



**EDNAP and 34<sup>th</sup> ENFSI DNA WG Meeting**  
**April 6-8, 2011 – Brussels, Belgium**



# NIST Update

**John M. Butler**

**NIST Human Identity Project Team**  
National Institute of Standards and Technology  
Gaithersburg, Maryland USA





# NIST Human Identity Project Teams within the Applied Genetics Group

Manuel **Fondevila**  
Alvarez

## *Forensic DNA Team*

## *Data Analysis Support*

## *DNA Biometrics Team*



John  
Butler



Mike  
Coble



Becky  
Hill



Margaret  
Kline



Dave  
Duewer



Pete  
Vallone



Erica  
Butts



Kristen Lewis  
O'Connor

Funding from the **National Institute of Justice (NIJ)**  
through NIST Office of Law Enforcement Standards

Funding from the **FBI S&T Branch**  
through NIST Information Access Division

Workshops  
& Textbooks

Concordance  
& LT-DNA

Software Tools &  
Data Analysis

ABI 3500  
& DNA  
Extraction

Mixtures,  
mtDNA & Y

SRM work,  
variant alleles &  
Cell Line ID

Rapid PCR  
& Biometrics

D12/vWA  
& Kinship  
Analysis



<http://www.cstl.nist.gov/biotech/strbase/NISTpub.htm>

[john.butler@nist.gov](mailto:john.butler@nist.gov)

001-301-975-4049



# Presentation Topics

- SRM 2391c to be available mid-2011
- STR kit concordance studies
- SE33 sequence information
- New STR loci characterized
- Tri-allelic patterns
- ABI 3500 open letter and NIST validation studies
- Training workshops & information
- *Advanced Topics in Forensic DNA Typing* (3<sup>rd</sup> edition)
- New STRBase sections: LT-DNA, mixtures, kinship

# Standard Reference Material (SRM) 2391c

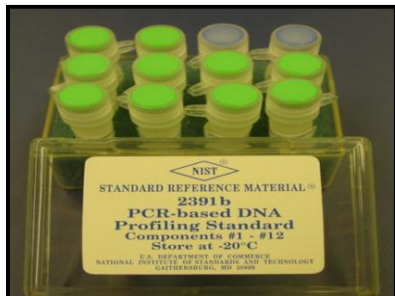
<http://www.nist.gov/srm>

*Traceable standards to ensure accurate and comparable measurements between laboratories*



Standards Reference Material

**Calibration with SRMs enables confidence in comparisons of results between laboratories**



**SRM 2391b**  
48 autosomal STRs  
characterized across  
12 DNA samples

## SRM 2391c Details

- 6 components
- **4 liquid genomic DNA components (PFA tubes) + 2 punches** (cells on **FTA** & 903 paper)
- **Certified values: 23 autosomal STR loci and 16 Y-STRs (all loci in kits)** either through sequencing alleles or examining multiple STR kits
- **Reference values: 26 autosomal STRs** (NIST 26plex + FFFL)
- 5 single source + **1 mixture**
- 3 males + 2 females (unique)
- All new samples
  - no 9947A or 9948
- **Will be available in mid-2011**

# STR Kits Tested with SRM 2391c

## Applied Biosystems (12)

Identifiler  
Identifiler Plus  
**NGM**  
**NGM SElect**  
COfiler  
Profiler  
Profiler Plus  
Profiler Plus ID  
**SGM Plus**  
**SEfiler Plus**  
MiniFiler  
Yfiler

## Promega (9)

PowerPlex 16  
PowerPlex 16 HS  
PowerPlex Y  
**PowerPlex ESX 17**  
**PowerPlex ESI 17**  
**PowerPlex ES**  
PowerPlex S5  
PowerPlex Y  
FFFL

## Qiagen (2)

**ESSplex**  
IDplex

## NIST assays

26plex  
miniSTRs

## Alleles sequenced:

SE33  
D12S391  
D1S1656  
Penta D  
Penta E  
D8S1115

**23 commercial STR kits examined**

**NIST developed 26plex and miniplexes**

**No discordant results observed on SRM 2391c samples**

# Commercially Available STR Kits

## Applied Biosystems (17)

- ~~AmpFISTR Blue (1996)~~
- ~~AmpFISTR Green I (1997)~~
- Profiler (1997)
- Profiler Plus (1997)
- COfiler (1998)
- SGM Plus (1999)
- **Identifiler** (2001)
- Profiler Plus ID (2001)
- ~~SEfiler (2002)~~
- **Yfiler (2004)**
- MiniFiler (2007)
- SEfiler Plus (2007)
- Sinofiler (2008) – China only
- **Identifiler Direct** (2009)
- NGM (2009)
- **Identifiler Plus** (2010)
- NGM SElect (2010)

## Promega Corporation (13)

- PowerPlex 1.1 (1997)
- PowerPlex 1.2 (1998)
- PowerPlex 2.1 (1999)
- **PowerPlex 16** (2000)
- PowerPlex ES (2002)
- **PowerPlex Y (2003)**
- PowerPlex S5 (2007)
- **PowerPlex 16 HS** (2009)
- PowerPlex ESX 16 (2009)
- PowerPlex ESX 17 (2009)
- PowerPlex ESI 16 (2009)
- PowerPlex ESI 17 (2009)
- PowerPlex 18D (2010)

## Qiagen (2010)

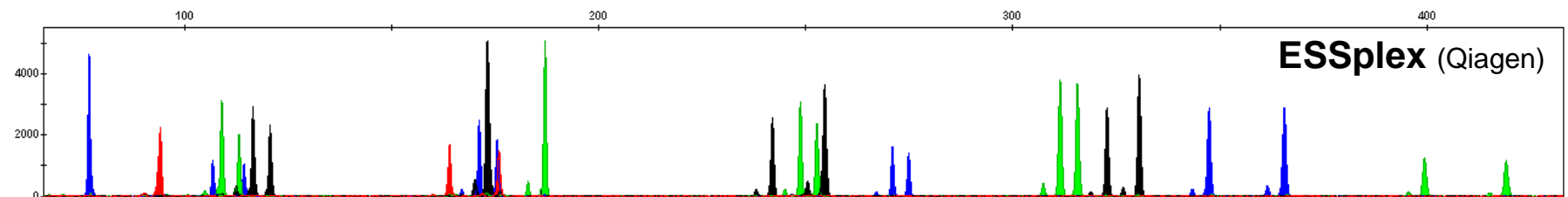
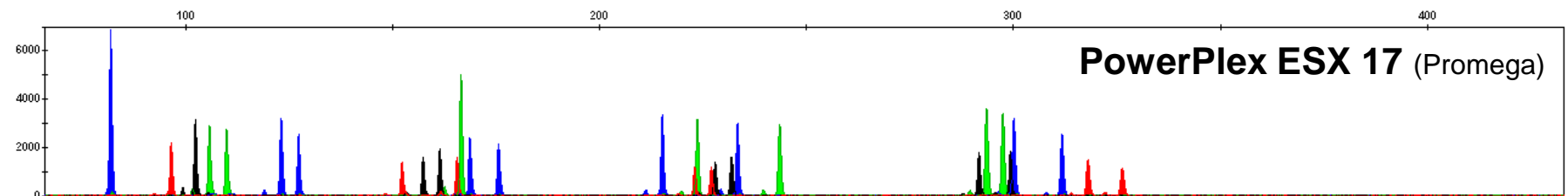
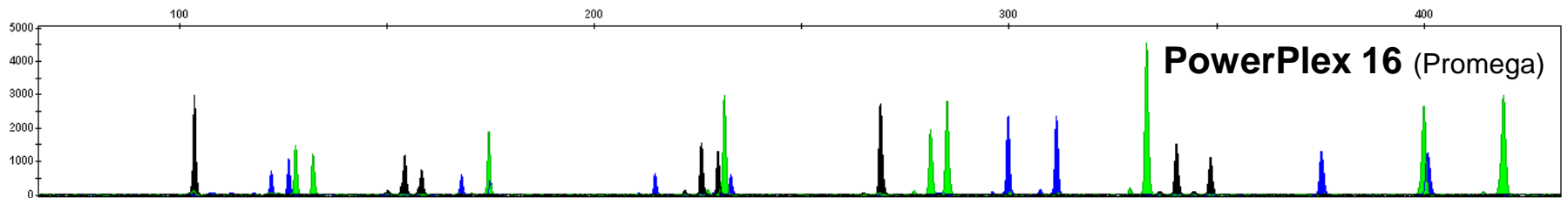
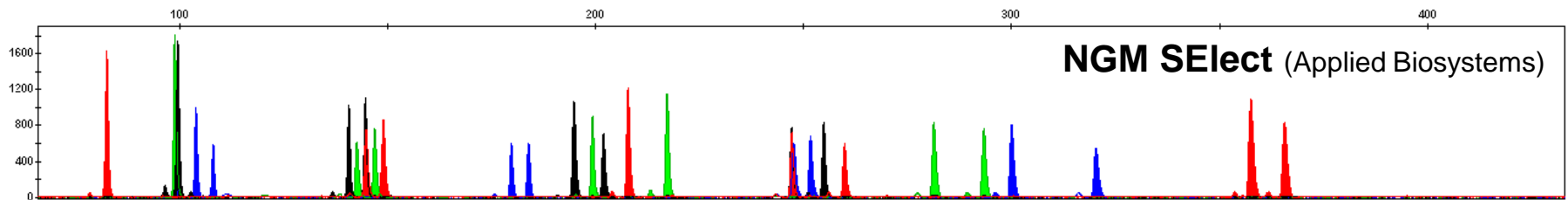
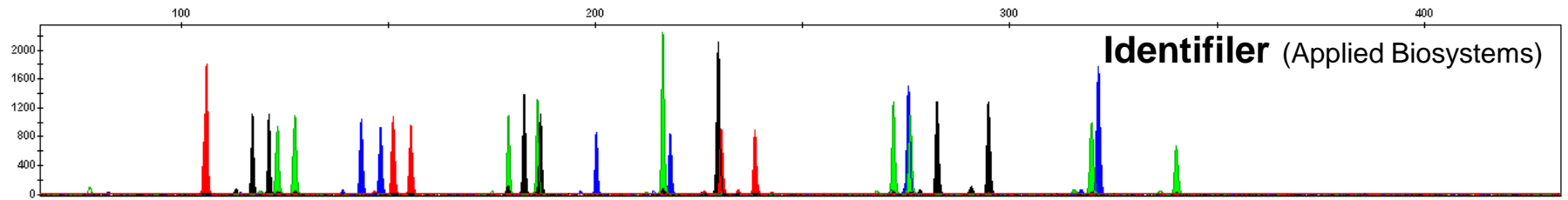
*Primarily selling kits in Europe  
Due to patent restrictions  
cannot sell in U.S.*

- ESSplex
- ESSplex SE
- Decaplex SE
- IDplex
- Nonaplex ESS
- Hexaplex ESS
- HD (Chimera)
- Argus X-12
- Argus Y-12
- **DIplex (30 indels)**

**~1/3 of all STR kits were  
released in the last two years**



# Same DNA Sample Tested with Five STR Kits





# STR Kit Concordance Testing

*Profiles in DNA* Article Published April 2010

Article Type: Feature

Volume 13 No. 1, April 2010

## Strategies for Concordance Testing

Carolyn R. Hill, Margaret C. Kline, David L. Duewer and John M. Butler

National Institute of Standards and Technology, Biochemical Science Division, Gaithersburg, Maryland, USA

### 4 S's of Concordance Testing

**Standard samples** (data on same samples)

**Software** (to check data concordance)

**Sequencing** (to understand null alleles)

**STRBase** (sharing with the community)

*Concordance evaluation or "null alleles" present commercial short tandem markers available to the kits because the primer (PCR) product sizes. W may occur due to primer-binding-site mutations that affect one set of primers but not another.*

[http://www.promega.com/profiles/1301/1301\\_08.html](http://www.promega.com/profiles/1301/1301_08.html)

# NIST Pipeline for STR Kit Analysis

Work by Becky Hill and Dave Duewer

- Concordance testing with standard samples
  - Sequence analysis of any null alleles to understand differences
- Locus characteristics
  - Heterozygote peak height ratios
  - Stutter percentages (including allele-specific)
- Allele frequencies for all new loci
  - Across U.S. Caucasian, Hispanic, African American, and Asian
- Probability of identity for different locus sets

# Summary of NIST Samples Evaluated

- **U.S. Population Samples (657 samples)**
  - Previously studied with Identifiler, MiniFiler, Yfiler, PP16, miniSTRs, and many additional assays (>200,000 allele calls)
  - 260 African Americans, 260 Caucasians, 140 Hispanics, and 3 Asians

<http://www.cstl.nist.gov/biotech/strbase/NISTpop.htm>
- **U.S. Father/Son pairs (786 samples)**
  - Previously studied with Identifiler, MiniFiler, Yfiler
  - **~100 fathers/100 sons for each group:** African Americans, Caucasians, Hispanics, and Asians
- **NIST SRM 2391b** PCR DNA Profiling Standard (**12 samples**)
  - Components 1-10 (includes 9947A and 9948): *well characterized*
  - ABI 007 and K562

**Total number of samples = 1455**

**1443 population samples**

# Kit Concordance Comparisons

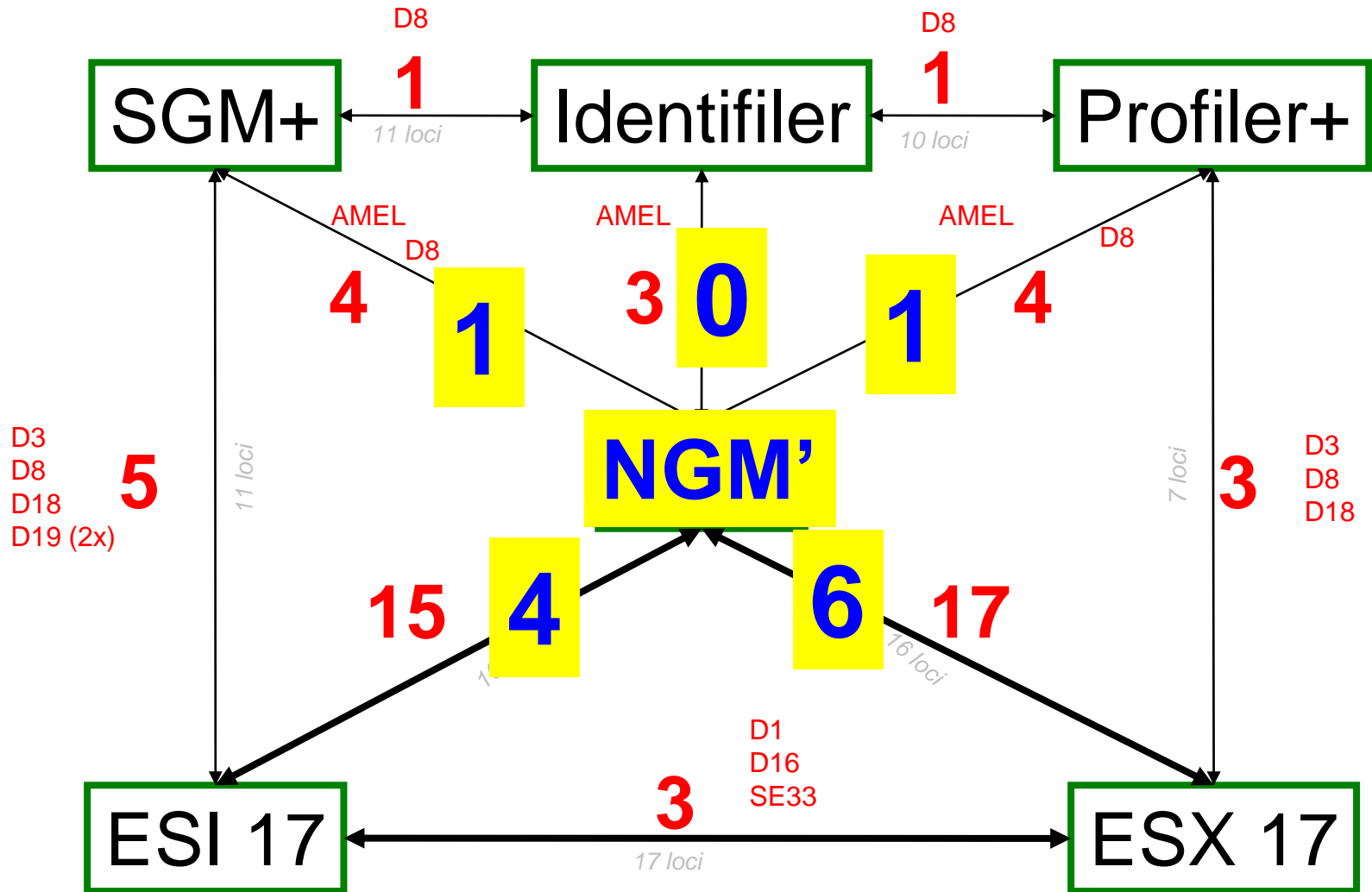
<u>Kits compared</u>	<u>Samples</u>	<u>Loci compared</u>	<u>Comparisons</u>	<u># Differences</u>	<u>Concordance (%)</u>
SGM-ID	1436	11	15,796	1	99.994
ID-ProPlus	1427	10	14,270	1	99.993
ID-IDplex	669	16	10,704	19	99.822
ID-PP16	662	14	9,268	4	99.957
ID-MiniFiler	1308	9	11,772	27	99.771
SGM-NGM	1436	11	15,796	1	99.975
ID-NGM	1449	11	15,939	1	99.981
ProPlus-NGM	1427	11	15,939	1	99.972
SGM-ESI	1436	11	15,796	1	99.968
ProPlus-ESX	1427	11	15,939	1	99.970
ESI-ESX	1455	11	15,939	1	99.939
ESI-ESSplex	1445	11	15,939	1	99.875
ESX-ESSplex	1445	16	23,120	30	99.870
ESI-NGMSElect	715	17	12,155	17	99.860
ESX-NGMSElect	715	17	12,155	7	99.942
ESS-NGMSElect	663	17	11,271	17	99.849
		TOTAL	240,156	186	99.923

**240,156 comparisons**  
**186 total differences**  
**99.92% concordance**  
*(many corrected now)*

*Kits (except Identifiler) were kindly provided by **Applied Biosystems, Promega, and Qiagen** for concordance testing performed at NIST*

# Initial Concordance Testing Summary

## Number of Discordant Results Observed

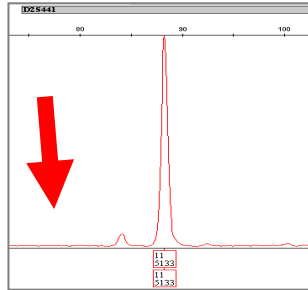


# Extra (Degenerate) Primers Added with NGM SElect

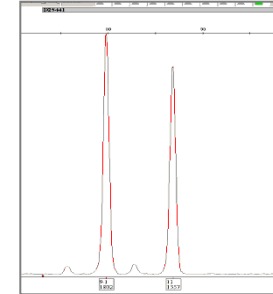
**NGM (original)**

**NGM SElect  
and NGM'**

**D2S441**



11,11



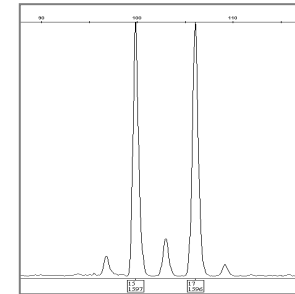
9.1,11

9.1 allele missing in 7 Asians

**D22S1045**



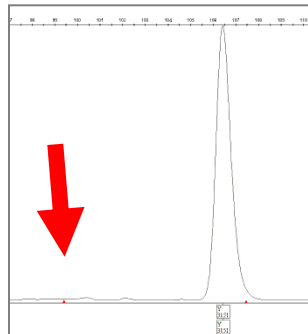
17,17



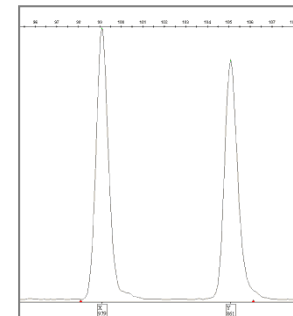
15,17

15 allele missing in 4 samples

**Amelogenin**



Y,Y



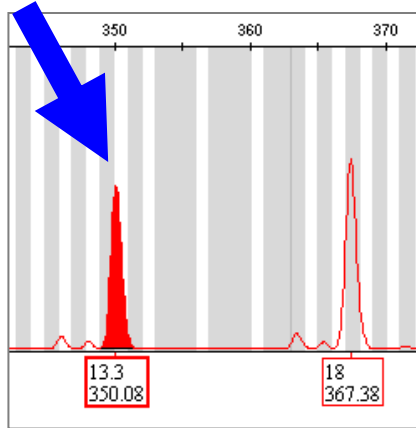
X,Y

X allele missing in 3 samples

# SE33 Differences

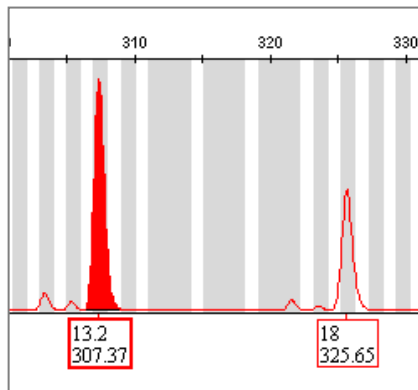
NGM Select/PP ESX 17 vs PP ESI 17

# Impact of SE33 Primer Positions



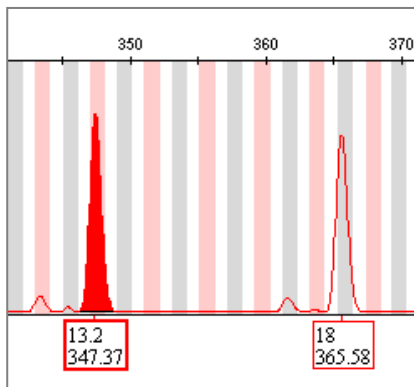
PowerPlex **ESI 17** (30 cycles)

**“13.3”, 18**



PowerPlex **ESX 17** (30 cycles)

**13.2, 18**



NGM **SElect** (29 cycles)

**13.2, 18**



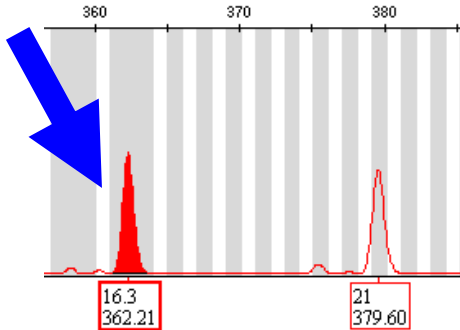
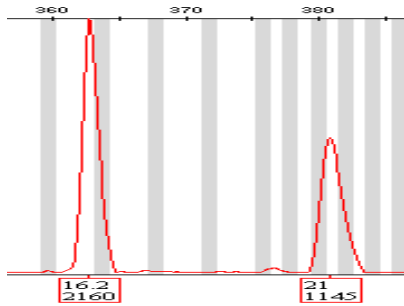
# SE33 Seq Differences

- Total African American samples tested:
    - 46 Blood samples
    - 258 Population samples
    - 190 Father/Son samples
- 494 AA samples total**
- 12 seq variations found out of 494 samples
    - **9 from earlier ESI/ESX data (not detected previously due to poor resolution of SE33 alleles)**

**2.43% NIST AA samples exhibit ESI difference**

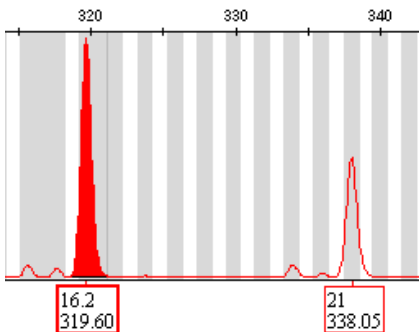
# Review of Our SE33 Data

Original ESI 17 data – incorrectly designated “16.2, 21”  
(broad peaks due to poor 3130xl resolution)



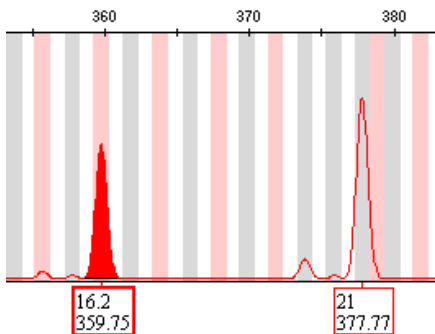
PowerPlex **ESI 17** (30 cycles)

**“16.3”, 21**



PowerPlex **ESX 17** (30 cycles)

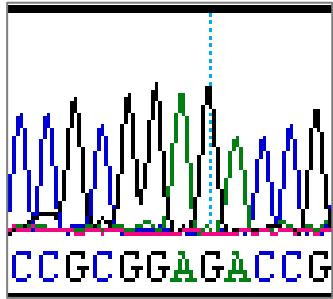
**16.2, 21**



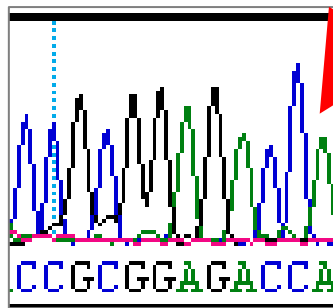
NGM **SElect** (29 cycles)

**16.2, 21**

# SE33 Sequence Reason for Migration Shift



**Normal SE33 allele flanking region**  
(57-68 bases downstream of repeat)



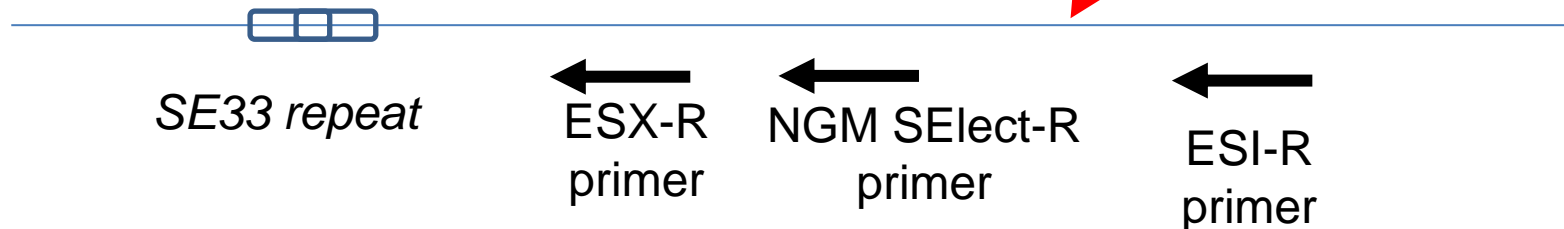
**Mutant SE33 Allele**

**G → A 68 bp**

downstream of SE33 repeat

**(no length difference)**

***Relative primer positions***



Collaboration with Promega (Bob McLaren and Doug Storts)

PCR products for sequencing kindly provided by Ingo Bastisch (BKA) and Volker Weirich (LKA-MV)

# The 10 STR Loci Beyond the CODIS 13

5 new European loci

STR Locus	Location	Repeat Motif	Allele Range*	# Alleles*
<b>D2S1338</b>	2q35	TGCC/TTCC	10 to 31	40
<b>D19S433</b>	19q12	AAGG/TAGG	5.2 to 20	36
<b>Penta D</b>	21q22.3	AAAGA	1.1 to 19	50
<b>Penta E</b>	15q26.2	AAAGA	5 to 32	53
<b>D1S1656</b>	1q42	TAGA	8 to 20.3	25
<b>D12S391</b>	12p13.2	AGAT/AGAC	13 to 27.2	52
<b>D2S441</b>	2p14	TCTA/TCAA	8 to 17	22
<b>D10S1248</b>	10q26.3	GGAA	7 to 19	13
<b>D22S1045</b>	22q12.3	ATT	7 to 20	14
<b>SE33</b>	6q14	AAAG <sup>‡</sup>	3 to 49	<b>178</b>

\*Allele range and number of observed alleles from Appendix 1, J.M. Butler (2011) *Advanced Topics in Forensic DNA Typing: Methodology*; <sup>‡</sup>SE33 alleles have complex repeat structure

Loci sorted on Probability of Identity ( $P_I$ ) values

STR Locus	Alleles Observed	Genotypes Observed	Het. (obs)	$P_I$ value <b>N = 938</b>
SE33	53	292	0.9360	0.0069
Penta E*	20	114	0.8799	0.0177
D2S1338	13	68	0.8785	0.0219
D1S1656	15	92	0.8934	0.0220
D18S51	21	91	0.8689	0.0256
D12S391	23	110	0.8795	0.0257
FGA	26	93	0.8742	0.0299
Penta D*	16	71	0.8754	0.0356
D21S11	25	81	0.8358	0.0410
D19S433	16	76	0.8124	0.0561
D8S1179	11	45	0.7878	0.0582
vWA	11	38	0.8060	0.0622
D7S820	11	32	0.8070	0.0734
TH01	8	24	0.7580	0.0784
D16S539	9	28	0.7825	0.0784
D13S317	8	29	0.7655	0.0812
D10S1248	12	39	0.7825	0.0837
D2S441	14	41	0.7772	0.0855
D3S1358	11	30	0.7569	0.0873
D22S1045	11	42	0.7697	0.0933
CSF1PO	9	30	0.7537	0.1071
D5S818	9	34	0.7164	0.1192
TPOX	9	28	0.6983	0.1283

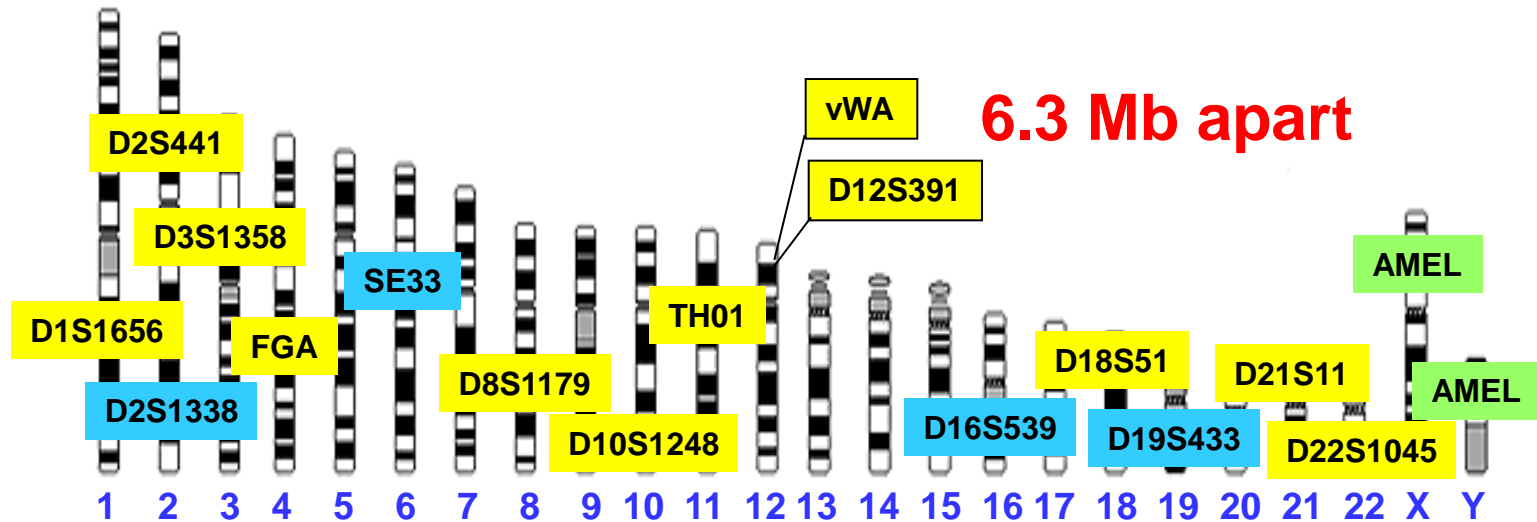
23 STR Loci  
present in STR kits  
rank ordered by their  
variability

Better for  
mixtures (more  
alleles seen)

There are several loci  
more polymorphic  
than the **current  
CODIS 13 STRs**

Better for kinship  
(low mutation  
rate)

# Chromosomal Positions for the European Standard Set and Other Common STR Markers Used



**European Standard Set** + D16S539, D2S1338, D19S433, SE33

**Can vWA and D12S391 be used with the product rule  
for match probability calculations? YES**

# Re-testing has shown no LD

G Model  
FSIGEN-643; No. of Pages 3



**ARTICLE IN PRESS**

Forensic Science International: Genetics xxx (2010) xxx–xxx

Contents lists available at ScienceDirect

Forensic Science International: Genetics

journal homepage: [www.elsevier.com/locate/fsig](http://www.elsevier.com/locate/fsig)



Short communication

Linkage disequilibrium analysis of D12S391 and vWA in U.S. population and paternity samples<sup>☆</sup>

Kristen Lewis O'Connor\*, Carolyn R. Hill, Peter M. Vallone, John M. Butler

*Biochemical Science Division, National Institute of Standards and Technology, 100 Bureau Drive, Gaithersburg, MD 20899-8312, United States*

**Original abstract:** No significant evidence of linkage disequilibrium was observed between the loci in the population samples. However, *significant linkage disequilibrium was detected in U.S. African American, Caucasian, and Asian father/son samples with phased genotypes.*

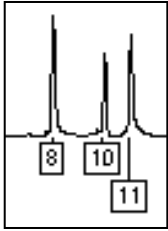
**An errata will be published with this article**

# Updated Conclusions on D12S391 and vWA

- **Single-locus genotype probabilities** may be **multiplied** for profile match probability calculations when **unrelated individuals** are involved
- **Recombination fraction and** the maximum likelihood estimates of the D12S391/vWA **haplotype frequencies** should be used for **kinship analysis**



TPOX



# Tri-Allelic Patterns

- Tri-alleles are Copy Number Variants (CNVs) in the human genome detected as three peaks at a single locus rather than the expected single (homozygous) or double (heterozygous) peak
- Observed at a rate of ~1 in every 1,000 DNA profiles with some loci having a higher rate
- With a million DNA profiles going into NDIS each year, **collectively U.S. DNA databasing labs will see approximately 1,000 tri-alleles this next year**

# Frequency of Tri-Allelic Patterns

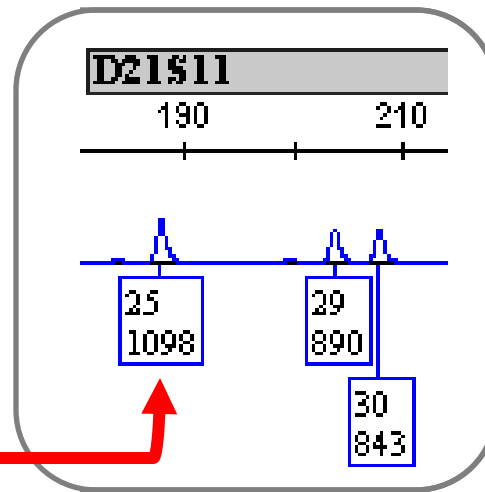
- Database Size:  
**69,000**
- Overall Average Occurrence:  
**1 in 1,000**

**Note:**  
This is Steven's  
summary  
of Missouri's data.  
You won't find this  
table on STRBase.

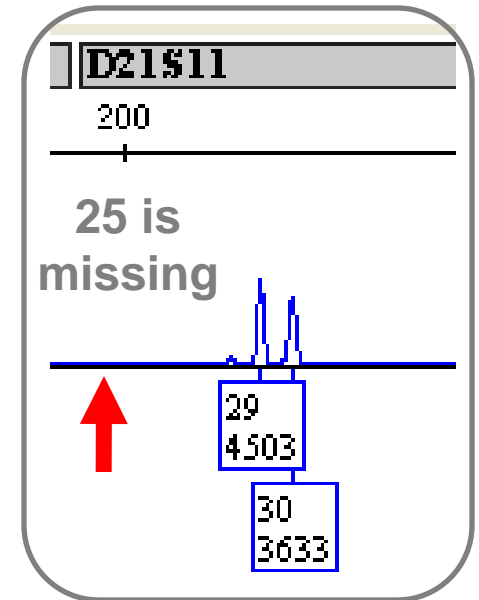
Locus	Observations	1 in...
D3S1358	2	35,000
VWA	10	6,900
FGA	11	6,300
D8S1179	2	35,000
D21S11	9	7,700
D18S51	3	23,000
D5S818	1	69,000
D13S317	4	17,000
D7S820	0	
D16S539	3	23,000
TH01	0	
TPOX	9	7,700
CSF1PO	1	69,000
Penta D	3	23,000
Penta E	10	6,900
<b>Combined</b>	<b>68</b>	<b>1,000</b>

# How Do You Characterize Your Tri-Allelic Patterns?

## Identifiler



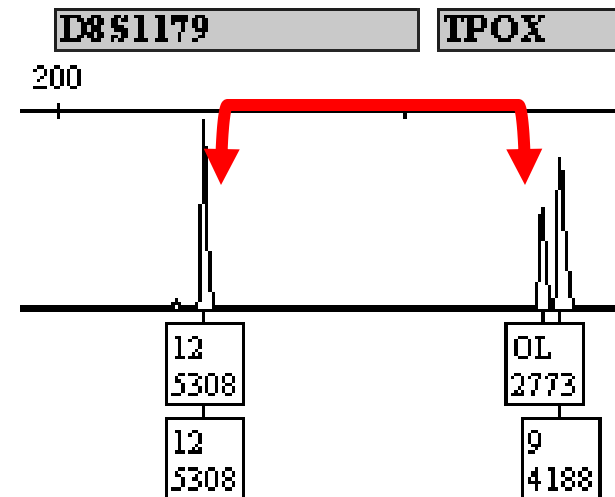
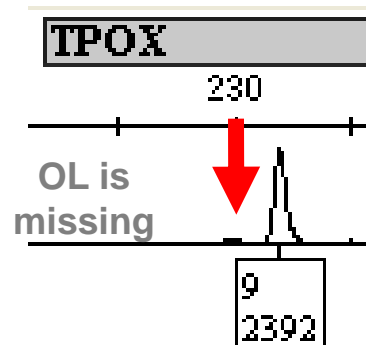
## PowerPlex 16 HS



You re-amplify it...  
It's Reproducible!

Check STRBase...  
It has never been  
observed before!

A New Large D8S1179  
Allele is Discovered –  
with “24” repeats!  
(sequence analysis shows  
duplication in flanking region)



# D8S1179

All Previously  
Known Alleles

Many alleles  
sequences  
are not  
known

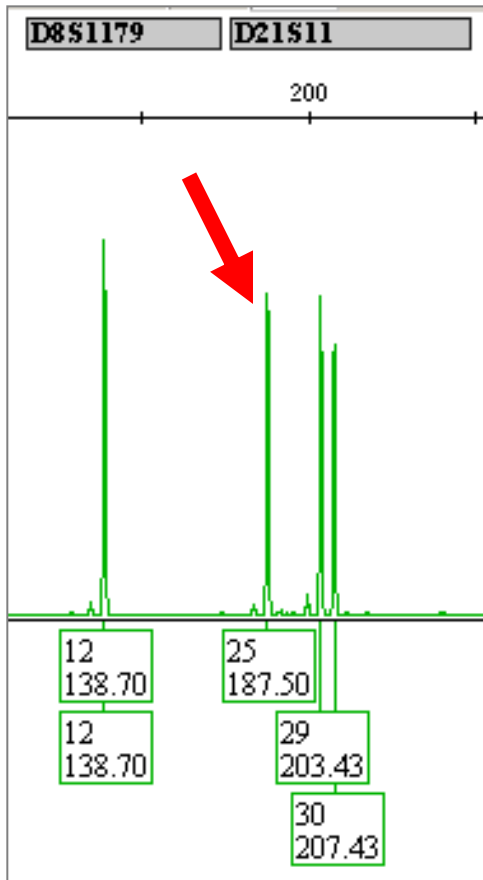
We just set the  
new world record  
for the largest D8  
allele ("24")

Allele (Repeat #)	Promega PowerPlex 16	ABI Identifiler	Repeat Structure [TCTR] <sub>n</sub>	Reference
6	199 bp	119 bp	Not published	STRBase
7	203 bp	123 bp	[TCTA] <sub>7</sub>	Griffiths <i>et al.</i> (1998)
8	207 bp	127 bp	[TCTA] <sub>8</sub>	Barber and Parkin (1996)
9	211 bp	131 bp	[TCTA] <sub>9</sub>	Barber and Parkin (1996)
10	215 bp	135 bp	[TCTA] <sub>10</sub>	Barber and Parkin (1996)
10.1	216 bp	136 bp	Not published	STRBase
10.2	217 bp	137 bp	Not published	STRBase
11	219 bp	139 bp	[TCTA] <sub>11</sub>	Barber and Parkin (1996)
12	223 bp	143 bp	[TCTA] <sub>12</sub>	Barber and Parkin (1996)
12.1	224 bp	144 bp	Not published	STRBase
12.2	225 bp	145 bp	Not published	STRBase
12.3	226 bp	146 bp	Not published	STRBase
13 (a)	227 bp	147 bp	[TCTA] <sub>11</sub> [TCTG] <sub>1</sub> [TCTA] <sub>11</sub>	Barber and Parkin (1996)
13 (b)	227 bp	147 bp	[TCTA] <sub>2</sub> [TCTG] <sub>1</sub> [TCTA] <sub>10</sub>	<b>Kline <i>et al.</i> (2010)</b>
13 (c)	227 bp	147 bp	[TCTA] <sub>1</sub> [TCTG] <sub>1</sub> TGTA[TCTA] <sub>10</sub>	<b>Kline <i>et al.</i> (2010)</b>
13 (d)	227 bp	147 bp	[TCTA] <sub>13</sub>	<b>Kline <i>et al.</i> (2010)</b>
13.1	228 bp	148 bp	Not published	STRBase
13.2	229 bp	149 bp	Not published	STRBase
13.3	230 bp	150 bp	Not published	STRBase
14	231 bp	151 bp	[TCTA] <sub>2</sub> [TCTG] <sub>1</sub> [TCTA] <sub>11</sub>	Barber and Parkin (1996)
14.1	232 bp	152 bp	Not published	STRBase
14.2	233 bp	153 bp	Not published	STRBase
15	235 bp	155 bp	[TCTA] <sub>2</sub> [TCTG] <sub>1</sub> [TCTA] <sub>12</sub>	Barber and Parkin (1996)
15.1	236 bp	156 bp	Not published	STRBase
15.2	237 bp	157 bp	Not published	STRBase
15.3	238 bp	158 bp	Not published	STRBase
16	239 bp	159 bp	[TCTA] <sub>2</sub> [TCTG] <sub>1</sub> [TCTA] <sub>13</sub>	Barber and Parkin (1996)
16.1	240 bp	160 bp	Not published	STRBase
17	243 bp	163 bp	[TCTA] <sub>2</sub> [TCTG] <sub>2</sub> [TCTA] <sub>13</sub>	Barber and Parkin (1996)
17.1	244 bp	164 bp	Not published	STRBase
17.2	245 bp	165 bp	Not published	STRBase
18	247 bp	167 bp	[TCTA] <sub>2</sub> [TCTG] <sub>1</sub> [TCTA] <sub>15</sub>	Barber and Parkin (1996)
19	251 bp	171 bp	[TCTA] <sub>2</sub> [TCTG] <sub>2</sub> [TCTA] <sub>15</sub>	Griffiths <i>et al.</i> (1998)
20	255 bp	175 bp	Not published	<b>STRBase</b>

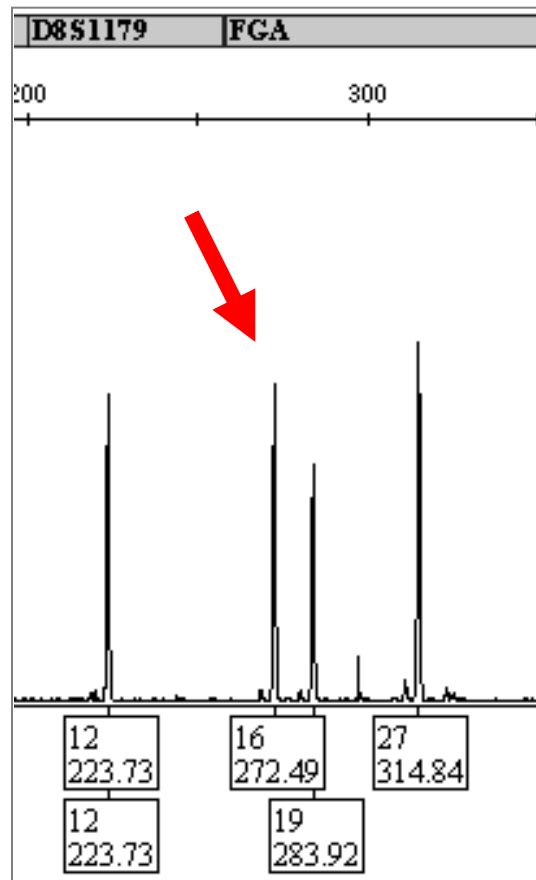


# Result with This Large D8S1179 Allele Using European STR Kits

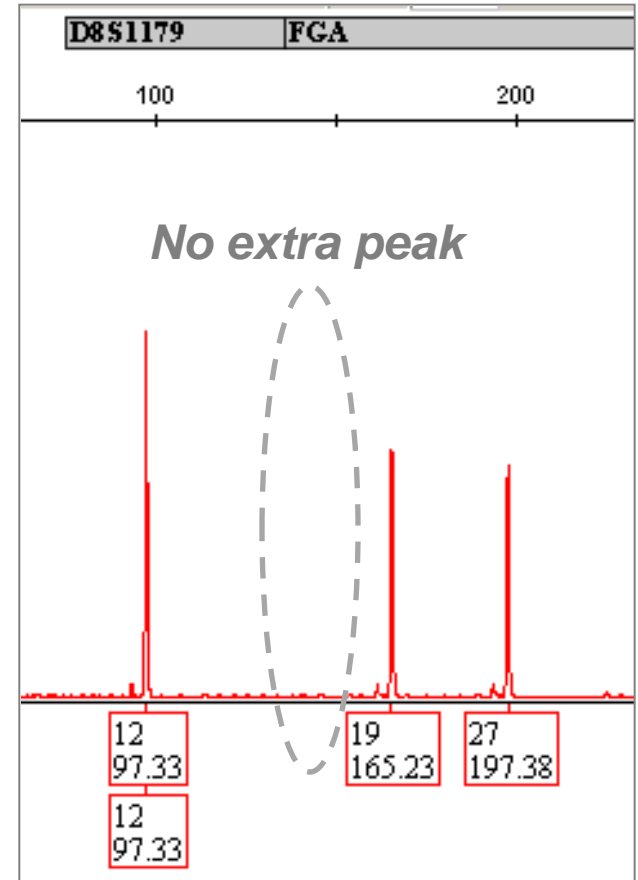
## NGM SElect



## PP ESX 17



## PP ESI 17



**False D21S11 tri-allele**

**False FGA tri-allele**

*Reverse primer internal to duplicated flanking region*

**STR Allele Sequencing Has Been Provided  
Free to the Community for the Past Ten Years  
Thanks to NIJ-Funding**



Short communication

STR sequence analysis for characterizing normal, variant, and null alleles

Margaret C. Kline<sup>\*</sup>, Carolyn R. Hill, Amy E. Decker<sup>1</sup>, John M. Butler

*National Institute of Standards and Technology, 100 Bureau Drive, M/S 8312, Gaithersburg, MD 20899, USA*

**111 normal and variant alleles** sequenced (at 19 STR & 4 Y-STRs)  
**17 null alleles** sequenced (with impact on various STR kit primers)

**Provides primer sequences for 23 autosomal STRs & 17 Y-STRs**  
**Provides full protocol for gel separations and sequencing reactions**  
***Primer positions are outside of all known kit primers***

# TrueAllele Studies Underway at NIST

- We purchased the software in September 2010.
- Mike Coble and Pete Vallone completed a three day training at Cybergenetics (Pittsburgh, PA) in October.
- Software runs on a Linux Server with a Mac interface.
- **We are in the process of validating the software with 2, 3 and 4 person mixtures.**



# First Impressions of TrueAllele

- Powerful software that makes full use of the data.
- Will need to educate the U.S. forensic community (accustomed to using RMNE) how to use LR's and probabilistic methods so they can explain this to a jury.
- **Computer intensive – can take several hours to run one sample.**

# ABI 3500 Genetic Analyzer

Open Letter to Applied Biosystems  
&  
NIST Validation Studies Underway

# DNA Community Moving to ABI 3500s

## Advantages

- Smaller footprint and 110V power requirement
- Better polymer delivery and temperature control
  - Improved success rates?
- New capabilities
  - between instrument normalization
  - 6-dye detection (bigger kits with more loci)
- Simpler software

## Disadvantages

- Up-front cost of new instruments
  - In the U.S., federal government (NIJ) will likely be expected to foot the bill
- Generates .hid files
  - Requires new analysis software
- Validation down-time
  - New RFU thresholds
- Higher per run cost with RFID tags & limited expiration
  - many labs cannot purchase reagents rapidly throughout the year
- Creating technicians not scientists
  - Plug and play approach leading to loss of understanding for process
  - Less flexible (*impacts research with it*)

# Cost for the Forensic DNA Community to Switch from ABI 3100s to 3500s

## 1. Instrument up-front cost

- Within the U.S. funding requests will likely come from federal grants

## 2. New software purchase

- Will likely be requested from federal grant funds (NIJ)
- new .hid file format will not work on current software (GMIDv3.2)
- 3500 will not create .fsa files with 36cm arrays (HID applications)

## 3. Validation time & expense

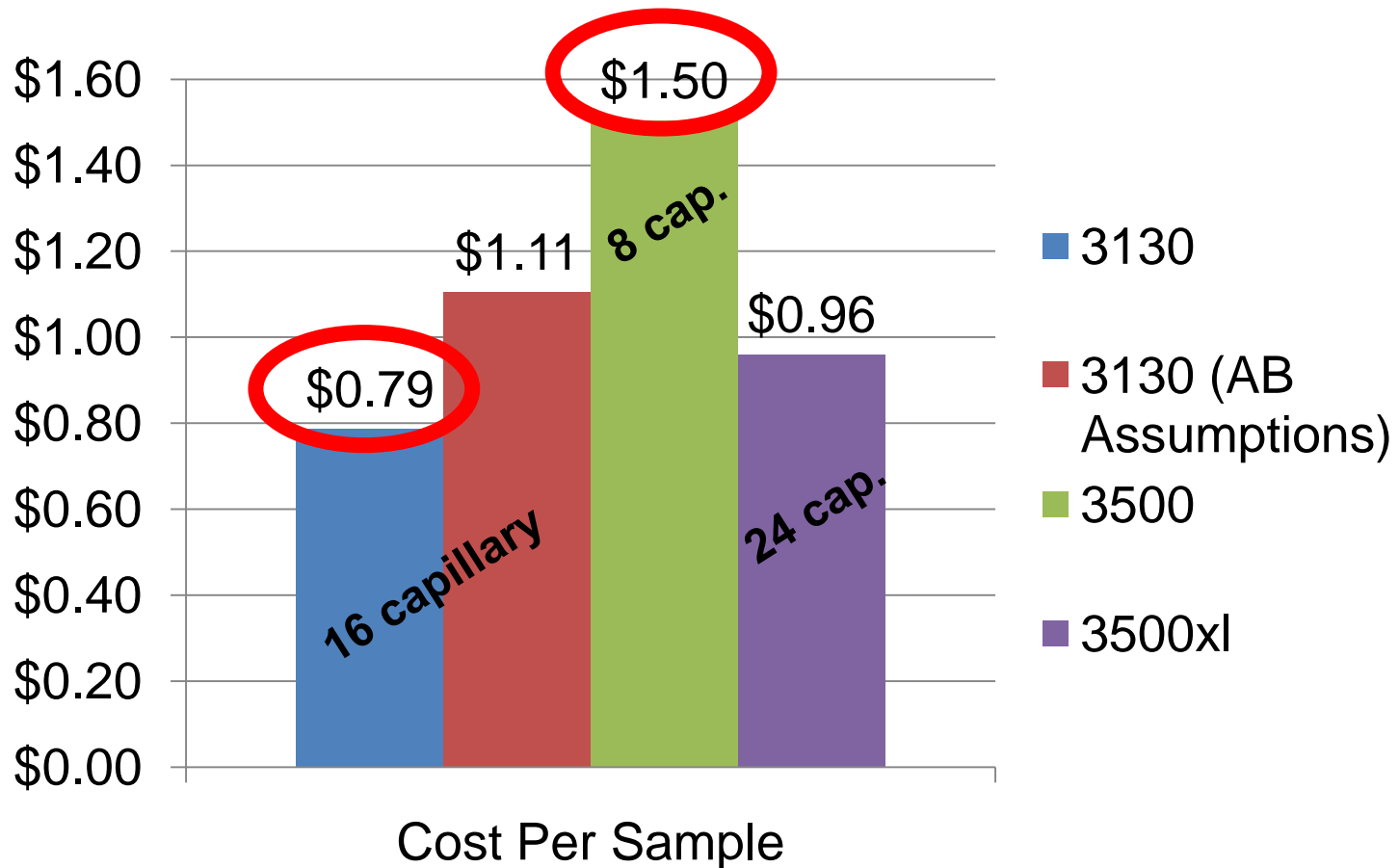
- Relative fluorescent scales are completely different...

## 4. Operational cost

- ABI claims that the running costs are equivalent to 3130s...

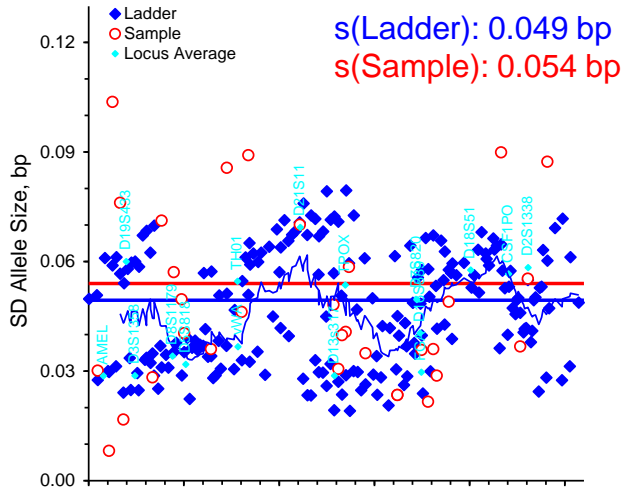
# NIST Calculated Cost per Sample for ABI 3130xl vs. 3500 and 3500xl Reagents

Running two plates per day (10 plates per week)

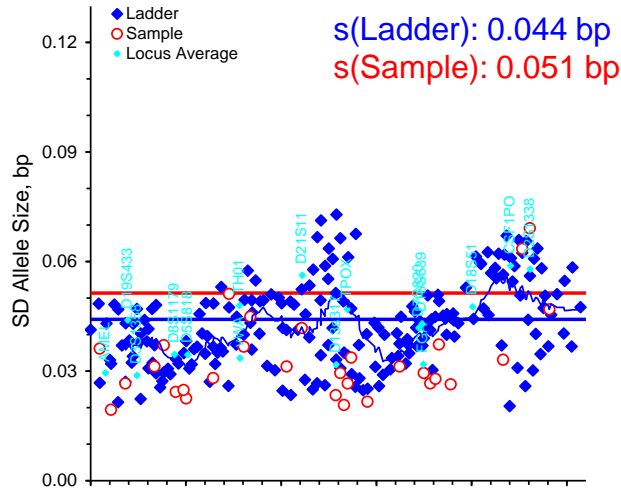


# Precision Studies at NIST

## ABI 3130xl (Identifiler)

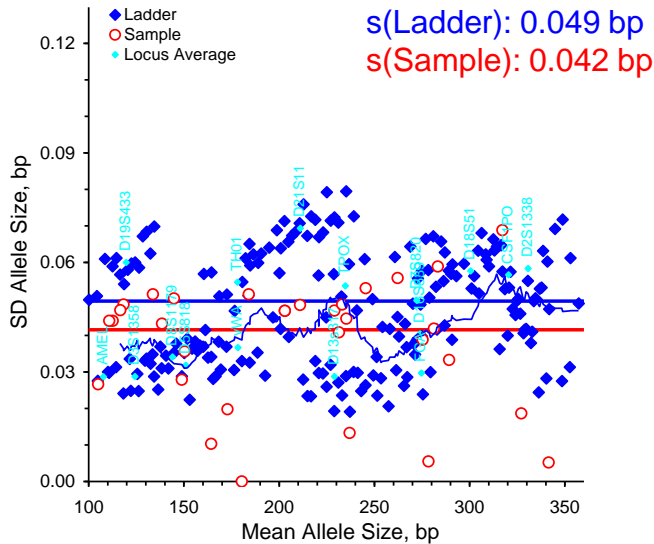


## ABI 3130xl (Identifiler Plus)

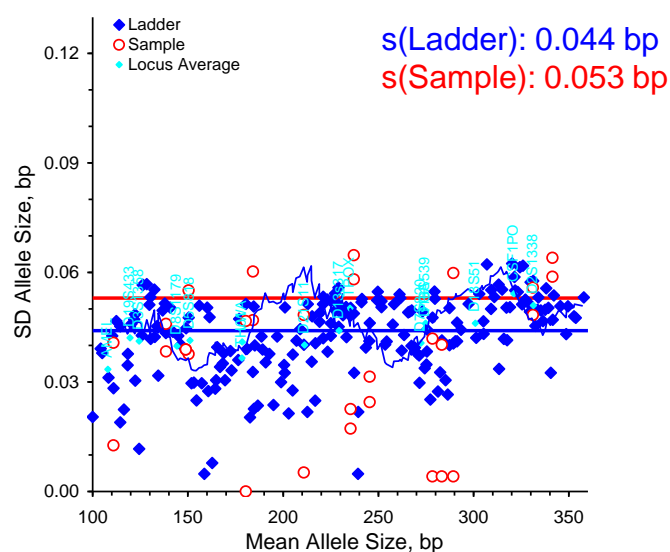


No significant difference between the 3130xl and 3500 for precision

## ABI 3500 (Identifiler)



## ABI 3500 (Identifiler Plus)



n=16 ladders  
n=6 samples

# Open Letter to Applied Biosystems on Concerns with ABI 3500

- **3/14/11 - emailed ~900 forensic DNA scientists** (SWGDM, forens-dna, ENFSI, EDNAP) inviting them to sign onto a letter that will be sent to Applied Biosystems expressing concern with ABI 3500
- **Very positive response with 101 who agreed to sign the letter**
- Letter was sent March 31 to the president of ABI and scientists involved with the ABI 3500
- **Community will be notified of ABI's response**

# Concerns Expressed in Open Letter



- RFID tags
- New .hid file structure requires new software
- Short shelf life of reagents – would like to see data for expiration times

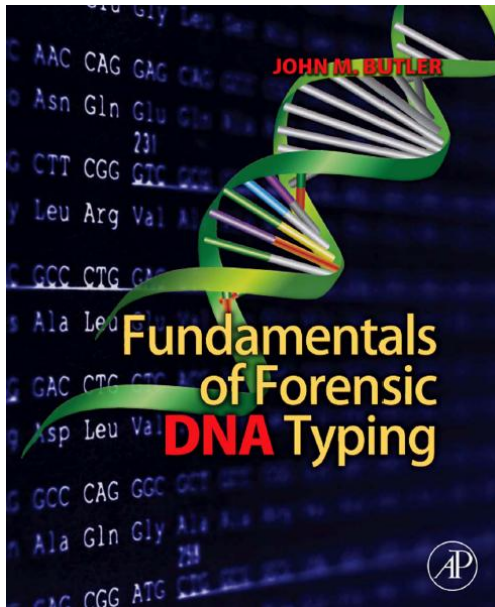
**Hopefully a change will result...**

**A desire for greater communication with the community – the 3500 FAQ sheet is a good start but does not directly address all of the concerns raised**



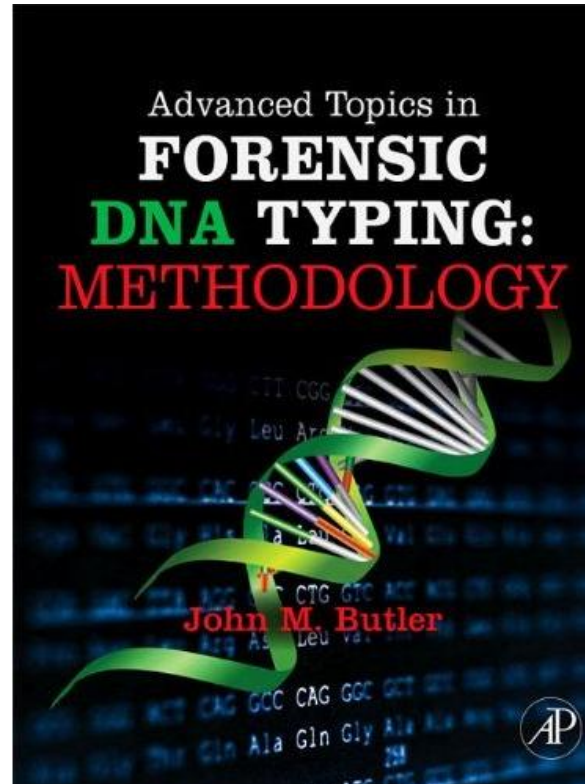
# Forensic DNA Typing Textbook

## 3<sup>rd</sup> Edition is Three Volumes



**Sept 2009**

~500 pages



**Sept 2011**

~700 pages

Currently being written

Advanced Topics in  
Forensic  
DNA Typing:  
**INTERPRETATION**

**Fall 2012**

~400 pages

# New STRBase Sections

## Forensic STR Information

- [STRs101: Brief Introduction to STRs](#)
- [Core Loci: FBI CODIS Core STR Loci and European Core Loci](#)
- [STR Fact Sheets \(observed alleles and PCR product sizes\)](#)
- [Multiplex STR kits](#)
- [Sequence Information \(annotated\)](#)
- [Variant Allele Reports](#) ◆
- [Tri-Allelic Patterns](#) ◆
- [Mutation Rates for Common Loci](#)
- [Published PCR primers](#)
- [Y-chromosome STRs](#) ◆
- [Low-template DNA Information](#) **Updated**
- [Mixture Interpretation](#) **NEW**
- [Kinship Analysis](#) **NEW**
- [miniSTRs \(short amplicons\)](#) ◆
- [Null Alleles](#) - discordance observed between STR kits ◆
- [STR Reference List](#) - now 3400 references ◆

# Upcoming Workshops



- ISFG (August 30, 2011)
  - **CE Fundamentals and Troubleshooting**



- Int. Symp. Human Ident. (October 3, 2011)
  - **Mixture Interpretation**



- Int. Symp. Human Ident. (October 6, 2011)
  - **Troubleshooting Laboratory Systems**

# Thank you for your attention

**Acknowledgments:** Applied Biosystems, Promega, and Qiagen for STR kits used in concordance studies

## Contact Information

**John Butler**

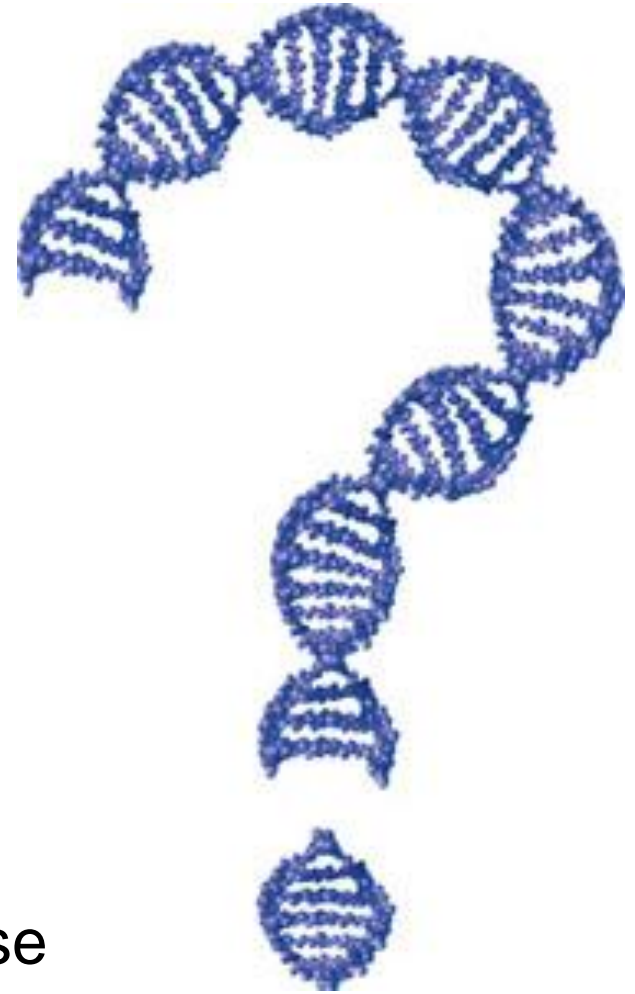
NIST Fellow

Group Leader of Applied Genetics

[john.butler@nist.gov](mailto:john.butler@nist.gov)

301-975-4049

<http://www.cstl.nist.gov/biotech/strbase>



**Our team publications and presentations are available at:**  
<http://www.cstl.nist.gov/biotech/strbase/NISTpub.htm>