The ANDE Rapid DNA System in Law Enforcement, Military, and Disaster Victim Identification Applications

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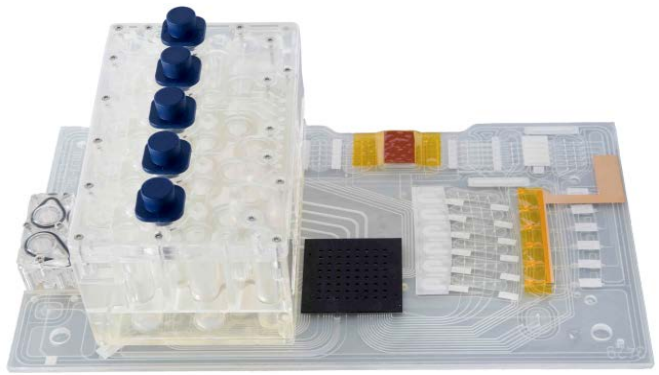
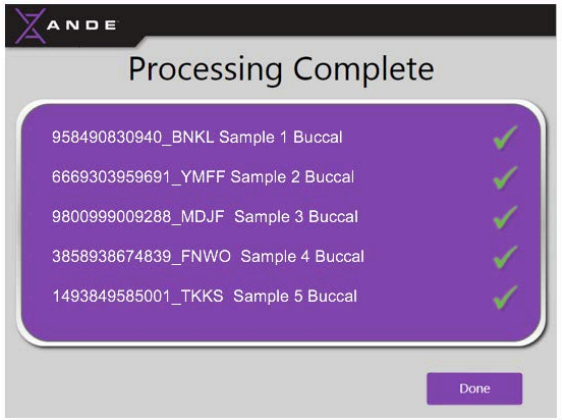
Founder and Chief Scientific Officer

ANDE Overview



Chip

ANDE



Integrated Expert System

"Rapid DNA describes the fully automated (hands free) process of developing a CODIS Core STR profile from a reference sample buccal swab. The "swab in – profile out" process consists of automated extraction, amplification, separation, detection and allele calling without human intervention."

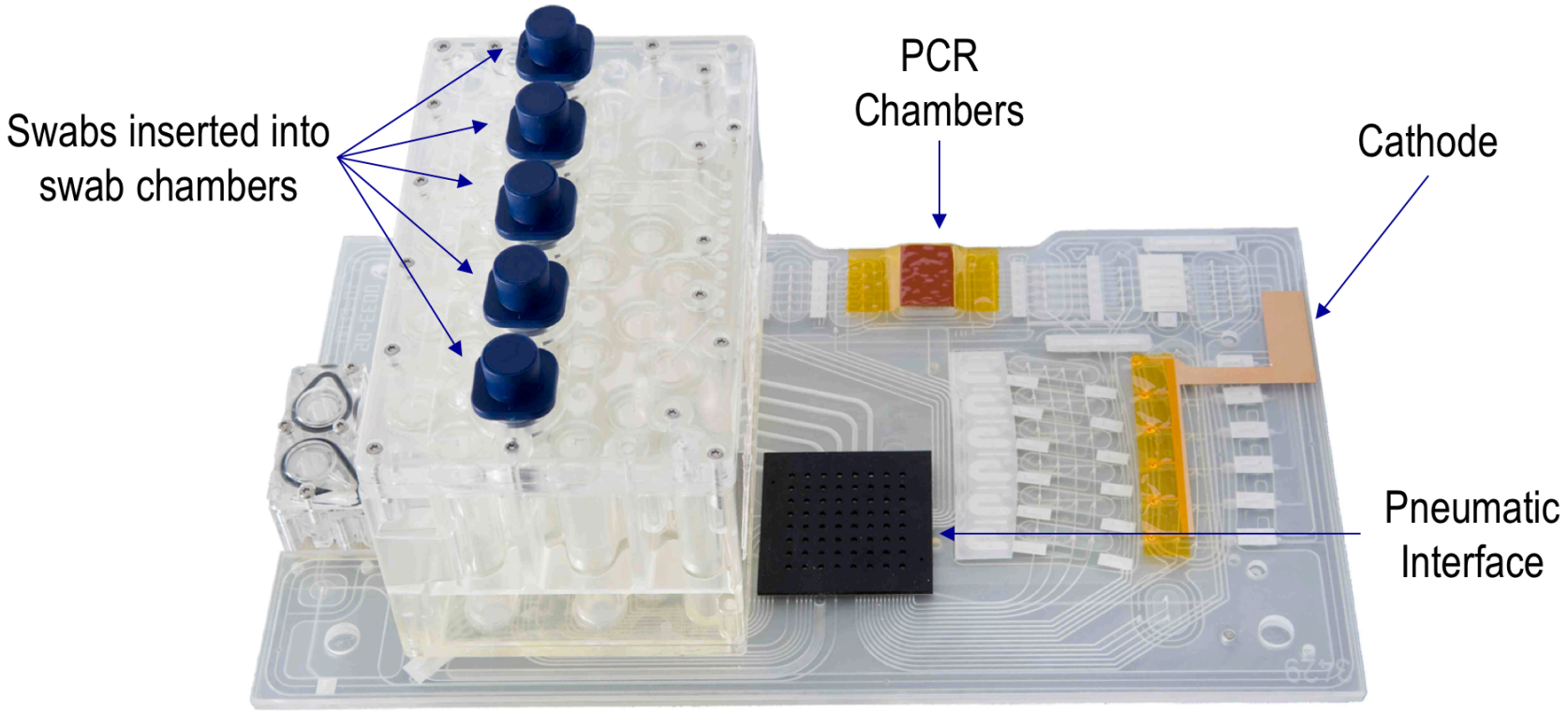
https://www.fbi.gov/services/laboratory/biometric-analysis/codis/rapid-dna-analysis

The ANDE 6C Instrument

- Capable of running the Flexplex™ 27 assay based on 6-color detection
- On board Expert System
- Purification, PCR, electrophoresis identical to NDIS-approved system
- Enhanced ruggedization for mobile applications in austere environments (MIL STD 810G, including 2-foot drop)
- Operates outdoors and on generator power

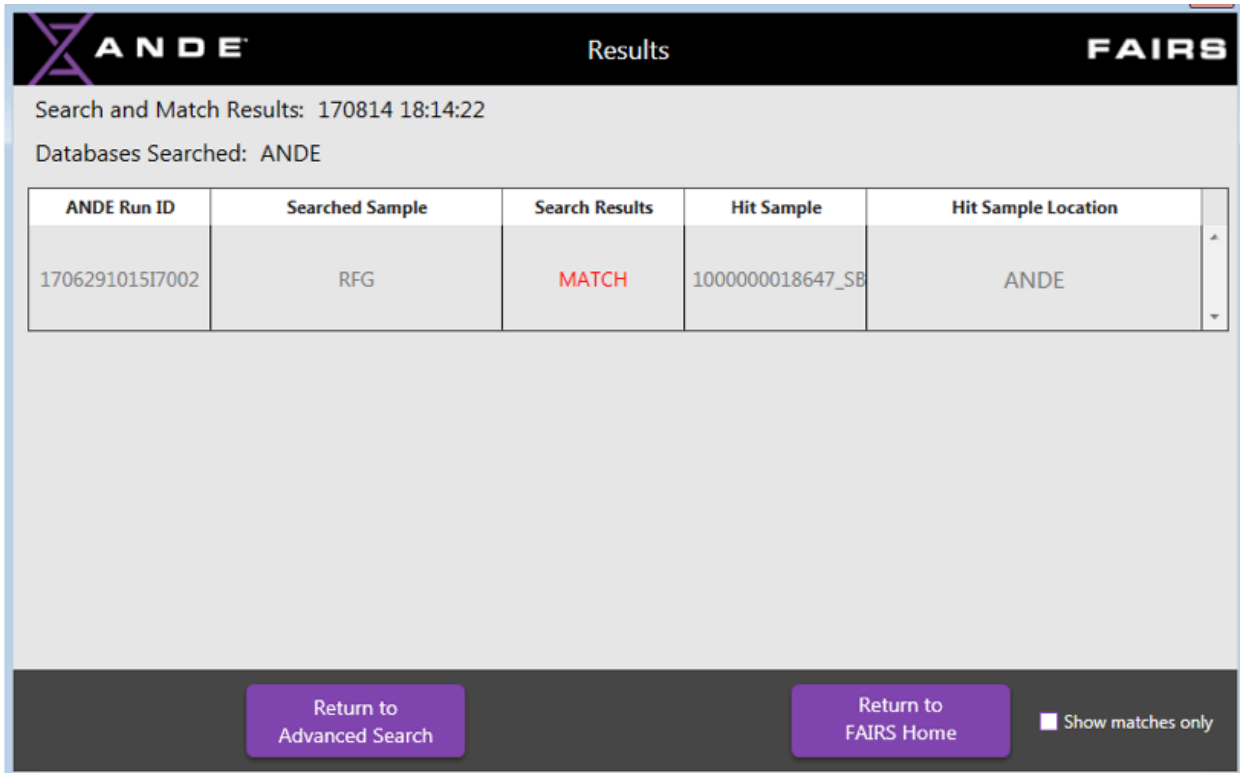
The FlexPlex A-Chip and I-Chip

- Integrate all process steps required for DNA analysis in a single consumable.
- A-Chip: Single-use plastic disposable processes up to five buccal swabs per run.
- I-Chip: Single-use plastic disposable processes up to four casework/DVI/SSE samples per run.
- All reagents on board and stable for 6 months at room temperature; no refrigeration required.
- Identical structure to the NDIS-approved PP16 Chip



FAIRS Software

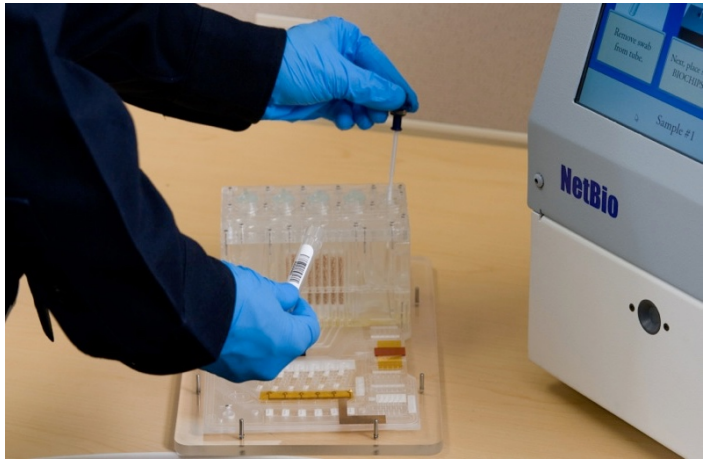
- Fully-integrated software package
- Database creation (import from ANDE and existing lab databases) and export
- Allows connection of a federation of instruments
- Search and match software
- Kinship software
- Full reporting functionalities



Simplicity of Operation for Lab or Field

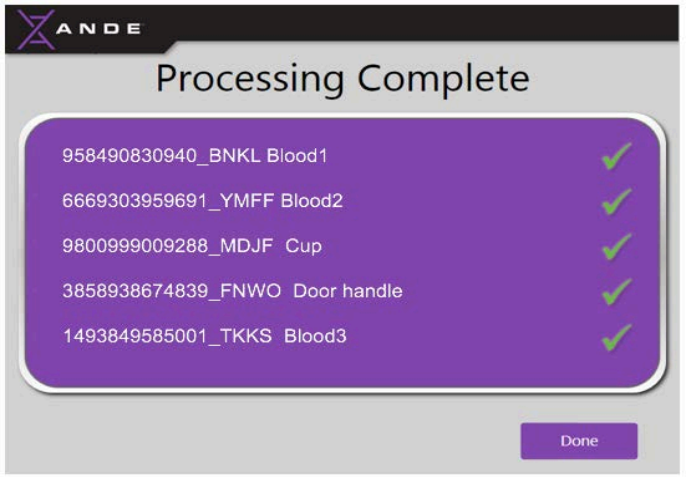
1. Collect sample

2. Scan RFID on sample cap

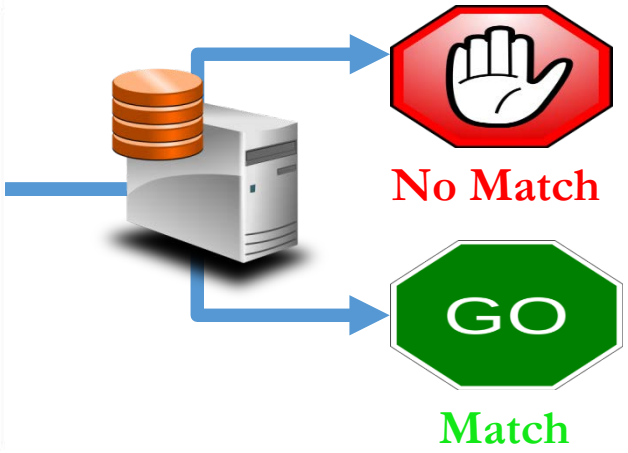


3. Load samples in Chip

4. Load Chip into ANDE and close door to start



5. In less than two hours, processing is complete



6. Search database—Actionable Results

Controlling DNA ID Generation Outside the Lab

Minimize points of potential system failure:

- Single consumable designed for ease of use
- Ensure sample tracking (e.g. RFID embedded in swab)
- Focus on minimizing contamination by user (e.g. locking swabs)
- Ruggedize system to avoid damage at the booking station
- Eliminate routine instrument calibration/adjustment: avoid service techs interfering with normal booking station process flow

Controlling DNA ID Generation Outside the Lab

Minimize mishandling of data

- User classes with tiered privileges
  - Operator: Police Officer/Warfighter/First Responder; no access to STR data
  - Admin: Forensic Analyst; access to STR data
  - SuperAdmin: Supervisor; access to STR data, ability to delete data and configure system
- Encryption—Booking station sample processing, successful run completion, and data upload performed without disclosing the STR profiles themselves

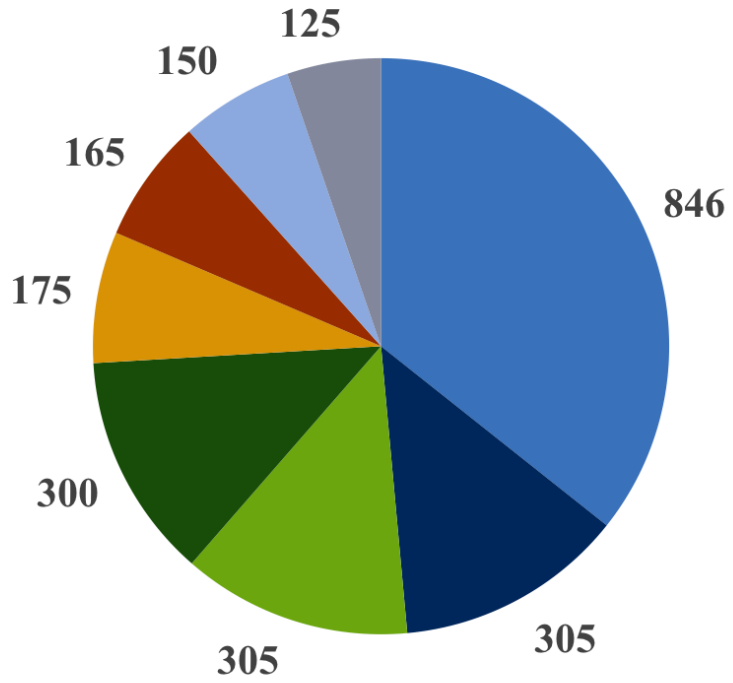
Developmental Validation Goals

- Demonstrate reliability, reproducibility, and robustness of the system. NDIS-approved system is based on CODIS core 13 loci; DV for CODIS core 20 loci (FlexPlex 27) is in progress
- Validate a full Rapid DNA System including an on-board Expert System for use with single source reference buccal swabs
- Incorporate knowledge and expertise of NDIS-participating forensic laboratories
- Utilize real-world buccal samples whenever possible
- Comply with the NDIS Operational Procedures, FBI's Quality Assurance Standards and SWGDAM guidelines
- Obtain the first NDIS Approval of a Rapid DNA System

NDIS Approval for use by accredited forensic laboratories is a critical step for adoption in police booking stations

Developmental Validation Test Sites

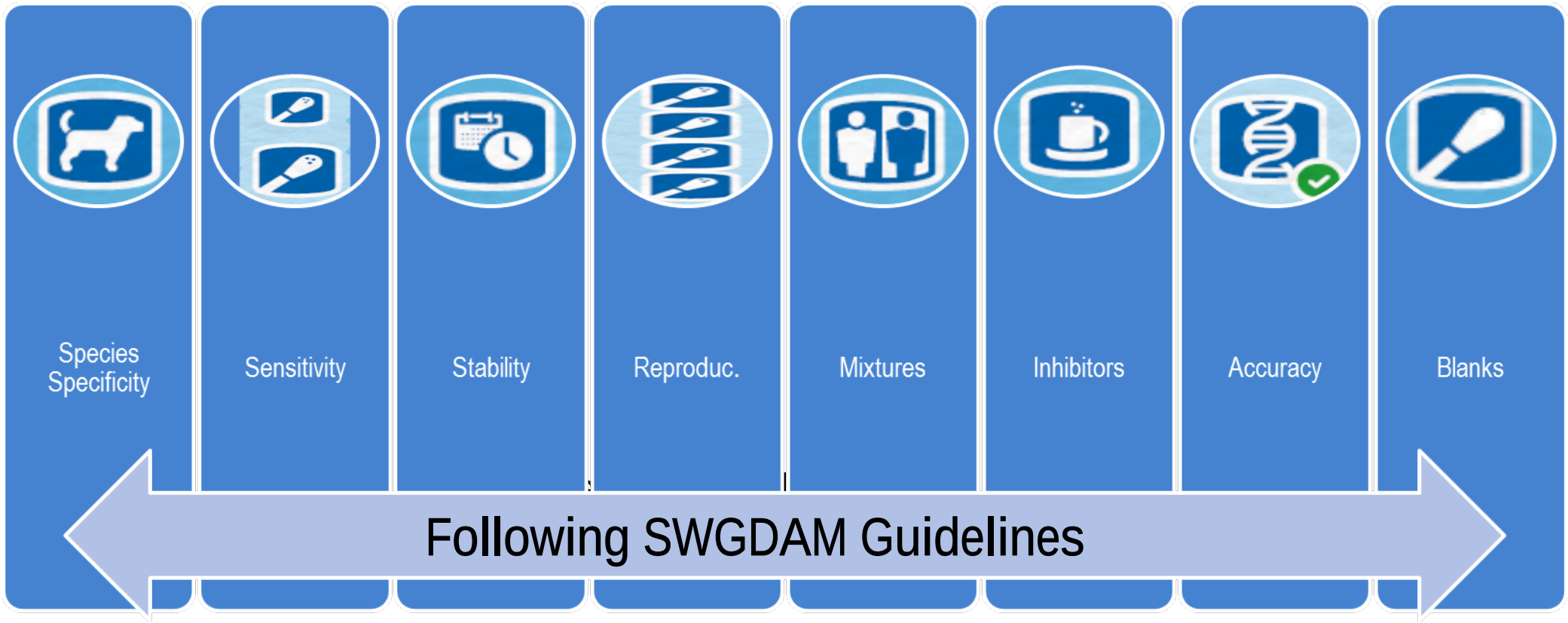
Laboratory Testing Sites
Alabama Department of Forensic Sciences
Michigan State Police
Florida Department of Law Enforcement
Pennsylvania State Police
Dubai Police GHQ, Gen. Dept. Forensic Sciences & Criminology
Defense Forensic Science Center, Office of Chief Scientist
National Institute of Standards and Technology
ANDE



> 2,000 swabs

- ANDE worked closely with the labs, SWGDAM, and the FBI to evaluate reliability, reproducibility and robustness for the forensic community.

Developmental Validation: Experimental Studies



Reproducibility

Buccal Swabs

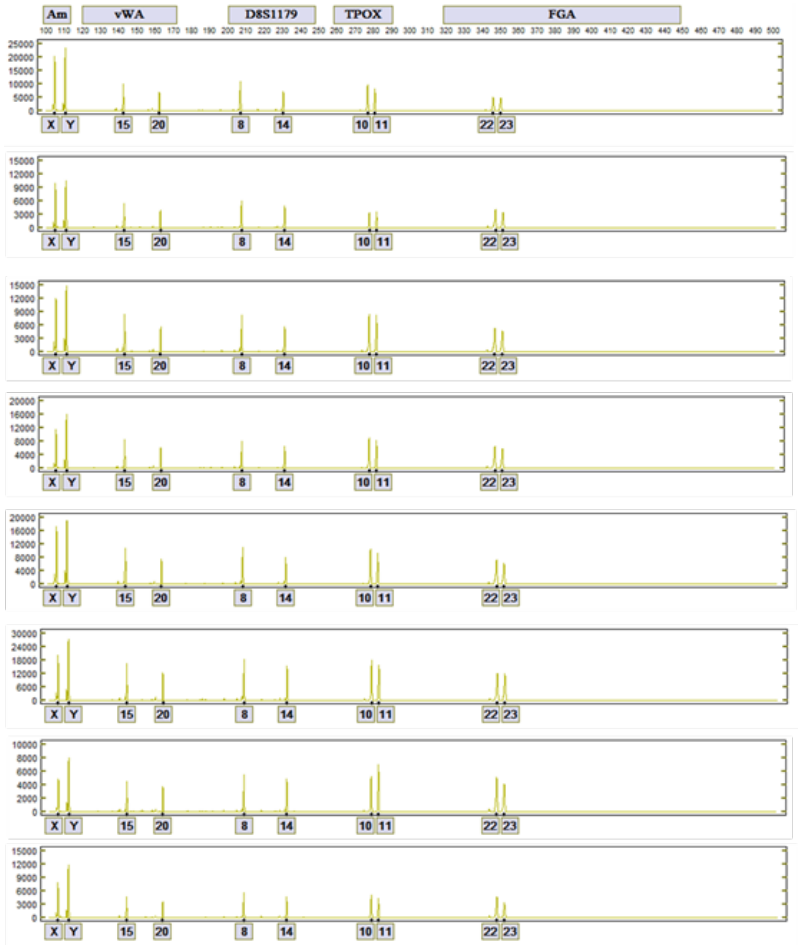
- 10 unique individuals
- 2 replicates per lab
- 2 different lot numbers

Purified DNA

- 3 unique individuals
- 2 replicates per lab
- 2 different lot numbers

NIST SRM

- NIST-traceable swab was created



- Full concordance among replicates and with conventional lab testing

Inhibitors

- 20 different substances in duplicate

Beer	Lipstick
Bloody swab	Mint
Cheetos	Mouthwash
Chocolate	Soda (2 brands)
Cigarette	Sugar
Coffee (2 brands)	Tea
Gum (2 brands)	Tobacco Dip
Lip Balm	Toothpaste (2 brands)

System was robust in presence of common oral inhibitors

Appropriate Controls: Blanks/Contamination

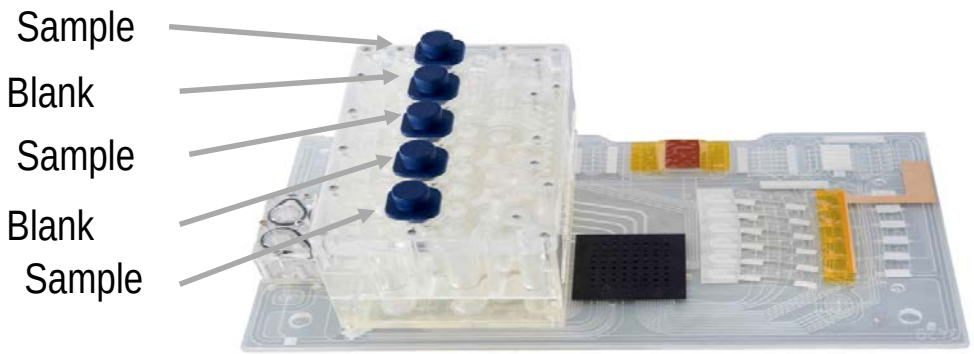
- 4 configurations (128 blank swabs total)

Sample-blank-sample-blank-sample

Blank-sample-blank-sample-blank

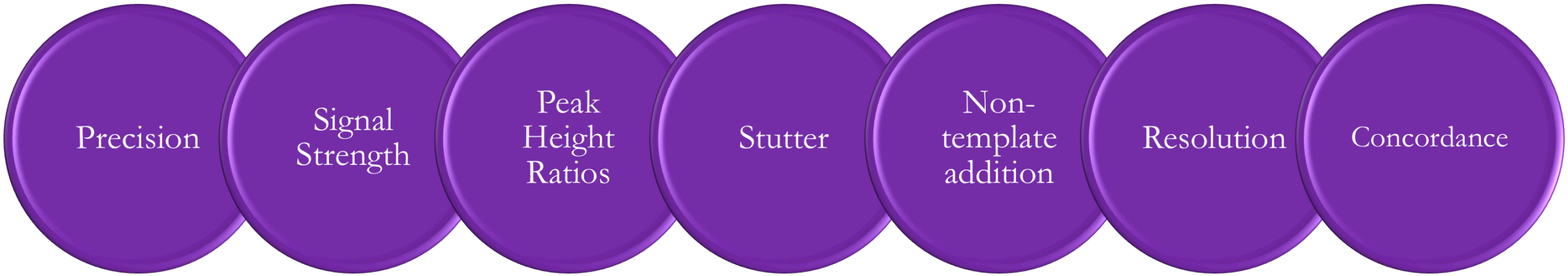
Sample-sample-sample-sample-blank

Blank-sample-sample-sample-sample



No blanks had called peaks

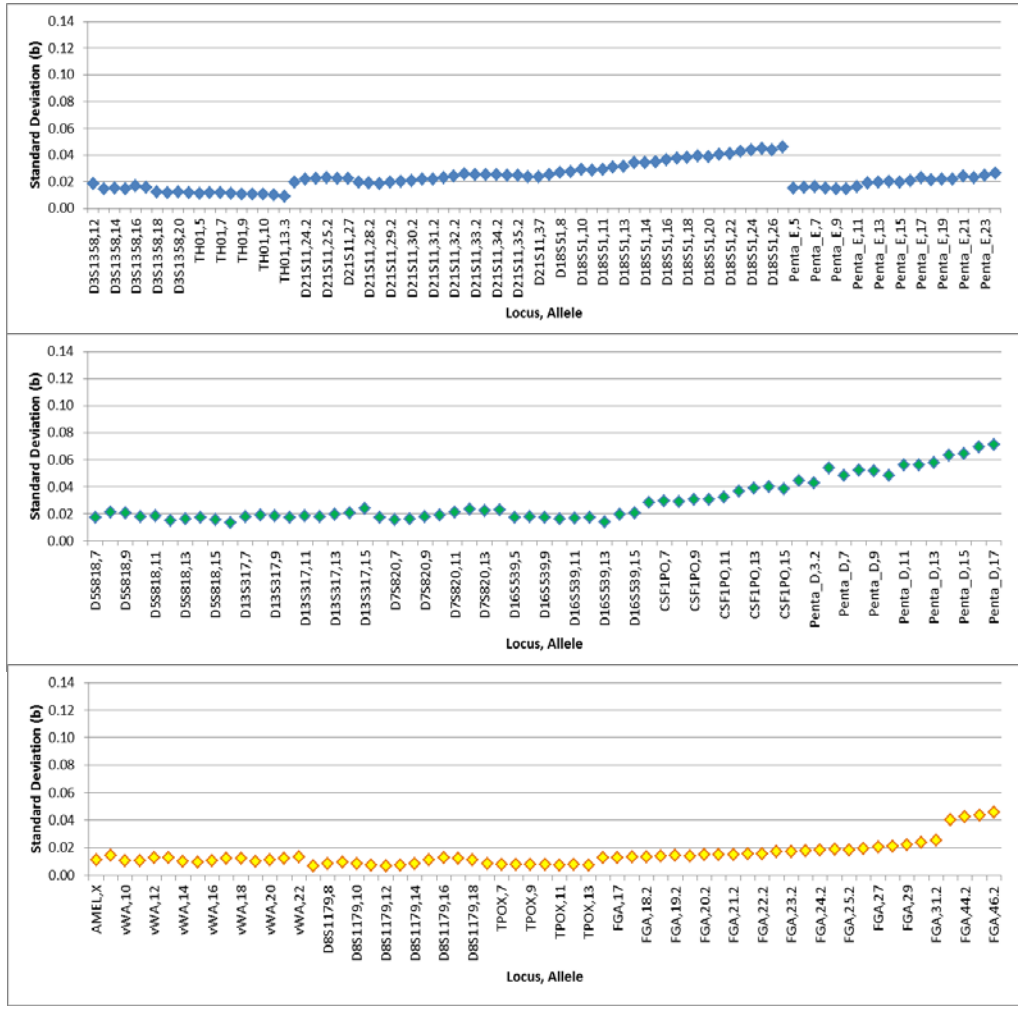
Accuracy Sample Set



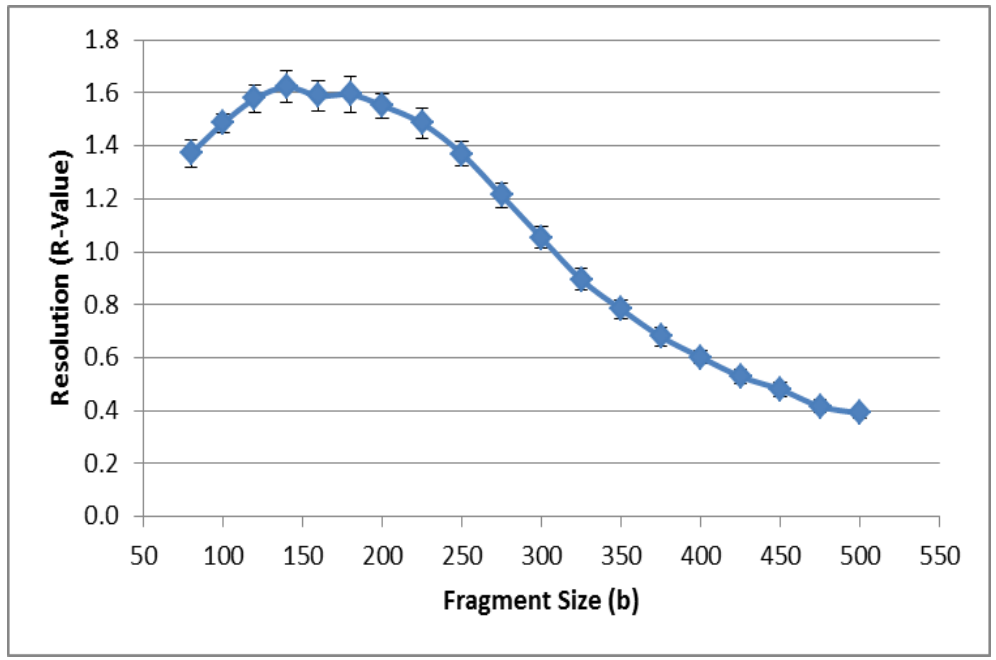
- 1398 unique individuals typed
- Conventional STR Typing with PowerPlex® 16 HS
- Performed by all 8 laboratories

Accuracy: Precision and Resolution

(n=418 allelic ladders across 14 instruments)



Variation is well below target value of 0.5 bases



In electrophoresis:
Resolution: $R = (2/\Delta b)^*(t_2 - t_1)/(w_1 + w_2)$

In Gaussian Distribution:
$w = 4\sigma$, $FWHM = 2(\ln 2)^{0.5} = 2.355\,\sigma$
$\sigma = Area/[(2\pi)^{0.5} * Height]$

So,
$R = [(2\ln 2)^{0.5}/\Delta b]^*(t2 - t1)/(FWHM_1 + FWHM_2)$
$= [1/(2^*\Delta b)](t_2 - t_1)/(\sigma_1 + \sigma_2)$

t – migration time
w – baseline width
FWHM – full width at half maximum
σ - standard deviation
Δb - base pair difference

R values ≥ 0.2 indicates single base resolution

Accuracy: Concordance

	Number of Samples	Percentage
Samples Assessed for Concordance	1368	
Concordant Samples	1367	99.93%
Concordant Alleles	35567	99.997%
Discordant Samples	1	0.07%
Discordant Alleles	1	0.0028%

- First pass success rate for the CODIS core 13 (including Expert System review) = **91%**

NDIS Approval: Conclusions

Study Design

- Partner support
- Buccal swabs
- No user modifications
- SWGDAM Guidelines
- Included fully-integrated Expert System

System Performance

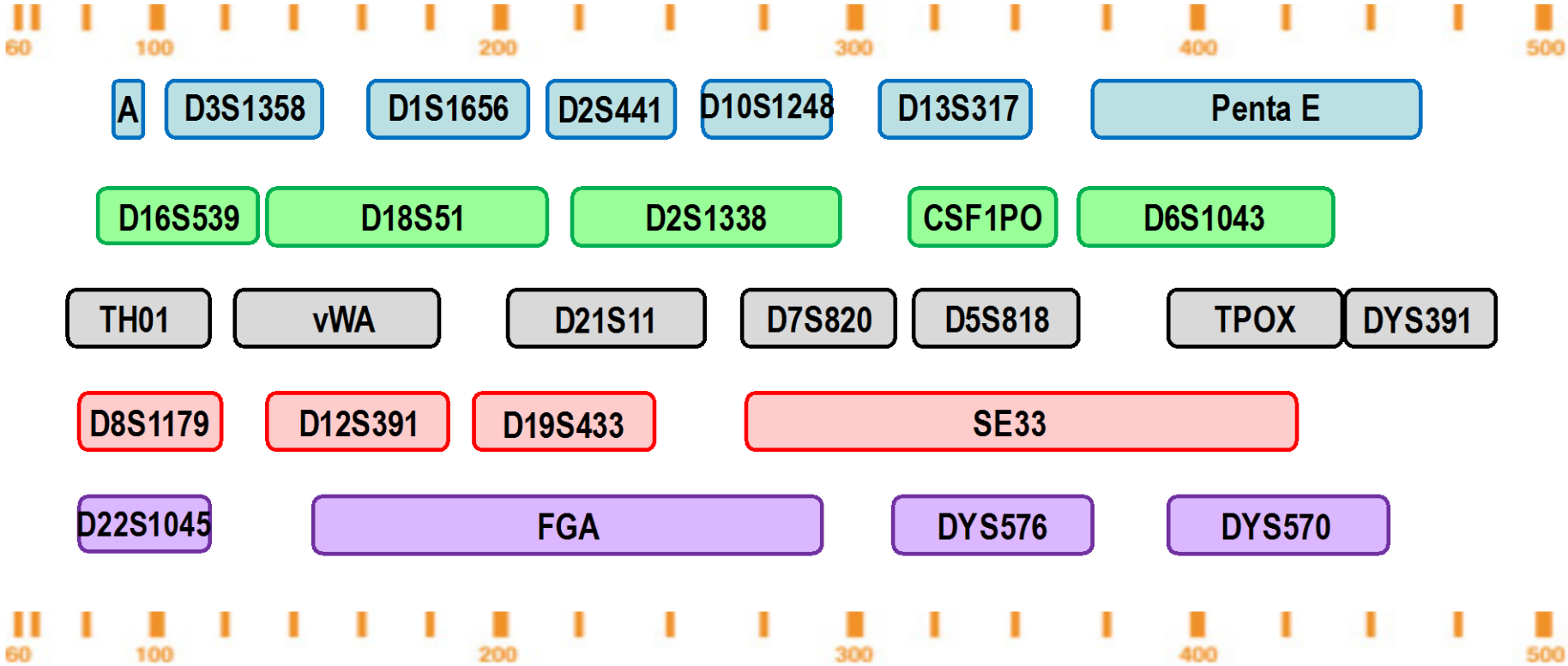
- Concordant with lab results
- Mixtures flagged
- Reproducible at 8 locations
- Single base resolution

Successful Results

- First pass success 91%
- Concordance over 99.99%
- NDIS approval in March 2016

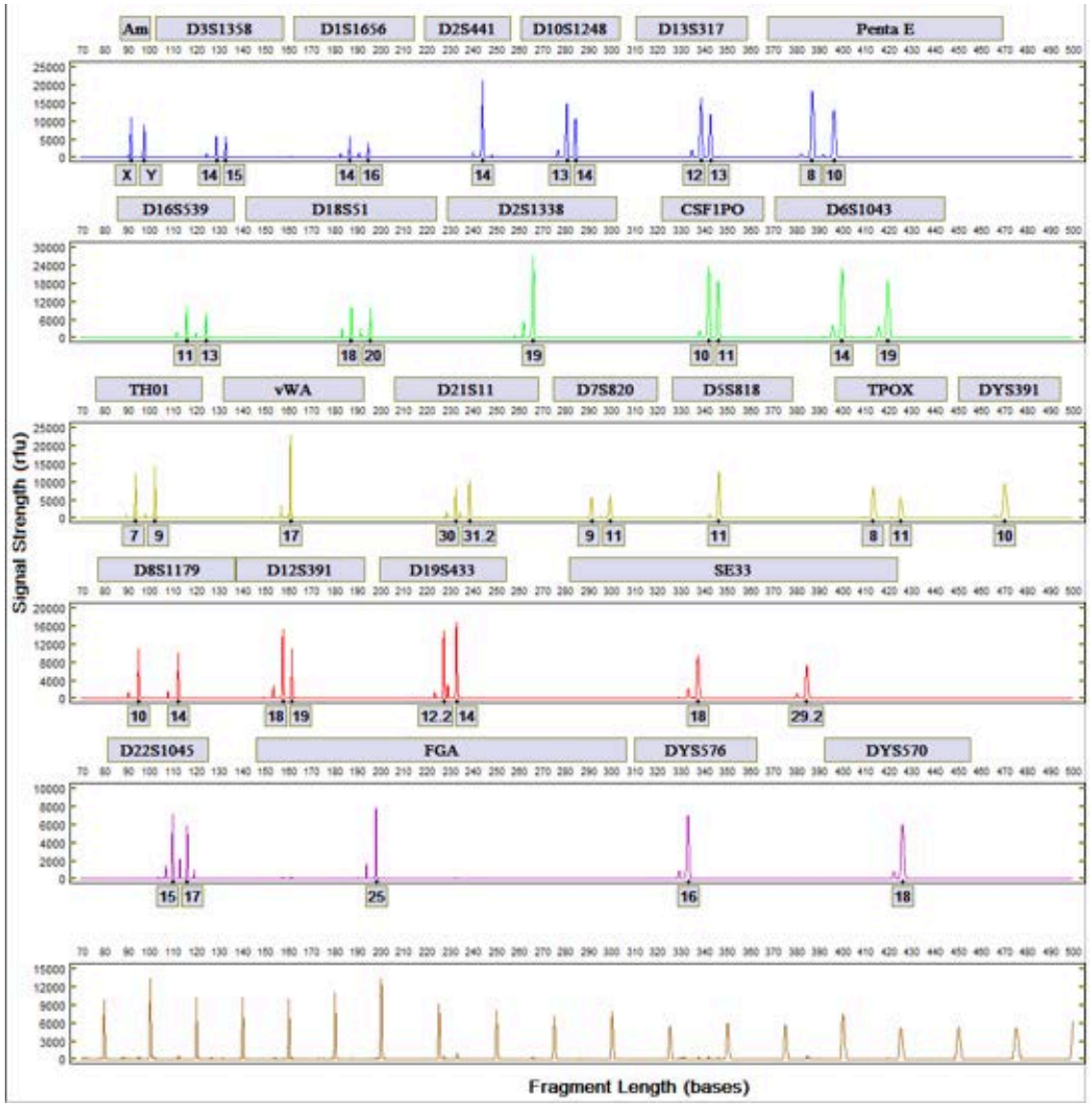
- **March 2016**: First and only **NDIS approved Rapid DNA Analysis System** for complete Rapid DNA Analysis, included Expert System for automated allele calling (PP16, buccal swabs)
- **August 2016:** Developmental Validation study for the FlexPlex assay initiated to be compliant with CODIS core 20 standard

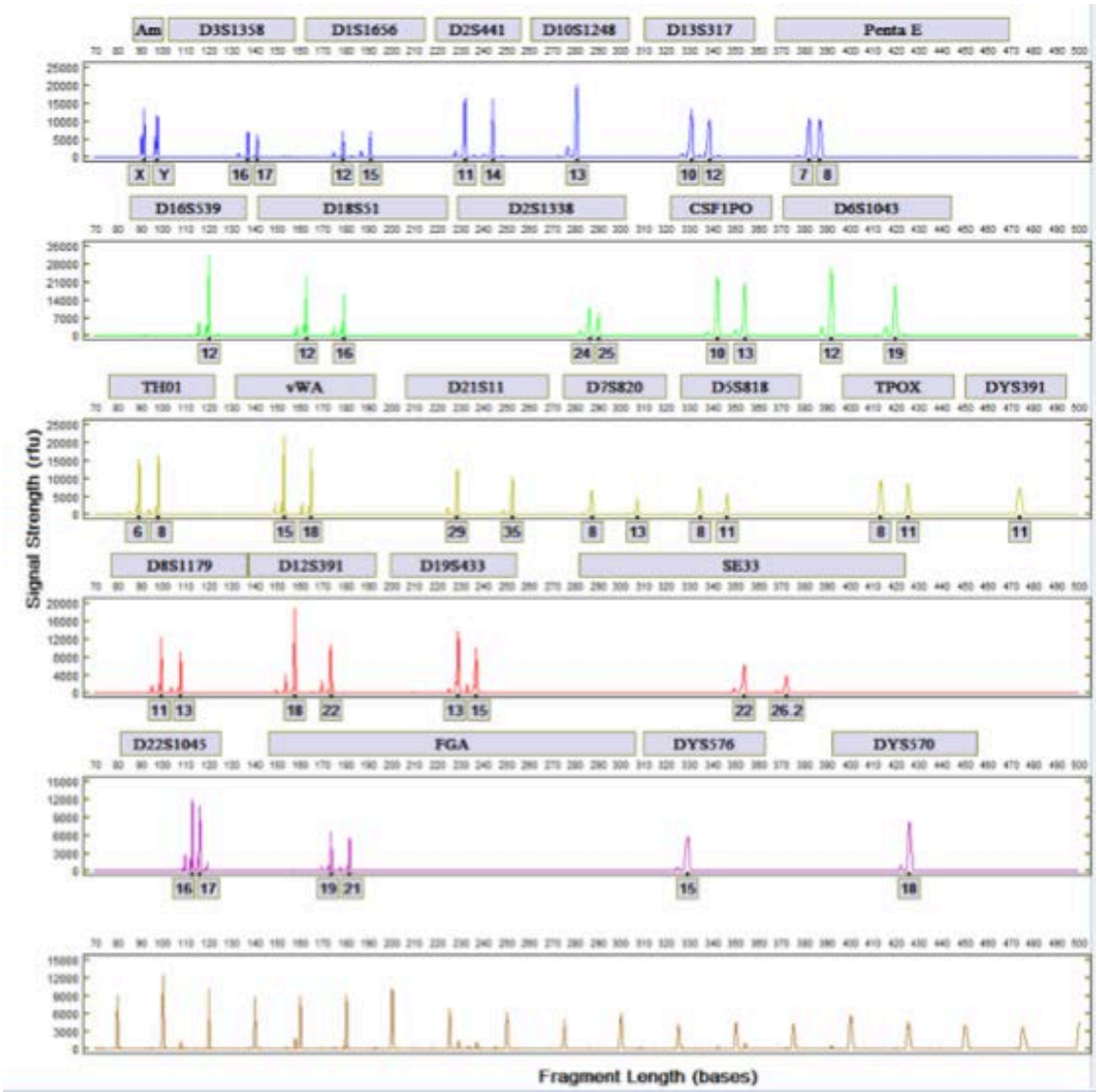
The FlexPlex™ Rapid DNA Assay



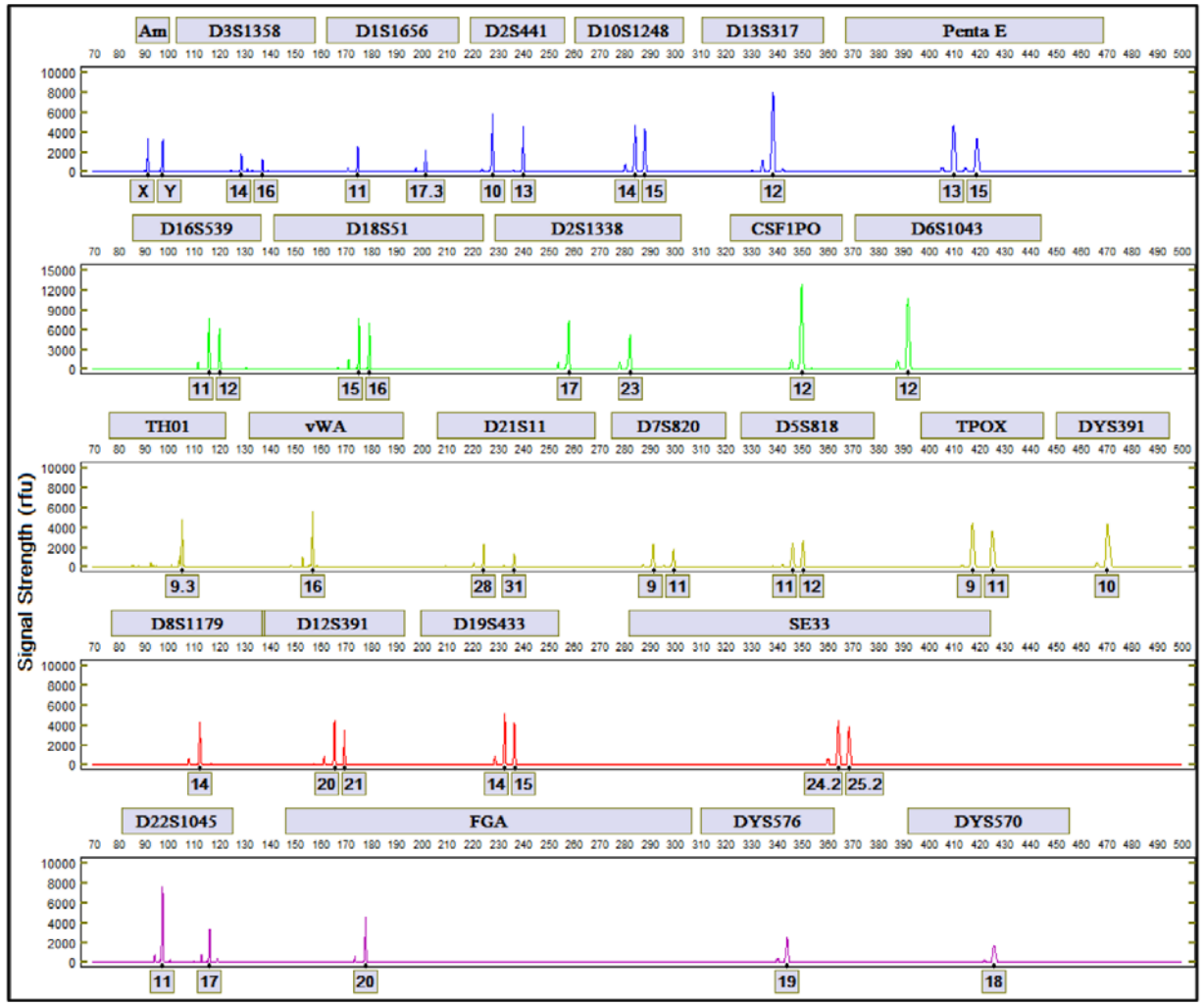
- Global compatibility: Contains all CODIS 20, European Standard, UK, Interpol, German, Australian STR loci; also contains D6, broadly used in China
- Based on Promega's Fusion 6C chemistry

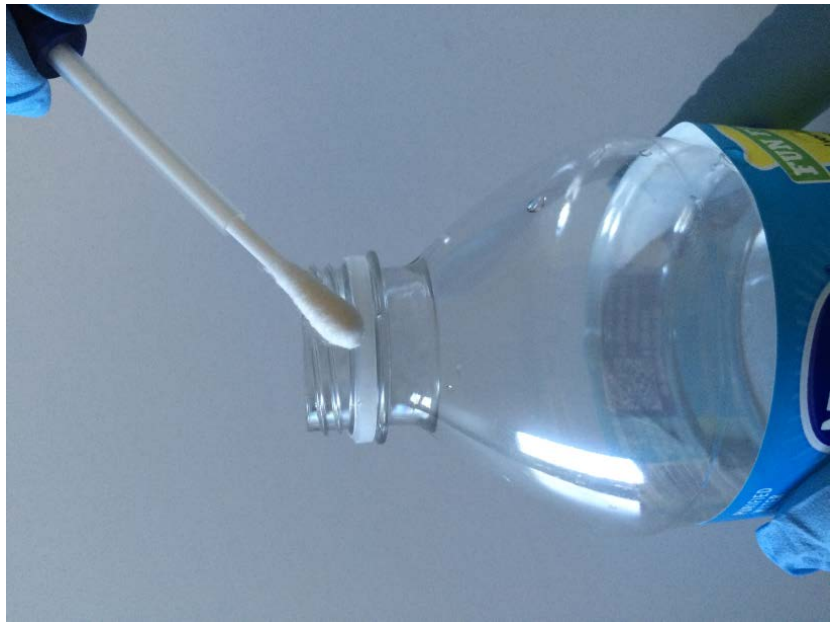
Processing Casework Samples: Blood on FTA (L) and Tile (R)



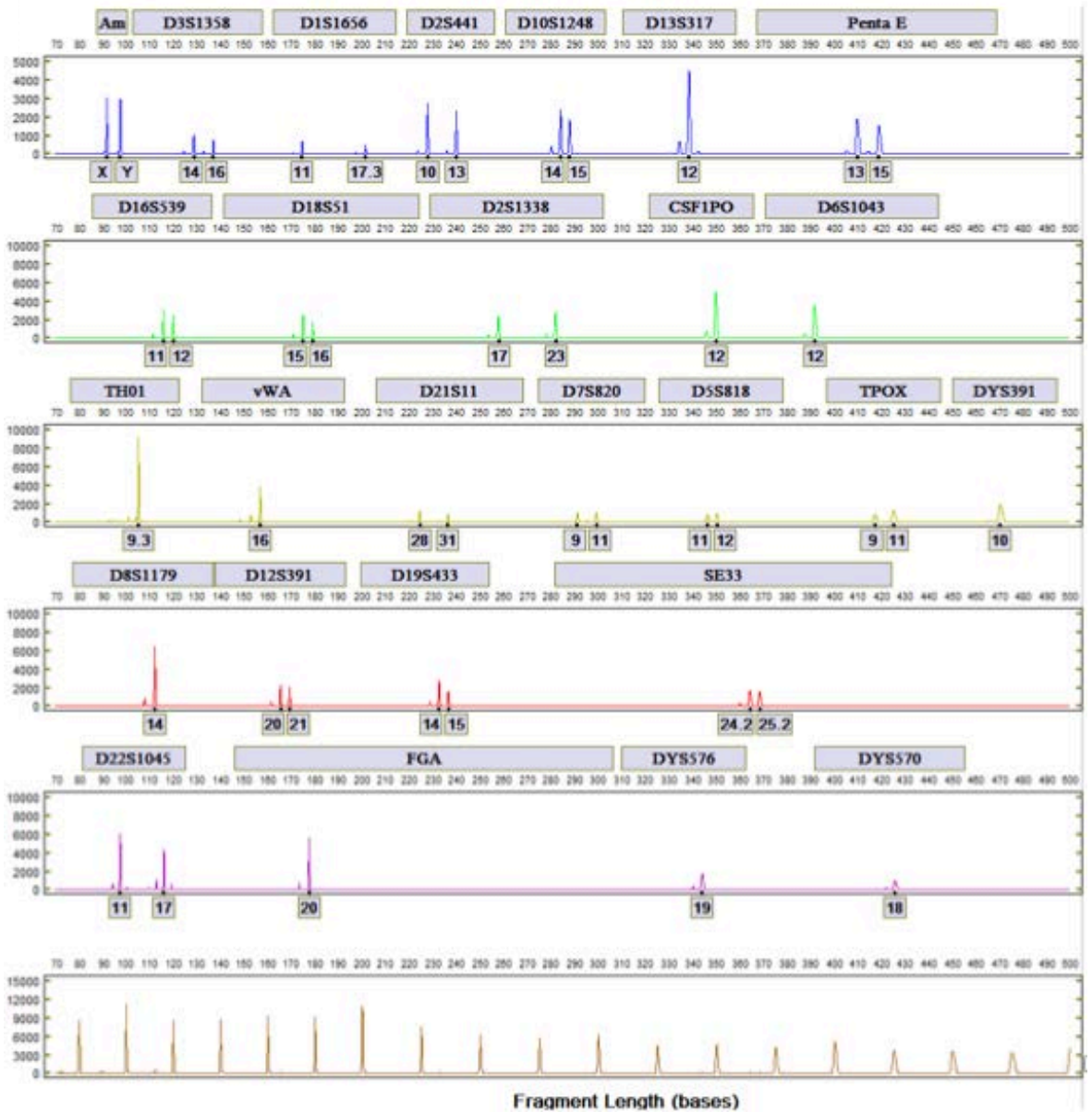


Processing Casework Samples: Oral Epithelial Cells





Processing Touch Samples: Gum



Third Party ANDE Ruggedization Testing

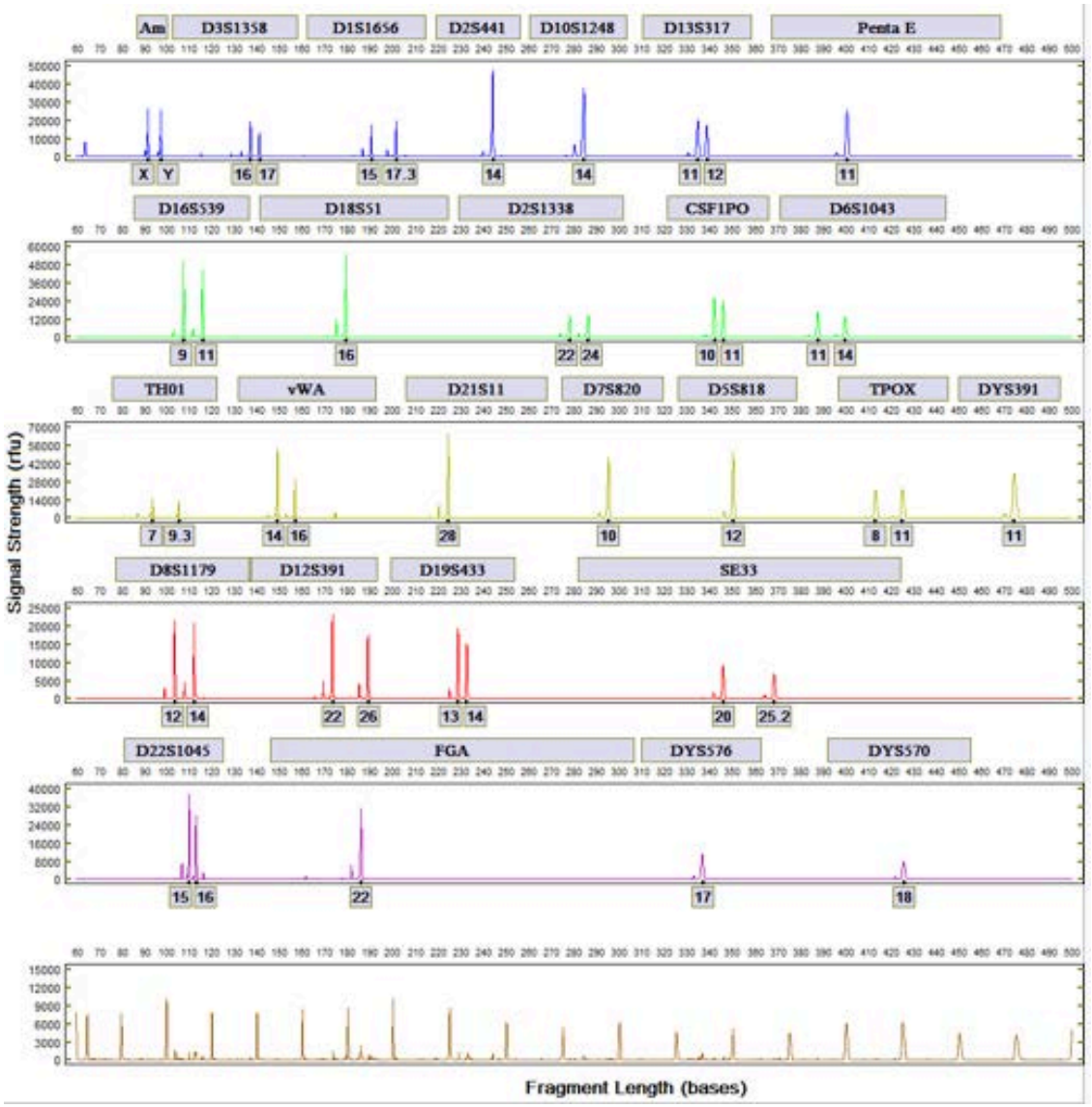
- Instrument
  - Shock (MIL-STD-810G)
  - Vibration (MIL-STD-810G)
  - Operating altitude (>8000 feet)
  - Operating humidity (>90%)
- Chips
  - Shock (MIL-STD-810G)
  - Vibration (MIL-STD-810G)
  - Storage temperature (0 – 45°C)
  - Storage altitude (>8000 feet)
- Chips subjected to environmental and transport testing performed at > 93% success and no test condition impacted performance.

Disaster Victim Identification

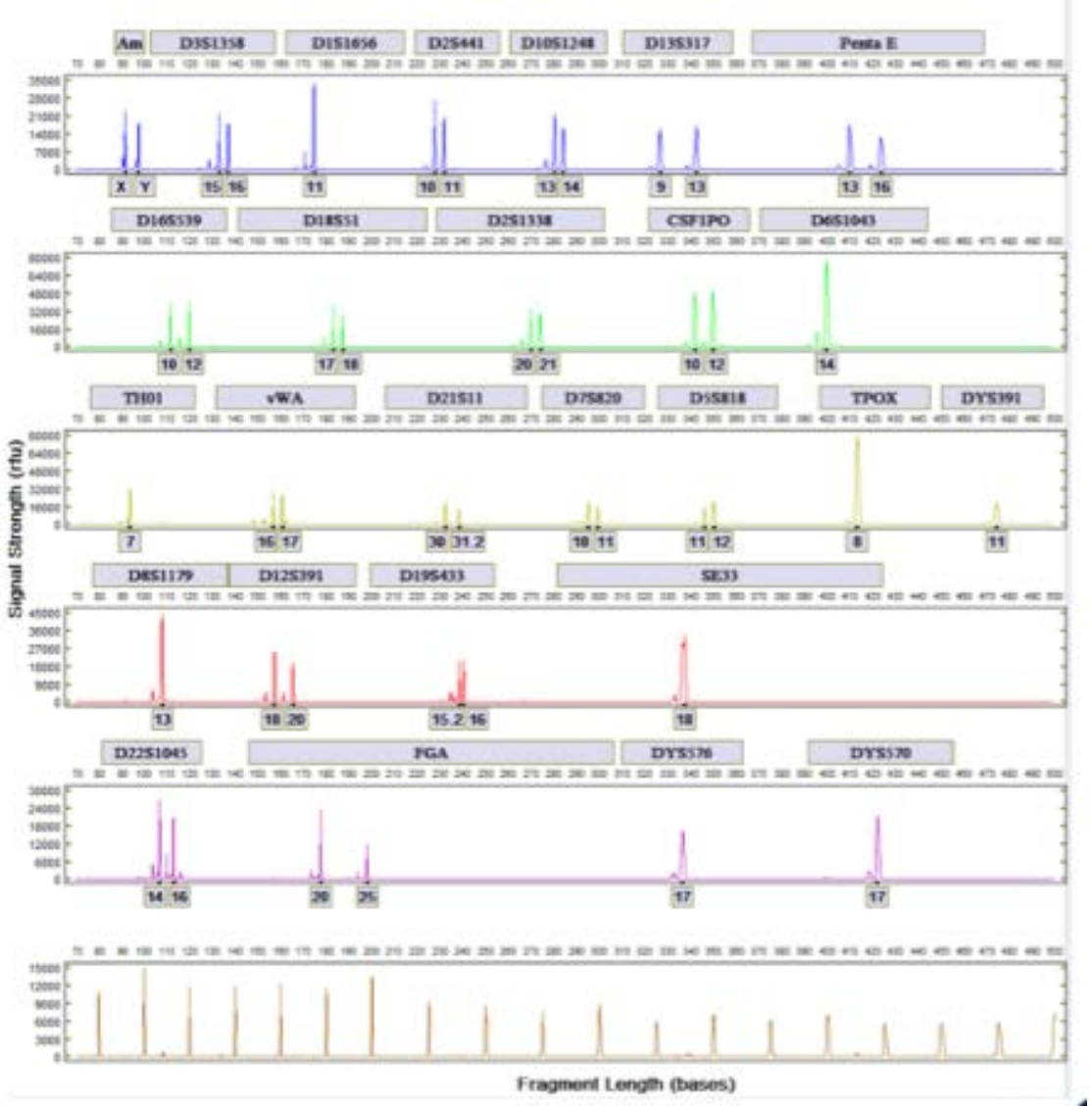
Rationale for Rapid DNA

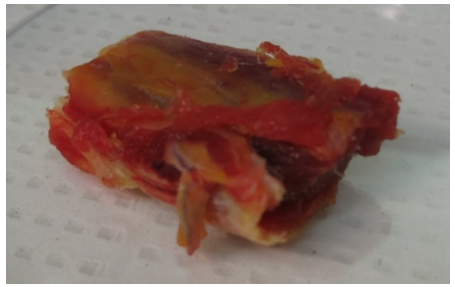
- Conventional processing of DVI samples requires sophisticated equipment, highly skilled technical operators, complex data interpretation, and kinship analysis—all of which require significant time.
- Local laboratories may be rendered non-functional by the disaster, and other laboratories may be overwhelmed by the volume of samples to be analyzed.
- Even a relatively small disaster such as a plane crash can take years for body parts to be identified.
- When large mass disasters occur, bodies may be unidentified for years or decades. The 2004 Indian Ocean earthquake and tsunami is a tragic example—more than a decade later, thousands of victims remain unidentified.

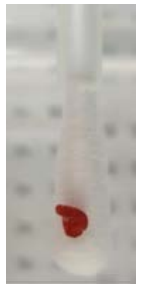
Processing Bone: Two Hours Outside the Lab



Processing Muscle

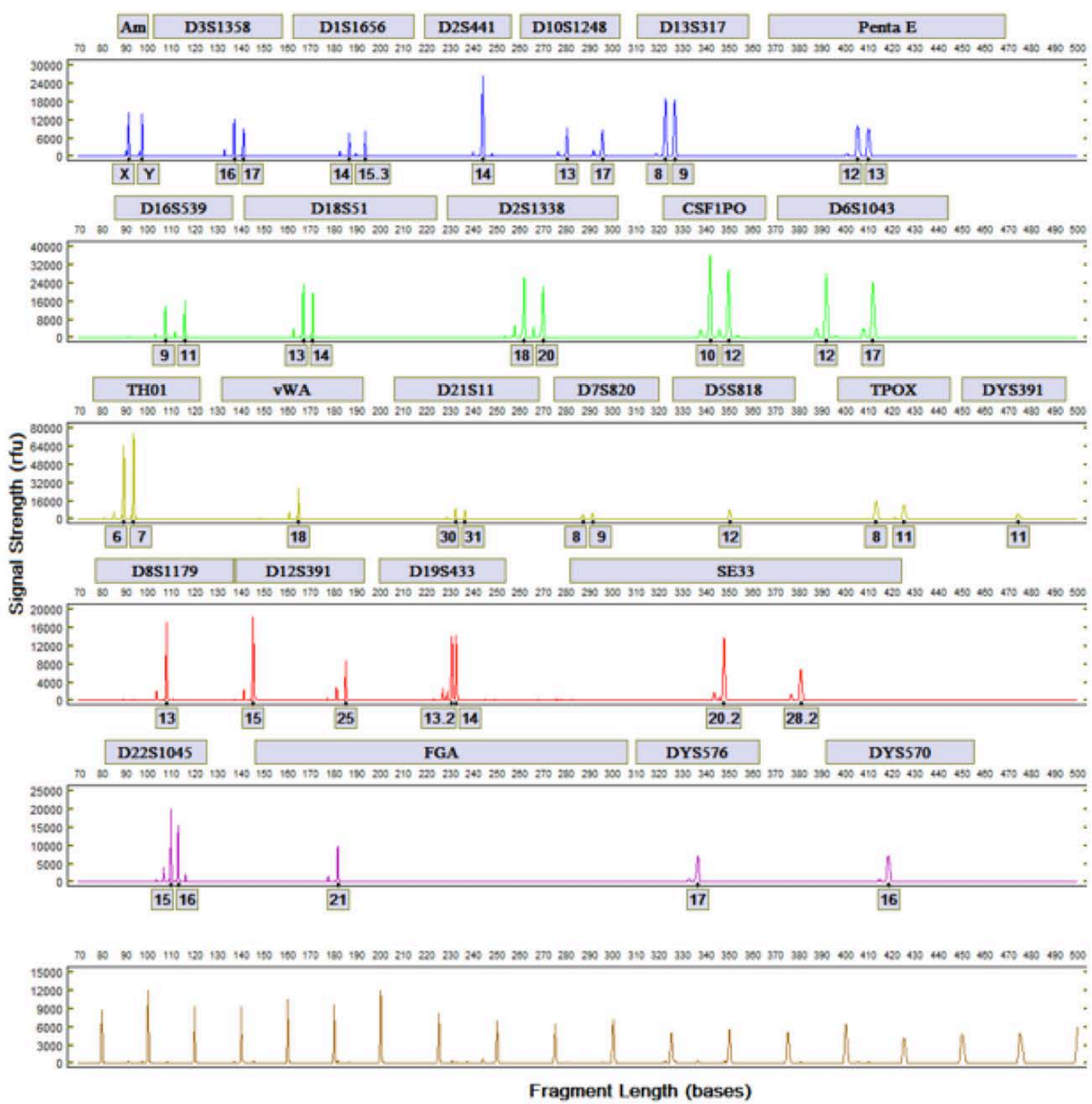


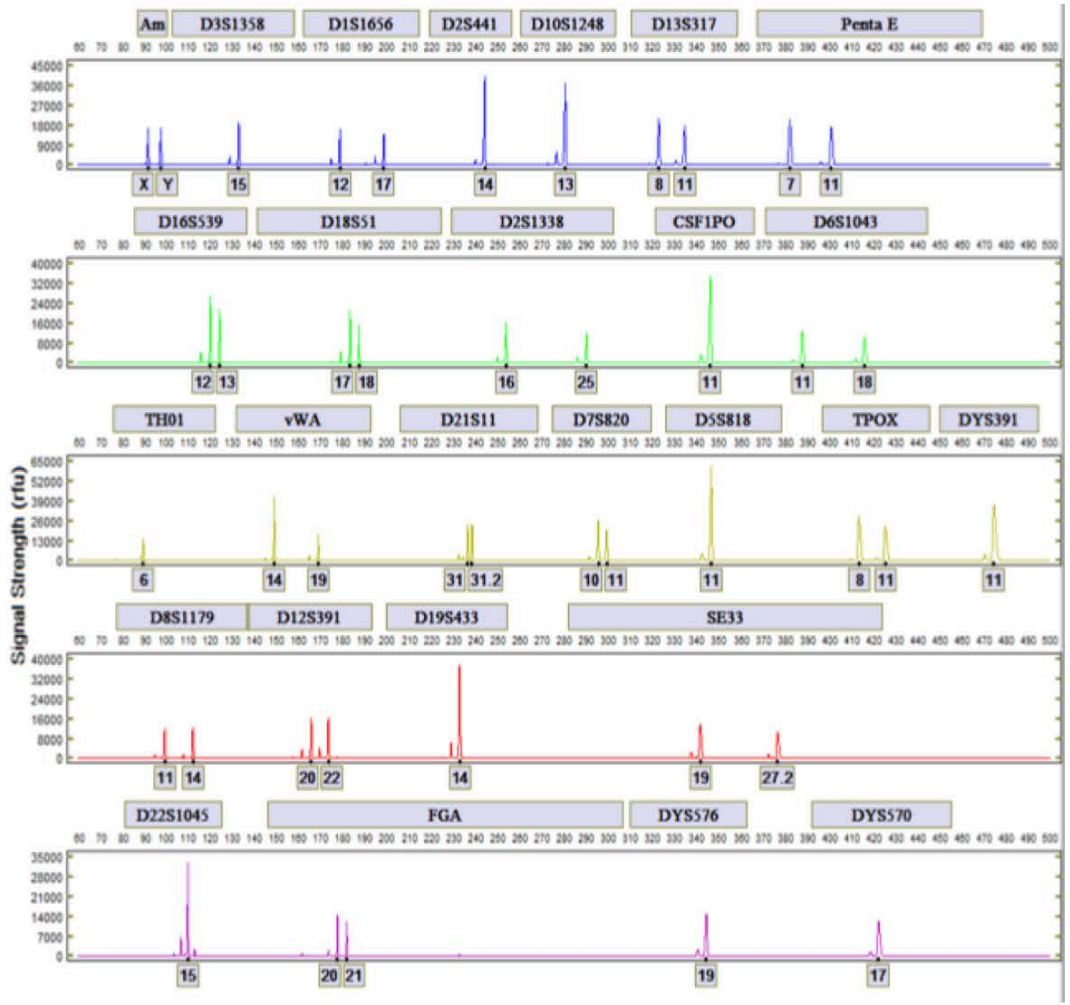




- Very small quantities required with fresh tissue (about the size or a grain of rice)
- Surveyed a wide range of muscle types

Other Tissue Types





Brain (left) and Liver (Right)

Kinship Analysis: Benefits of FlexPlex

		Cases with Probability of Kinship >99.5%		
Relationship	Total Cases	CODIS 13	CODIS 20	FlexPlex 27
Full Sibling	N = 50	38	44	46
Half Sibling	N = 7	1	2	3
Grandparent-Grandchild	N = 22	3	4	8

- The FlexPlex Assay, based on 27 STR loci, is the most powerful kinship assay currently available. FlexPlex allows high probability determinations where other assays fail.
- FAIRS Kinship Software performs calculations on FlexPlex data generated by ANDE and other assays generated in the lab
- ANDE is ruggedized, portable, and runs off generator power, and, in tandem with FAIRS, allows kinship determination in embassies, at borders and ports, and at disaster sites.

Conclusions

- First and Only NDIS approval for a Rapid DNA System
  - Buccal swabs, CODIS 13 approval received March 2016. Integrated Expert System capable of database searching without analyst data interpretation.
  - FlexPlex 27 DV in progress
- ANDE integrates all DNA analysis processes into a single desktop-sized unit that can easily be operated by non-technical users.
- The FlexPlex 6-color, 27-locus assay provides improved discrimination power of autosomal STR loci for increased compatibility with international databases and kinship determination.
- Chips provide an efficient means for analysis of reference, casework, disaster victim identification, and sensitive site exploitation samples.
- FAIRS software allows database management, search and match, and kinship modules.
- ANDE is ruggedized, portable, and runs off generator power, allowing DNA analysis in compromised environments with minimal infrastructure.
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