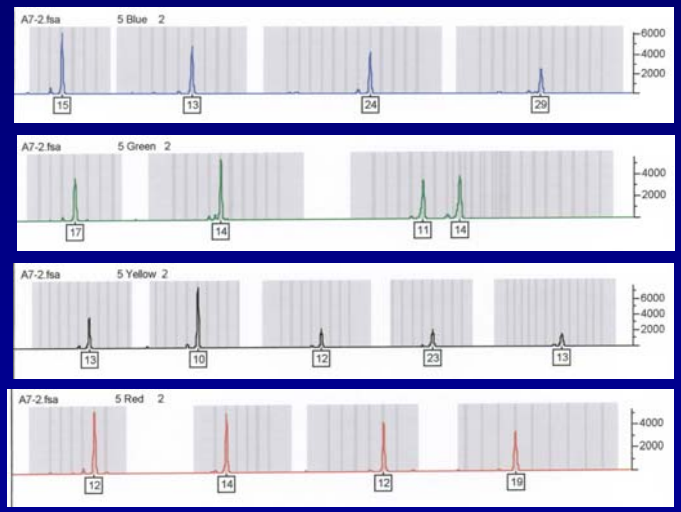
Demonstrating the Utility of the AmpFlSTR Yfiler PCR Amplification Kit While Conducting Internal Validation Studies



Future Trends in
Forensic DNA Technology
Phoenix, AZ
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Spencer Laboratories, Inc.

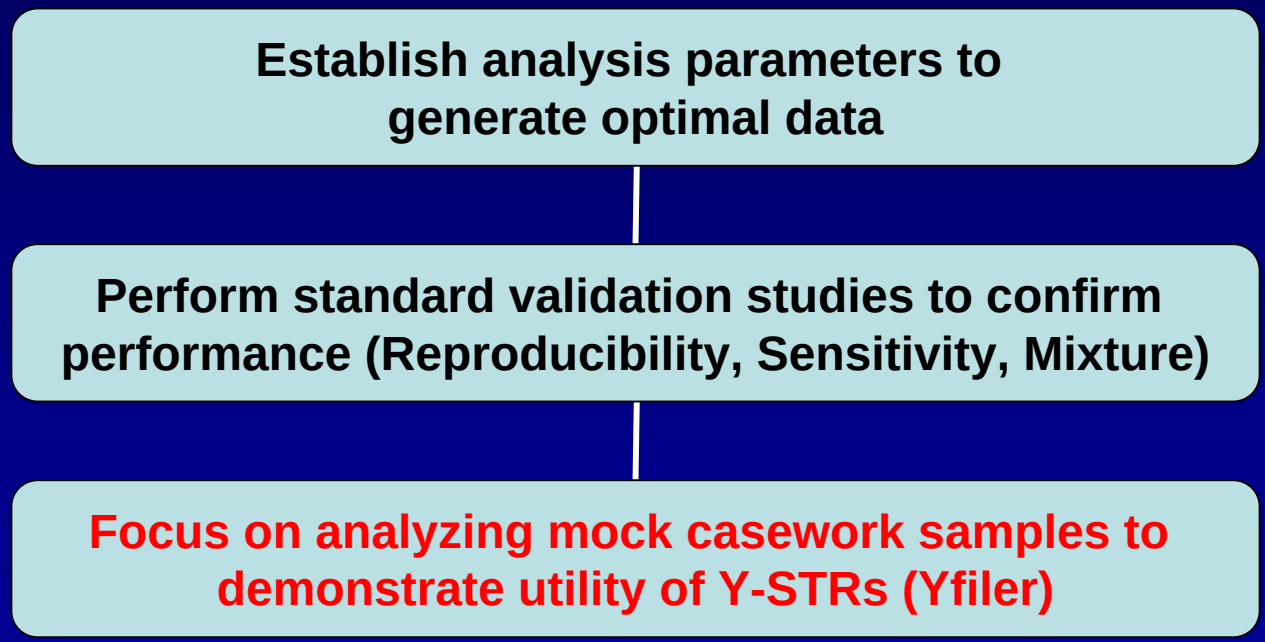
Presentation Overview

- Evaluation of the utility of Y-STRs (Yfiler)
- Development of validation strategy
- Highlights of internal validation studies
- Validation summary
- Casework example
- “Future trends”

Evaluation of Y-STRs

- **Major Forensic Advantage**
  - **Useful with mixtures of DNA containing a small quantity of male DNA in a larger background of female DNA (i.e. oral assaults, fingernail clippings from female victim)**
    - **These types of mixtures often yield complex, ambiguous or inconclusive results with autosomal STRs**
- **Major Forensic Limitation**
  - **Sharing of Y-STR haplotype among paternal male relatives; moderate discriminatory power among unrelated men**

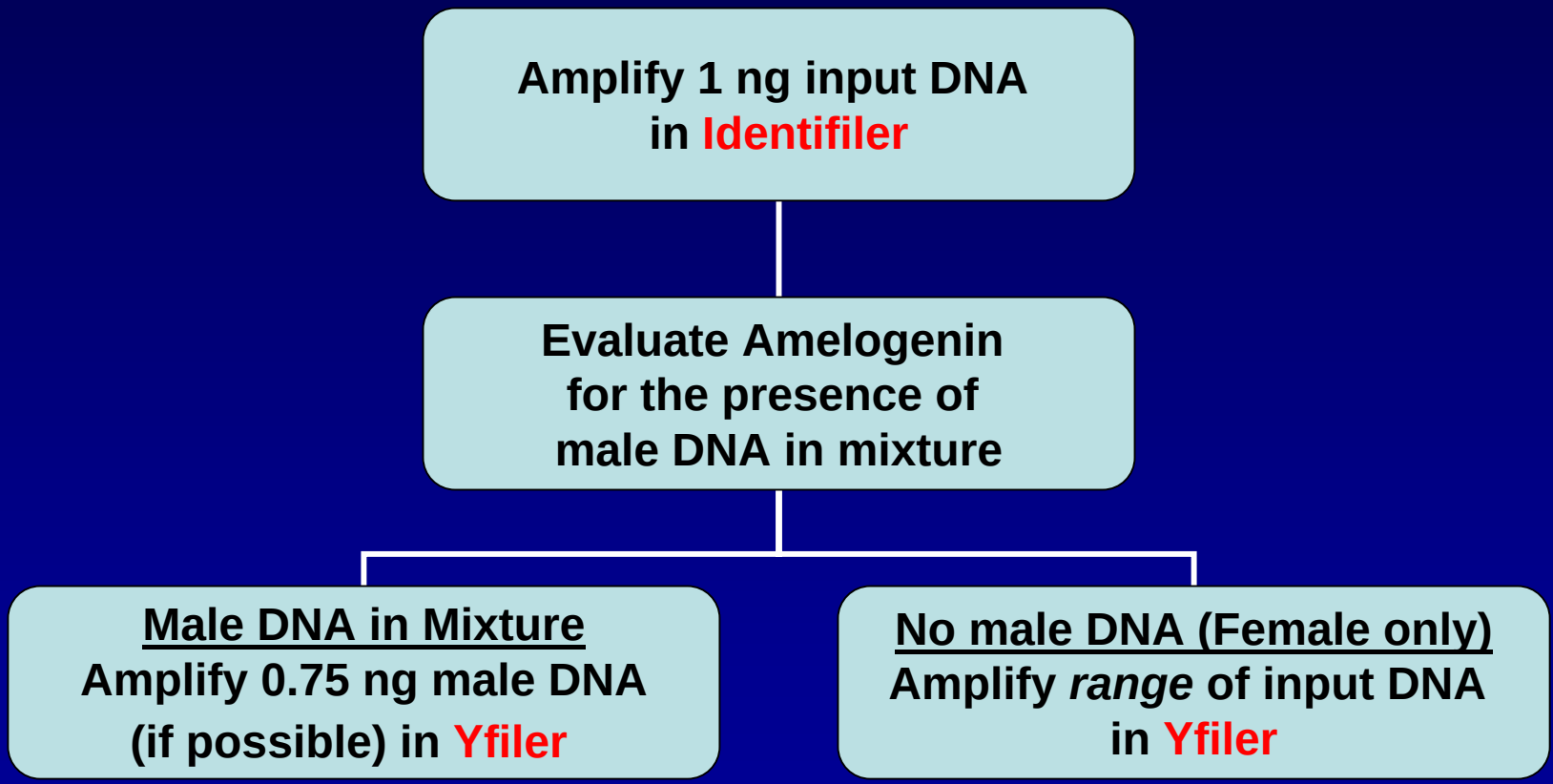
Development of Validation Strategy



Sample Preparation for Simulated Oral Assault Evidence

- Collected swabbings of neck and breasts of 2 females 30-60 minutes after each area was licked by male
- Performed DNA extraction according to standard organic extraction procedure
- Expecting high female: low male DNA mixture

Analysis Strategy



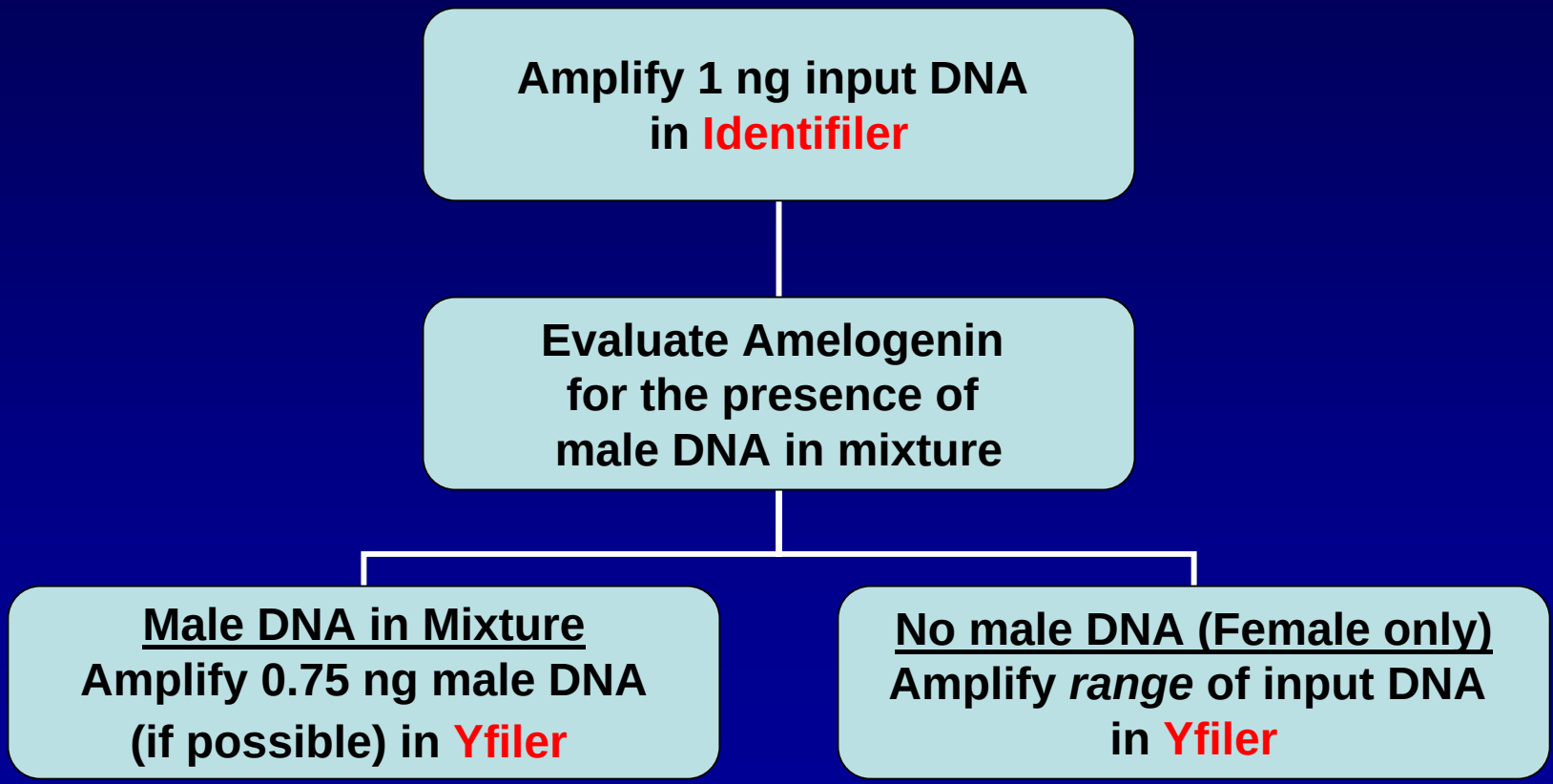
Identifiler Results for Simulated Oral Assault Evidence

- Single source male profile detected; no presence of mixture
- Yfiler analysis of samples would not demonstrate the utility of Y-STRs; samples were not amplified for Yfiler!

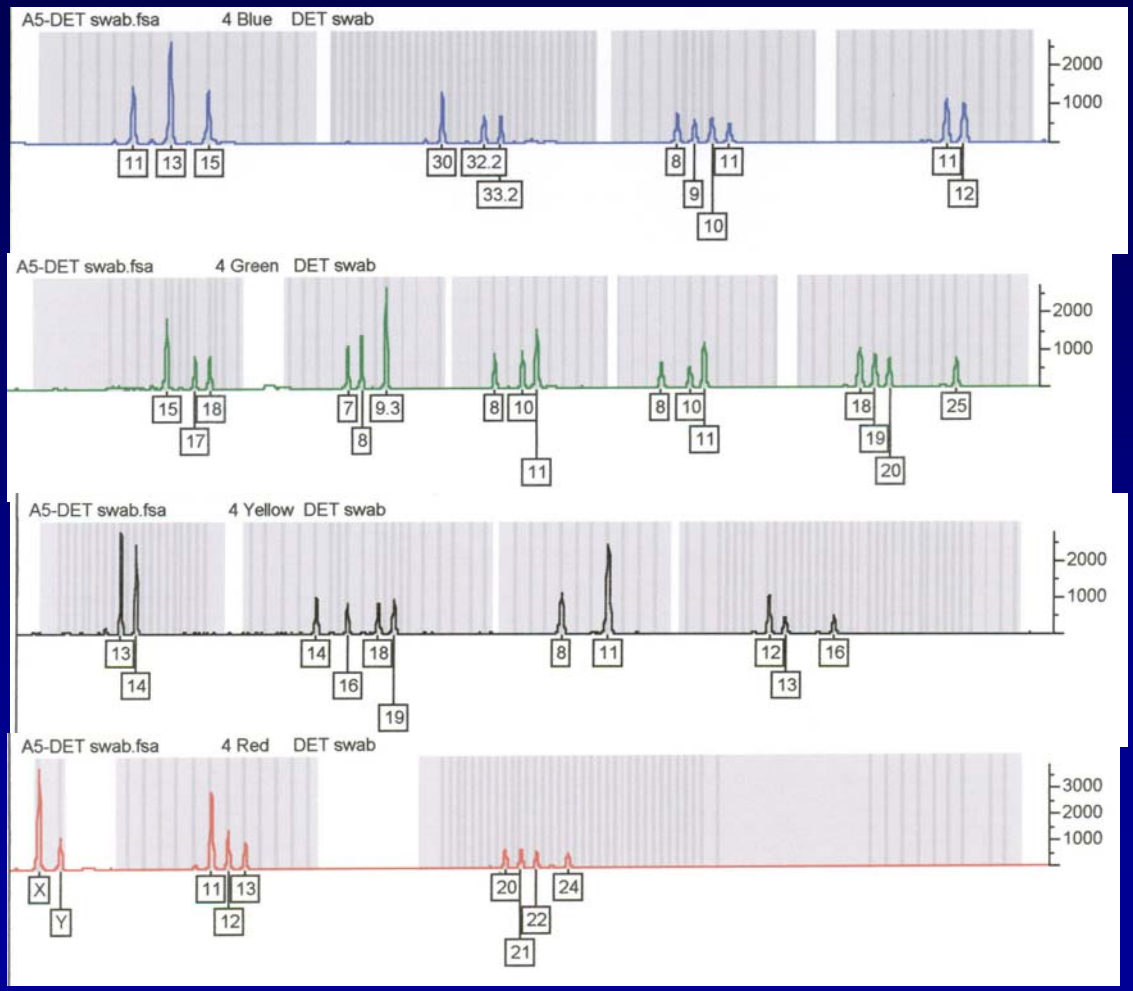
Sample Preparation for Simulated Sexual Assault Evidence

- Collected oral swab from 2 females
- Dipped each oral swab in very dilute semen
- Performed DNA extraction (w/o differential) to simulate a sperm cell fraction with a high degree of epithelial cell carryover

Analysis Strategy



Identifiler Analysis of Swab 1



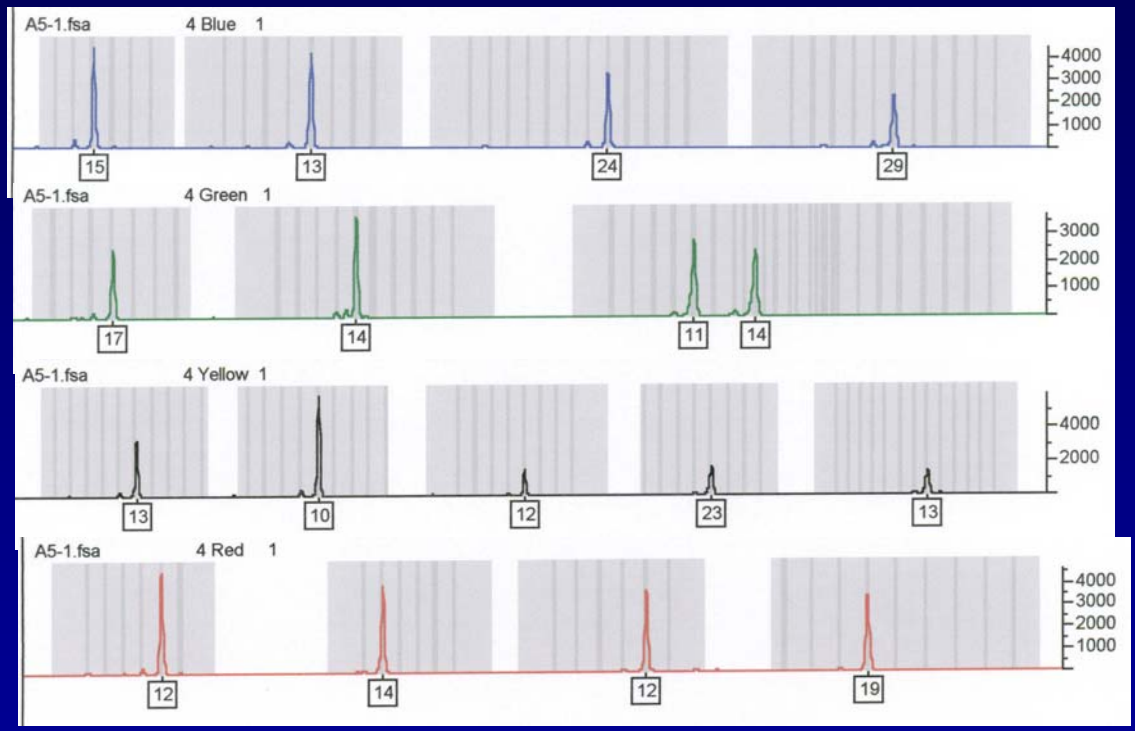
Identifiler Results for Swab 1

- Mixture of male and female detected
- All alleles from male contributor were detected

Yfiler Strategy for Swab 1

- **Based on evaluation of Amelogenin, male contributor was present at approx. 50%**
- **TOTAL DNA concentration determined by QuantiBlot was multiplied by 50% to arrive at the approx. concentration of *male* DNA in the extract**
- **0.75 ng male input DNA was targeted and amplified in Yfiler**

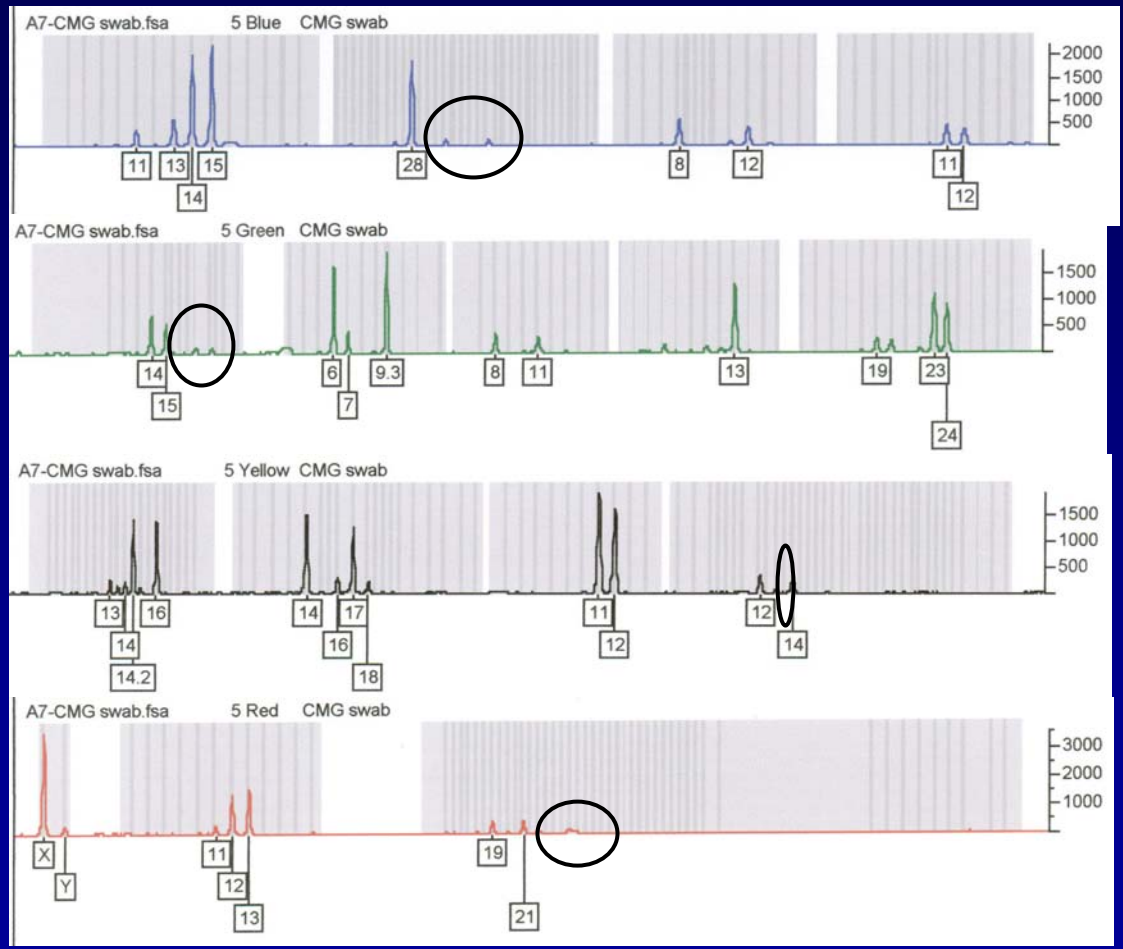
Yfiler Analysis of Swab 1



Yfiler Results for Swab 1

- Single source Y haplotype detected
- Consistent with known male contributor (data from reference not shown)
- “Cleaned-up” the ID data; simplified data interpretation

Identifiler Analysis of Swab 2



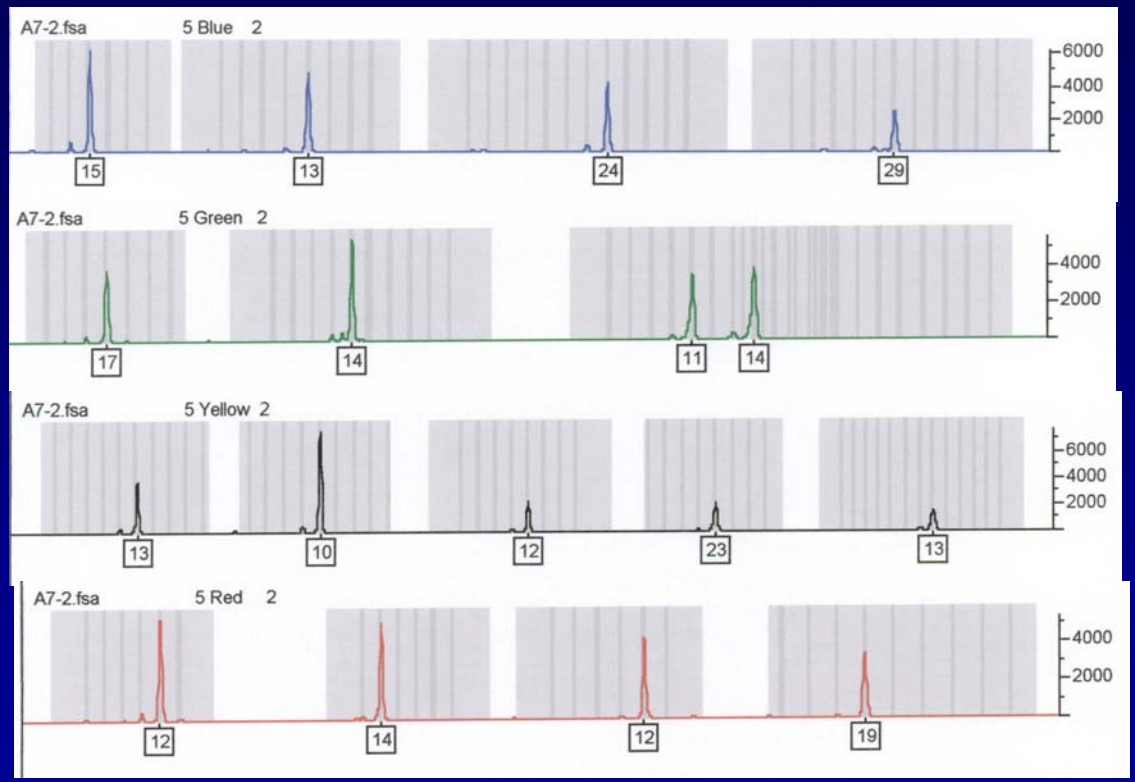
Identifiler Results for Swab 2

- Mixture of male and female detected
- Numerous alleles from male contributor were not detected (partial result; subject to more complex interpretation)

Yfiler Strategy for Swab 2

- **Based on evaluation of Amelogenin, male contributor was present at approx. 20%**
- **TOTAL DNA concentration determined by QuantiBlot was multiplied by 20% to arrive at the approx. concentration of *male* DNA in the extract**
- **0.75 ng male input DNA was targeted and amplified in Yfiler**

Yfiler Analysis of Swab 2



Yfiler Results for Swab 2

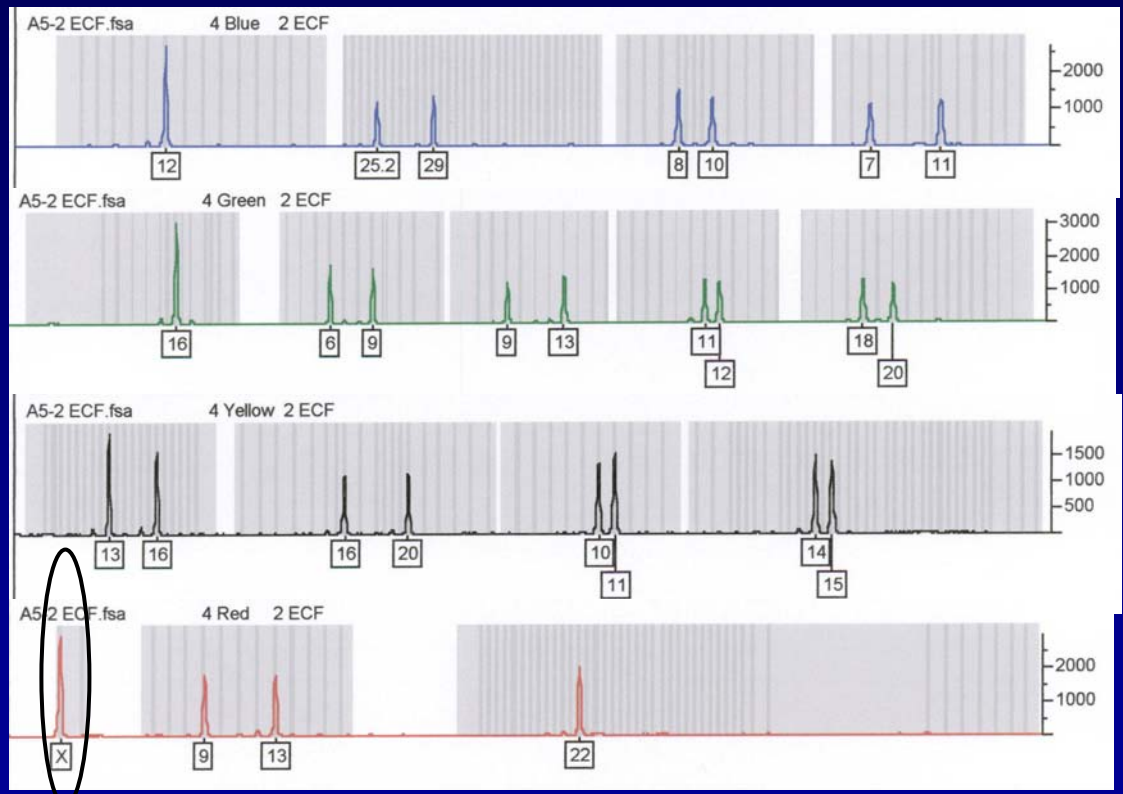
- **Single source Y haplotype detected**
- **Consistent with known male contributor (data from reference not shown)**
- **Simplified data interpretation; generated more *conclusive* result?**

Validation Summary

- Yfiler is a highly effective tool with complex mixtures of male and female DNA
- Studies with simulated casework samples (although difficult to make) were most helpful in determining true utility of assay
- Yfiler is currently best used in conjunction with Identifiler

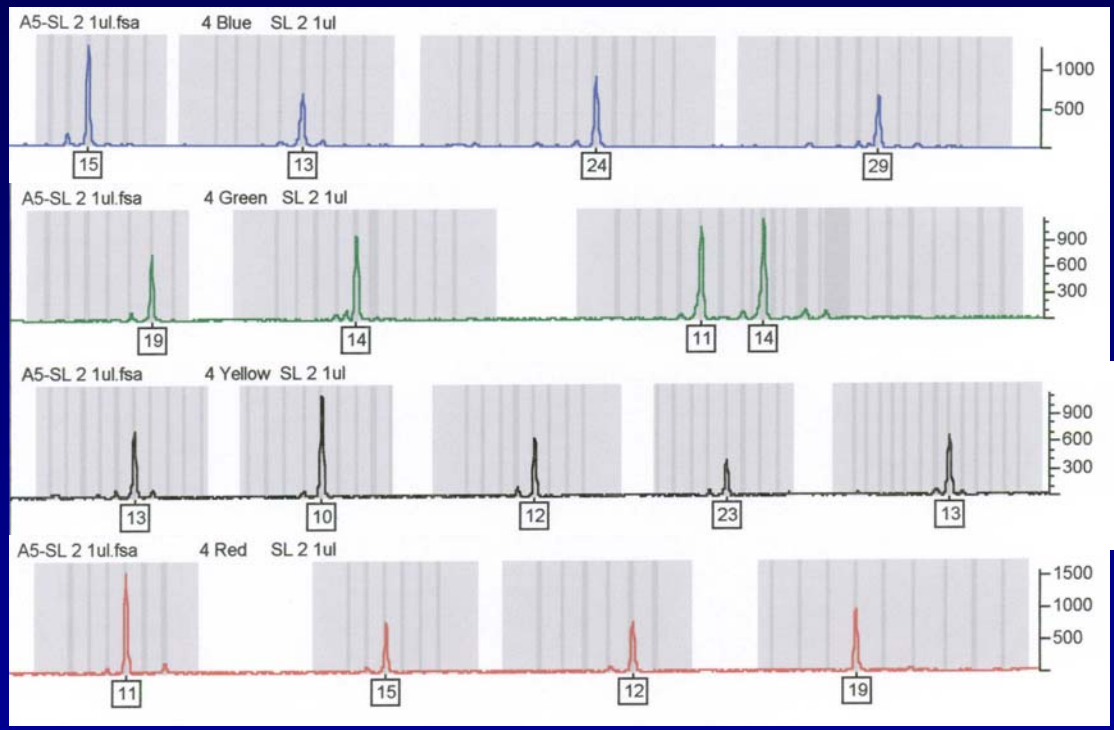
Identifiler Analysis of Casework Sample

* Lip/breast swabbing from female victim of oral assault



* Since no male DNA detected, a range of input DNA will be amplified in Yfiler

Yfiler Analysis of Casework Sample



*** Currently awaiting reference sample from suspect**

“Future Trends”

- **Use of male-specific quantitation, along with total DNA quantitation (Quantifiler, Quantifiler Y), to determine ratio of male:female DNA**

- **Choose appropriate DNA typing system**
  - **Autosomal STRs**
  - **Y-STRs**

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Thank you very much!