From Lab to Verdict

Navigating the Intersection of Forensic Science, Media, and Legal Decision-Making

The DNA Analyst's Perspective

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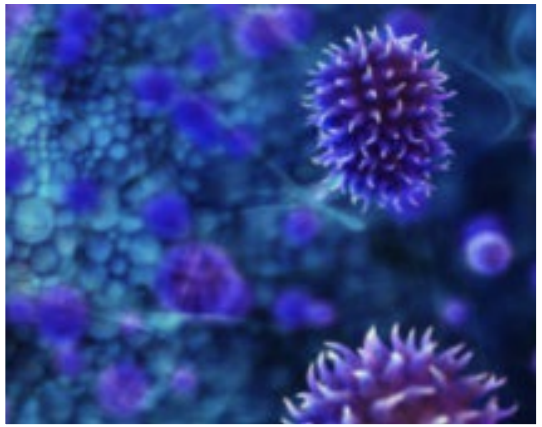
Technical Leader

Signature Science Forensic DNA Laboratory • Austin, Texas

Introduction to Signature Science

- Multi-disciplinary scientific services company since March 2001
  - ~200 employees in four locations across the U.S.
- Forensics/public safety
- Biosecurity & emerging threats
- Infection disease modeling/forecasting
- Chemical threat collection and detection
- Lab QA/data science/bioinformatics
- CBRNE training/exercises



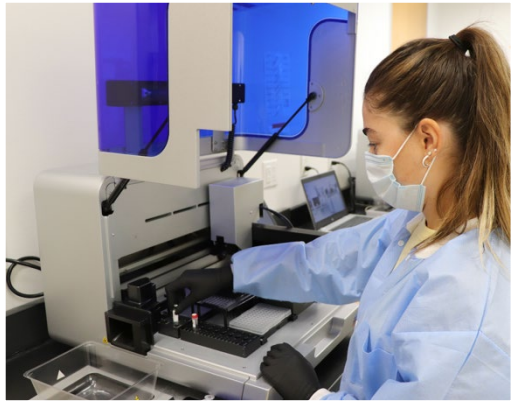
Center for Advanced Genomics™

- FGG Laboratory based in Charlottesville, Virginia
  - ForenSeq® Kintelligence / MiSeq FGx™
  - Infinium® Global Screening Array (GSA) / Illumina iScan™
  - Whole genome sequencing / Illumina NextSeq™ 200

[GEDmatch] PRO™

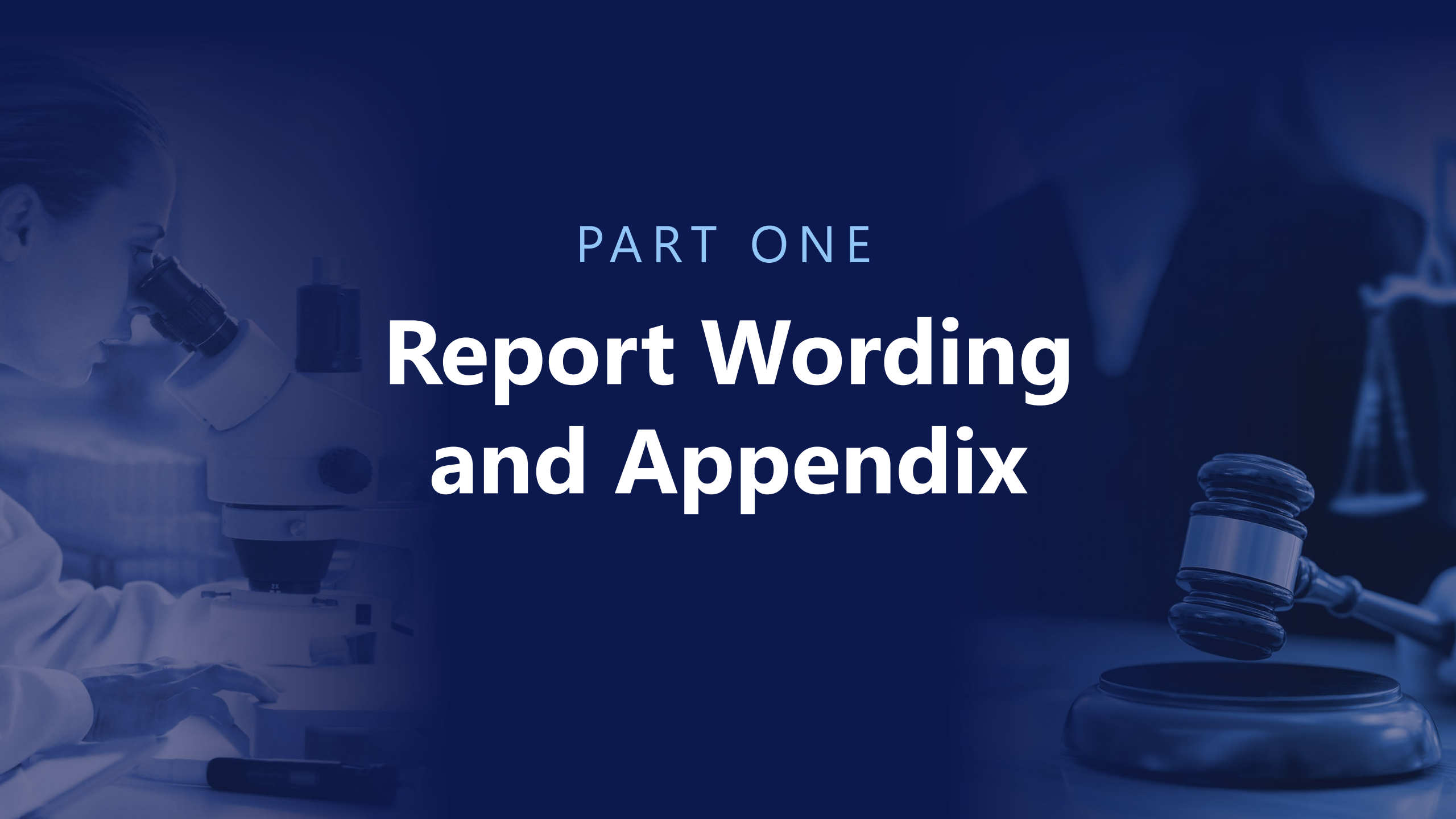
Center for Advanced Genomics™

- Forensic DNA casework laboratory based in Austin, Texas
  - ~30 employees
    - 16 DNA Analysts (9 remote)
    - 8 DNA Technicians
  - Applied Biosystems GlobalFiler™ and Yfiler™ Plus
  - Qiagen Investigator®24plex
  - Coming in 2025, Promega PowerPlex® Fusion 6C
  - Probabilistic genotyping - STRmix™ or manual interpretation



Focus Topics

Part One	**Report Wording and Appendix**
Part Two	**Explanation of Lab Results by the DNA Expert**
Part Three	**Future Considerations**



PART ONE

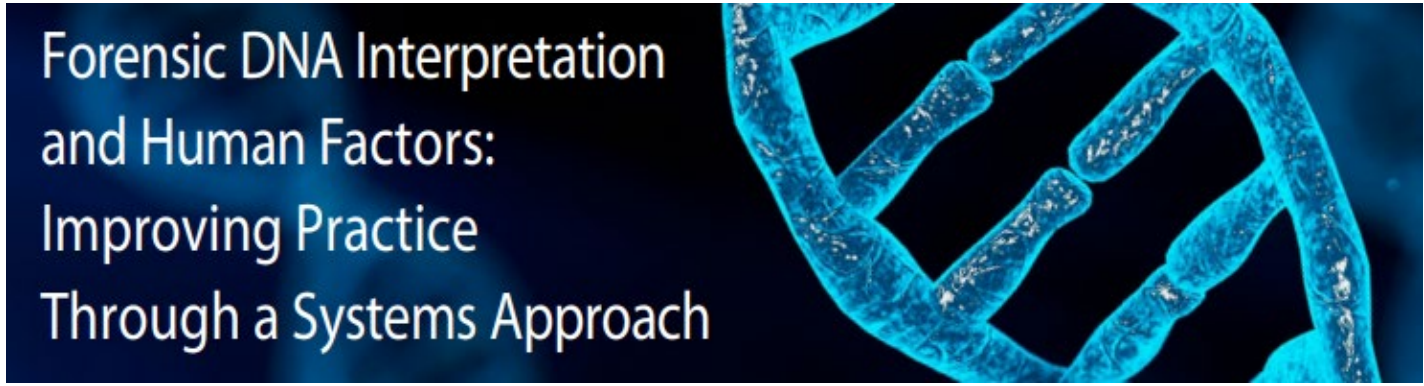
Report Wording and Appendix

Report Appendix

- Goes in every report, regardless of results
- Purpose →
  - Add common statements to condense what analysts had to manually add to different sections of the report
  - Define common serological and DNA terms
  - **Provide limitations to the testing performed**

Report Appendix

- Satisfies SigSci report writing SOP requirement
  - *The conclusions should clearly state appropriate qualifications or limitations on the evidence interpretation*



- NISTIR 8503 (https://doi.org/10.6028/NIST.IR.8503)
  - Published in May 2024
  - Report of the Expert Working Group on Human Factors in Forensic DNA Interpretation

Report Appendix

Appendix A

Data reported in this case were determined by procedures that have been validated according to standards established by the Scientific Working Group on DNA Analysis Methods (SWGDAM) and adopted as Federal Standards.

All items submitted were received properly sealed unless otherwise noted and described in the case record.

IDENTIFICATION OF BLOOD AND SEMEN

A presumptive test is a non-confirmatory test used for detecting the possible presence of a biological fluid.

A confirmatory test is a test that verifies the presence of a biological fluid.

A negative presumptive or confirmatory test may mean the biological fluid is not present in the tested sample or not present above the detection limit of the test.

DNA TESTING

DNA testing involves several steps, including DNA extraction, DNA quantitation, PCR amplification, and separation and detection of DNA. Appropriate positive and negative controls are used throughout the analysis.

When semen is present or possibly present on an item, the item is extracted using a two-step method referred to as a Differential Extraction that first recovers DNA from non-sperm cells (designated E) and then recovers DNA from sperm cells, if present (designated S). Incomplete separation can occur and fractions may contain both non-sperm cell DNA and sperm cell DNA. This terminology does not imply the presence or absence of spermatozoa in this case.

DNA isolated from all tested items was quantified using the Qiagen Investigator Quantiplex Pro Kit. DNA quantitation is an estimation of the amount of DNA in a sample. The absence of DNA may mean that DNA is not present in the tested sample or not present above the detection limit of the quantitation assay. Background fluorescence from the instrument used to quantitate DNA or fluctuations within the standard curve used to estimate the amount of DNA may result in detectable signal(s) that indicate low level amount(s) of DNA in a sample in the absence of DNA.

In the absence of assumed contributors for applicable items, the analyst evaluated the unknown item(s) to identify characteristics (e.g., alleles in a DNA profile) suitable for comparison and, if applicable, suitable for statistical rarity calculations prior to comparison to the known item(s).

Based on the DNA results detected in a sample, the number of contributors represents the best described number of individuals contributing to a sample as determined by the analyst.

The amount of DNA for the major contributor in a DNA mixture is present at a higher proportion than other contributors in the mixture.

The evaluation of a DNA comparison cannot conclusively identify an individual as the source of the DNA.

An exclusion to a DNA profile may mean an individual's DNA is not present in the tested sample or not present above the detection limit of the test.

This report does not provide any information about how or when the DNA was deposited.

STRMIX™

If STRmix™ was performed in this case, see the Component Interpretation table of STRmix™ Deconvolution Report for summary of potential individual contributors. Please contact this analyst if additional likelihood hypothesis testing is required.

STATISTICS

Random Match Probability, Combined Probability of Inclusion, and Likelihood Ratio statistics are generated using National Institute of Standards and Technology 1036 Revised U.S. allele frequencies (July 2017).

Y-STR statistics are calculated using the Y-Chromosome Haplotype Reference Database (YHRD) currently available from yhrd.org.

Table 1 below is adapted from recommendation 1.2 of the Scientific Working Group on DNA Analysis Methods (SWGDAM) Ad Hoc Working Group on Genotyping Results Reported as Likelihood.

Table 1. Scale of Verbal Qualifier for Reporting Likelihood Ratios (LR)

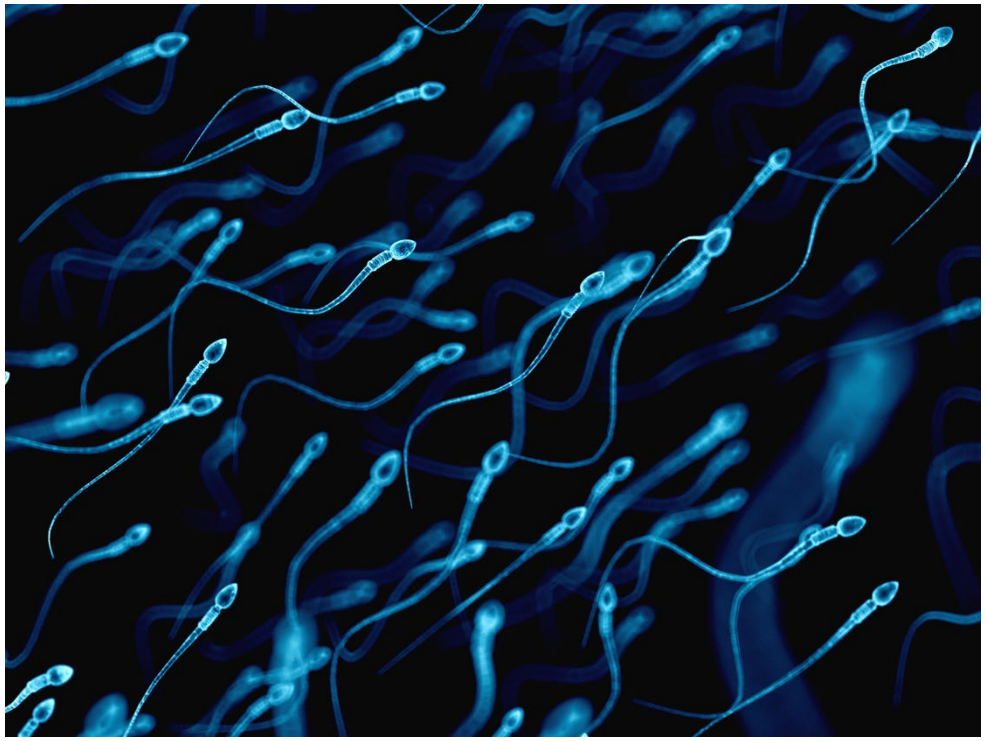
	Likelihood Ratios	Verbal Qualifier	
LR for H1 support	≥1,000,000	Very Strong Support	Increasing support in favor of H1 ↑
	10,000-999,999	Strong Support	
	100-9,999	Moderate Support	
	2-99	Limited Support	
Equal support for H1 and H2	1	Uninformative	Equal support for H1 and H2
1/LR for H2 support	2-99	Limited Support	Increasing support in favor of H2 ↓
	100-9,999	Moderate Support	
	10,000-999,999	Strong Support	
	≥1,000,000	Very Strong Support	

H1: The DNA originated from the person of interest and N-1 unknown individuals, where N is the number of contributors in the profile

H2: The DNA originated from N unknown individual(s), where N is the number of contributors in the profile

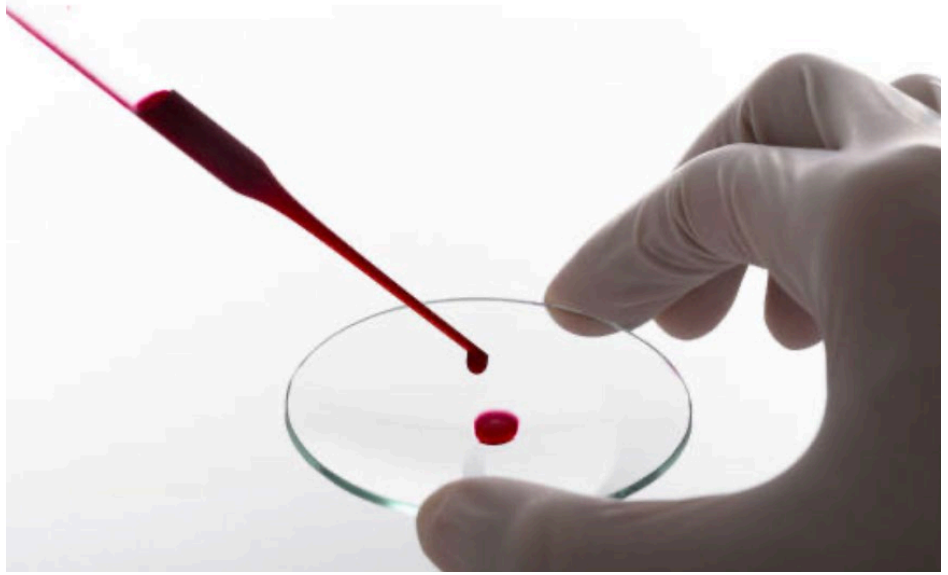
Reporting — Semen Example

- A ***presumptive test*** for the presence of semen was negative on the following item(s):
- Semen was indicated on the following item(s); however, no spermatozoa were identified to ***confirm*** the presence of semen:
- Spermatozoa were identified on the following item(s) thus ***confirming*** the presence of semen:



Appendix — Identification of Blood and Semen

- A **presumptive test** is a non-confirmatory test used for detecting the possible presence of a biological fluid.
- A **confirmatory test** is a test that verifies the presence of a biological fluid.
- A negative presumptive or confirmatory test may mean the biological fluid is not present in the tested sample or not present above the detection limit of the test.



Reporting — Differential Extraction Example

- 1B.1-S Vaginal swabs (sperm cell fraction)

  The DNA profile obtained from this item was interpreted as a single source male DNA profile.

  Note: SigSci opted to add the limitations of sperm fraction and non-sperm fraction terms to the Appendix based on NIST 8503 Recommendation 5.2.

Appendix — DNA Testing → Differential Extraction

- When semen is present or possibly present on an item, the item is extracted using a two-step method referred to as a Differential Extraction that first recovers DNA from non-sperm cells (designated E) and then recovers DNA from sperm cells, if present (designated S). Incomplete separation can occur and fractions may contain both non-sperm cell DNA and sperm cell DNA. This terminology does not imply the presence or absence of spermatozoa in this case.

Reporting — Stop at Quant Example

- After interpretation of quantification results, no further testing was performed on the following item(s) because no human DNA was detected:

  1D.1-S Anal swabs (sperm cell fraction)

Appendix — DNA Testing → Quantitation

- The absence of DNA may mean that DNA is not present in the tested sample or not present above the detection limit of the quantitation assay. Background fluorescence from the instrument used to quantitate DNA or fluctuations within the standard curve used to estimate the amount of DNA may result in detectable signal(s) that indicate low level amount(s) of DNA in a sample in the absence of DNA.

Reporting — DNA Profile Results

- The DNA profile obtained from this item was interpreted as a single source female DNA profile. John Doe is excluded as the contributor of this single source DNA profile.

Appendix — DNA Testing → DNA Profile Results

- Based on the DNA results detected in a sample, the number of contributors represents the best described number of individuals contributing to a sample as determined by the analyst.
- The evaluation of a DNA comparison cannot conclusively identify an individual as the source of the DNA.
- An exclusion to a DNA profile may mean an individual's DNA is not present in the tested sample or not present above the detection limit of the test.
- **This report does not provide any information about how or when the DNA was deposited.**

Appendix — Pros and Cons

Pros

- Eliminates the need to add specific statements to a report based on the type of testing and results (e.g., adding differential extraction statement only when the samples in the case are differentially extracted)
- Gives a basic overview of serology and DNA testing
- Provides limitations of testing if the analyst is not called to testify (e.g., plea deal, stipulation to DNA results by defense)

Cons

- At the end of report so may be missed by readers
- Includes information that may not apply to that specific case, which can lead to confusion
- Adds potential to be questioned on topics that do not pertain to that specific report

Discussion

- Does your laboratory add any kind of limitations in your report?
  - If so, how are the limitations captured?
  - Any additional limitations your lab reports contain that were not covered here?
- If your laboratory has an appendix, how has it been received by customers?
  - Is it ignored? Is it discussed?
  - Have you ever been cross-examined about it on the stand?

PART TWO

Explanation of Lab Results by the DNA Expert

Case Scenario

- A woman was walking in the park and was pulled off the walkway into a grassy area by an unknown male. He grabbed the ends of her scarf and choked her with it as he sexually assaulted her. She believes he was wearing a condom.

Lab Results

- The sexual assault kit yielded no or insufficient male DNA. No semen was found on the scarf. The ends of the scarf were sampled for DNA. The DNA profile obtained from the scarf sample was interpreted as a mixture of three individuals with at least one male contributor. Both the victim and a suspect were compared to the scarf sample.
  - Obtaining this mixture profile is approximately 22 quadrillion times more likely if the DNA originated from Jane Doe (victim) and two unknown, unrelated individuals than if the DNA originated from three unknown, unrelated individuals.
  - Obtaining this mixture profile is approximately 700,000 times more likely if the DNA originated from Joe Schmoe (suspect) and two unknown, unrelated individuals than if the DNA originated from three unknown, unrelated individuals.

Quantitation Results

Lab Result

- The sexual assault kit yielded no or insufficient male DNA.

Explanation

- We determine how much human DNA, which is female and male DNA, is in a sample, in addition to just male DNA.
- In a sexual assault case with a female victim and a male suspect, the amount of male DNA in the sample is the determining factor for if the sample proceeds on in the DNA testing process.
- In this case there was no male DNA or too small of an amount of male DNA based on thresholds established by the laboratory.

Serology Results

Lab Result

- No semen was found on the scarf.

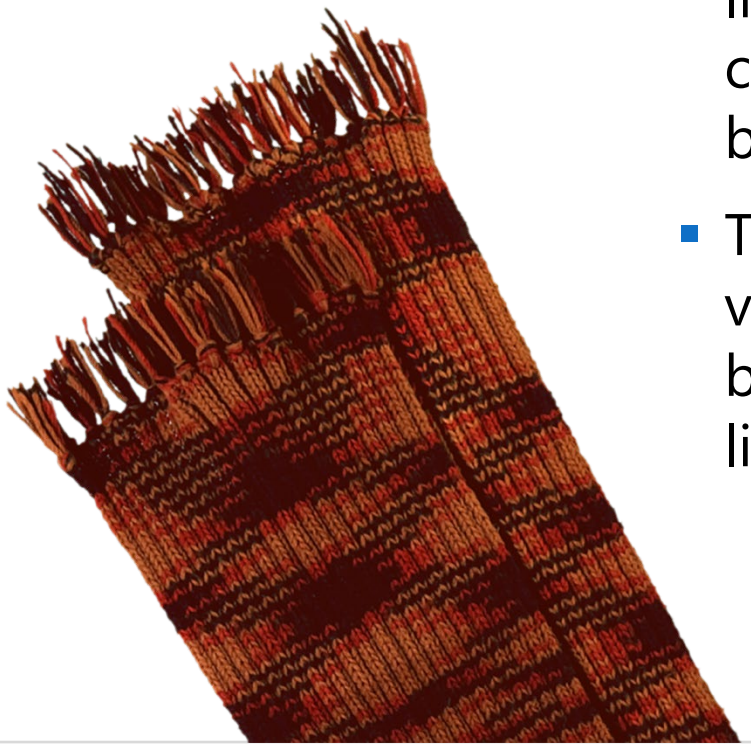
Explanation

- For sexual assault cases with male suspects, semen is commonly tested for since body fluids such as semen typically have a higher concentration of DNA.
- In this case, no semen was found which means semen is not present in the tested sample or not present above the detection limit of the test.
- It could also mean there were stain(s) present that were too small to be visible to the naked eye.

Sampling for DNA

Lab Result

- The ends of the scarf were sampled for DNA.



Explanation

- In the absence of semen, an item can be sampled for other sources of DNA, like DNA that is left behind when you contact an item, such as DNA left behind from wearing an item.
- The scarf was used to choke the victim, so the ends were sampled because that would be the areas most likely touched by the suspect.

DNA Results

Lab Result

- The DNA profile obtained from the scarf sample was interpreted as a mixture of three individuals with at least one male contributor.

Explanation

- A mixture is when you have more than one individual in a sample.
- In this case, the sample was interpreted as a mixture of three contributors with at least one male contributor.
- The true number of contributors is never known; this number of contributors represents the best described number of individuals determined based on analyst training and experience.

Statistics

Lab Result

- Both the victim and a suspect were compared to the scarf sample.

Explanation

- A visual comparison was performed between the DNA profiles from individuals of interest to the mixed DNA profile on the scarf.
- An expert software was used to generate a statistic known as a likelihood ratio.
- The analysis performed by the expert software was evaluated to ensure it was intuitive based on the visual comparisons made to the DNA profiles.

Statistics

Lab Result

- Obtaining this mixture profile is approximately 22 quadrillion times more likely if the DNA originated from Jane Doe (victim) and two unknown, unrelated individuals than if the DNA originated from three unknown, unrelated individuals.

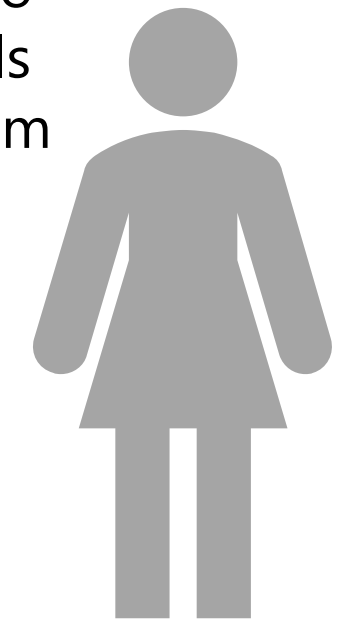
Explanation

- First explain the statistic
  - A likelihood ratio is a ratio of probabilities that looks at two competing scenarios and gives a numerical value that gives strength of support for one scenario over another.
  - In the case of DNA, in simple terms, it is looking to see, given the evidence, if it is more likely that the DNA originated from a person of interest than if the DNA originated from an unknown, unrelated person.

Statistics

Lab Result

- Obtaining this mixture profile is approximately 22 quadrillion times more likely if the DNA originated from Jane Doe (victim) and two unknown, unrelated individuals than if the DNA originated from three unknown, unrelated individuals.



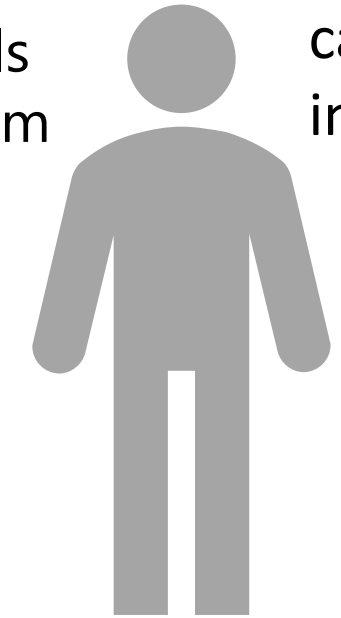
Explanation

- State statistic
- Put 22 quadrillion into perspective
  - Number of zeroes?
  - Explain verbal scale?
  - Go into validation trends (e.g., non-contributor study)?

Statistics

Lab Result

- Obtaining this mixture profile is approximately 700,000 times more likely if the DNA originated from Joe Schmoe (suspect) and two unknown, unrelated individuals than if the DNA originated from three unknown, unrelated individuals.



Explanation

- State statistic
- Put 700,000 into perspective
- The evaluation of a DNA comparison cannot conclusively identify an individual as the source of the DNA.

This comparison cannot explain how or when the DNA was deposited.

Other Relevant Information

- The DNA expert is there to explain to the jury what DNA can and cannot tell you
- Educate on DNA transfer
- What can be said about DNA transfer related to the case?
  - NISTIR 8503 and Texas Forensic Science Commission (TFSC) Recommendations

DNA Transfer Related to the Case

- Activity level propositions
  - Recommendations in NISTIR 8503
    - Defines activity-level propositions as “Statements that are formulated to help answer questions related to disputed activities and the presence or absence of biological material.”
  - Recommendations made by TFSC in response to TFSC complaint No. 23.67; TIFFANY ROY; (TIMOTHY KALAFUT, PH.D.; EVALUATION OF BIOLOGICAL/DNA RESULTS GIVEN ACTIVITY LEVEL PROPOSITIONS)

NISTIR 8503

Recommendations for DNA analysts:

- Avoid discussing the possibility of direct or indirect transfer in a case
- "How" and "When" questions should only be answered by those appropriately trained and are distinct from the "Who" question that is what the typically trained DNA analyst can answer

https://doi.org/10.6028/NIST.IR.8503

Table 7.1: Proposed responses to questions about how or when the DNA was deposited

Example of Questions Posed to DNA Experts	Proposed Ways for the Expert to Respond
In your opinion, is direct transfer more likely than indirect?	• DNA analysis does not allow a scientist to directly answer how the DNA was deposited (direct or indirect transfer). The DNA results presented in my report regard the comparison of DNA profiles and can only help answer questions about whose DNA may be present or not. • My testimony about the value of the DNA comparison is only meaningful to help the jury determine who the source of the DNA was. That testimony does not provide any information that addresses the issues of how or when.
Could this [alleged activity] have happened?	• Offering an opinion on this question would amount to speculating on what is alleged. It is not my role as a scientist to speculate about or determine what happened.
Is it possible that the DNA was deposited when the Person of Interest (POI) [engaged in an activity at the scene prior to or after the alleged event]?	• It is not my role to discuss the possibility of the alleged event (or any other event). My expertise is based upon DNA profile comparisons which can only assist in helping you answer questions about whose DNA is present or not. • Agreeing that something is "possible" is not the same as offering an opinion about the probability of the results in the context of case-specific circumstances. • Discussing whether something is possible does not help me convey the significance of the results in the context of this case. For example, getting struck by lightning or flipping a coin and getting "heads" are both possible but have very different probabilities.
Are there other explanations for the presence (or absence) of this DNA?	• It would be inappropriate and speculative for me to discuss why the DNA was or was not detected. • Answering this question would not allow me to convey a balanced assessment of the findings in the context of this case. • The only way I can evaluate the results is by considering at least two opposing views.

Kaitlin Armstrong Murder Trial

https://www.youtube.com/watch?v=wiSxFoBVlzY

TFSC Complaint No. 23.67

- "The DPS analyst…testified the DNA evidence was 224 billion times more probable if Wilson, Armstrong, and an unrelated, unknown person were contributors to the mixture from the bicycle seat than if Wilson and two unrelated, unknown people were contributors."
- Complaint filed regarding Dr. Timothy Kalafut's testimony in the Kaitlin Armstrong murder trial in Texas
  - "In rebuttal, the State called Dr. Timothy Kalafut, and his testimony included an evaluation of the DNA evidence given competing activity level propositions prepared in advance of trial. Dr. Kalafut opined that the DNA evidence was 'much more likely' if Armstrong had a direct interaction with the bicycle than if the DNA was transferred through a series of indirect activities."
  - References NISTIR 8503 Report

https://www.txcourts.gov/media/1458950/final-report-complaint-2367-roy-tiffany-073024_redacted.pdf

TFSC Complaint No. 23.67 Recommendations

- Recommendations made in the following areas:
  - Appropriate responses to hypothetical questions regarding activity in a case
  - Evaluating foundational basis of evaluations given activity level propositions
  - Education/training; quality control; reporting/testimony
  - Expectations for experts outside of accredited laboratories testifying in Texas

TFSC Complaint No. 23.67 Recommendations

- A diverse group of agencies (NIST, legal, statistics, human factors experts, etc.) should "conduct a scientific foundational-type review including a public comment phase, to evaluate and report on the state of published literature and offer recommendations"
- Recommendations also address implementation in the areas of education, training, quality assurance, reporting, and testimony

TFSC Complaint No. 23.67 Recommendations

- Of note, TFSC will request that ANAB and A2LA add the following to the Texas accreditation checklist

(1) When asked hypothetical questions that require the consideration of transfer, persistence, prevalence, and recovery (TPPR) testifying witnesses should endeavor to communicate that the DNA comparison results (or lack thereof) do not answer the questions of "how" or "when" DNA was deposited or speak to its absence. Such testimony could potentially lead to evidence being misleading, overvalued, or undervalued.

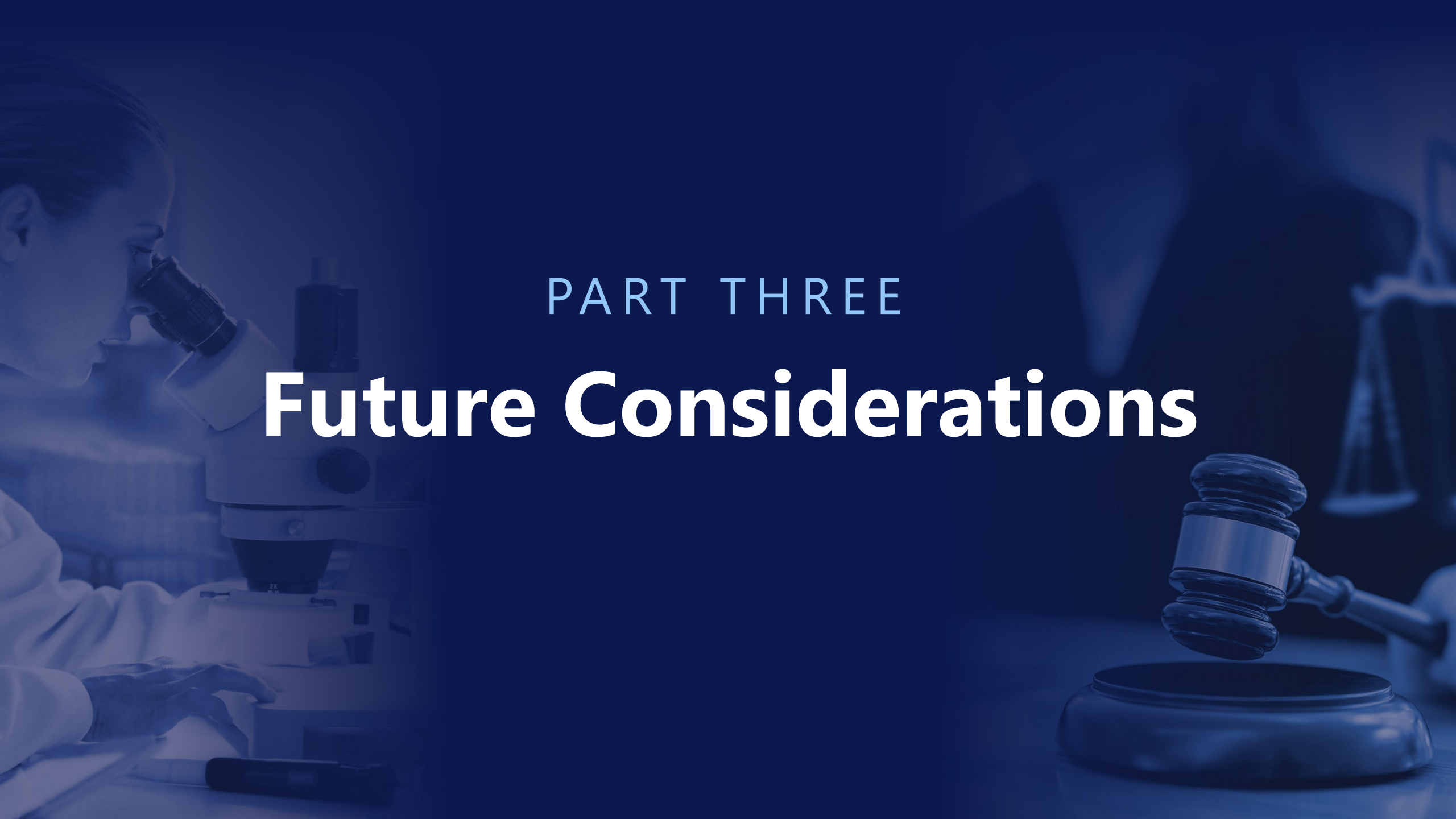
TFSC Complaint No. 23.67 Recommendations

- Of note, TFSC will request that ANAB and A2LA add the following to the Texas accreditation checklist

(2) Whenever possible, testifying experts should reiterate that while they may be able to provide limited general information about TPPR, answering questions about how or when the DNA was deposited (or is absent) in the particular case, is outside the testifying witness' purview. To help address questions about how or when the DNA was deposited in the case, a separate evaluation would be needed.

Discussion

- How do you put a likelihood ratio into perspective?
- How are labs handling questions on DNA transfer?
- What kind of training do you have for transfer testimony?
- Do you agree with the NIST/TFSC recommendations?



PART THREE

Future Considerations

NISTIR 8503 Recommendation 4.2

- Express likelihood ratios as an order of magnitude or to one significant figure

CURRENT WORDING

Obtaining this mixture profile is approximately **45,300** times more likely if the DNA originated from Wonder Woman and one unknown, unrelated individual than if the DNA originated from two unknown, unrelated individuals.

POSSIBLE NEW WORDING

Obtaining this mixture profile is approximately **40,000** times more likely if the DNA originated from Wonder Woman and one unknown, unrelated individual than if the DNA originated from two unknown, unrelated individuals.

NISTIR 8503 Recommendation 4.3

- Implement a cap to the statistic that is being reported (**1 billion for LR**/1 in 1 billion for RMP/CPI)

CURRENT WORDING

Obtaining this mixture profile is **approximately 93.5 quintillion** times more likely if the DNA originated from Superman and two unknown, unrelated individuals than if the DNA originated from three unknown, unrelated individuals.

POSSIBLE NEW WORDING

Obtaining this mixture profile is **estimated to be greater than 1 billion** times more likely if the DNA originated from Superman and two unknown, unrelated individuals than if the DNA originated from three unknown, unrelated individuals.

NISTIR 8503 Recommendation 4.4

- Clearly report that the propositions are reversed when reporting 1/LR

CURRENT WORDING

Obtaining this mixture profile is approximately 80.2 thousand times more likely if the DNA originated from two unknown, unrelated individuals than if the DNA originated from Batman and one unknown, unrelated individual.

POSSIBLE NEW WORDING

Obtaining this mixture profile is approximately 80.2 thousand times more likely if the DNA originated from two unknown, unrelated individuals than if the DNA originated from Batman and one unknown, unrelated individual. **This statistic indicates that the DNA results support the alternative proposition that only unknown, unrelated individuals, and not Batman, contributed to the DNA mixture.**

NISTIR 8503 Recommendation 5.2

- Remove terms that may be misinterpreted (e.g., ***major contributor***, sperm fraction)

CURRENT WORDING

- The DNA profile obtained from this item was interpreted as a mixture of two individuals with **a major male contributor.**
- Limited DNA results were obtained from the **minor component** of this mixture. These results are insufficient for comparisons to known reference samples; therefore, no further conclusions can be made.

POSSIBLE NEW WORDING

- The DNA profile obtained from this item was interpreted as a mixture of two individuals with **at least one male contributor. The DNA profile of a male contributor was determined and is suitable for comparison.**
- Limited DNA results were obtained from the **remaining contributor** to this mixture. These results are insufficient for comparisons to known reference samples; therefore, no further conclusions can be made.

NISTIR 8503 Recommendation 5.2

- Remove terms that may be misinterpreted (e.g., major contributor, ***sperm fraction***)

CURRENT WORDING

1B.1-**S** Vaginal swabs (**sperm cell fraction**)
1B.1-E Vaginal swabs (non-sperm cell fraction)

POSSIBLE NEW WORDING

1B.1-**1** Vaginal swabs (Fraction **1**)
1B.1-2 Vaginal swabs (Fraction 2)

OR

1B.1-**A** Vaginal swabs (Fraction **A**)
1B.1-B Vaginal swabs (Fraction B)

Discussion

- Have you incorporated any NISTIR 8503 recommendations into your reporting?
  - Expression of likelihood ratios
  - Cap on reported statistic
  - Reporting 1/LR
  - Removing terms that may be misinterpreted

Questions?

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512-583-2262

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