

# **Blood testing**

- Kastle-Meyer test
  - Indicates presence of blood, does not identify presence of blood
  - Color change test
  - Not species specific
  - Common items tested for blood: knives, clothing, scene swabs



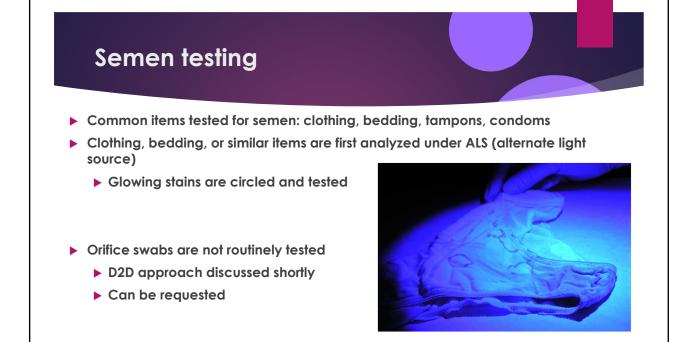


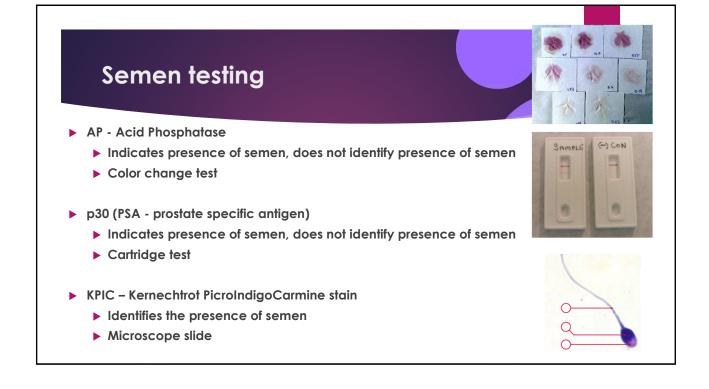


#### Blood testing in the field

- BLUESTAR and Leucomalachite Green are most popular
- We strongly encourage that you don't do this unless absolutely necessary
  - Once tested, it can not be retested
  - If stain is not marked, we will not know where to swab
- If you must...
  - Swab the glowing stain and submit the swabs
  - Circle the glowing stain before submitting to the lab









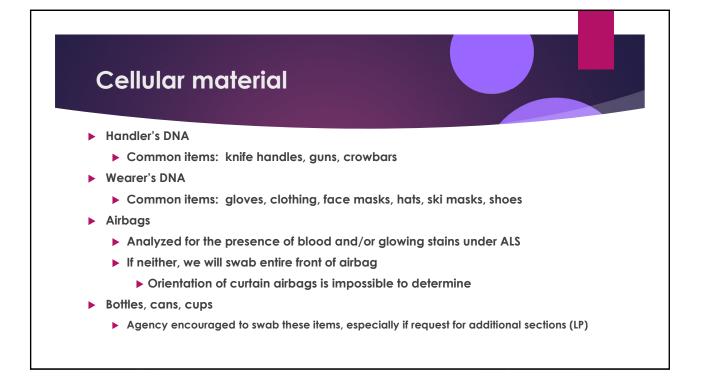
## To semen test...or not to semen test

- Direct to DNA (D2D) approach for orifice swabs
  - Swabs are not tested for the presence of semen but instead sent directly onto DNA analysis
  - More efficient and better turnaround times
  - Quantitation is a better indicator of sample information than biology analysis
  - No separate biology report
- Samples from kit are chosen based on case scenario
  - ▶ Generally, half of each probative sample is preserved for DNA
- If case question is not answered in the first round of D2D analysis, additional kit swabs or clothing may be sent for biology analysis and/or DNA
  - ► This will be reflected in the report

## Saliva testing

#### Phadebas test

- Indicates the presence of saliva, does not identify presence of saliva
- Color change test
- Not species specific
- Most common item tested for saliva: clothing in sexual assault cases based on scenario
- Testing is almost always not done on items thought to have saliva on them such as bottles, cups, cigarettes
  - > These items are swabbed/preserved and sent directly to DNA
  - Saliva testing will be taken into consideration if request is made up front, prior to analysis

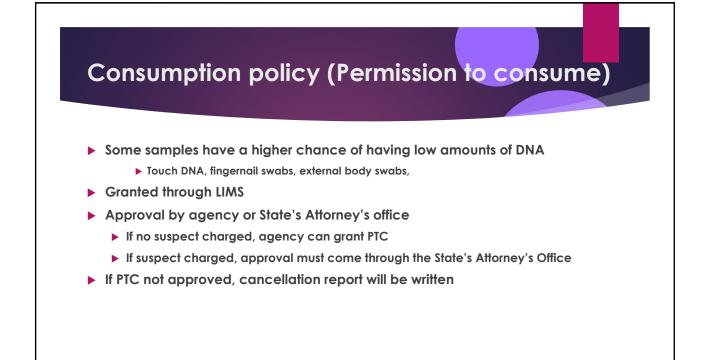


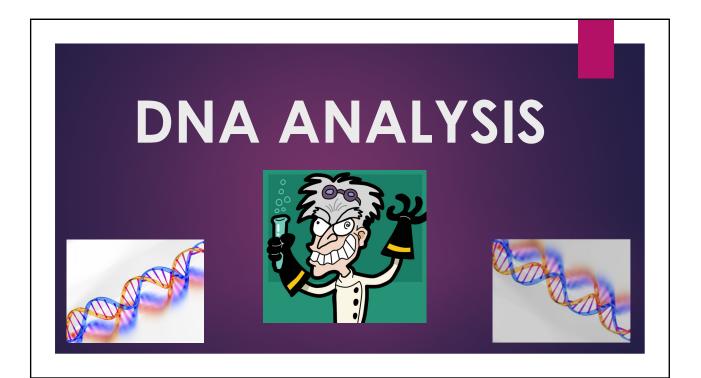
## Collection and storage of samples

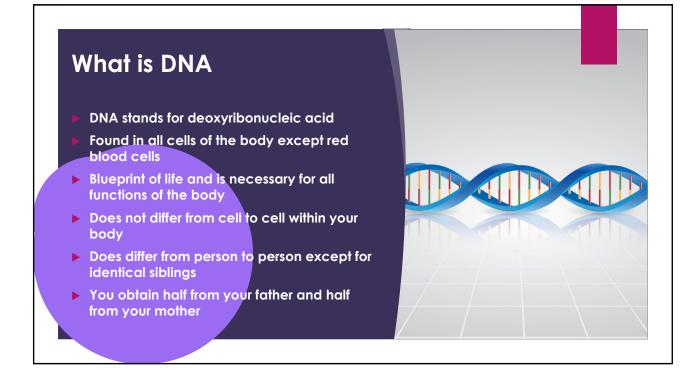
- Ensure Clean Technique is used during collection
  - Gloves, masks, sterile swabs/water
- Concentrate sample onto as few swabs as possible
  - > 1 swab for most items, can use 2 if area to be swabbed is large
- Allow swabs to dry completely
  - Store in paper, not plastic
  - Room temperature
  - Out of direct sunlight
- Ensure location collected/ownership properly documented
  - Location collected
  - Owner (if known)
  - Elimination standards

#### Scene swabs

- Car swabs
  - ► Limit submission to best evidence: steering wheel, gear shift, driver compartment
- Building/residence swabs
  - > Point of entry of private residence, items touched for a decent amount of time, blood
- Swabs not routinely accepted:
  - Items touched once for access or to be moved (door pulls, cords)
  - Drug packaging
  - Items taken from person if that's whose profile you are looking for
    - ► This includes their car and residence, unless additional information is provided
  - > Public area items (store door handles, counter tops, common housing areas)







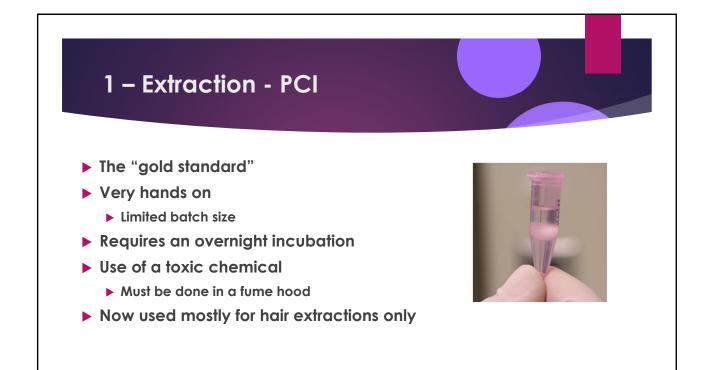
## What are the steps for DNA analysis

- ▶ 1 Extraction
  - ▶ Get the DNA off the sample
- 2 Quantitation
  - How much DNA is present
- 3 Amplification
  - Making copies of the DNA
- 4 Capillary Electrophoresis
  - Separate the DNA to create a profile



## 1 - Extraction

- ▶ The sample is soaked in a master mix of chemicals and exposed to heat
- ▶ This releases the cells from the samples and breaks the cell walls open
- ▶ The sample is "cleaned up" so that you end with a purified liquid DNA sample
- We use 3 main methods to complete extraction:
  - PCI
  - Maxwell robot
  - Tecan robot



## 1 – Extraction – Maxwell robot

- Samples are preprocessed to release the cells off the substrate and into a liquid
- This liquid is added to a cartridge and placed onto the instrument
- The instrument utilizes DNA IQ technology which uses magnetic beads that attract the DNA
- These beads move through the wells of the cartridge into a series of lysis and washes
- 26 minutes later, you have a purified DNA sample
- Each instrument can process 48 samples
  - Chicago has 6 Maxwell 48s

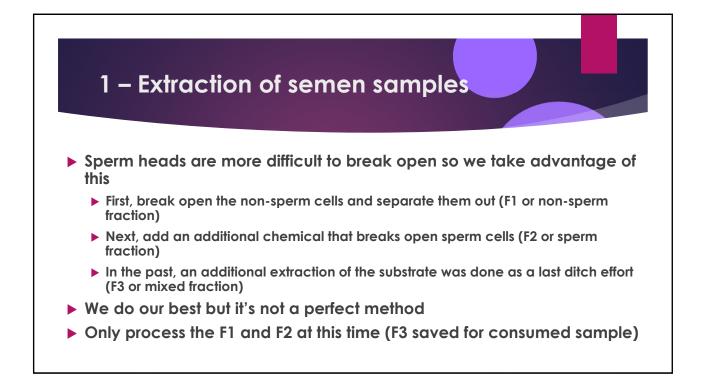


#### 1 – Extraction – Tecan robot

- Like the Maxwell robot:
  - Samples are preprocessed prior to putting on the instrument
  - Uses DNA IQ technology
- Unlike the Maxwell robot:
  - Tecan moves the liquid from step to step, not the beads
  - Runs up to 96 samples per batch
  - Also used to set up the next two steps in DNA analysis: quant and amp
- Utilizing Tecans has greatly reduced our backlog







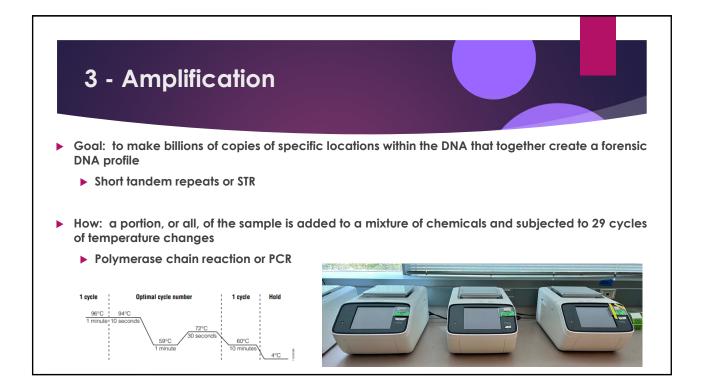
## 2 - Quantitation

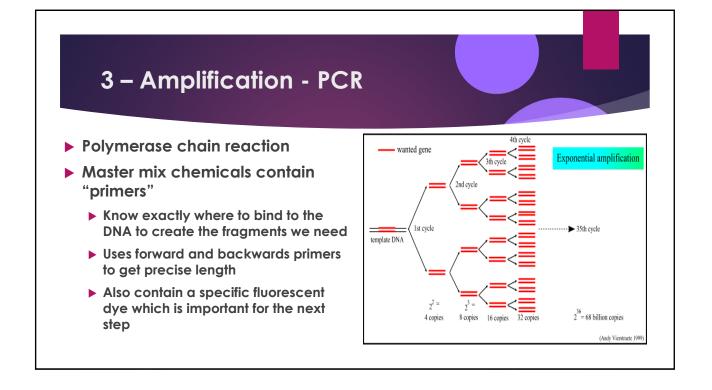
- qPCR (quantitative PCR)
- Tells us:
  - How much human DNA is present
  - How much male DNA is present
    Use both to calculate MTFR
  - How degraded the DNA is (quality of the sample)
- All this information is used to determine how to proceed with the sample



# Examples of qPCR results

Well	Sample Name	Dilution	Target Name	Task	СТ	Quantity(ng/µl)	MTFR	DI	
G4	DFS22		Autosomal	UNKNOWN	32.6401	0.0112	0.9649	2.7317	G4 – A sample that is
G4	DFS22		Degradation	UNKNOWN	34.2318	0.0041			~1:1 MTFR with good
G4	DFS22		IPC	UNKNOWN	20.3866				quality
G4	DFS22		Y	UNKNOWN	33.4417	0.0057			7
32	DFS22-		Autosomal	UNKNOWN	Undetermined	0.0000			
32	DFS22-		Degradation	UNKNOWN	Undetermined	0.0000			B2 – A sample with n
32	DFS22-		IPC	UNKNOWN	20.2806				human DNA detecte
32	DFS22-		Y	UNKNOWN	Undetermined	0.0000			
G6	DFS21		Autosomal	UNKNOWN	34.9151	0.0023		3.8333	G6 – A sample with
G6	DFS21		Degradation	UNKNOWN	37.0301	0.0006			only female DNA wit
G6	DFS21		IPC	UNKNOWN	20.3819				good quality
G6	DFS21		Y	UNKNOWN	Undetermined	0.0000			





### 3 – Amplification - STR

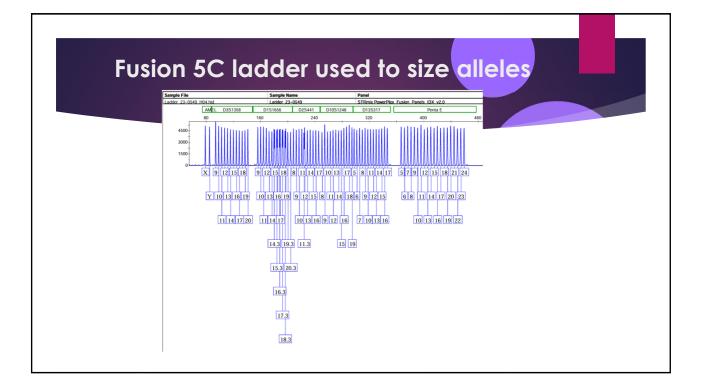
- Short tandem repeats
- DNA genome is made of 4 nucleobases: adenine, cytosine, guanine, thymine
- STRs are repeats of the same sequence of these bases
  - The number of repeats directly determines how we refer to information in a person's DNA profile
- Found in "junk DNA" which is located In between important genes
  - Loci or locations
- Our current profiling kit uses 23 STR loci plus a sexdetermining location

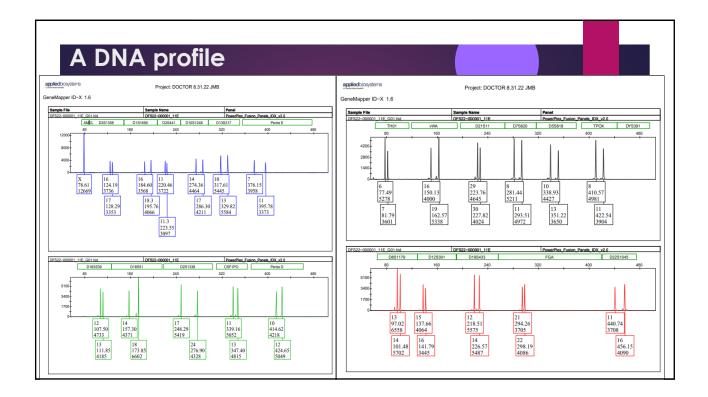
Short Tandem Repeats (STRs)

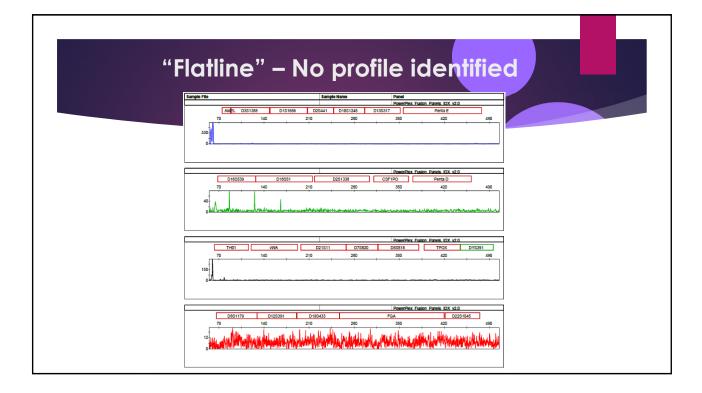
#### 4 – Capillary electrophoresis

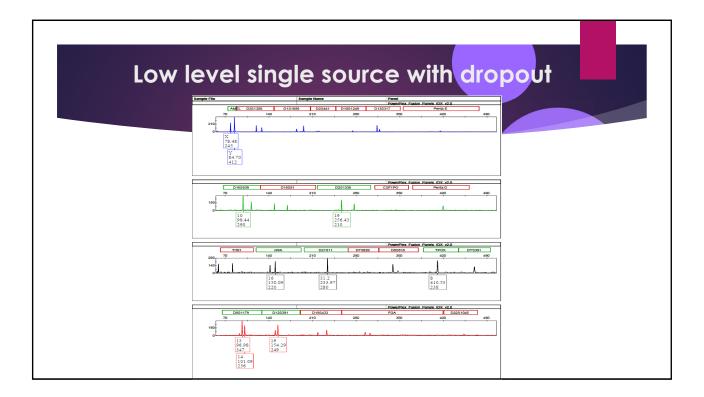
- Amplified product is loaded into the instrument
- DNA is injected into extremely thin tubes via electric charge – 36 cm long
- Tubes (aka capillaries) are filled with polymer
- As DNA moves through the polymer, it naturally separates out by size
  - Smaller fragments navigate through the polymer faster
- At the end, the DNA passes by a very small area containing a window, a laser, and a camera. The laser excites the fluorescent tag attached during amp and this is captured by a camera.
- Combination of the size of the DNA fragment and the color of the fluorescent tag helps put together a DNA profile

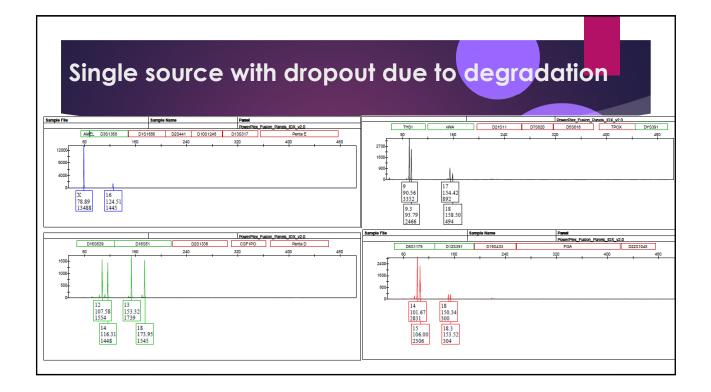


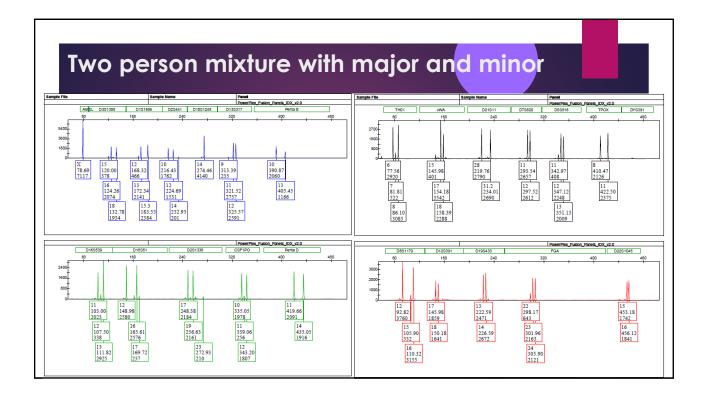


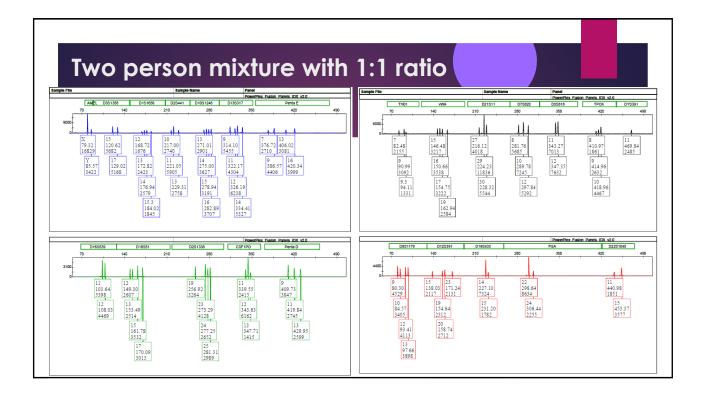


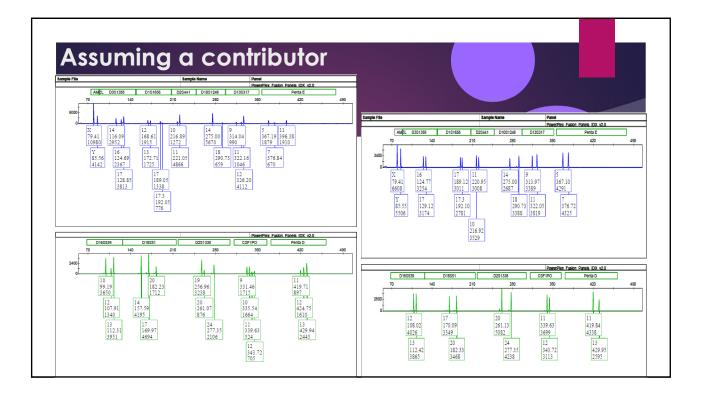


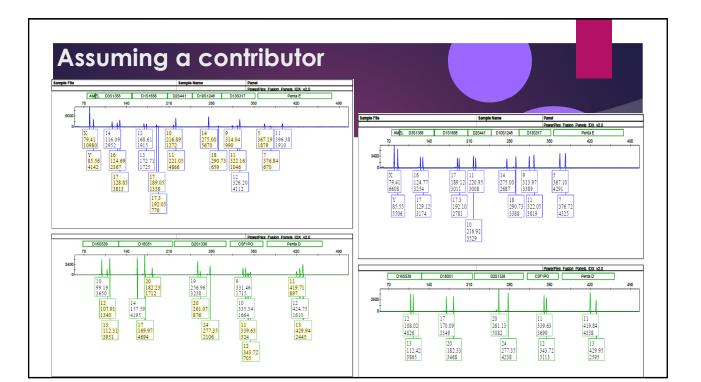


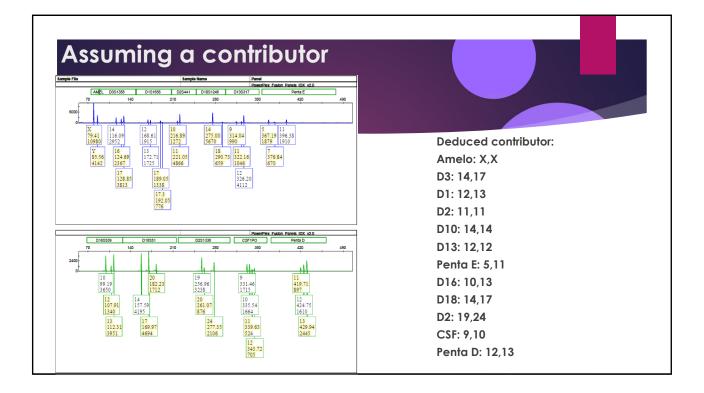


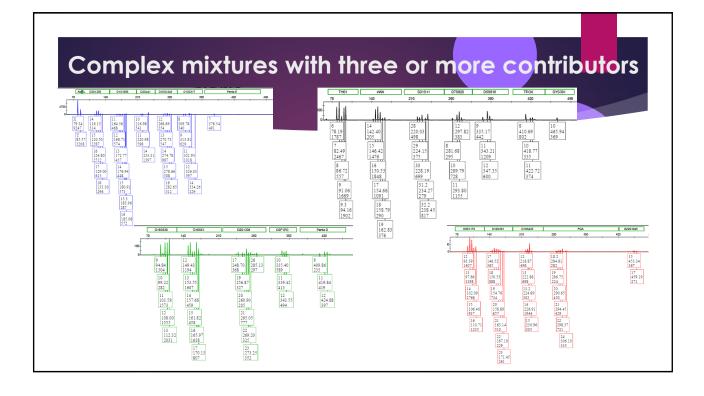




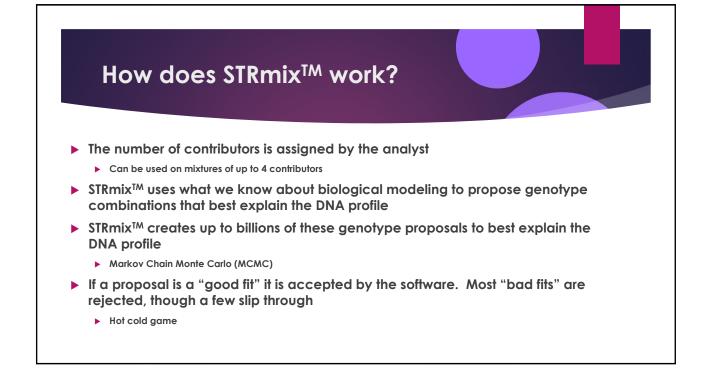


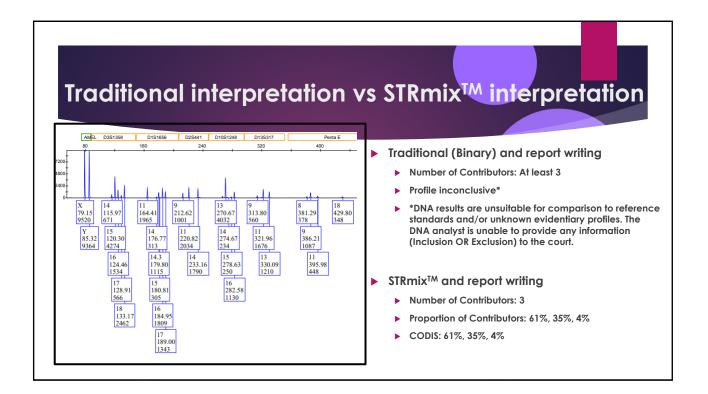












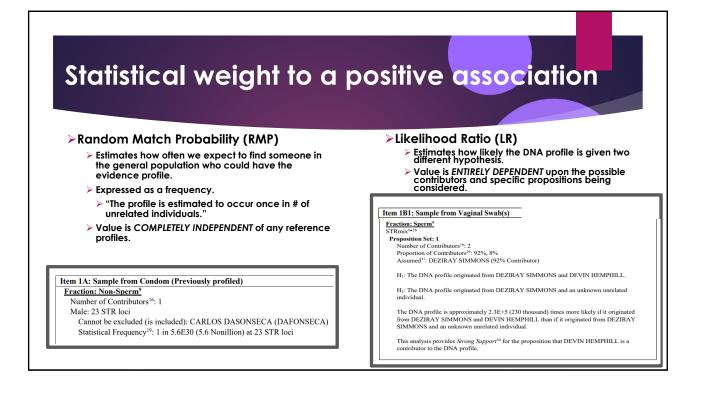
#### The realities of STRmix<sup>TM</sup>

#### **Advantages**

- ► STRmix<sup>TM</sup> can quickly and easily analyze complex mixtures
- The software reduces variability in interpretation.
- Using more data enhances the ability to distinguish between true donors and non-donors

#### Limitations

- ► STRmix<sup>™</sup> analysis cannot be done on all DNA samples. Samples not suitable for STRmix<sup>™</sup> include:
  - Very low level profiles including those with data at only one location.
  - Samples in which the number of contributors cannot be determined.
  - Samples which have more than 4 contributors.



ikelihood verk.	bal scale
To help understand the mo	agnitude of the numbers
Likelihood Ratio	Verbal Qualifier
Likelihood Ratio	Verbal Qualifier Uninformative
Likelihood Ratio 1 2-99	
1	Uninformative
1 2-99	Uninformative Limited Support



## State of Illinois v Dwright Doty

- > 9 year old boy killed in an alley while playing basketball in 2015
- Evidence included a basketball, two guns, car swabs
  - Basketball was swabbed in quadrants that were profiled separately
- CCSAO requested STRmix be used
  - ESR helped with ISP validation
  - Data was sent to ESR for analysis
- Frye hearing was held to gain admissibility
  - Multiple "big names" came in to testify



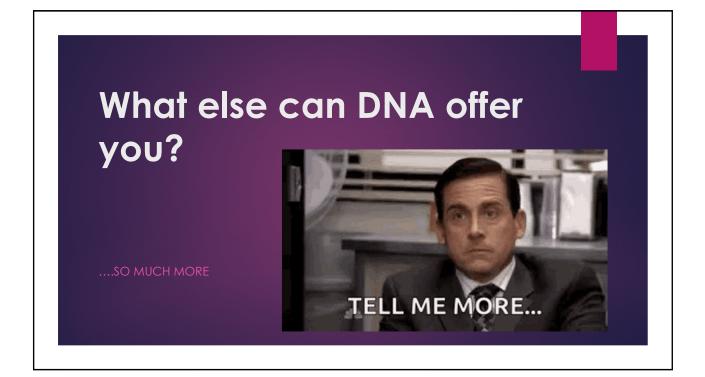
#### State of Illinois v Dwright Doty

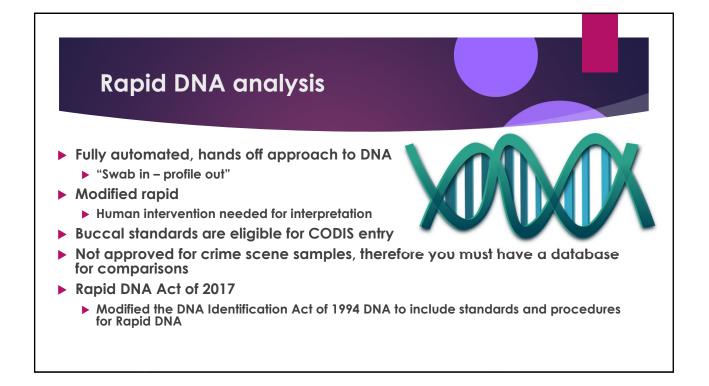
#### Traditional (Binary) interpretation

- Quadrants of basketball:
  - Mix of at least 3 major matches victim
  - Mix of at least 2 major matches victim
  - Mix of at least 3
  - Mix of at least 2
- Various car swabs Dwright Doty included in 2 sets of swabs

#### STRmix interpretation

- Quadrants of basketball:
  - The DNA profiles on the basketball are at least 6x10<sup>5</sup>, 3x10<sup>12</sup>, 1x10<sup>16</sup>, 1x10<sup>6</sup> times more likely if Mr. Doty is a donor than if he is not.
- Various car swabs Both inclusions corroborated by STRmix





# NDIS (CODIS) approved Rapid DNA Instruments

#### ANDE 6C

- NDIS approval for lab use in June 2018
- NDIS approval for booking stations in February 2021
- 5 samples at a time
- 27 STR loci
- ~90 minute run time
- ▶ 85% first run passing rate reported
- ▶ ISP had 65% passing rate in 632 samples
  - February 2020 May 2021

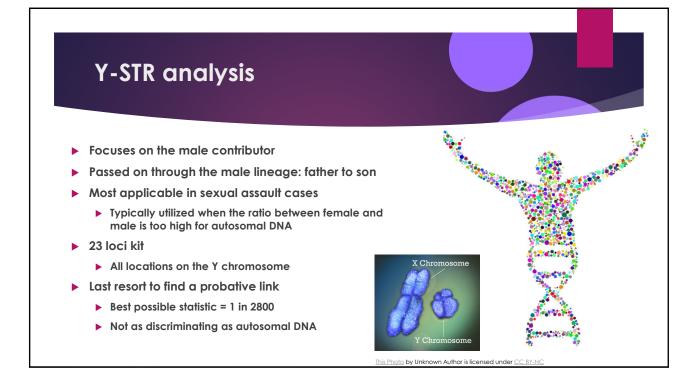
#### **Applied Biosystems RapidHIT**

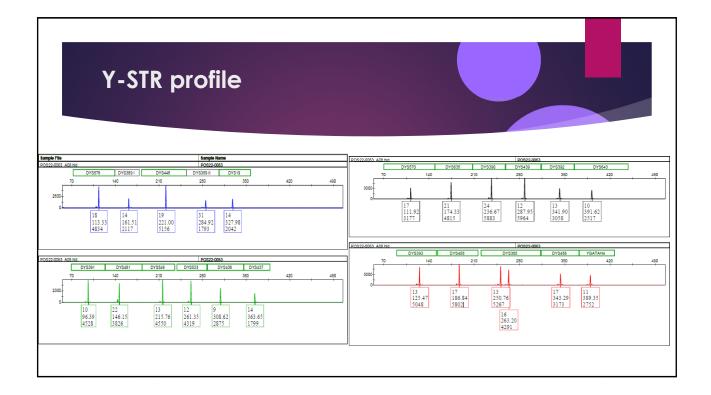
- NDIS approval for lab us in September 2020
- NDIS approval for booking stations in July 2021
- 1 sample at a time
- 23 STR loci
- ~90 minute run time
- 85% first run pass rate reported
- ISP has 76% passing rate in 441 samples
  - August 2022 Current

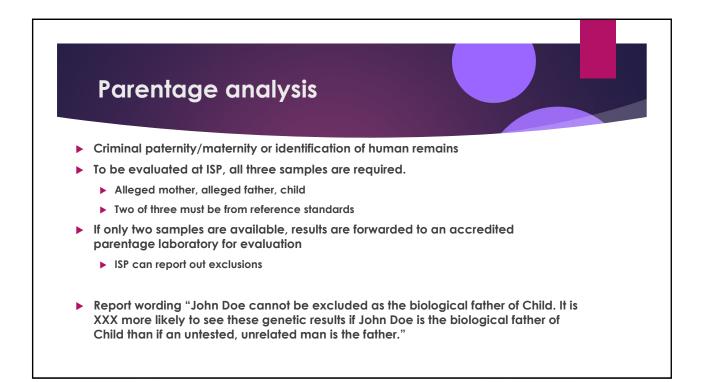


## **Bone extractions**

- When nothing else remains
- Requires a clean bone from the ME's office
  - Whole bone is absolute best, we request the ME does not cut it in half
  - Prefer a femur
- A portion of the bone is sawed into dust, purified, and extracted
- Unidentified human remains

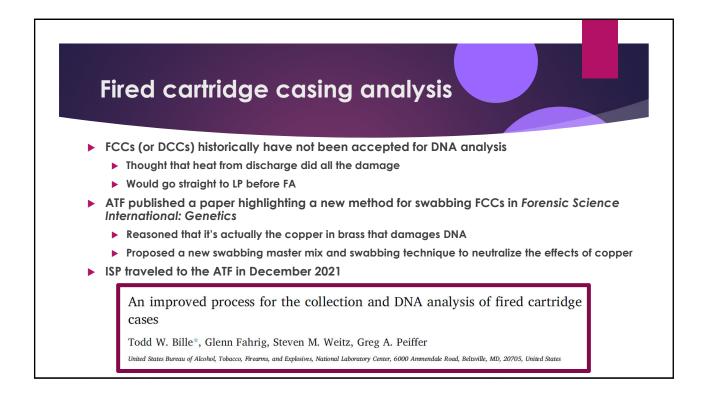






## Familial analysis

- A search of CODIS to determine if a close biological relative of a convicted offender (in the Illinois DNA Index) could be the source of the DNA profile from an unsolved case
- Non-routine and must meet strict criteria to be considered
  - Approval needed from FSC Familial Search Committee
- Two pedigree trees are created to represent a parent/offspring relationship or a full sibling relationship
- The profile to be searched is associated to each tree as a known relative and candidates from each search will be ranked from highest to lowest
- Important to remember: THESE NAMES ARE NOT THE PERSON THAT LEFT DNA BEHIND AT THE SCENE, they are possible relatives. Deeper agency investigation is needed.



## CODIS – the end game to all of our work (mostly)

- Combined DNA Index System
  - Created and maintained by the FBI
- Houses several databases
  - Local, State, and National level databases
  - Convicted offender, arrestee, forensic, forensic mixture, forensic partial, suspect standards, homicide victim standards (legal), unidentified human remains, staff, etc
- ▶ Goal of casework is to get a profile suitable for entering and searching in CODIS
- Samples need to meet certain criteria and level of completeness to be eligible
  - ▶ Must be criminal case, thought to be offender's profile, not found on offender's person
  - > At least 6 loci for SDIS and 10 for NDIS as well as meet rarity threshold
  - ▶ Elimination standards are HIGHLY recommended and must be at least requested before entry

### CODIS database sizes as of 12/12/23

#### Local (LDIS)

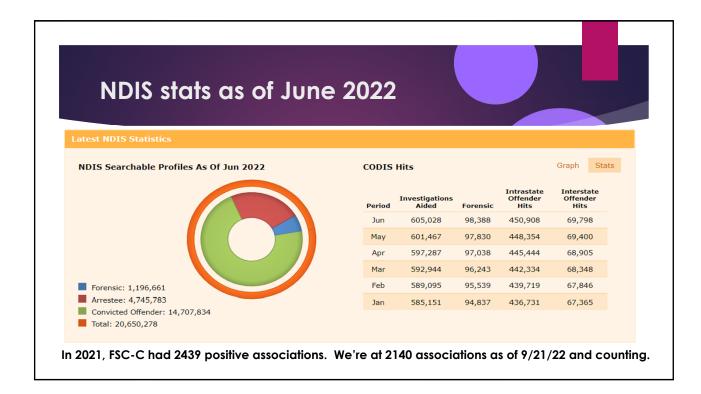
Forensic: 20,118 Forensic mixture: 4,786 Forensic partial: 6,774 Legal: 8,247 Missing person: 7 Unidentified human remains: 32 Staff: 882 Suspect: 9,215

#### State (SDIS)

Arrestee: 914 Forensic: 47,603 Legal: 12,181 Missing person: 55 Offender: 696,178 Staff: 881 Suspect: 29,549

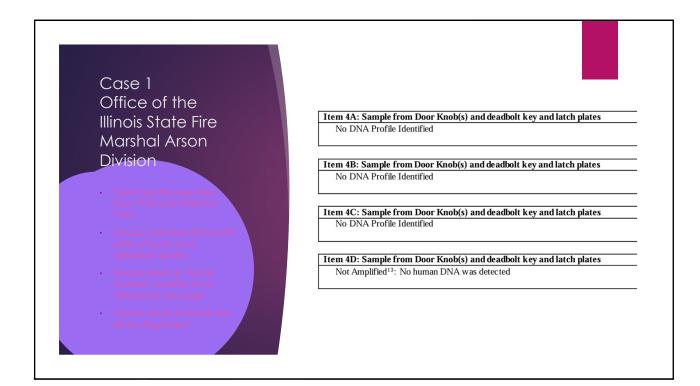
#### National (NDIS)

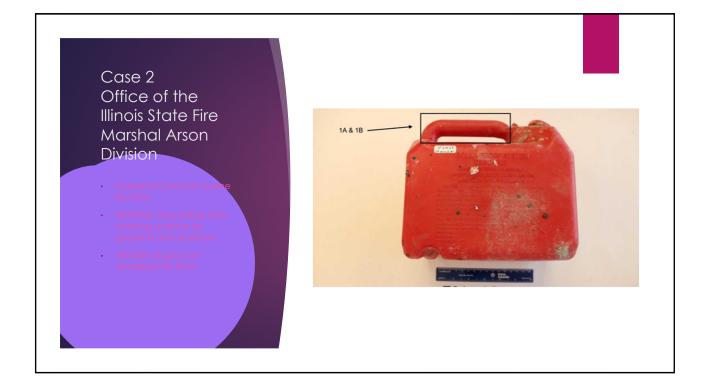
Arrestee: 5,303,917 Convicted offender: 14,502,179 Detainee: 1,484,033 Forensic: 874,621 Forensic mixture: 154,738 Forensic partial: 206,602 Legal: 134,522

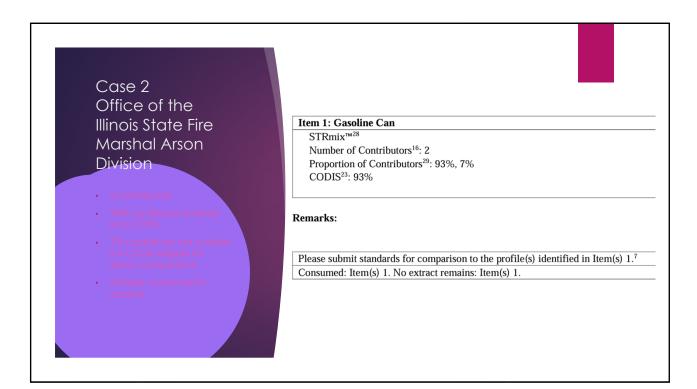




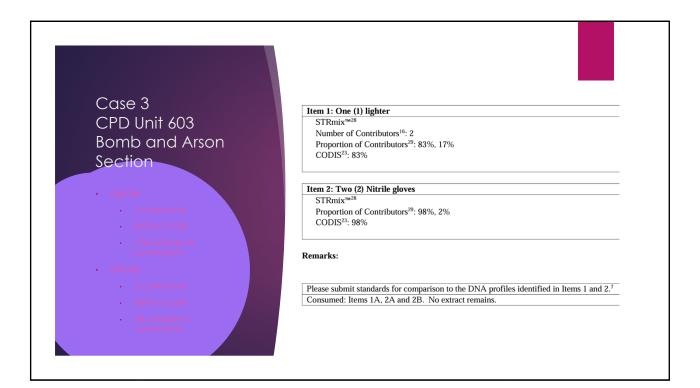


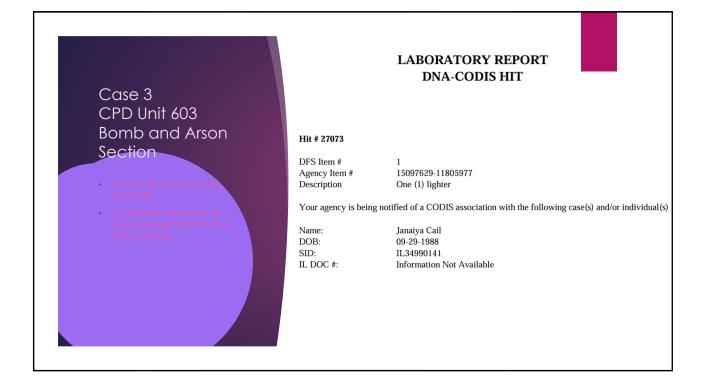


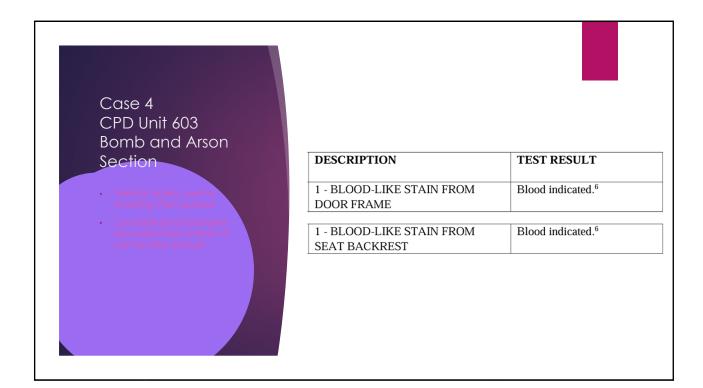


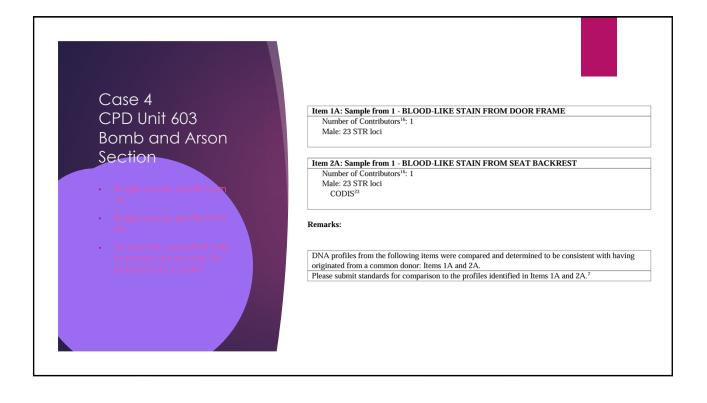


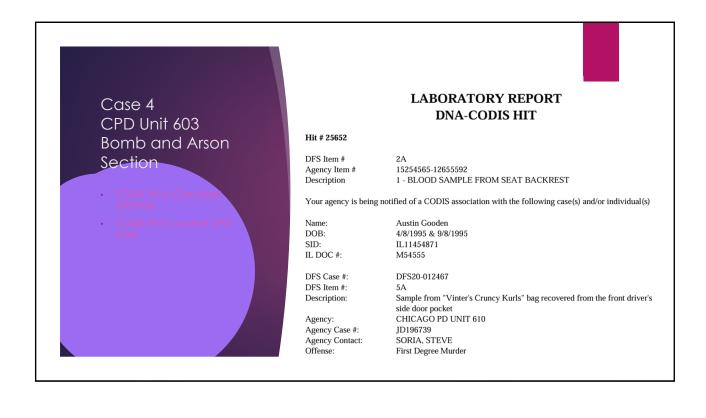


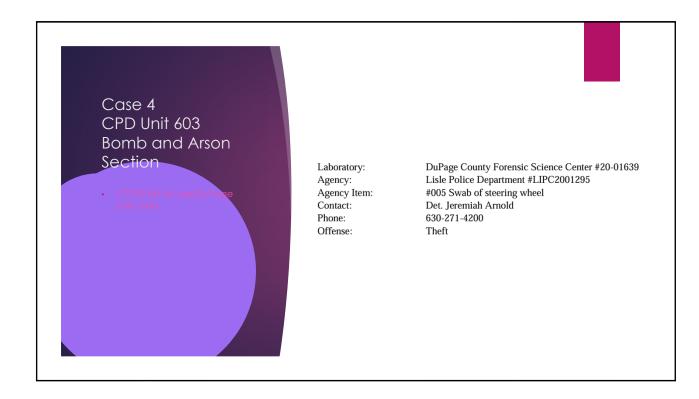


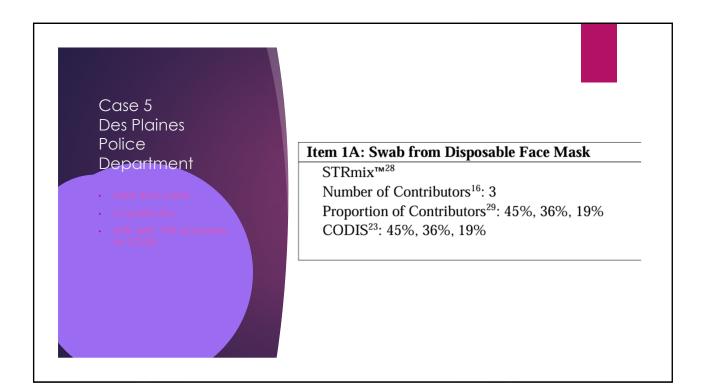


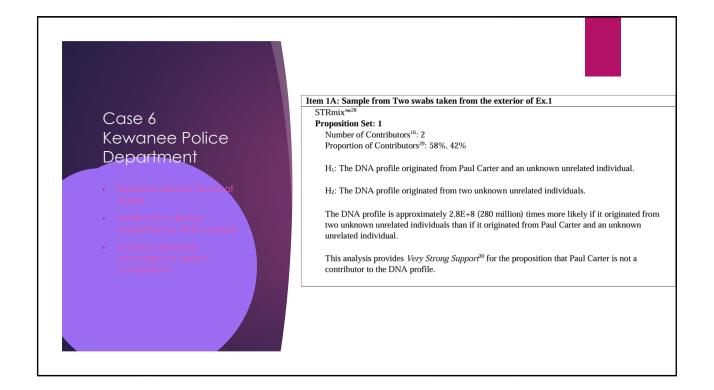


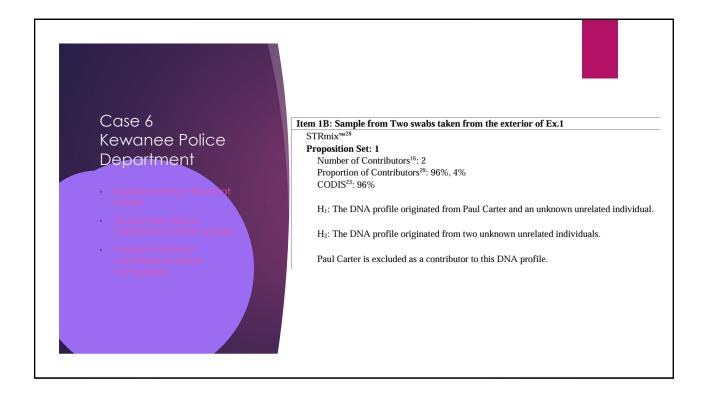












# Forensic DNA analysis has its limitations

- ▶ We cannot tell you how a DNA profile was left at the scene
- We cannot tell you when a DNA profile was left at the scene
- We cannot tell you the last person to touch an object
- The process takes time
  - Rush 2-4 weeks
  - Priority 1-3 months
  - CSA up to 6 months
  - General turnaround time up to 10 months
- ▶ We cannot work every last piece of evidence collected
- We prioritize violent crimes and crimes against people

