^1 O_{10}^1 + E_{9A}^1 + FSR_8^1 O_8^1}$$

And the forward stutter component is:

$$\mathbf{F}_{9}^{1} = \frac{FSR_{8}^{1}O_{8}^{1}}{SR_{10}^{1}O_{10}^{1} + E_{9A}^{1} + FSR_{8}^{1}O_{8}^{1}}$$

So that $A_9^1 + S_9^1 + F_9^1 = 1$

 $p(O_9^1 \mid E_9^1)$ is then modelled by $N(0, \frac{S_9^1 k_{a-1}^2 + \frac{A_9^1 c^2}{C_{10}^1} + \frac{E_9^1 k_{a+1}^2}{E_9^1})$. Mathematically for locus 1:

I note from Justin's email "stutter being smaller peaks could be influenced more readily by stochastic effects". The model that STRmix uses states that the variance in stutter peaks has a direct relationship to the height of the allelic peak, it doesn't suggest that the stutter peak itself is subject to stochastic effects. From page 31 of the user's manual:

In words, this model states that as the expected peak height (or the parent peak height for a stutter) decreases, then the peak height variability increases. This relationship should be intuitive for most forensic scientists. However, there are sensible limits to this relationship,

From Kylie's email: "Other people think that there is no empirical data to suggest that stutter on top of stutter causes a push up in stutter peak height". This is a concern as we move forward to more continuous modelling, including the modelling of all stutter types, as this is the model that we will be using.

Please let me know if you would like me to contact Duncan to clarify/confirm any of this information.

Thanks

Emma

Emma Caunt

Scientist

Forensic DNA Analysis, Police Services Stream, Forensic & Scientific Services

Health Support Queensland, Queensland Health

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From: Justin Howes

Sent: Wednesday, 24 June 2020 3:36 PM

To: Emma Caunt

Subject: FW: taking into account combined stutter

Ηi

Please see the thread herein. Could you please consult StatsPWG on this point?

Thanks

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

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From: Kylie Rika

Sent: Wednesday, 24 June 2020 3:33 PM

To: Justin Howes

Subject: RE: taking into account combined stutter

No objections 😂

From: Justin Howes

Sent: Wednesday, 24 June 2020 3:31 PM

To: Kylie Rika

Subject: RE: taking into account combined stutter

Hi

Unless you object, I will actually forward this thread to Emma for her to summarise in her question to the PWG.

Regards Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

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From: Kylie Rika

Sent: Wednesday, 24 June 2020 3:29 PM

To: Justin Howes

Subject: RE: taking into account combined stutter

Yes that sounds good. Thanks Justin

Kylie

From: Justin Howes

Sent: Wednesday, 24 June 2020 3:20 PM

To: Kylie Rika

Subject: RE: taking into account combined stutter

Hi

I know we do have information regarding considering cumulative stutter, but am also not aware of any literature around this. In theory, I can understand it but similarly, stutter being smaller peaks could be influenced more readily by stochastic effects alone and not with the assumption that there is stutter from one or both of the peaks observed.

I will task Emma as the StatsPWG rep to consult other jurisdictions. It will have to wait for her to return, but I think this is an appropriate action I can task the PWG rep to take.

Does this task make sense to you?

Regards

Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

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From: Kylie Rika

Sent: Wednesday, 24 June 2020 3:11 PM

To: Justin Howes

Subject: taking into account combined stutter

Hi Justin

Over the last couple of months I've noticed that there appears to be two schools of thought around what to do with cumulative stutter.

Some people think that stutter on top of stutter would push the stutter peak height up. Therefore if there is this type of combined stutter above stutter t/hold then its OK to still consider it stutter.

Other people think that there is no empirical data to suggest that stutter on top of stutter causes a push up in stutter peak height.

Depending on what school of thought you align with, can cause different calls in number of contributors etc...

Please let me know if/how you would like me (or someone else) to address this so that we have staff on the same page with this.

Thanks

Kylie



Kylie Rika

Senior Scientist - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

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JH-71

Justin Howes

From: Justin Howes

Sent: Wednesday, 8 July 2020 11:38 AM **To:** Sharon Johnstone; Kylie Rika

Subject: combined stutter

Hi Kylie and Sharon

I understand there are some differing views on the considerations relating to observations of potential stutter at the same location and the potential cumulative effect if stutters are coinciding eg. -2rpt at same size as +1rpt for a different parent allele. I had asked our StatsPWG rep (Emma) to consult the PWG but decided there was no need given there is a point about considering the effect in our SOP 33773 and in the STRmix manual.

I have asked for more information to be added to the future interpretation SOP that Emma has been tasked.

In the meantime, could you please ensure that your staff do consider this effect and can consult the manual for the equations that are considered (pg 40).

Thanks Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

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JH-72

Justin Howes

From: Kylie Rika

Sent: Wednesday, 9 September 2020 4:54 PM

To: Allison Lloyd; Sharon Johnstone

Cc: Justin Howes

Subject: RE: Combined stutter

Hi Allison

I think a FRIT seniors meeting including Justin, sounds like a great idea – we haven't had one in a while.

In my view, the "tension" is nothing more than there being differences of opinion, but I acknowledge the impact this has had on you and it's not good that you feel so uncomfortable. As a FRIT senior group and as a FRIT team, we need to find better ways to manage the differences of opinion so that people don't feel as though things are personal and so that people don't feel disrespected. I am looking forward to the communication workshops with Tess.

Thanks Kylie

From: Allison Lloyd

Sent: Wednesday, 9 September 2020 2:09 PM

To: Kylie Rika Sharon Johnstone

Subject: RE: Combined stutter

Hi both,

I believe that the FRIT seniors should probably get together and have a bit of a discussion about some specific topics to do with this before having a full on meeting with FRIT.

Look, I don't know what's going on between the two of you but it is starting to get ridiculous. We're all supposed to be on the same team and work together. This year has been especially hard for everyone but I am tired of defending each of you to members of FRIT and to members of your own teams when they ask me what's going on with you both because the tension is obvious. It makes me uncomfortable and is one of the reasons I will most likely not agree to a contract extension if it is offered.

Thanks AL

From: Kylie Rika

Sent: Wednesday, 9 September 2020 1:39 PM

To: Sharon Johnstone ; Justin Howes

Allison Lloyd

Subject: RE: Combined stutter

Hi Sharon

Justin asked us to take the information from our separate teams discussions and send out to each group for further comment – which is what I have done.

RT2 has provided further comment on the points raised in RT1's document. The red text is RT2's feedback and it would not have been fair/appropriate/respectful to edit these thoughts or to not allow my team a voice. Therefore I

have sent my teams thoughts on unedited, they are not necessarily my views, they are the views of my team and it is not up to me to change that even if others find it confronting.

In my view, a next good step towards consistency, is a FRIT team meeting to discuss. In the absence of this, the only way to discuss is by back and forth in a document – which is fine but I am not going to dilute/alter the commentary – it is what it is, and all of us need to appreciate that robust discussions on science are healthy and necessary and should not be taken personally.

With regards to your point about reworks being used if there is any doubt in the interpretation, I have seen many instances where the debate on whether a peak is combined stutter or not is prior to reworks and/or final interpretations.

I will await Justin's advice/direction on next steps here and am also interested in Allison's thoughts.

Thanks Kylie

From: Sharon Johnstone

Sent: Wednesday, 9 September 2020 11:24 AM

To: Kylie Rika Allison Lloyd

Subject: RE: Combined stutter

Hi Kylie,

I have read the documents provided on this topic, however I have not shared this information with my team. The document that I shared with the FRIT management team was an effort to document common trains of thought on the topic in an effort to move forward. The dot points in the document were a combination of ALL of the individual responses that I had received in my team and therefore I believed that it was reasonable that many of your team members would also have common thoughts on many of the points listed.

The responses received to these dot points in red text appear to be in opposition with all of the common ideas in R1. I found this format confronting and I did not believe that the content nor the current format would assist in getting all case managers "on the same page". I was hoping that it would be clear which points were common thought across case managers and which points were not. This is not the case from the response as there appears to conflicting information under various points. I believe that the opinions I documented applied to final interpretations once reworking was complete as I believe it is well accepted that any doubt in interpretation is sort to be clarified through the use of reworks.

I would like for you to reconsider your team's response to this document and perhaps make it clear which points your team as a whole had a consistent thought with, and which points your team as a whole did not. This will then make it easier to determine the next steps towards consistency for both teams.

Regards, Sharon



Sharon Johnstone

Senior Scientist – Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream Health Support Queensland, Queensland Health Please note that I may be working from a different location during the COVID-19 pandemic. The best contact method is via email.

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From: Kylie Rika

Sent: Tuesday, 1 September 2020 9:24 AM

To: Justin Howes ; Sharon Johnstone

Allison Lloyd

Subject: RE: Combined stutter

Morning all

Reporting 2 have discussed the content in the document from Reporting 1.

Attached are two documents. The first is information that was discussed within RT2 on 16 July. The second contains responses from discussions with RT2 in relation to the discussion RT1 had.

RT2 is happy for both documents to be shared with RT1 if it will help in getting all case managers on the same page with combined stutter.

Thanks Kylie

From: Justin Howes

Sent: Monday, 24 August 2020 3:02 PM

To: Kylie Rika ; Sharon Johnstone up>; Allison

Lloyd

Subject: RE: Combined stutter

Hi

Yes, with outcomes from both Reporting Teams discussions now being established, the information from these team discussions can now go out to each group for further comment.

Thanks Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

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From: Kylie Rika

Sent: Monday, 24 August 2020 1:56 PM

To: Justin Howes Sharon Johnstone

Allison Lloyd

Subject: RE: Combined stutter

Hi all

In addition to sharing the outcome of RT2's discussion (see my earlier email), I have also been thinking on the points raised by RT1 and Sharon's email.

I agree with Sharon's email below and see that sharing of understanding between teams would be a good start to collaboration on the way forward.

I note that RT1 has approved for the document that contains their understanding to be shared. Without the two teams sharing info with each other, I can't see how an agreed way forward can be achieved.

Are we able to please re-consider sharing all the info about this topic to all case managers?

Thanks Kylie

From: Justin Howes

Sent: Friday, 21 August 2020 4:10 PM

To: Kylie Rika Sharon Johnstone >; Allison

Lloyd

Subject: RE: Combined stutter

Hi

I understand both Reporting teams have had the opportunity to discuss in meetings (or otherwise) cumulative/combined stutter, and have feedback. I had mentioned in communications that there is information that mentions combinations of stutter to be considered, and that staff do consider this effect (attached).

I haven't mentioned or provided direction on how it could be considered and applied. I think this is where we are at on this point now and this is for senior-level collation. Kylie are you able to please share the outcome of RT2 discussion on this? I understand you mentioned it was for discussion on Thurs 16 July but I am unable to find the minutes.

With all of this information, I think this can be thought of within the senior group for now.

Thanks Justin



Justin Howes

Team Leader - Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Forensic & Scientific Services, Health Support Queensland, Queensland Health

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From: Kylie Rika

Sent: Friday, 21 August 2020 2:15 PM

To: Sharon Johnstone >; Justin Howes

Allison Lloyd

Subject: RE: Combined stutter

Thanks Sharon

Does anyone mind if I send the document to Emma to look over given the content around STRmix and number of contributors guidelines?

Thanks

Kylie

From: Sharon Johnstone

Sent: Friday, 21 August 2020 1:23 PM

To: Kylie Rika ; Justin Howes ; Allison Lloyd

Subject: Combined stutter

Hi All,

The topic of combined stutter has been one that members of my team have identified to me as a source of a difference of opinion. In order to assist in the understanding of the problem, I have sort and gathered the opinions of all of my team individually. In doing so I was able to see the commonality within the team and identify nuances on the topic that would benefit from an agreed way forward. It must be noted that the possibility that stutter artefacts could combine is not in debate, rather the process by which we consider and evaluate a profile for such an event. Please read the attached document that has been developed and approved by my team to use as a sharing of understanding of my team and a basis for which improvements can be made and further considered.

I see this as a starting point for further collaboration towards a clearer way forward for all reporters.

Regards, Sharon



Sharon Johnstone

Senior Scientist – Forensic Reporting and Intelligence Team

Forensic DNA Analysis, Police Services Stream

Health Support Queensland, Queensland Health

Please note that I may be working from a different location during the COVID-19 pandemic. The best contact method is via email.



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JH-73

All other labs are modelling post stutter in STRmix and therefore don't have this issue.

STRmix has always modelled cumulative peaks. With the first version of STRmix this was only peaks consisting of allele and stutter, however they were modelled as a purely additive effect. This is the model that we are currently using.

The most recent versions of STRmix model all stutter types and it still considers them as additive.

From page 40 of the user's manual:

Where T_9^1 is the total allelic product generated from the mass parameters and genotype set. The proportion of allelic component in E_9^1 is:

$$\mathbf{A}_{9}^{1} = \frac{E_{9A}^{1}}{SR_{10}^{1}O_{10}^{1} + E_{9A}^{1} + FSR_{8}^{1}O_{8}^{1}}$$

And the proportion of stutter component is:

$$S_{9}^{1} = \frac{SR_{10}^{1}O_{10}^{1}}{SR_{10}^{1}O_{10}^{1} + E_{9A}^{1} + FSR_{8}^{1}O_{8}^{1}}$$

And the forward stutter component is:

$$\mathbf{F}_{9}^{1} = \frac{FSR_{8}^{1}O_{8}^{1}}{SR_{10}^{1}O_{10}^{1} + E_{9,4}^{1} + FSR_{8}^{1}O_{8}^{1}}$$

So that $A_9^1 + S_9^1 + F_9^1 = 1$

 $p(O_9^1 \mid E_9^1)$ is then modelled by $N(0, \frac{S_9^1 k_{a-1}^2 + \frac{A_9^1 c^2}{O_{10}^1} + \frac{F_9^1 k_{a+1}^2}{E_9^1} + \frac{F_9^1 k_{a+1}^2}{FSR_8^1 O_8^1})$. Mathematically for locus 1:

The model that STRmix uses states that the variance in stutter peaks has a direct relationship to the height of the allelic peak, it doesn't suggest that the stutter peak itself is subject to stochastic effects. From page 31 of the user's manual:

In words, this model states that as the expected peak height (or the parent peak height for a stutter) decreases, then the peak height variability increases. This relationship should be intuitive for most forensic scientists. However, there are sensible limits to this relationship,

As we move forward to more continuous modelling, including the modelling of all stutter types, as this is the model (that peak heights are cumulative) that we will be using.

I have checked the SOPs and have found the following:

QIS 17117 doesn't mention anything about combining stutters

QIS 33773 Section 23.5 states:

"A combination of stutter thresholds can be considered when determining the number of contributors for loci where +1 / -1 / -2 repeat stutter peaks of different alleles overlap."

This isn't very detailed, but does make the point.

The number of contributors guidelines (in the new Basics of Interpretation SOP) does talk about the removal and combining of stutters, but I feel that the information is quite outdated now that we are using new and improved stutter models with v2.7 (the guidelines were written when we were using v2.0.6). In its current form it is quite confusing.

Further to this, the journal article *The interpretation of single source and mixed DNA profiles (Taylor et al 2013)* states:

Expected peak heights are assumed to be additive when there are multiple contributions to a peak, whether from multiple alleles or a combination of alleles and stutter.

Empirical data suggests that the variance in a stutter peak in a model based on LUS follows a different pattern to an allelic peak. In general, stutter peaks show less relative variance than allelic peaks.

Thanks

Emma

JH-74

The potential for a peak to possibly be affected by the combination of multiple sources of stutter was recognized by everyone. It was also agreed widely that the consideration of combined stutter as the sole source of a peak was rarely applied for interpretation in the team. Where use was supported the following conditions were common in the group:

- The whole profile needs to be taken into consideration

 Stutter at one locus within a profile does not influence the presence of stutter at another locus as all loci behave differently. From Project #170 776 +1 rpt stutter peaks were observed at D6, whereas only 45 were observed at D2 and only 8 observed at Th01. For -2 rpt stutter, 327 peaks were observed at D1 but no instances of -2rpt stutter were observed at PE and only 1 at each of PD, Th01 and TPOX. We also know that stutters within a locus can behave differently depending on the size of the parent allele. Therefore it is not possible for the profile as a whole to provide information about what might be happening at a particular locus with respect to stutter
- Also, The possibility of combining -1 rpt stutter and +1 rpt stutter thresholds is generally supported but there was no support to combine -2rpt stutter thresholds with other stutter thresholds unless modelled by STRmix
 Evidence to indicate why -2rpt stutter would be different would be good, particularly since +1 rpt stutter isn't currently modelled by STRmix either. SOP 33773 suggests that combinations including -2 stutter repeats can be considered when determining number of contributors. However, given our current version of STRmix, care needs to be taken when considering whether a peak should remain labelled (STRmix may be forced to consider a peak as an allele 100% of the time, when this peak may be alternatively considered as combined stutter).
- The presence of stutter elsewhere in the profile increased the support for combined stutter
 thresholds to be considered
 See first point and whilst the presence of stutter elsewhere in the profile may add
 confidence, it should not negate or exclude the possibility for stutter to be considered for a
 particular locus being considered. An isolated incidence (eg in a mixture) may act as a flag
 for consideration of rework however.
- The use of combined stutter thresholds was supported with regard to single source profiles
 but extreme hesitation was expressed with the use in mixed DNA profile interpretation
 Information/explanation as to why a different approach would be used for single source
 profiles and mixtures would be good. Whilst additional care needs to be taken with respect
 to mixed DNA profiles, (and additional runs would be recommended), keeping an artefact on
 in a mixture means that you can falsely exclude someone on it.
- High peak heights of the parent peaks are more likely to increase the presence of stutter throughout a profile
 Yes, because all stutter products are proportional to the height of the parent peak, however the <u>variability</u> in the heights of these stutters is different. The height of the parent pk is important to assess whether it's reasonable that it is high enough to be producing +1 and -2 stutter. The heights of the parent peaks can come into play when making decisions and it is very much a profile by profile sort of thing.

- Combined stutter could be considered as an explanation for a peak to slightly exceed the limit of a single threshold, however any peak nearing the limit of the combined threshold was generally not accepted to be the most reasonable explanation for that peak If they didn't occur in the same position but were separated they would both be considered to be stutter – so information/explanation as to why this would be different if they are in the same position would be good. Also, smaller peak heights induce greater variability and therefore it is quite likely that higher than expected stutter would be seen for all stutter products. This also depends on a number of factors, including the type of combination stutter being observed, and the allowance given for each of the individual stutter types with respect to the peak observed, as well as the peak height of the parent peaks and consideration of the profile as a whole. Whilst it is reasonable to combine n-1 and n+1 stutters (particularly when the combined stutter falls well below the combined threshold), a general rule shouldn't be applied where combined stutter would only be accepted if the peak is only slightly over the single stutter threshold and reject it if the peak approaches the whole amount of combined stutter. Depending on the profile, this may add to consideration for rework etc, but not necessarily an outright blanket rejection.
- Overall, the concept of using combined stutter to remove a peak from interpretation was to be used not as a blanket rule but with extreme caution

 In what circumstances would it be used? There are times when we assess pullup and stutter combining together and choose to call it high stutter due to pull up rather than an allele, is there much difference to that and 2 stutter pks, it's essentially considering two artefacts in combination. The use of combined stutter does not necessarily remove a peak from the profile or interpretation. Its use is in considering recognized amplification issues when assessing likely number of contributors. As with other considerations in interpreting DNA profiles, the use depends largely on the information present within the profile, (including whether there are other indications of an additional contributor), the quality of the profile and sometimes the ability to rework. In situations where the peak/locus in question may be the only indication of an additional contributor, one may wish to consider multiple runs to be especially useful in determining the application of combined stutter and ultimately the likely number of contributors.

Hesitations in using combined stutter as a reasonable explanation for a peak generally came down to the lack of a documented process with supportive guidelines on how to use this approach, in combination with the lack of in-house testing. Influencing factors also include:

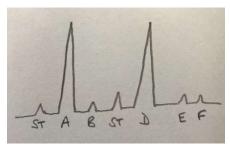
• Our laboratory has not verified nor implemented the ability for STRmix to model combined stutter
STRmix v2.7.0 has been validated for use with PP21 and the 3500 for modelling both -1 and +1 rpt stutter. The reasoning of adding stutter pks to each other is not different to adding stutter pks to allelic pks in the biological model. Current and future versions of STRmix do consider peak heights as cumulative. Additionally, depending on the combination of stutter observed and how we have determined number of contributors, we have the ability to check the considerations of STRmix from a decon as to whether it has considered a peak to be an allele or stutter and whether this fits with any assumptions we have made regarding stutter thresholds.

 -1rpt stutter threshold are allele specific and -2 rpt stutter and +1 rpt stutter thresholds are locus specific

That is due to a number of factors, but has no relevance in this instance. It is acknowledged in the STRmix Users' Manual that locus specific thresholds will most likely be the best approach for a laboratory for these stutter types.

The number of contributors work recognizing that it is better to nominate n+1 contributors
than it is to nominate n-1 contributors for the interpretation of a mixed DNA profile due to
the potential to falsely exclude or to load incorrectly to NCIDD.
 Under the right conditions, any artefact left labelled on the epg (stutter peaks - combined or
otherwise mutations pullsup) that is considered to be an allele will cause false exclusions.

otherwise, mutations, pull-up) that is considered to be an allele will cause false exclusions and incorrect loads to NCIDD this is why any artefacts that are not modelled by STRmix should be removed.



Consider the locus above from a 3p mix. Peak B is over the -2 rpt threshold for peak D and over the the +1 rpt threshold for peak A but below the combined thresholds. If the peak is left labelled STRmix will consider it to be an allele all of the time. If my reference sample is D,E they will be excluded. Also, if E and F are slightly higher then B may well be resolved for upload to NCIDD. So if B is a true artefact we will have a false exclusion and a potential false upload to NCIDD. Stutter peaks in -1 position are not generally utilized in considering an exclusion of a reference sample. Peaks in other stutter positions may be used in excluding a reference sample and/or included in a profile loaded to NCIDD, hence it is particularly important to be confident the peak is a genuine allele and not a result of combined stutter, otherwise mis-matches on NCIDD and/or false exclusions may occur with the nominated number of contributors.

A number of people have suggested further work that could be conducted to help characterize the effect of combined stutter. These suggestions will be considered. However, this work may or may not be performed and therefore a way forward may have to be sort without further testing. The main issues seem to be around when the label of a peak is removed; when a peak in combined stutter position may not be intuitive when comparing a reference sample, or included in an NCIDD load with the nominated number of contributors. For a certain subset of combined stutter profiles, one possible path could be to consider the combined stutter peak as a possible indication of an additional contributor, but not necessarily utilizing it when comparing a reference fit against the number of nominated contributors, particularly in instances where STRmix has accepted it as stutter some of the time. That is, to not further elevate the nominated number of contributors where a discrepancy lies only with a peak in combined stutter position. Worthy to note however is that while this might be OK if the peak is in a -1rpt stutter position or below LOR, caution needs to be applied if the peak is in a -2 or +1 rpt stutter position and labelled because STRmix will consider it to be an allele all of the time and will include it in the LR calculation. If we have different considerations or impacts for the different types of combination stutter, we may benefit from elaborating on the guidelines for combined stutter types in some format.

A CLINICAL AND STATEWIDE SERVICE

The Examination for and of Spermatozoa

1 PURPOSE AND SCOPE

This method describes the microscopic examination of smears for the presence of spermatozoa. It includes the examination of items contained within the Sexual Assault Investigation Kits (SAIKS) that are assembled by either QHSS or other external companies such as Medi-Redi (owned by House with No Steps).

This SOP also includes workflow diagrams pertaining to examination of items in alleged sexual assault cases. These diagrams show the steps necessary in these examinations which include AP and PSA screening, along with microscopic examination for spermatozoa.

2 ACTIONS

2.1 Interpretation

- 1 The basophile haematoxylin stains the deoxyribonucleic-acid (DNA)/histone rich base of the sperm head deep purplish-blue. The acidophile eosin stains the acrosomal cap pink and, in intact-spermatozoa, also stains the tail pink.
- 2 The use of counterstaining differentiates spermatozoa from most cell debris and can assist in the differentiation of human spermatozoa from common animal spermatozoa.
- 3 Confusion with yeasts, especially monilia, can occur and extreme care must be taken when monilial infections such as thrush are suspected. With experience, spermatozoa and yeasts can be distinguished by size and/or the presence of cell walls.

2.2 Slide Preparation (for AP positive stains and Sexual Assault kits with no slides)

- 1 Use new slides and clean with ethanol. Label with the sample ID, date, case number and sampler's initials using a pencil only.
- 2 Use clean, flamed instruments.
- 3 Create a suspension from the exhibit by one of the following methods,
 - Scrape the stained area into a 1.5ml eppendorf tube. Add drops of distilled water to the tube until the scraping is covered. Vortex thoroughly.
 - II. Excise the stained area and cut into small pieces. Place pieces into a 1.5ml eppendorf tube and add drops of distilled water to the tube until the pieces are covered. Vortex thoroughly.
 - III. If slide is being prepared from a swab, excise the cotton from the swab and cut the cotton into small pieces. Place the pieces of cotton into a 1.5ml eppendorf tube and add drops of distilled water to the tube until the pieces are covered (approx 150-300µl). Vortex thoroughly.
- 4 Add a drop of the recently vortexed suspension to the labelled slide.



5 Dry the slide on a heat block. If a heat block is not available, heat-fix the slide by passing it over a flame with the material to be stained uppermost.

2.3 Slide Stainer

The slide can be stained in the automatic slide stainer in Histology. If this is not available, see manual staining procedure in Appendix 1.

2.4 Microscopic Examination

Examine slide using the x40 or x100 (oil immersion) objective. Score the number of spermatozoa observed (use the standard microscopy form, <u>17037</u> or the Sexual Assault Investigation Kit form, <u>17032</u>).

0	(0)	None seen
<+	(<1+)	Very hard to find (Use vernier)
+	(1+)	Hard to find
++	(2+)	Easy to find
+++	(3+)	Very easy to find
++++	(4+)	Abundant

- Note whether spermatozoa are intact (heads and tails) or non-intact (heads only). Look for epithelial cells and whether there are bacteria or yeast present. Human spermatozoa are distinguished from non-human mammalian sources by their morphology and by their behaviour toward HE, resulting in a purple base and clear cap (see Section 2.5).
- 3 If limited sperm are located, not the location on the slide as per the current laboratory procedure and/or take photographs.

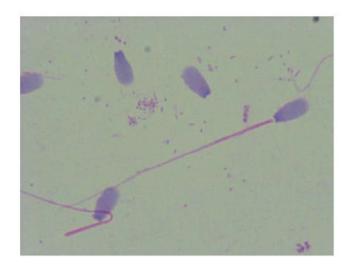
2.5 Animal Semen

- 1 Identification or differentiation of animal spermatozoa is an extremely unusual request, however its possible presence should always be considered. Microscopy and our molecular biology protocols will differentiate human from animal spermatozoa.
- 2 Special stains are available to assist in the differentiation/identification of human/animal spermatozoa. However H/E and routine molecular biology techniques will resolve the issue, and if non-human reactions are obtained there will be support for an opinion.

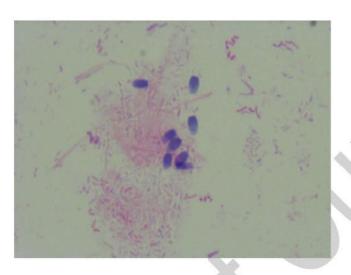
2.6 Animal Spermatozoa Images

- 1 The images of animal spermatozoa given below are observations from a limited number of samples. It must be borne in mind that there may be variations in shape and size according to age, breed, or individuality of animal concerned.
- 2 The following images are meant to assist in the formation of an opinion. Do not attempt to diagnose an animal spermatozoa species and do not rely on immune antisera. If animal non-human reactions (i.e. negative reactions) are given at DNA quantification stage, a typical opinion could be, "spermatozoa present did not appear to be of human origin. No human DNA was detected".

The Examination for and of Spermatozoa



Bull Sperm at x1000 magnification. (Oil immersion) Sperm head lengths ~6-7μm.

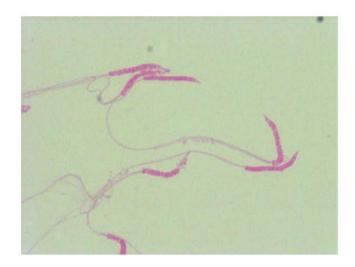


Cat Sperm at x1000 magnification. (Oil immersion) Sperm head lengths ~3-3.5µm.

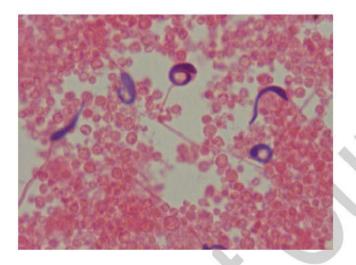


Human Sperm at x1000 magnification. (Oil immersion) Sperm head lengths ~3-4µm.

The Examination for and of Spermatozoa



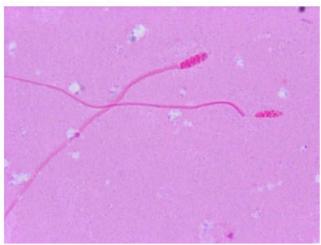
Kangaroo Sperm at x1000 magnification. (Oil immersion) Sperm head lengths ~9-11μm.



Koala Sperm (uncurled) at x1000 magnification. (Oil immersion) Sperm head lengths ~8-9µm.



Pig Sperm at x1000 magnification. (Oil immersion) Sperm head lengths ~6-6.5μm.



Possum Sperm at x1000 magnification. (Oil immersion) Sperm head lengths ~4-5µm.

Photos from DNA Analysis.

2.7 Spermatozoa Interpretation

If slides are stained properly spermatozoa should be easily distinguished from epithelial cells, cellular debris, fibres etc. Spermatozoa heads can look similar in shape and colour to yeasts. If in any doubt consult an experienced examiner.

The recovery of semen is dependent on a number of factors but not limited to

- The amount of spermatozoa in the ejaculate
- II. The amount of ejaculate
- III. The environment the ejaculate is deposited on
- IV. Washing
- V. Douching
- VI. Menstruation
- VII. Efficiency of the sampling process
- VIII. Time between ejaculation and sampling
- IX. Storage of the samples
- X. Natural drainage or degradation of spermatozoa in certain environments

With respect to the above influences, the time since ejaculation has occurred can only be estimated. A number of studies have been conducted regarding the persistence of spermatozoa in the vagina. References to these studies can be found in Appendix 6.2.

2.8 Penile Swabs

Submit for DNA testing even if the semen on the penile swabs is of no evidentiary value. Unprotected sexual intercourse with no ejaculation may leave foreign cells (vaginal, anal, oral) on the penis. Depending on the time of swab collection and the effectiveness of the swab, female DNA on the penile swab may be successfully profiled from an unwashed penis.

Penile swabs may be submitted for Differential Lysis if the case history is such that the complainant had intercourse with another male prior to the alleged offence. Transfer of semen from the previous partner to the alleged offender may occur and DNA may be obtained in the sperm fraction.

2.9 Examination of Sexual Assault Swabs

1 If serum coated, charcoal swabs or other unsuitable swabs/media are submitted, the client must be notified. These swabs should still be examined. The serum coated or transport media swabs can be submitted for analysis. Add 'Manual DNA IQ' as the processing comment and email the Analytical Senior Scientist with the barcode numbers. If the supernatant is to be retained the sample will be processed by off-deck lysis and Manual DNA IQ. Register as per normal and email the Analytical Senior Scientist with the numbers. No processing comment is required.

- 2 If a smear has not been received, one will need to be made. See Section 2.2. Stain the smears with the H&E stainer in Histology. Examine the smears for spermatozoa noting the presence or absence of intact sperm, sperm heads, epithelial cells, white blood cells, bacteria and any other cells seen.
- 3 If no spermatozoa are found, perform AP tests on suspensions made from the swabs.
- 4 If a smear is negative for spermatozoa and positive for AP it is necessary to perform a PSA test to characterize the biological material as semen.
- 5 If smears are received with paper labels attached, photograph the slide with the label, remove the label and relabel with the Statmark Pen or a diamond pencil and rephotograph the slide with the new labels. See 20080 Photography of Exhibits in DNA Analysis.
- 6 Submit swabs in separate sterile 1.5mL tubes for differential lysis extraction if not already in a tube as part of the slide preparation.

2.10 Examination of Items Previously Screened by QPS

QPS Scientific Officers will sometimes perform AP tests on items. Confirmatory testing should be performed in this case, and re-AP testing or feedback on screening results may be required. (See flow diagram in Appendix 6.3)

3 REFERENCES

- 1 Biology Methods Manual, Metropolitan Police Forensic Science Laboratory, Great Britain, 1978.
- 2 Allard, J.E (1997). "The collection of data from findings in cases of sexual assault and the significance of spermatozoa on vaginal, anal and oral swabs." Science and Justice V37(2): April; 99-108.
- 3 Allery, J.P., Telmon, N., Mieuset, R., Blanc, A., Rouge, D. (2001). "Cytological Detection of Spermatozoa: Comparison of Three Staining Methods." *Journal of Forensic Sciences* V46(2): 349-351.
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- 6 Khaldi, N., Miras, A., Botti, K., Benali, L., Gromb, S. (2004) "Evaluation of Three Rapid Detection Methods for the Forensic Identification of Seminal Fluid in Rape Cases." *Journal of Forensic Sciences* July; 49(4):749-753.

- 6 Maher, J., Vintiner, S., Elliot, D., Melia, L. (2002) "Evaluation of the BioSign PSA Membrane Test for the Identification of Semen Stains in Forensic Casework." The New Zealand Medical Journal Feb 8:115(1147):48-49.
- 8 Montagna, C.P. (1996). "The recovery of seminal components and DNA from the vagina of a homicide victim 34 days postmortem." *Journal of Forensic Sciences* July 41(4): 700-702.
- 9 Randall, B. (1987). "Persistence of vaginal spermatozoa as assessed by routine cervicovaginal (Pap) smears." Journal of Forensic Sciences May 32(3): 678-683.
- 10 Ricci, L. R., Hoffman, S.A., (1982). "Prostatic acid phosphatase and sperm in the post-coital vagina." Annals of Emergency Medicine 11(10): 530-534.
- 11 Silverman, E. M., Silverman, A.G. (1978). "Persistence of spermatozoa in the lower genital tracts of women." *JAMA: The Journal of the American Medical Association* 240(17): 1875-1877.
- 12 Willott, G.M. and Allard, J.E. (1982). "Spermatozoa their persistence after sexual intercourse." Forensic Science International 19(2): 135-154.

4 ASSOCIATED DOCUMENTS

QIS: 20080 - Photography of Exhibits in DNA Analysis

QIS: 17185 - Detection of Aspermic Semen in case Work Samples using the Biosign

PSA11-WB Rapid Test for Prostate Specific Antigen

QIS: 17186 - The Acid Phosphatase Screening Test for Seminal Stains

QIS: 17037 - Microscopy of Smears Form

5 AMENDMENT HISTORY

Version	Date Issue	Author/s	Comment
1	Unknown	Unknown	Unknown
2	Unknown	Unknown	Unknown
3	Unknown	Unknown	Unknown
4	27 Nov 2002	V lentile	Format updated, manual staining to appendix. Removed notes on examination of swabs, removed unpublished paper, as work wasn't completed.
5	19 Nov 2003	L Freney	Updated references
6	12 Jul 2006	J Howes/A Williamson	"Reference" put after "Actions".
7	05 Aug 2006	J Howes	Added in Sexual Assault Investigation Flowcharts, examination of SAIK Swabs, Photograph or Witness required for ++ (1+) sperm and PSA test.
8	23 Oct 2006	J Howes	Reporting results Eg. ++ or 2+
9	25 Jun 2007	J Howes	Unified grading scale comments. Added Crimelite flowchart.
9	13 Mar 2008	QIS2 Migration	Headers and Footers changed to new CaSS format. Amended Business references from

The Examination for and of Spermatozoa

	0.0	Project	QHSS to FSS, QHPSS to CaSS and QHPS to Pathology Queensland
10	16 July 2010	A Lloyd	Removal of Crimelite in scope and the Crimelite flowchart. Changed section 2.2 to include use of suspensions. Removal of section 2.8 – Vaginal Secretions. Changes to section 2.10 to remove AP testing on smears positive to spermatozoa. Photograph or locations required for smear with 1 or 2 sperm seen. Clarification of flowchart regarding previously screened items by QPS. Changes to SAIK flowchart. Removal of animal sperm diagrams and insertion of photographs of animal sperm.

6 APPENDICES

- 1 Preparation of H & E stain and manual staining procedure
- 2 How long do spermatozoa remain in the vagina?
- 3 Workflow Charts

6.1 Preparation of H & E Stain

CHEMICALS

Absolute alcohols

WARNING: Ethanol liquid and vapour are combustible. May irritate eyes and skin. Health effects well known – substance of abuse.

Eosin (yellowish)

WARNING: Eosin (yellowish) can cause serious damage to the eyes. Avoid contact, wear PPE.

Haematoxylin

WARNING: Haematoxylin: the toxicological properties have not been investigated. Prevent contact with skin and eyes. Do not inhale or ingest. Wear PPE.

Sodium iodate

WARNING: Sodium iodate causes burns and is harmful if inhaled or swallowed. Protect eyes and skin.

Chloral hydrate (SLR)

WARNING: Chloral hydrate causes burns and is harmful if inhaled or swallowed. Protect eyes and skin.

Citric acid

WARNING: May cause skin irritation. Inhalation may cause irritation to mucus membranes.

Avoid skin contact

Acetic acid

WARNING: Acetic acid is extremely corrosive and is harmful if inhaled or swallowed. Protect eyes and skin.

Hydrochloric acid

WARNING: Hydrochloric acid causes burns and is harmful if inhaled or swallowed. Protect eyes and skin.

NOTE: All the above chemicals are available from the Histology Section

PREPARATION OF REAGENTS

NOTE: All reagents prepared in the laboratory shall bear a label:

....(enter details eg 10% NaOH)....
Prepd from Lot/batch:....
Date: ../../. Initials:
Expires:../../. Store at:....°C
WARNING: Contains

Mayer's Haematoxylin

Preparation of solution:

Haematoxylin 1g
Distilled water 1000mL
Potassium or ammonium alum 50g
Sodium iodate 0.2g
Citric Acid 1g
Chloral hydrate SLR 50g

The haematoxylin, potassium alum and sodium iodate are dissolved the distilled water by warming and stirring or by allowing to stand overnight. The chloral hydrate and citric acid are added and mixture is boiled for 5 minutes then cooled and filtered.

This mixture is then ready for use and has a shelf life for over one year at room temperature.

Eosin

Eosin Y (eosin yellowish, eosin water soluble) C. I. No 45380 (C. I. Acid Red 87)

Use as a 1% solution in distilled water. Add 0.5mL of acetic acid to 1000mL of eosin Y

- NOTE: This method provides a useful stain for spermatozoa and vaginal and buccal epithelium. It results in a dark purple nucleus with pink cytoplasm.
- Place slide on staining rack over sink, flood with haematoxylin and leave for 5-10 minutes.
- Wash well in running tap water until smear "blues" (5 minutes or less)
- Differentiate in 1% acid alcohol (1% HCl in 70% alcohol) for 5-10 seconds.
- Wash well in tap water until smears are "blue" again (5 minutes or less).
- Counterstain in 1% eosin for 10 minutes.
- Wash in running tap water for 1-5 minutes.
- Allow slide to dry on hot plate or on filter paper on the bench
- Mount in depex if desired.



6.2 How long do Spermatozoa remain in the vagina?

6.2.1 Other references mentioning persistence of spermatozoa in the vagina -

- O.J. Pollack. 1963 Arch. Pathology 35 p140-184
 non-motile sperm 3-24 hrs.
- 2 Noble Sharp 1963 Canada. Med. Ass. J. 89
 - non-motile sperm 7-12 hrs, exceptionally 18-24 hrs, unique case 3-4 days.
- Gordon, Turner and Price 1965 Medical Jurisprudence
 3-4 days
- Morrison 1972 Brit. J. Vener. Dis 48 p141
 up to 9 days or 12 days in the cervix, sometimes after menstruation.

6.2.2 How Long Do Spermatozoa Remain Alive (Motile) In the Vagina?

The period for which spermatozoa may remain alive after deposition in the vagina may best be reviewed by quoting from the following workers in this field.

O.J. Pollak: 'The number of motile spermatozoa discernible in the vagina may be normal after one hour and markedly decreased after 2 hours; after 3 hours normally no spermatozoa are found.

Menstruation often prolongs motility in the vagina to as long as 4 hours compared with the normal period of 30 to 45 minutes.

Concerning human spermatozoa Weisman in his book "Spermatozoa and Sterility" (1941) summarises the periods of motility as follows:

Vagina...2 to 3 hours. Cervix...48 to 110 hours.

6.2.3 Semen and Seminal Stains, Arch. Path., 1943, 35, 140.

Samuel L. Siegler: 'Normally 10% of the spermatozoa are alive in the vagina at the end of 2 hours post coitum. Variations in number and motility depend upon the pH of the vagina and semen, quantity of semen deposited, bacteria and flora of the vagina and the time examined post-coitally. The author has seen motile spermatozoa in the vaginal pool after 8 hours. "Fertility in Women" (Wm.Heinmann Medical Books Ltd., 1945).

Lane-Roberts, Sharman, Walker, Wiesner and Barton: 'In most cases all intra-vaginal spermatozoa cease to move irreversibly within a few hours of coitus (Seguy and Vimeux, 1933; Hartman, 1932). Huhner (1928, 1937), who has paid much attention to the problems involved, regards 30 minutes to 3 hours as a common measure of survival in the human subject, reduction in this period signifying an abnormal condition involving lowered fertility. Our own observations support the view that spermatozoa survive in the vagina for only a few hours and that even in fecund couples the variations are considerable.

In several cases in which repeated examinations were possible before conception occurred, all motility ceased within one hour after intercourse. A fall of motility to 10% within 30 minutes is compatible with fecundity. On the other hand, spermatozoa may continue to move for 3 hours in a normal untreated vagina.' (Sterility and Impaired Fertility, Hamish Hamilton Medical Books, 1948).

Sidney Smith: 'The evidence points to a comparatively short life of the spermatozoa in the female tract and the period appears to get shorter with the number of observations. It is at present believed that the life of the spermatozoa in the vagina is a matter of hours.' (Forensic Medicine, 1955).

Gonzales, Vance, Helpern and Umberger: 'The motility of the spermatozoa in the specimen may give a clue to their length of stay as they remain motile from 30 to 60 minutes after deposition in the vagina.' (Legal Medicine, 1954).

Louis Portnoy and Jules Saltman: 'As for motility, the sperms are mostly found non-motile or dead. This is to be expected because, after a lapse of one to three or more hours, all the sperms will normally have been killed by the acidity of the vaginal secretions. There may be live sperms but more likely not.' (Fertility in Marriage, Signet Book, 1951).

6.2.4 How Long Do Spermatozoa (Non-Motile) Remain In the Vagina

Pollak in his comprehensive paper "Semen and Seminal Stains", states:

"Although the various authors give the period of their presence in the vagina as from 30 minutes to 17 days, one may safely consider the period for non-motile spermatozoa in the vagina after coitus to be 30 minutes to 24 hours."

Gonzales and others in their book "Legal Medicine" state:

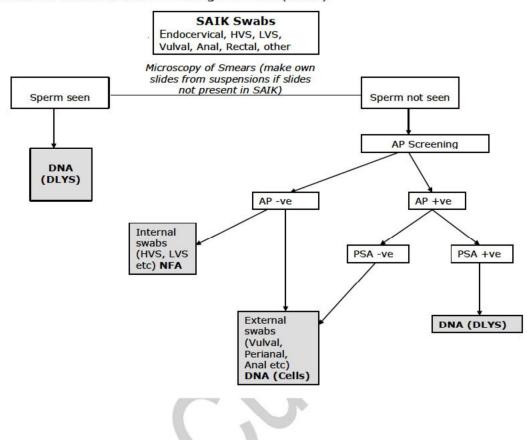
"Some have claimed that spermatozoa may be demonstrated in the vaginal contents of non-fatal cases from 45 minutes to several days after the last coitus, but these considerations do not offer a satisfactory basis for estimating the duration of their residence in the vagina in as much as the time of the last coitus cannot be determined with precision. If they are non-motile, it might be difficult to determine the length of time they have been in the female tract."

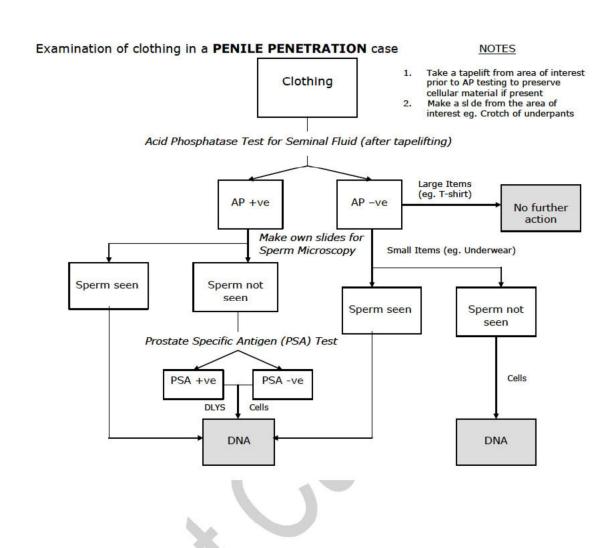
Gordon, Turner and Price in their book "Medical Jurisprudence" make this comment:

"The finding of spermatozoa on examination of a vaginal smear is indicative of an ejaculation into the vagina but it affords no evidence of the time of the ejaculation. In charges of rape, therefore, particularly in the case of married women, it becomes necessary to exclude the possibility of sexual intercourse having taken place before the assault. In this connection it should be noted that spermatozoa could be recovered from the vagina 3 to 4 days after their introduction. Some authorities claim that they may be recovered after a lapse of even longer periods."

6.3 Workflow Charts

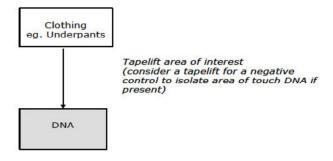
Examination of Sexual Assault Investigation Kits (SAIKs)



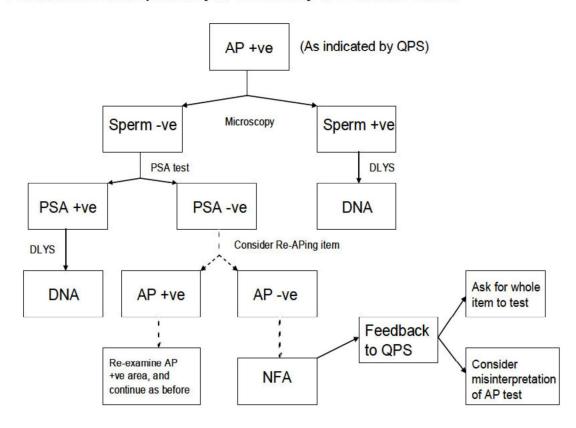


NOTES Examination of a DIGITAL and PENILE PENETRATION case Take a tapelift from area of interest (ie. Crotch) prior to AP testing to preserve cellular material if present Clothing eg Underpants Make a slide from the area of interest (AP +ve area or likely area eg. Crotch of underpants) Acid Phosphatase Testing (after tapelifting) AP +ve AP -ve Make own slides for Sperm Microscopy Sperm seen Sperm not Sperm seen Sperm not seen seen Prostate Specific Antigen (PSA) Test Tapelift for trace DNA (cells) DLYS PSA +ve PSA -ve Tapelift for trace DNA (cells) DLYS DNA

Examination of clothing in a DIGITAL PENETRATION ONLY Case



Examination of Items previously AP screened by QPS Scientific Officers



JH-76

Erin Shearer

From: Cathie Allen

Sent: Monday, 6 March 2017 1:17 PM

To: Justin Howes **Subject:** Re: Next week

So sorry to hear your sad news. Of course TOIL is fine for that day. Take care Cathie

Sent from my iPhone

- > On 6 Mar 2017, at 6:37 am, Justin Howes <Justin.Howes@health.qld.gov.au> wrote:
- > Hi Cathie, no problems and can understand. You shouldn't be really emailing on day off anyway....
- > Paul didn't send email and fair call. I spoke to whoever was here at 4pm on Fri from R1 and there were two R2 people left, so included them. No dramas. See what happens.
- > I am ok thanks but something terrible happened personally which I have sent a TOIL request for this Thurs to you on (funeral). A good friend committed suicide last Sun we found out as Christy was going to work last Mon and me to Melb so not great to leave at a terrible time, but lucky friend's wife is surrounded by good, helpful people. Two boys similar ages to ours tough week last week.
- > Anyway, see you tomorrow.
- > > jah >
- >

> > p |

- > Justin Howes> Team Leader Forensic Reporting and Intelligence Team
- > Forensic DNA Analysis, Forensic & Scientific Services, Health Support
- > Queensland, Department of Health



> HSQ's vision | Delivering the best health support services and solutions for a safer and healthier Queensland.

> Queensland Health acknowledges the Traditional Owners of the land, and pays respect to Elders past, present and future.

> -----Original Message-----

> From: Cathie Allen [mailto:cja.07@hotmail.com]

> Sent: Friday, 3 March 2017 8:21 PM

> To: Justin Howes

> Subject: Re: Next week

>

>

> Hi Justin

>

```
> Sorry for the late message, I think Paul is reluctant to have comms to entire team, based on HR
advice but will be interesting to see what he's said.
> Hope you're ok.
> Cheers
> Cathie
>
> Sent from my iPhone
>> On 3 Mar 2017, at 8:51 am, Justin Howes <Justin.Howes@health.qld.gov.au> wrote:
>> Hi, yes got the news late yesterday. I just delivered to Mgt Team, and was asked to give JMW
heads up (both done). On from Mgt is to prevent mixed messages and Chinese whispers, to have a
comms from Paul in similar way to your emails about returning staff. I think best to have a msg from
Paul to the whole team to set a baseline for everything. I am going to pop over and see if he can do
this, but yesterday the msg to me was to speak to R1 and Mgt.
>>
>> Any thoughts for now??
>>
>> jah
>>
>>
>> Justin Howes
>> Team Leader - Forensic Reporting and Intelligence Team
>>
>> Forensic DNA Analysis, Forensic & Scientific Services, Health Support
>> Queensland, Department of Health
>>
>>
>> HSQ's vision | Delivering the best health support services and solutions for a safer and healthier
Queensland.
>>
>> Queensland Health acknowledges the Traditional Owners of the land, and pays respect to Elders
past, present and future.
>>
>>
>> -----Original Message-----
>> From: Cathie Allen [mailto:cja.07@hotmail.com]
>> Sent: Friday, 3 March 2017 10:44 AM
>> To: Justin Howes
>> Subject: Next week
>>
>> Hi Justin
>>
>> Paul has been in contact with me regarding Amanda's return next week. I understand that HR
have provided you with some advice to verbally provide to the team. Did they also say that we could
tell the management team? So that if other staff have a question, the line managers can all say the
same thing?
>>
>> Hope you're ok, see you on Tuesday,
>> Cathie
>>
>> Sent from my iPhone
```

- >> *
- >> ****** This email, including any attachments sent with it, is
- >> confidential and for the sole use of the intended recipient(s). This confidentiality is not waived or lost, if you receive it and you are not the intended recipient(s), or if it is transmitted/received in error.
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- >> Unless stated otherwise, this email represents only the views of the sender and not the views of the Queensland Government.
- >> *********
- >>

JH-77

Erin Shearer

From: Allan McNevin

Sent: Thursday, 12 May 2016 12:39 PM

To: Justin Howes

Subject: RE: Diff lysis slide investigation

thanks

From: Justin Howes

Sent: Thursday, 12 May 2016 11:35 AM

To: Allan McNevin **Cc:** Kirsten Scott

Subject: FW: Diff lysis slide investigation

Hey Al

Some reporters were discussing ideas re ER and DLYS slides and Amanda came across them and asked them to write them down so that they could potentially be considered in any reviews/projects that might develop.

Below are the ideas explored by a few reporting staff worth considering.

JAH

- -Identifying a staining/fixing issue:
 - -Suggest collecting run off from slide washing, centrifuging and making a slide to stain and observe possible presence of cellular material that has washed off
 - -Is liquid added to the slide spread out to increase surface area to facilitate drying? Or just left as a drop?
 - -Suggest making sure that slides are fixed properly on the hot block prior to staining. An experiment to identify how long is sufficient should be conducted. The SOP does not clarify how long slides are left, what temperature the hot block is on, or how much sample is added these variables should be investigated for best outcome and then fixed.
- -Suggestion of agitating swab and water with pipette prior to vortexing and waving slide through flame prior to staining.
 - -Suggest having a parallel duplicated study using methanol as a slide fixative before the addition of stain.
- -Datamining (which may or may not include the examination of diff slides not done at the time) of past samples including:
 - -Result of diff slide from micro neg/PSA pos samples
 - -Result of diff slide from micro pos/low sperm count samples
 - -Result of diff slide from high quant/ low micro samples
 - -Collection of epithelial number data and sample type for these datamined samples also (with the thought that a low number of epi cells in certain samples could indicate the loss of sample)
- -Investigation of the amount of liquid added to the swabs
 - -Too little added may mean that sperm present may not be expelled from the swab
 - -Too much added may mean that the sample is too diluted
- -Suggest making control swabs and checking the process step by step

The major overarching concerns of this issue are the fact that in certain circumstances we may not have sent samples for DNA profiling at all (micro, AP and PSA neg) and therefore have missed evidence. Also, occasionally we are asked in court specifically about the number of sperm seen in a sample – if we know that this number is unreliable, how happy will reporters be to quote numbers?

Justin Howes BSc BA MSc (For Sci)
Team Leader – Forensic Reporting & Intelligence Team
Forensic DNA Analysis | Police Services Stream
Forensic and Scientific Services | Health Support Queensland
Department of Health | Queensland Government











JH-78

Report for QIS Audit as of 30/09/2022 8:41:46 AM

Report for QIS Audit -

8752 Audit of all extraction batches from

Audit Contact Details

Contact Cathie ALLEN	onal Unit/s Evidence Recovery and Q	/ original change of
	sational	1/ 04:3

Audit (Lead/Internal) Contact Details

Auditor (Internal/Lead)	Susan BRADY
rganisational Unit/s	Intelligence
Site/Locations	Coopers Plains

Audit Details

Date Audit Performed 28/07/2008	28/07/2008
Audit Type Process	Process
Audit Status	Closed
Audit Subject	Audit Subject Audit of all extraction batches from implementation of DNAIQ and use of robots (23.10.2007 to present)to identify any contamination events.
Audit Objective No Objective	No Objective
Audit Scope No Scope	No Scope
Audit Criteria No Criteria	No Criteria

Audit Outcome

All initia	t
Audit Findings	

All initial results obtained from casework samples processed with DNA IQ and the MultiPROBE II PLUS HT EX platforms between the 23 October 2007 and the 28 July 2008 were investigated, this included a total of 216 extraction batches. Of these, 202 batches were released (94%), six batches were placed on hold pending the release of results (3%) and five

30/09/2022

batches were placed on hold due to the involvement in previous OQIs (2%). An additional three batches were quarantined through the identification of new contamination events that resulted in three new OQIs (1%).

assist in strengthening the confidence scientists have in reporting results. This quality tool could have applications outside the The methodology applied in this audit provides scientists with a new tool to reinforce the existing quality system. It may also scope of this audit and be applied to other types of batches.

Possible recommendations from this audit are listed below:

- The Batch Comparison Macro could be applied to all new extraction batches to assist in the identification of any adverse events prior to the release of results.
- If an adverse event is identified, a streamlined process needs to be in place to address the issue effectively with efficient laboratory communication (eg. e-mail alert system).
- Availability of AUSLAB functions to export all results from any batch type, facilitating other types of audits and quality measures.
- A previously identified system fault in AUSLAB needs to be addressed to ensure that all samples progress from extraction to quantification.

Contact Comments

Associations

No Associations found

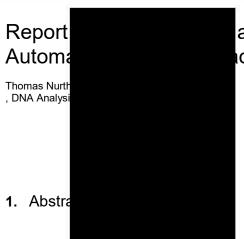
Records

No Records found

8752 Audit of all extraction batches from Copyright \circledcirc 2015, Health Services Support Agency, Queensland Health - All Rights Reserved

JH-79

CaSS | Forensic and Scientific Services



ation of contamination in the action protocol

2. Introduction

Extraction of DNA using the Automated DNA IQ protocol began on the x October 2007. Between this date and Feb 2008 at total of x number of batches were extracted on both PerkinElmer MultiPROBE II HT EX with Gripper Integration platforms.

Several Opportunities for Quality Improvements (OQIs) had been raised but no obvious link was established...

15 OQI in total have been raised.

3. Aim

To document the well to well contamination in automated DNA IQ extraction batches.

4. Equipment and Materials

MultiPROBE[®] II PLUS HT EX with Gripper™ Integration Platform (PerkinElmer, Downers

Crove II LISA)

Deption with Mettler SAG285/L balance (Mettler-Toledo,

Greifensee, Switzerland)

- DN
 pmega Corp., Madison, WI, USA)
- Extraction buller (TomM Tris, 1mM EDTA, 100mM NaCl, 20% SDS)
- SlicPrep™ 96 Device (Promega Corp., Madison, WI, USA)
- Nunc™ Bank-It tubes (Nunc)
- 175µL non-conductive sterile filter RoboRack tips (PerkinElmer, Downers Grove, IL, USA)
- 1000µL Conductive sterile filter Robotix tips (MBP)
- ABI Prism[®] 7000 SDS (Applied Biosystems, Foster City, CA, USA)
- Quantifiler™ Human DNA Quantification kits (Applied Biosystems, Foster City, CA, USA)
- AmpFℓSTR® Profiler Plus Amplification kits (Applied Biosystems, Foster City, CA, USA)
- GeneAmp® 9700 thermalcycers (Applied Biosystems, Foster City, CA, USA)
- ABI Prism[®] 96-well optical reaction plates (Applied Biosystems, Foster City, CA, USA)
- ABI Prism[®] 3100 Genetic Analyzer (Applied Biosystems, Foster City, CA, USA)
- GeneScan[™] 500 ROX[™] Size Standard (Applied Biosystems, Foster City, CA, USA)
- Hi stems, Foster City, CA, USA)
- osystems, Foster City, CA, USA)
- Fo
 - Inc., Florham Park, NJ, USA)Vire & Equipment, Corsham, Wiltshire, England)



5. Metho

5.1 Autom
As per QIS
Casework s

5.2 Re-ext
As per QIS
Casework s

Method of Extracting DNA from Reference and Section of Extracting DNA from Refe

5.3 Manual DNA IQ of substrates

As per QIS 24897 Automated DNA IQ Method of Extracting DNA from Reference and Casework samples Section 18.4

5.4 BRES export analyser mask

A Batch Analyser mask was configured to output the 9PLEX result from an extraction batch.

5.5 Low threshold with GeneMapper IDx

Samples were analysed at 50 rfu and 20 rfu in GeneMapper IDx by a competent scientist.

5.6 PE review and program alteration

The PerkinElmer Liquid handling specialist was asked to review the automated DNA IQ protocol.

5.7 Audit

An Audit of the whole DNA extraction procedure was performed

5.9 Macro

A Microsoft Excel macro was created to check the DNA profiles within a batch for matching DNA profiles

6. OQIs

18 OQIs have been raised in relation to these events. See Table 1 for the list of OQIs raised and the batches that were affected.

Table 1-List of OQis and batches

Table 1-L	St of Outs and patches	
OQI#	Ext Batch ID	
19330		
19349		
19477		
19767		



19768	
20231	
20351	
20422	
20437	
20615	
20617	
20690	
20925	
21222	
21309	
22880	
22882	
19703	

6.1 19330

This batch of reference samples (FTAEXT20080205_01) appears to have a single contamination event.

- Position D2 (12) contains a mixture. The source of this mixture was found to be Position A4 (Pos 25).
- Re-punch and extraction of a different area of the FTA yielded a single source DNA profile.
- Re-extraction of the stored lysate D2 (12) (Store plate) yielded a single source DNA profile that matched the expected reference profile.
- Re-extraction of the original FTA card D2 (12) yielded single source DNA profile that matched the expected reference profile.
- Direction of contamination right to left.

Contamination of the sample has occurred at a point after the removal of the lysate in the extraction process, as the lysate profile is a single source profile matching the expected reference profile. The right to left direction of contamination is consistent with robotic movement. Contamination may have occurred by robotic dripping during elution, during capping/decapping, during storage or during preparation of amplifications.

6.2 19<u>349</u>

This batch ples (CWIQEXT20080225_02) appears to have a single contamination event.

- The negative extraction control (B1/2) was found to contain a DNA profile. The source of this DNA profile was found to be the extraction positive control in position A1 (Pos 1).
- Re-extraction of the stored lysate (B1/2) (Store plate) did not yield a DNA profile for the negative extraction control.
- No substrate exists for a negative extraction therefore no substrate extraction profile for comparison
- Direction of contamination top to bottom of plate.

Contamina extraction to bottom

ed at a point after the removal of the lysate in the for the negative extraction control is NSD. The top onsistent with robotic movement. Contamination



luring elution, during capping/decapping, during may have d storage or ations. 6.3 This batch EXT20080402 01) appears to have multiple contaminat A1/1) was found to contain a mixed DNA profile. The The contain a profile consistent with the DNA in 6 D4/28, G4/31, H4/32). pos 1/1) (Store plate) yielded a mixture consistent with Re the strate (A1/1) yielded a single source DNA profile Re

- Position (B5/34) was found to contain a mixed DNA profile. The mixed profile was found to contain a profile consistent with the DNA in 6 positions (A4/25, B4/26, C4/27, D4/28, G4/31, H4/32).
- Re-extraction of stored lysate (B5/34) (Store plate) yielded a mixture consistent with the initial mixture obtained.
- Re-extraction of the original substrate (B5/34) yielded a partial single source DNA profile
- Position (B6/42) was found to contain a mixed DNA profile. The mixed profile was found to contain a profile consistent with the DNA in 6 positions (A4/25, B4/Pos26, C4/27, D4/28, G4/31, H4/32).
- Re-extraction of stored lysate (B6/42) (Store plate) yielded a mixture consistent with the initial mixture obtained.
- Re-extraction of the original substrate (B6/42) yielded a partial single source DNA profile
- Position (C6/43) was found to contain a mixed DNA profile. The mixed profile was found to contain a profile consistent with the DNA in 6 positions (A4/25, B4/26, C4/27, D4/28, G4/31, H4/32).
- Re-extraction of stored lysate (C6/43) (Store plate) yielded a DNA profile consistent
- Re-extraction of the original substrate (C6/43) yielded an NSD profile with under the contract.
- Position (D6/44) was found to contain a DNA profile consistent with the DNA in 6 positions (A4/25, B4/26, C4/27, D4/28, G4/31, H4/32).
- Re-extraction of stored lysate (D6/44) (Store plate) yielded a DNA profile consistent with the original contamination
- Re-extraction of the original substrate (D6/44) yielded a partial profile (low RFU) which was not consistent with the original 9PLEX or lysate DNA profiles.
- Position (C7/51) was found to contain a DNA profile consistent with the DNA in 6 positions (A4/25, B4/26, C4/27, D4/28, G4/31, H4/32).
- Report reaction of stored breats (27/51) (Store plate) yielded a DNA profile consistent will
- Restrate (C7/51) yielded an NSD profile with ur



- ition (C11/83) contains a mixture.
- inal profile, lysate and substrate have same alleles referred for mixture repretation to confirm uncontaminated profile.

 Inpled at different times, places and by different people.

- Direction and left to right.

A contamin before the contaminat contaminat contaminat agitation or

urred at a point during the extraction process r removal of the substrate, as the lysate profiles are PLEX contamination). The right to left direction of tic movement; however the left to right may have occurred during storage, during plate of plate seals.

6.4

This batch of casework samples (CWIQEXT20080417 01) appears to have multiple contamination events.

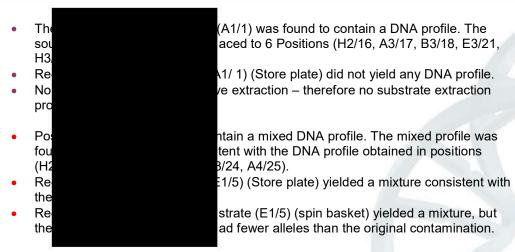
- The extraction negative control (A1/1) was found to contain a DNA profile. The source of the DNA profile was traced to Pos (A4/25).
- Re-extraction of stored lysate (A1/1) (Store plate) yielded a DNA profile consistent with the initial profile obtained.
- No substrate exists for a negative extraction therefore no substrate extraction profile for comparison
- Position (A2/9) was found to contain a profile consistent with the DNA profile obtained in Pos (A4/25)
- Re-extraction of stored lysate of (A2/9) (Store plate) did not yield any DNA profile
- Re-extraction of the original (A2/9) substrate (spin basket) did not yield any DNA profile
- Position (G2/15) was found to contain a profile consistent with the DNA profile obtained in Pos (A4/25)
- Re-extraction of stored lysate of (G2/15) yielded a partial DNA profile consistent with (A4/25).
- rates for (G2/15) did not yield any DNA profile.
- original substrate for A4 (25) yielded a DNA profile consistent with the initial prome, re-extraction of the lysate yielded a consistent partial profile.
- Direction of contamination right to left.

A contamination of the samples has occurred at a point during the extraction process before the removal of the lysate but after removal of the substrate, as some of the lysate profiles are contaminated (consistent with original 9PLEX contamination). The right to left direction of contaminations are consistent with robotic movement. Contamination may have occurred during storage, during plate agitation on the robot, by robotic dripping, or by the removal of plate seals.

6.5 This batch contamina

EXT20080225 02) appears to have multiple





- Position (H1/8) was found to contain a profile consistent with the DNA profile obtained in Pos (H2/16, A3/17, B3/18, E3/21, H3/24, A4/25).
- Re-extraction of stored lysate (H1/8) (Store plate) yielded a profile consistent with the original contamination.
- Re-extraction of the original substrate (H1/8) yielded an NSD profile with undetermined Quant.
- Direction of contamination right to left.

A contamination of the samples has occurred at a point during the extraction process before the removal of the lysate but after removal of the substrate, as some of the lysate profiles are contaminated (consistent with original 9PLEX contamination). The right to left direction of contaminations are consistent with robotic movement. Contamination may have occurred during storage, during plate agitation on the robot, by robotic dripping, or by the removal of plate seals.

6.6 19768

This batch of casework samples appears to have a single contamination event.

- The extraction negative control (A1/Pos 1) was found to contain a DNA profile. The
 ed to 2 Positions (G4/31 & H4/32).
- Re-extraction or stored tysate (AT/Pos 1) (Store plate) yielded a partial DNA profile
- No substrate exists for a negative extraction therefore no substrate extraction profile for comparison
- Direction of contamination right to left.

Contamination of the extraction negative control has occurred at a point during the extraction process before the removal of the lysate, as the lysate is also contaminated (consistent with original 9PLEX contamination). The right to left direction of contamination is consistent with robotic movement. Contamination may have occurred during storage, during plate agitation on the robot, by robotic dripping, or by the removal of plate seals.

6.7 2
This batch s to a single contamination event.



re. The mixture was consistent with the DNA profile Pos fou k H3/24). Re e (G1/7) of the contaminated sample yielded a th the initial mixture obtained. mix strate (G1/7) yielded a single source DNA profile. Re - Direction Contaminat ed at a point during the extraction process before val of the substrate, as the lysate profile is the remova contaminat PLEX contamination). The right to left direction of movement. Contamination may have occurred contaminat during store the robot, by robotic dripping, or by the removal of plate seals.

6.8 19767

This batch of reference samples appears to have a single contamination event.

- Position A9 (65) was found to contain a mixture. The source of this mixture was found to be position A4 (25).
- Re-extraction of the stored lysate (A9/65) of the contaminated sample (Store plate) yielded a mixed DNA profile consistent with the initial mixture obtained.
- Re-punch and extraction of a different area of the FTA yielded a single source DNA profile.
- Re-extraction of the original FTA card substrate (A9/65) yielded single source DNA profile consistence with the single source profile obtained from the re-punch of a different area of the FTA card.
- Direction of contamination left to right.

Contamination of the sample has occurred at a point during the extraction process before the removal of the lysate but after removal of the substrate, as the lysate profile is contaminated (consistent with original 9PLEX contamination). The left to right direction of contamination is not consistent with robotic movement. Contamination may have occurred turing storage, during plate agreement are robot, or by the removal of plate seals.

6.9 20617

This batch of casework samples appears to have multiple contamination events.

- Position F6 (46) contains a mixture. The source of this mixture was found to be position G6 (47).
- Re-extraction of the stored lysate (F6/46) of the contaminated sample (Store plate) yielded a mixed DNA profile consistent with the initial mixture obtained
- Re-extraction of the original substrate(F6/46) yielded a single source DNA profile
- Potential The source of this mixture was found to be
- e (G7/55) of the contaminated sample (Store plate) yie sistent with the initial mixture obtained



Re pro
 strate (G7/55) yielded a single source partial DNA

- Direction et and bottom to top of plate.

A contamin before the reprofiles are top directio right contart plate agitat

urred at a point during the extraction process r removal of the substrate, as some of the lysate th original 9PLEX contamination). The bottom to istent with robotic movement, however the left to ation may have occurred during storage, during ripping, or by the removal of plate seals.

6.10 20 This batch s to have multiple contamination events

- Position E7 (53) is a pooled sample which contains a mixture. The mixture was consistent with the DNA profile found in 4 positions (A7/49, B7/50, C7/51, and D7/52).
- Re-extraction of the stored lysate (E7/53), which had not been pooled, was found to contain a mixture with fewer alleles than the original pooled profile.
- Re-extraction of the original substrate (E7/53), was found to contain a mixture with fewer alleles than the original pooled profile but matches the lysate mixture profile.
- Position F7 (54) was found to contain a partial profile (low RFU peaks). The partial profile is consistent with the pooled DNA profile in (E7/53).
- Re-extraction of the stored lysate (F7/54) was found to contain a partial profile (low RFU peaks), consistent with the DNA profile found in 3 positions (E7/53, G7/55 and H7/56 – which are later pooled).
- Re-extraction of the original substrate (F7/54) yielded an NSD profile with undetermined Quant.
- Position G7 (55) is a pooled sample with (E7/53): Refer above for details.
- Re-extraction of the stored lysate (G7/55) which had not been pooled, was found to contain a mixture with fewer alleles than the original pooled profile.
- rate (G7/55), was found to contain a mixture with rewer arrests man the original pooled profile. The substrate profile was consistent with the breat mixture profile, but did have 3 additional alleles due to higher RFU values in the substrate mixture profile in comparison with the lysate mixture profile.
- Position H7 (56) is a pooled sample with (E7/53): Refer above for details.
- Re-extraction of the stored lysate (H7/56) which had not been pooled, was found to contain a mixture with fewer alleles than the original pooled profile.
- Re-extraction of the original substrate (H7/56), was found to contain a mixture with fewer alleles than the original pooled profile but matched the lysate mixture profile.
- Position A8 (57) is a pooled sample with (E7/53): Refer above for details.
- Re-extraction of the stored lysate (A8/57) which had not been pooled, was found to contain a mixture consistent with the initial pooled mixture which was
- strate (A8/57) yielded a mixture, but the mixture from the lysate re-extraction.



ure. The mixture was consistent with the DNA Pos 19, B7/50, C7/51, and D7/52). pro Re e (A9/65) (Store plate) was found to contain a mix I mixture profile. strate (A9/65) yielded a single source DNA profile Re - Direction and top to bottom of plate. traction plate may have occurred at more that one Contaminat ome samples appear to have become point during lysate from the magnetic beads, with other samples contaminat ate has occurred after the removal of the lysate indicating c on of contaminations are consistent with robotic from the pla taminations are not. Contaminations may have movement, itation on the robot, by robotic dripping, or by the occurred di removal of plate seals.

6.11 20437

This batch of casework samples appears to have multiple contamination events

- Position F1 (6) yielded an NSD profile with undetermined Quant
- Re-extraction of the stored lysate (F1/6) (Store plate) was found to contain a profile consistent with the DNA profile obtained in Pos (F2/14).
- Re-extraction of the original substrate (F1/6) yielded an NSD profile with undetermined Quant
- NOTE: Operator of robot noted that 8tip arm contacted the lysate plate at location between position 6 and 14 on this plate.
- Position G1 (7) was found to contain a profile consistent with the DNA profile obtained in Pos (E1/5)
- Re-extraction of the stored lysate (G1/7) (Store plate) was found to contain a profile consistent with the DNA profile obtained in Pos (E1/5).
- Re-extraction of the original substrate (G1/7) yielded an NSD profile with a Quant value of 0.000995.
- Position B2 (10) contains a mixture.
- Re-mission file stored lysate (B2/10) (Store plate) was found to contain a mixture consistent with the initial mixture profile.
- Re-extraction of the original substrate (B2/10) was found to contain a mixture consistent with the initial mixture profile.
- This sample has produced consistent profiles from substrate, lysate and initial amplification. This sample may not be contaminated.
- Position E2 (13) was found to contain a full DNA profile
- Re-extraction of the stored lysate (E2/13) (Store plate) was found to contain a profile consistent with the initial DNA profile.
- Re-extraction of the original substrate (E2/13) yielded an NSD profile with an
- Popularin a partial DNA profile.
- e (F2/14) (Store plate) was found to contain a pr



- Re-extraction of the original substrate (F2/14) yielded an NSD profile with an undetermined Quant value.
- NOTE: Operator of robot noted that 8tip arm contacted the lysate plate at location between position 6 and 14 on this plate.
- Direction of contamination top to bottom of plate.

Contamination of the samples on this extraction plate has likely occurred at more that one point during the extraction process. Some samples appear to have become contaminated before the removal of the lysate from the magnetic beads (lysate profile consistent with original 9PLEX contamination). However the sample in position F1 (6) on this extraction plate is NSD in the original 9PLEX and in the substrate, with the lysate profile being the only contaminated step. The contamination of the lysate in this position can be explained by the operator of robot noting that 8tip arm contacted the lysate plate at location between position 6 and 14 on the plate.

The top to bottom direction of contaminations are consistent with robotic movement. Contaminations may have occurred during storage, during plate agitation on the robot, by robotic dripping and jamming, or by the removal of plate seals.

6.12 20615

This batch of casework samples appears to have multiple contamination events.

- Position F3 (22) contains a mixture. The mixture was consistent with the DNA profile found in 4 positions (D4/28, E4/29, F4/30, and G4/31).
- Re-extraction of the stored lysate of the contaminated sample (F3/22) yielded a DNA profile consistent with the original contamination.
- Re-extraction of the original substrate (F3/22) yielded a mixed DNA profile.
- Position D4 (28) was found to contain a partial DNA profile. (referred to Tom for allele confirmation)
- Re-extraction of the stored lysate (D4/28) (Store plate) was found to contain a
 mixture which had alleles not present in either the initial product or substrate reextraction.
- Re-extraction of the original substrate (D4/28) yielded a full profile consistent with the initial partial profile.
- Direction right to left.

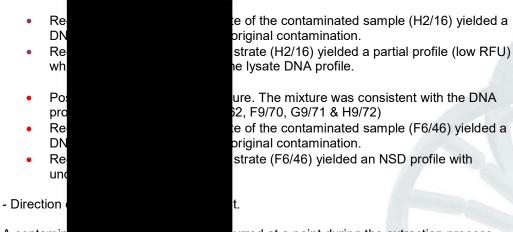
Contamination of the sample has occurred at a point during the extraction process before the removal of the lysate but after removal of the substrate, as the lysate profile is contaminated (consistent with original 9PLEX contamination). The right to left direction of contamination is consistent with robotic movement. Contamination may have occurred during storage, during plate agitation on the robot, by robotic dripping, or by the removal of plate seals.

6.13 20925

This batch of casework samples appears to have multiple contamination events.

 Position H2 (16) contains a mixture. The mixture was consistent with the DNA profile found in 2 positions (G3/23 & H3/24).





A contamination and surred at a point during the extraction process before the removal of the lysate but after removal of the substrate, as some of the lysate profiles are contaminated (consistent with original 9PLEX contamination). The right to left direction of contaminations are consistent with robotic movement. Contamination may have occurred during storage, during plate agitation on the robot, by robotic dripping, or by the removal of plate seals.

6.14 21222

This batch of casework samples appears to have a single contamination event.

- Position F11 (86) contains a mixture. The mixture was consistent with the DNA profile found in 5 positions (B10/74, C10/75, F10/78, G10/79, and H10/80).
- Re-extraction of the stored lysate of the contaminated sample (F11/86) yielded a DNA profile consistent with the original contamination.
- Re-extraction of the original substrate (F11/86) yielded an NSD profile with an
- Direction of contamination left to right.

A contamination of the samples has occurred at a point during the extraction process before the removal of the lysate but after removal of the substrate, as some of the lysate profiles are contaminated (consistent with original 9PLEX contamination). The right to left direction of contaminations are consistent with robotic movement. Contamination may have occurred during storage, during plate agitation on the robot, by robotic dripping, or by the removal of plate seals.

6.15 21309

This batch of casework samples (CWIQEXT20080531_01) appears to have multiple contamination events.

Positive
 pr
 xture. The mixture was consistent with the DNA pr
 38).



e of the contaminated sample (G11/87) yielded a Re original contamination. DN Re strate (G11/87) yielded a partial DNA profile (low RF Pos xture. The mixture was consistent with the DNA pro e of the contaminated sample (H12/96) yielded a Re DN original contamination. strate (H12/96) yielded an full DNA profile Re

- Direction to top of plate.

Contaminate the removal of the system but after removal of the substrate, as the lysate profile is contaminated (consistent with original 9PLEX contamination). The left to right direction and bottom to top direction of contamination is not consistent with robotic movement. Contamination may have occurred during storage, during plate agitation on the robot, or by the removal of plate seals.

6.16 22880

This batch of casework samples appears to have a single contamination event.

- Position G7 (55) contains a mixture. The mixture was consistent with the DNA profile found in 5 positions (B10/74, C10/75, F10/78, G10/79, and H10/80).
- Re-extraction of the stored lysate of the contaminated sample (F11/86) yielded a DNA profile consistent with the original contamination.
- Re-extraction of the original substrate (F11/86) yielded an NSD profile with an undetermined Quant value.
- Direction of contamination left to right.

Contamination of the sample has occurred at a point during the extraction process before the removal of the lysate but after removal of the substrate, as the lysate profile is contaminated (consistent with original 9PLEX contamination). The left to right direction of c movement. Contamination may have occurred during storage, during plate agitation on the robot, or by the removal of plate seals.

6.17 ?19703?

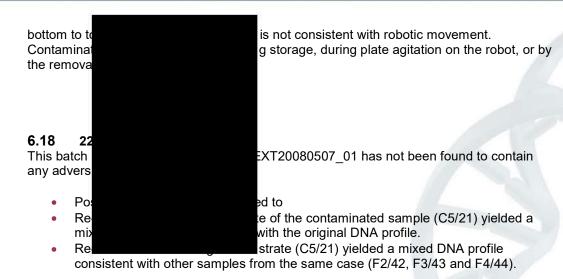
This batch of casework samples appears to have a single contamination event.

- Position F5 (45) contains a mixture. The mixture was consistent with the DNA profile found in 3 positions (E5/37, E6/38, and E7, 39).
- Re-extraction of the stored lysate of the contaminated sample (F5/45) yielded a DNA profile consistent with the original contamination.
- Re-extraction of the original substrate (F5/45) yielded a full profile consistent with other samples from the same case (F2/42, F3/43 and F4/44).

Contamina the remov contamina

ed at a point during the extraction process before /al of the substrate, as the lysate profile is PLEX contamination). The left to right direction and





Three separate events are believed to have occurred:

The non concordant DNA profiles between the original sample (334116178) and the spin basket (342253513) can be attributed to consumable contamination. This statement is based on the following assumptions:

- The sample was extracted on a manual DNA IQ extraction batch with only samples from the same case and controls. The DNA profile obtained does not match any of these DNA profiles
- The DNA profile obtained from this sample was checked against all other samples and is a unique DNA profile.
- If the source of the DNA profile was the reagents; then the other samples in this
 extraction batch including the controls should display the same profile or mixtures
 which they did not.
- There is no database of plastics manufacturers to confirm if the DNA profile's source was the plastic.

The non concordant DNA profiles between the original sample (C5/21) and the lysate (103500111) can be attributed to user error. The profile obtained is consistent with the user rotating the otors plate 105. The DTA profile matches sample (76/D10).

The mixed ned for the lysate (342254626) is consistent with the original sample (334116178) and contamination from the adjacent well (Pos 29/E4) 301864386 and possibly other wells (minor contributors). Contamination of the sample has occurred at a point during the extraction process before the removal of the lysate but after removal of the substrate, as the lysate profile is contaminated (consistent with original 9PLEX contamination). The left to right direction and bottom to top direction of contamination is not consistent with robotic movement. Contamination may have occurred during storage, during plate agitation on the robot, or by the removal of plate seals.





7. Discus

The resulting type of every state of the contamination events. Broadly the events fit

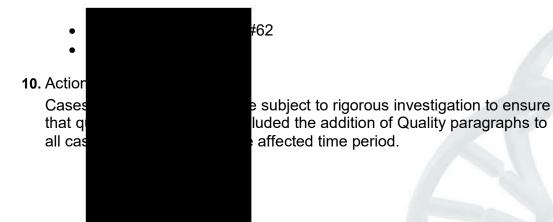
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An extension and the ken to evaluate the performance of the DNA IQ method performed on the PerkinElmer MultiPROBE II extraction platforms.

An external audit was performed by Wiley & Sloots

- 8. Summary
- 9. References
 - Letter from Crown Law & Opinion from Solicitor General.pdf
 - OQI19330 Report
 - OQI 19349 Report
 - OQI 19477 Report
 - OQI 19767 Report
 - OQI 20231 Report
 - eport
 - OQI 20422 Report
 - OQI 20437 Report
 - OQI 20615 Report
 - OQI 20617 Report
 - OQI 20690 Report
 - OQI 20925 Report
 - OQI 21222 Report
 - OQI 21309 Report

 - Audit OQI 21718
- Queensland Government







Appendices

Original OQI 19930 FTAEXT20080205_01

CWIQEXT20080225 02 **OQI 19349**

346790253	6	17	25	33	41	49	25	99			88
346790262	10	18	26	34	42		28	99			06
3	11	19	27	35	43		69	29			91
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2	13	21	29	37	45	16	61	69			93
9	14	22	30	38	46	54	62	02	78	98	94
7	15	23	31	39	47		63	71			95
8	16	24	32	40	48		64	72			96

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9 17 320124335 334116189 41 49 57 65 73 10 18 320124326 34 42 50 58 66 74 11 19 27 35 43 51 59 67 75 12 20 28 36 44 52 60 68 76 13 21 320124371 37 45 53 61 69 77 14 22 30 38 46 54 62 70 78 15 23 31 47 55 63 71 79 22012436 24 24 56 64 72 80 78	ı											
10 18 320124326 34 42 50 58 66 74 75 11 19 27 35 43 51 59 67 75 12 20 28 36 44 52 60 68 76 13 21 320124371 37 45 53 61 69 77 14 22 30 38 46 54 62 70 78 15 23 31 39 47 55 63 71 79 220124349 24 320124362 40 48 56 64 72 80	7	6	17	320124335	334116189	41	49	22	65	73	81	88
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12 20 28 36 44 52 60 68 76 13 21 320124371 37 45 53 61 69 77 14 22 30 38 46 54 62 70 78 15 23 31 39 47 55 63 71 79 320124349 24 320124362 40 48 56 64 72 80	3	11	19	27	35	43	51	69	29	22	83	91
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14 22 30	30		38	46		62	70	78		94
15 23 342270241	342270241		39	47		63	71	62		95
16 24 342270230	342270230		40	48	99	64	72	80	88	96

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3	11	19	27	35	43	51	69	289009815	334742062	83	91
4	12	20	28	36	44	25	09	89	92	84	92
9	13	21	29	37	45	23	61	69	22	85	93
9	14	22	30	38	46	54	62	70	82	86	94
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	49	90	51	52	23	54	29	99
	41	42	43	44	45	46	47	48
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Appendix 2. Quality Paragraphs used

Category A - samples tested during the period of Oct 07 to July 08 but no adverse event associated with the results of this

Testing for this case has been conducted in a period where some results were the subject of adverse events. This period was between October 2007 and July 2008. Testing for this case was not the subject of any adverse results. This conclusion has been reached by assessing the individual results by comparison to all other results from samples processed along side each

Include the following if Retesting has been conducted: Retesting has been conducted on identified samples which have confirmed the alleles in the original result. These samples have been reported as they have been assessed as no adverse event having been detected and the results have passed all quality assurance checks. Category B - samples tested during the period and an adverse event occurred on those samples - results cannot be reported (QC failure). Testing for this case has been conducted in a period where some results were the subject of adverse events. This period was between October 2007 and July 2008. Within this case, the adverse event is demonstrated to have affected a result or results and the integrity cannot be verified, then this result has therefore been reported as follows: "These samples did not pass our Quality System requirements at the DNA Analysis stage and therefore the DNA profiling results relating to these samples cannot be reported".

Category C - samples tested during the period, an adverse event has occurred on that sample/s, retesting has been conducted and the retesting results can be reported

751

extracted portion), retesting has been conducted, using an alternative manual extraction method. The retesting has confirmed Testing for this cas<mark>e</mark> has been conducted in a period where some results were the subject of adverse events. This period was between October 2007 and July 2008. Testing for some samples within this case has been the subject of an adverse event The cause of the adverse event was identified to have occurred within the extraction process performed on an automated platform. Where sample is remaining (which could include, but is not limited to, additional stain remaining or previously the alleles in the original result. These samples are reported as they have been assessed as no adverse event has been detected and the results have passed all quality assurance checks. 752