

Report concerning the provision of expert advice concerning Sperm Microscopy at QHFSS

Clint Cochrane 10<sup>th</sup> October 2022

**Introduction**

1. I have been engaged by the Commission of Inquiry into DNA testing in Queensland to review general and specific concerns raised regarding the scientific testing methods undertaken for sperm microscopy in the QHFSS DNA Analysis Unit. More specifically I was asked to evaluate an identified inconsistency between the levels of sperm detected during the initial evidence recovery (ER) and the microscope slides created during the differential extraction process, and the laboratory response to the issue.
2. I am the laboratory manager of the Forensic Biology/DNA laboratory, part of the NSW Health Pathology Forensic & Analytical Science Service. I have over 21 years working in Forensic Biology/DNA analysis. I have particular expertise in forensic biology analysis of sexual assault casework and have also published a number of scientific papers in this field. I have attached my curriculum vitae as Appendix A.
3. I have prepared this report on my own behalf, and none of my comments or opinions may be attributed to NSW Health.
4. I have attached my instructions and an index identifying the material with which I was briefed by the Commission as Appendix B.
5. During late 2015 an issue was identified by reporting scientific staff at QHFSS that the DNA results (strong male DNA profiles recovered) were inconsistent with the lack of sperm identified from ER slides. When the reporting staff requested that the slide created during the differential extraction be examined; sperm were found, sometimes in high density (2+ or above). The high density of sperm on the differential slides is consistent with the DNA results recovered. During the first half of 2016 this issue was identified on multiple occasions in different cases, with specific occasions being notified to Line Managers or above. A small analysis on 2014/15 data (1), of samples where ER slides had zero sperm detected then subsequently had differential slides examined, showed nearly 10% of the subsequent slides had high density of sperm (2+ or above).
6. Both correspondence and this data analysis flagged that the effectiveness of ER slide preparation was in question. It should be noted that the competence of the staff performing the microscopy was ruled out as the source of the issue. Of particular concern to reporting staff was that the ER slide was an examination component that influenced whether DNA testing would progress for sexual assault casework. Other components such as presumptive tests for semen, sample origin (e.g. condoms) and case context were also factors in DNA testing progression.
7. Prior to project #181 starting, on 8 August 2016, a change in laboratory procedure meant that all suspected semen samples were to be processed using a differential extraction with a slide being created, in addition to the ER slide. Both slides were to be considered in determining whether testing would progress and what further testing was required. This was a risk mitigation measure to ensure ineffective ER methods would not adversely affect the progression of samples to DNA testing.

8. A timeline listing key points in the raising of the spermatozoa discrepancy issue and project #181 are presented in appendix 1. Its contents are based on the provided correspondence, project documents and witness statements.

### **Project #181**

#### **Background**

9. Project #181 was originally designed to identify the possible cause of the sperm microscopy issue. The project appears to be the method of choice for the laboratory to resolve the microscopy issue, rather than a OQI or Adverse Event investigation.
10. Over a period of nearly four (4) years the focus of the project changed from trying to identify the issue, to attempting to improve the ER slide process, then to relying exclusively on the differential slides for sperm microscopy while preserving the ability to perform semen presumptive testing.
11. The project had seven parts of experimental testing involved.

<b>Date commenced</b>	<b>Project Part</b>	<b>Testing performed</b>	<b>Results/ Conclusions</b>
August 2016	Part 1	Investigation into the current microscopy method sensitivity using mock samples. Detection levels for presumptive tests Acid Phosphatase (AP) and p30 also examined.	Epithelial cells were transferred in significant quantities onto the ER slides, signifying that there was not a loss of material during slide staining. Likely cause of low sperm density on ER slides appears to be inefficient transfer of sperm from the substrate onto the slide.  Decision made to cease trying to identify issue and concentrate on improving ER slide microscopy method.
April 2017	Part 2	Alternate ER microscopy preparation method using a spin basket trialled.	This method did not show improvement on the ER method in routine practice. Decision made to divert project from improving ER sperm microscopy preparation methods, to modify protocols to rely on differential slides while preserving presumptive screening options.

May 2018	Part 3	Explored viability of varying ER sample suspension volumes to allow for retrospective semen presumptive screening while relying exclusively on differential slides for microscopy (i.e. ER microscopy not to be performed).	Microscopy and p30 detection sensitivity was superior using adapted method compared to the ER method.  The AP presumptive test was adversely affected by the adapted protocol.
March 2019	Part 4	Attempted to optimise ER suspension incubation conditions to maximise AP and p30 sensitivity, while not affecting microscopy of the differential slide.	Incubation conditions appeared to have minimal effect on sperm microscopy sensitivity.  AP testing remained poor using the adapted protocols.
June 2019	Part 5	Explored the effects of different variables on AP performance.	AP testing remained poor using the adapted protocols.  Microscopy and p30 largely unchanged.
June 2019	Part 6	Further attempts to optimise AP performance by reducing suspension volume and trialling other variables including physical agitation.	All attempted modified protocols did not overcome the continued poor performance of AP testing.  Microscopy and p30 largely unchanged.
June 2019	Part 7	Trialled method using different substrates such as swabs, fabric swatches and tapelifts. Also varied semen donor source to simulate the variability that may be encountered in casework.	Adapted method was superior across substrates when compared to the ER method.  AP results remained poor.
July 2020	Project completed	New modified protocol implemented. AP discontinued as screening method except when trying to localise stains on items.	

### Discussion

- Reporting staff concerns of the underperformance of the ER method for preparation of microscope slides were well founded. Sperm density variability between ER and differential slides could be partially explained by the increased concentration of sperm to liquid volume for the second slide, when the variability is modest. Large discrepancies (a difference of more than 1 sperm density grading) are unlikely to be the result of concentration, especially a

difference of 0 sperm to 2+ or more sperm. These discrepancies point to a sub-optimal method for creation of slides in ER.

13. In part 1 of project #181 it was established that the inefficient transfer of sperm from the substrate onto the slide was the likely cause of the ER sperm microscopy issue. After this testing it was decided that testing to determine the root cause of this inefficiency was likely to be difficult and time consuming and the need for improved method for sperm microscope slide creation was inevitable regardless. In my opinion the decision to pivot to method improvement was the most time and resource efficient way to resolve the issue. The main concern with not determining the root cause would be difficulty narrowing potentially affected cases, if a retrospective examination of previous casework was undertaken.
14. In part 2 the project only looked at 1 alternate method for creation of ER slides, using a spin tube. This is a very limited look at alternatives to the tube elution method used by QHFSS. Other potential options would include the medical examiners making microscope smears at the time of SAIK collection and ER making smears by using direct contact methods such as pressing damp slides, tapping or swirling (2). Of note, some of these methods are used in other Australasian jurisdictions and were previously in use in QHFSS prior to 2008. The literature review conducted as part of the project appeared to focus only on methods in use or close to what was being utilised in the laboratory. The laboratory could have benefitted by questioning the other Australasian laboratories through the Biology Specialist Advisory Group (BSAG). Alternatively, the consumables used in collection of SAIK evidence could have been evaluated, though this would probably be more time consuming as DNA recovery would also require testing.
15. Parts 3 to 7 of the project are focused on adopting the differential slide as the sole sperm microscopy point and creating optimal ER testing methods to preserve the presumptive semen tests, AP and p30, for azoospermic semen identification. It should be noted that other options of direct testing for AP could also have been considered. Ultimately this adapted method was adopted for sperm microscopy and p30, while AP testing was discontinued except for localising stains on items.
16. The results of parts 1 and 2 categorically support the superiority of the differential lysis slide over the ER slide when it comes to sperm microscopy. The quality and sperm recovery on the differential slide is undoubtedly better. Removal of ER microscope slide creation avoids duplication and improved quality.
17. The downsides of this decision include:
  - a. Differential extraction is a time-consuming process.
  - b. Submitting all samples looking for sperm for differential extraction is both less time and cost effective than stopping samples without sperm at ER.
  - c. Differential extraction is an inefficient extraction protocol when compared to kit-based extraction protocols (such as the Applied Biosystems PrepFiler Forensic DNA Extraction Kit). A lot of cellular material is lost over the course of the differential extraction process, which could affect the success of future DNA testing such as Y-STR profiling.

18. Overall the project appeared to take a long time, nearly 4 years, for the testing that occurred. This is especially the case considering the routine processing of samples with differential slides being enacted prior to the commencement of the project. Both ER and differential slides were being examined for casework for the entirety of the 4 years, which appears to be a prolonged unnecessary duplication of testing. Better project planning and dedicated commitment of resources could have reduced the project time, which would have benefitted the laboratory overall. Staff being dedicated to the project full time, rather than working around other laboratory duties would allow prompt delivery of project work.
19. Additionally the time spent on this project comes at an opportunity loss to pivot to other research, such as Y-STR testing. Y-STR testing is a valuable tool for sexual assault investigation that is not in place at QHFSS.

### **Questions asked from the Commission**

1. Whether the methods, systems and processes in relation to sperm detection, testing and analysis was consistent with international best practice when the issue arose in 2016;
20. The workflows in place for examination of sexual assault casework before the issue arose in 2015 was identified in section 6.3 of The Examination for and of Spermatozoa (document number 17189V10) from 20/09/2010. There were subsequent versions of this SOP and the workflow charts were removed between version 10 and 13 (I did not access versions 11 or 12). These workflow charts describe the progression of a sample through microscopy, presumptive tests (acid phosphatase and p30) and DNA testing. The results of the microscopy and presumptive tests could lead to 3 possible options:
  - a. DNA testing using a differential lysis method to separate sperm cells from other cellular material. This was performed where:
    - i. sperm was detected, or
    - ii. Sperm was not detected, but both AP and p30 testing was positive.
  - b. DNA testing using routine DNA extraction ('Cells') method. This was performed on external samples, such as vulval or perianal swabs or underwear, where sperm were not detected and either AP or p30 testing was negative.
  - c. No further action taken, on internal swabs such as vaginal samples.
21. Further examination strategies for sexual assault casework are presented in the Examination of Sexual Cases SOP (document number 32106V3) from 29/01/2015. This SOP details how the case context may modify the laboratory progression of samples.
22. The QHFSS workflow for sperm detection, testing and analysis as written were in line with best practice, assuming processes are working. The creation of microscope smears in ER was underperforming, which led to instances where sperm was not detected in ER but were plentiful on the differential slide. This underperformance flagged that this method was not

meeting international best practice in 2016. The laboratory response to attempt to improve the method (project #181), shows there is an issue they are trying to address.

23. It should be noted there is no defined international best practice in sexual assault investigation, other jurisdictions have strategies more suitable to their needs. In 2013 the National Institute of Justice in the USA published recommended guidelines, National Best Practices for Sexual Assault Kits: A Multidisciplinary Approach, which included the recommendation for a Direct to DNA approach. It should be noted that this recommendation revolves around efficiency gains and clearing backlogs in sexual assault casework in USA forensic laboratories. The adapted methodology at QHFSS has some commonalities with NIJ recommendations.
2. Whether the identification, investigation and resolution of the sperm microscopy issue, including Project 181 was appropriate and consistent with international best practice, including consideration of the following particular issues:
  - i. Whether it was sufficient for the sperm microscopy testing issue identified to be investigated as a project, or should the issue have been raised as an OQI or an Adverse Event;
24. In my opinion multiple instances of sperm not being found in ER slides than in abundance on a differential slide would warrant an OQI or an Adverse Event. The persistent reappearance of this issue shows the ER slide preparation protocol is not meeting expected standards. The frequency and scale of underperformance should have triggered a quality based investigation such as an OQI or Adverse Event.
25. Whether the action resulting should have been dealt with as a project or OQI/ Adverse event investigation seems largely irrelevant, as long as the investigation proceeds in a timely manner. Given the resources required to investigate, the decision to do as a project appears warranted.
- ii. Whether the response time and duration of Project #181, including from when the sperm microscopy issue was raised in and around 2016 to project finalisation, dealt with the issue with a reasonable time and with appropriate urgency;
26. Of more relevance is if it was considered that this issue was investigated promptly. It should be noted that there was approx. 6 months between the issue being initially flagged and the initial project request being written. It was then a further 2 months before the change to laboratory protocol to routinely test all potential semen samples with a differential extraction and also the project commencing. This meant that a sub-optimal method was used for approx. 8 months after concerns were initially voiced.
27. This appears to be an excessive period to initiate an investigation into an underperforming method that may prevent samples from being appropriately tested. The delay seems especially long considering the amount of correspondence to management highlighting:
  - a. problematic ER sperm microscopy results,
  - b. the considerable concern scientific staff expressed around the validity of ER sperm microscopy results, and
  - c. offers from teams outside ER to spare their team members to research the issue.

28. The workflow modification on 8/8/2016 largely resolved the ER sperm microscopy issue, ensuring that samples containing sperm progressed as required. Project #181 started after this decision was made.
29. As discussed earlier in paragraph 18, project #181 took almost 4 years to complete. This appears excessive considering the amount of testing performed. There appeared to be significant delays progressing between different parts of the project (each of the first 3 parts took approximately 1 year each).

*iii. Whether the project should have included a root cause analysis of what was causing the problem;*

30. The project came to the realisation that transfer of sperm during the creation of the ER slide was the issue. It was hypothesised that this was occurring because of the difference in dilution volumes between ER and differential slide.
31. I am not sure this adequately explains the vast difference in cell density in some slides. Ultimately the solution was to improve sperm recovery for microscopy. This could have been done through alternate means such as doctors creating slides at the time of administering the SAIK, different ER methods or creating a differential slide. QHFSS chose to do the latter. I have no issues with that decision based on the quality of slides but the adopted course may have created additional work, delay and/or expense. Note that differential extraction is an inefficient extraction method when compared to kit-based extraction protocols where sperm separation is not performed. Preservation of samples for Y-STR analysis may be more effective, especially in cases with very low sperm counts.
32. Wide testing of alternate ER methods or different consumables was not performed. I am not sure that a formal root cause analysis would have made a particular difference to the outcome of the method for sperm microscopy chosen.

*iv. Whether the project should have considered the issues of how long this issue had existed, what samples it may have affected, and what should be done about those samples;*

33. On 8/8/16 all suspected semen samples were to be processed through differential lysis with a smear to be made in ER and during the differential protocol.
34. The project considered the protocols directly prior to this decision, not historic options previously used within QLD (direct slide or doctor making slide). The assumption would be this issue would have persisted as long as this method was unchanged (assuming no changes to consumables etc). I understand that method was in place at the laboratory since 2008 (4). The effect with previous protocols was not tested.
35. Both project and laboratory appear to be focused on contemporaneous and future improvement rather than focusing on previous casework. Samples that were not tested but stored, could be retrospectively examined with improved technology.
36. The laboratory has an opportunity to consider re-testing of internal SAIK samples where no other evidentiary results were identified from other internal samples AND no further testing

was performed AND the case has avenues of progression (i.e. not finalised). The laboratory may benefit from additional information provided from Police and court. Additionally samples where presumptive testing was negative and sperm could be located using logic (e.g. crotch of underpants) may also be considered. The amount of cases that would fall into these categories would appear limited, based on the review of 738 samples between 8/8/16 and 28/3/17 (discussed in the Data analysis of modified sexual assault process (unfinished) – May 2017, further in iv). This “cold case” testing may also benefit from using Y-quantification screening of samples and potentially Y-STR analysis.

37. If practicable, the QHFSS should perform a data analysis to identify cases fitting the criteria stipulated in paragraph 36 prior to 8/8/2016. If easily identifiable, the amount of further testing required would likely be minimal.
38. If the cases are exceedingly difficult to identify through the laboratory information management system, QHFSS/Queensland Health will need make a policy decision to determine the priority of examining sexual assault ‘cold-cases’ for further forensic DNA evidence. Due to the likely limited impact on cases of the ER slide examination issue, other retesting options such as Y-STR testing may be more fruitful in providing new evidence to these historic cases. Cold case reviews are typically more resource intensive than contemporary casework and require experienced staff with a broad range of knowledge in historic and current casework options.

v. *Whether, and to what extent, any failure to consider what previous samples were affected by the issue means that useable DNA evidence may not have been obtained by the laboratory;*

39. According to the Examination of Sexual Cases v3.0 from 29/01/2015 QHFSS workflow encourages the upfront submission of all SAIK samples for each case. Sperm microscopy and presumptive testing are performed on all samples before deciding if/and what DNA testing is required. This policy means that there may be multiple opportunities within the same SAIK to unearth evidential body fluid and DNA results. This direction would limit the amount of cases where the ER slide issue could lead to cases where DNA results were not obtained. Granted there may be samples that provide unique evidence (e.g. oral samples) that other SAIK samples may not.
40. An alternate way of treating SAIK samples is by testing minimal samples, through progressive submission of samples, to provide the required evidence and storing the remaining samples for future testing opportunities. As an example, finding sperm and obtaining a male DNA on one internal vaginal sample, provides the same information that finding the same results on multiple swabs provides. The progressive model would submit 1 vaginal sample and wait for results prior to testing other vaginal samples. If the results were positive, other samples remain for future testing, if required. If results are negative, other samples may be tested or a decision could be made to preserve samples for improved future methods.



41. The benefit of upfront submission is processing efficiency, while it may limit decision making opportunities. Progressive submission allows testing to be targeted more effectively but is less efficient in the laboratory.
42. Since 8/8/2016 all possible semen samples went through differential extraction to develop a microscope slide. Differential extraction is ideal in samples where sperm is required to be separated from other cellular material. An example of this is a vaginal swab in a sexual assault. The sperm is the evidentiary biological fluid, the female's own vaginal cells are irrelevant and may complicate DNA analysis. Being able to preferentially target sperm in a DNA extraction is obviously advantageous in these circumstances.
43. The disadvantage of the differential extraction is it is an inefficient technique. This is not a major concern when sperm (a high DNA yielding body fluid) is present, but is problematic when it is absent and you are potentially looking for trace (typically low DNA yielding epithelial cells) DNA. It should be noted that it is not uncommon for sperm not to be detected in sexual assault matters.
44. QHFSS' excessive upfront submission for differential extraction may affect the prospects of future DNA testing success with alternate technology, such as Y-STRs. Y-STRs exclusively target male DNA on the Y-chromosome, effectively making the female DNA present in the sample irrelevant. This has obvious benefits in forensic DNA analysis for sexual assault matters. Y-STR testing can target trace amounts of male DNA in SAIK samples where semen has not been detected. Kit based extraction methods (such as the Applied Biosystems PrepFiler Forensic DNA Extraction Kit) that maximise DNA recovery are more beneficial for Y-STR DNA testing than the inefficient differential extraction method.
45. The duplication of DNA testing on multiple samples that would provide the same evidence appears to be over-testing that creates additional laboratory processing and reporting requirements. This additional testing would likely result in wasted time and resources.
46. Testing all potential semen samples upfront with a differential extraction protocol is not best practice for sexual assault cases where sperm is not detected. Y-STR testing has been used in forensic DNA testing for over a decade so casework examination workflows should have been designed to consider preserving samples and/or using efficient extraction methods for Y-STR typing. Access to Y-STR testing for sexual assault casework would be considered laboratory best practice.
47. The laboratory performed a review of 738 samples (3) tested between 8/8/16 and 28/03/17 where sperm were not identified on ER slides and a differential extraction slide was made as per routine protocol. Of the 738 slides:
  - a. 591 did not have sperm on the differential slide either;
  - b. 147 subsequently identified sperm on the differential slide.
48. An assessment was performed on these 147 samples to determine what would have previously occurred prior to the change in laboratory protocol on 8/8/16. Of the 147 samples:
  - a. 71 samples would have been tested by differential extraction anyway due to presumptive testing results, sample type or other SAIK results in case.

- b. 47 samples would have progressed through DNA testing using the routine 'Cells' protocol. For samples containing sperm this may be a less effective method for internal SAIK swabs due to the large cellular density of the internal cavities. It should be noted that this extraction would typically be more effective for Y-STR analysis though.
  - c. 29 samples would not have been DNA tested based on the previous workflow. Of these 29 samples:
    - i. 28 samples would not have recovered new evidential DNA profiles using the QHFSS kit. This is due to other SAIK results in the case or DNA results being unsuccessful or non-comparable.
    - ii. 1 sample would have recovered DNA evidence that would not have been tested in the previous workflow.
49. This data review would suggest there would be very limited occasions where DNA evidence was missed through the previous laboratory workflow. Only 1/738 samples would have been heavily affected.
50. I would place a couple of caveats on this data:
- a. This is assuming similar lab conditions and consumables prior to 8/8/16.
  - b. All of this work is using autosomal DNA not Y-STRs.
- vi. Whether the scope of the review conducted by ESR, and the materials and instructions provided to ESR for that review, adequately identified and addressed the issues identified by Amanda Reeves and others.*
51. It appears that there is very limited information related to the issues relating to the creation of Project 181 passed on in the Request/Terms Of Reference to ESR. The Request/Terms of Reference only stated; *"An issue has been raised specifically regarding spermatozoa negative, acid phosphatase negative sexual assault samples, however a review of the processing of SAIKs would be appreciated in the spirit of continuing quality improvement."*
52. The review appears to be commissioned directly looking at SOPS/procedures with a different report AP Paper – False Positive Investigation. I cannot comment if other communications addressed this issue.
53. I could not identify any material provided to ESR that mentioned the changed ER protocol introduced on 8/8/2016. The change to practice to examine all potential semen samples fit within the options available in the laboratory SOPs. The SOPs only required amendment after the completion of Project #181 when the process was changed so that the only slide examined would be the differential lysis slide.
54. It doesn't appear that ESR were specifically tasked with assessing the ER microscopy issue, its cause or potential solutions.

3. Whether the amended methods, systems and processes implemented in relation to sperm microscopy was consistent with international best practice;

55. The amended sperm microscopy methods allows reliable identification of sperm, however overuse of the differential extraction could hamper the success of Y-STR testing on intimate SAIK samples.
56. QHFSS could be more advanced with using Y-STR testing or Y-quantitation as a screening tool. While not relating to sperm microscopy, it would improve their sexual assault investigation capabilities. By 2020 being able to utilise Y-STR testing in sexual assault investigations is considered best practice.
57. Note that there is no universally agreed international best practice across the Forensic DNA field. Other peak bodies may recommend different sexual assault workflow strategies that better suits the requirements of their jurisdictions. The NIJ recommends only retrospective body fluid identification post-DNA testing, with the implementation of a Direct to DNA strategy. This strategy is designed for efficient sample throughput in laboratories backlogged with sexual assault casework. In the interest of laboratory efficiency, the direct to DNA recommendation by the NIJ encouraged upfront DNA testing on samples, bypassing biological fluid identification. If the biological source of the DNA testing is questioned, retrospective testing such as sperm microscopy may be performed. Note that this recommendation would encourage upfront differential extraction, which may negatively effect the chance of recovering Y-STR profiles.
58. Performing sperm microscopy at the differential lysis stage does hamper the ability to make time since intercourse assessments. Time since intercourse estimates can be provided by using the density of sperm on a microscope slide and the percentage of complete sperm (i.e., sperm with a tail) versus sperm heads. The differential lysis process will typically damage the fragile sperm tail, meaning only the sperm density can be relied on for the time since intercourse estimation. This can potentially increase the potential time frame in which sperm was deposited in an internal body cavity prior to the patient's forensic medical examination. Time since intercourse estimations are rarely encountered in court matters but may be valuable evidence in some cases.

4. If any deficiency in the methods, systems or processes for sperm microscopy or the resolution of the issue that arose in 2016 is found, the impact of that deficiency on:

i. Whether the obtaining of a useable DNA profile from a sample by the laboratory was reliable and accurate;

59. Samples that went through routine extraction, due to sperm not being detected in ER, might have been "swamped" by the complainant's own DNA. The likelihood of this would be dependent on where the sample was collected from.
60. Without the use of Y-STR testing, these samples may not be able to obtain an evidentiary DNA profile. As detailed in paragraph 44, Y-STR testing exclusively targeted male DNA on the Y-chromosome, effectively ignoring any background female DNA present in a sample. Y-STR testing does not have the same discriminatory power between individuals as routine

(autosomal) DNA testing, because the Y-chromosome is inherited along the paternal line meaning a Y-STR profile will be shared with paternally related males within a family.

*ii. Whether DNA profiles obtained by the laboratory are reliable and accurate.*

61. Prior to 2016, some samples may not have been processed for DNA which could have yielded a useable DNA profile.
  62. The reliability and accuracy of DNA results for the samples tested should not be affected.
  63. Samples that underwent differential extraction may have less chance of success with future Y-STR testing due to the inefficiency of the differential extract method losing cellular material.
5. *If any deficiency in the methods, systems or processes for sperm microscopy is found, what should be done now to rectify that issue.*
64. The current sperm microscopy methods allows reliable identification of sperm, however overuse of the differential extraction could hamper the success of Y-STR testing on intimate SAIK samples.
  65. SAIK workflows, especially the upfront examination of all potential semen samples, should be reviewed to consider the preservation of samples for Y-STR testing.
  66. QHFSS could be more advanced with using Y-STR testing or Y-quantitation as a screening tool. While not relating to sperm microscopy, it would improve their sexual assault investigation capabilities. Being able to utilise Y-STR testing in sexual assault investigations is considered best practice.
  67. If the laboratory introduces Y-STR testing, a review of SAIK workflow should occur.
  68. There is an avenue for identifying samples for retrospective analysis as discussed in my answer to 2iii.

Signed:



Clint Cochrane

Dated: 10 October 2022

**References:**

1. Project proposal #181 Investigation into the sensitivity of spermatozoa microscopy – August 2016
2. Spiker 2012, Thesis – Identification of Spermatozoa on Sexual Assault Swabs: A Comparative Analysis of Traditional Tube Extraction and Direct Slide Elution Methods. *Southern Illinois University*
3. Data analysis of modified sexual assault process (unfinished) – May 2017
4. Statement of Jacqueline Wilson to Commission of Inquiry – dated 21 Sept 2022.

**Appendix 1: Timeline**

<b>Date</b>	<b>Issue/ resolution</b>
Late 2015	Scientist first raises slide density discrepancy issue with Manager.
March 2016	Email to Howes discussing persistent nature of discrepancy. First provided written documentation of issue.
May to July 2016	Multiple additional occasions where the microscopy issue was identified was raised with managers.  Potential avenues for investigation raised with managers, including offers to provide staff to progress investigation.
May 2016	ER manager tasked to look into issue further.
02/06/2016	Initial project request
19/07/2016	Senior staff flag lack of urgency in addressing issue
8/8/16	Change in procedure to diff extract every sample suspected of containing sperm, with diff slide to be read as a matter of course.
August 2016	Initial Project plan written.  Project #181 Part 1 starts
02/05/2017	Part 2 proposal signed
22/05/2018	Part 3 (signed 05/06/2018)
13/02/2019	Part 4 (signed 8/3/2019)
14/06/2019	Part 4 – amended (signed 1/7/19)
22/7/2020	Final report (signed 5/8/2020)

**APPENDIX A – CURRICULUM VITAE (see attached)**

**APPENDIX B – INSTRUCTIONS AND INDEX TO MATERIAL (see attached)**

