

EDNAP and 32<sup>th</sup> ENFSI DNA WG Meeting  
April 13-15, 2010 – The Hague, The Netherlands

# NIST Update

**John M. Butler**  
and the NIST Human Identity Project Teams  
National Institute of Standards and Technology  
Gaithersburg, Maryland USA

## APPLIED GENETICS Group

Major Programs Currently Underway

- **Forensic DNA**
  - New loci and assays (26plex)
  - **STR kit testing**
  - Ancestry SNP assays
  - **Low-template DNA studies**
  - Mixture interpretation
  - STR nomenclature
  - **Variant allele cataloging** and sequencing
  - Expert systems review
  - Training workshops to forensic DNA laboratories
  - Validation information and **software tools**
  - **Textbook** – 3<sup>rd</sup> ed. (2 vol.)
- **Clinical Genetics**
  - CMV SRM
  - Huntington's SRM
- **Ag Biotech**
  - "universal" GMO detection/quantitation (35S promoter)
- **DNA Biometrics**
  - **Rapid PCR** methods
  - Efforts to standardize testing of future portable DNA systems
  - Kinship analysis
- **Cell Line Authentication**

### Topics to Address

- NIST Team Members and Projects
- SWGDAM STR Interpretation Guidelines
- Concordance studies with ESX/ESI and NGM kits
- Potential linkage with D12S391 and vWA
- Cell line authentication using STR markers
- Low template DNA work
- Rapid PCR & DNA biometrics work
- *Fundamentals* book published & *Advanced Topics* book underway

### NIST Human Identity Project Teams within the Applied Genetics Group

*Forensic DNA Team*

*DNA Biometrics Team*

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

*Data Analysis Support*

**In March 2010, Mike Coble returned to NIST after 4 years at AFDIL**

**New Staff and Projects**  
Erica Butts – DNA extraction  
Kristen Lewis - kinship analysis

Amy Decker left for AFDIL in Nov 2009

## SWGDAM STR Interpretation Guidelines Completed & Released

SWGDAM APPROVED 1/14/10

**SWGDAM Interpretation Guidelines for Autosomal STR Typing by Forensic DNA Testing Laboratories**

*Scientific Working Group on DNA Analysis Methods (SWGDAM)*

The Scientific Working Group on DNA Analysis Methods, better known by its acronym of SWGDAM, is a group of approximately 50 scientists representing federal, state, and local forensic DNA laboratories in the United States and Canada. During meetings, which are held twice a year, subcommittees discuss topics of interest to the forensic DNA community and often develop documents to provide direction and guidance for the community. A mixture interpretation subcommittee was formed in January 2007 and worked for several years to provide a guidance document on autosomal short tandem repeat (STR). This document was presented to the full SWGDAM group and received approval in January 2010.

[SWGDAM Interpretation Guidelines for Autosomal STR Typing by DNA Testing Forensic Laboratories \(pdf, 9mb\)](#)  
[CODIS \(Brochure, 3mb\) | Print version \(pdf\)](#)  
[DNA Fingerprint Act of 2005, Enforcement Policy](#)

[http://www.fbi.gov/hq/lab/html/codis\\_swgdam.pdf](http://www.fbi.gov/hq/lab/html/codis_swgdam.pdf)

### All Statistical Approaches Are Considered

**Table 1 – Suitable Statistical Analyses for DNA Typing Results**  
The statistical methods listed in the table cannot be combined into one calculation. For example, combining RMP at one locus with a CPI calculation at a second locus is not appropriate. However, an RMP may be calculated for the major component of a mixture and a CPE/CPI for the entire mixture (as referred to in section 4.6.2).

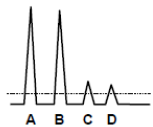
Category of DNA Typing Result	RMP	CPE/CPI	LR (1)
Single Source	✓		✓
Single Major Contributor to a Mixture	✓		✓
Multiple Major Contributors to a Mixture	✓ (2)	✓ (2)	✓
Single Minor Contributor to a Mixture	✓	✓ (3)	✓
Multiple Minor Contributors to a Mixture	✓ (2)	✓ (3)	✓
Indistinguishable Mixture	✓ (1)	✓	✓

(1) Restricted or unrestricted  
(2) Restricted  
(3) All potential alleles identified during interpretation are included in the statistical calculation

[http://www.fbi.gov/hq/lab/html/codis\\_swgdam.pdf](http://www.fbi.gov/hq/lab/html/codis_swgdam.pdf)

## Restricted vs Unrestricted

Are relative peak heights considered?



### Unrestricted

All combinations of alleles are deemed possible (relative peak height differences are not utilized)

AB + AC + AD + BC + BD + CD

### Restricted

Based on relative peak heights, alleles are paired only where specific combinations of alleles are deemed possible

AB + AC + AD + BC + BD + CD

Figure 1. Illustration of "restricted" versus "unrestricted" approaches based on relative peak heights (using an assumption of two donors with all peaks above the stochastic threshold).

[http://www.fbi.gov/hq/lab/html/codis\\_swgdam.pdf](http://www.fbi.gov/hq/lab/html/codis_swgdam.pdf)

## Evaluation of New European STR Loci

- U.S. population data collected using multiple kits
  - Examined **U.S. population data from 1443 individuals** (Caucasian, African American, Hispanic, Asian)
    - PowerPlex ESX 17 & ESI 17 Systems (Promega)**
    - AmpFISTR NGM Kit (Applied Biosystems)**
- Linkage analysis of vWA and D12S391
  - Located 6.3 Mb apart on chromosome 12
  - With unrelated individuals, no significant linkage in agreement with Phillips, C., et al. (2010)
  - With related individuals, linkage observed**
  - Recommending use of diplotypes with relatives – see Lewis, K.E., et al. (submitted)

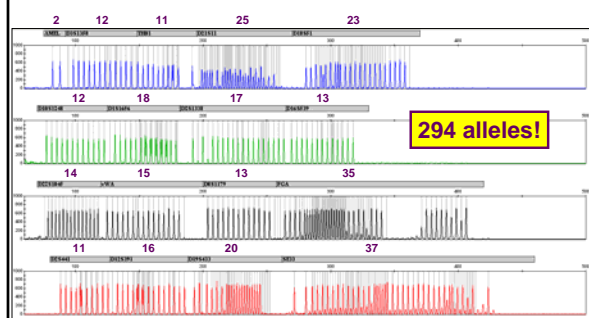
## STR Loci Present in Commercial Kits

U.S.			Europe			
PP16	Identifiler	MiniFiler	ESX/ESI17	NGM	SEfiler	SGM Plus
TPOX	TPOX					
CSF1PO	CSF1PO	CSF1PO				
D5S818	D5S818					
D7S820	D7S820	D7S820				
D13S317	D13S317	D13S317				
FGA	FGA	FGA	FGA	FGA	FGA	FGA
vWA	vWA		vWA	vWA	vWA	vWA
D3S1358	D3S1358		D3S1358	D3S1358	D3S1358	D3S1358
D8S1179	D8S1179		D8S1179	D8S1179	D8S1179	D8S1179
D18S51	D18S51	D18S51	D18S51	D18S51	D18S51	D18S51
D21S11	D21S11	D21S11	D21S11	D21S11	D21S11	D21S11
TH01	TH01		TH01	TH01	TH01	TH01
D16S539	D16S539	D16S539	D16S539	D16S539	D16S539	D16S539
	D2S1338	D2S1338	D2S1338	D2S1338	D2S1338	D2S1338
	D19S433	D19S433	D19S433	D19S433	D19S433	D19S433
	D12S391	D12S391	D12S391	D12S391		
	D1S1656	D1S1656	D1S1656	D1S1656		
	D2S441	D2S441	D2S441	D2S441		
	D10S1248	D10S1248	D10S1248	D10S1248		
	D22S1045	D22S1045	D22S1045	D22S1045		
		SE33			SE33	
	Penta D					
	Penta E					

**U.S. is looking to expand the core loci (18-20 total) to provide more international overlap**

## New STR Kits: Analysis and Concordance

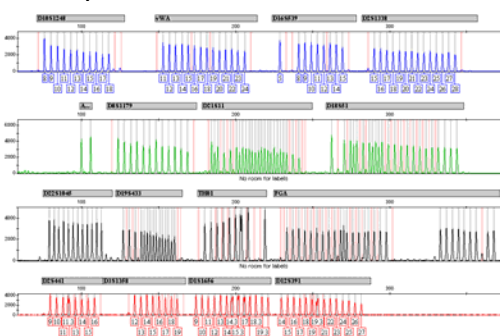
## PowerPlex® ESX 17 Allelic Ladders



Virtual bins added in for fairly commonly observed micro-variants (i.e. >4 mentions on STRBase)

ESX/ESI 17 kits provided by Promega Corporation

## AmpFISTR® NGM Allelic Ladders



NGM kits provided by Applied Biosystems

## 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 attempted = 1455**  
**1443 samples with complete profiles**

## FSI Genetics Forthcoming Article on PowerPlex ESX 17 and ESI 17 Systems

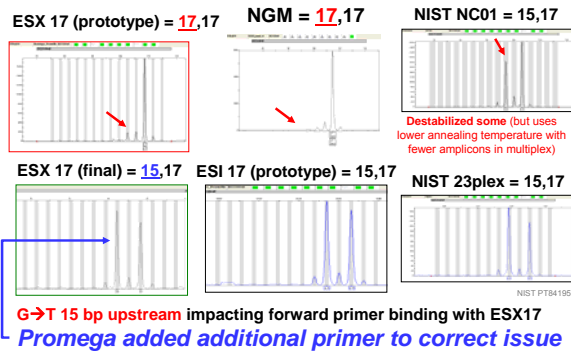


Concordance and population studies along with **stutter and peak height ratio analysis** for the PowerPlex<sup>®</sup> ESX 17 and ESI 17 Systems

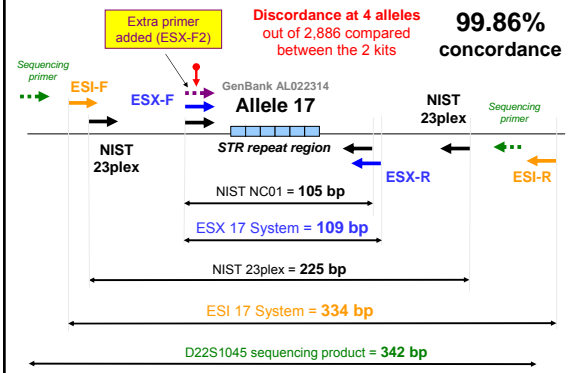
Carolyn R. Hill<sup>a,\*</sup>, David L. Dueser<sup>a</sup>, Margaret C. Kline<sup>a</sup>, Cynthia J. Sprecher<sup>b</sup>, Robert S. McLaren<sup>b</sup>, Dawn R. Rabbach<sup>b</sup>, Benjamin E. Krenke<sup>b</sup>, Martin G. Ensenberger<sup>b</sup>, Patricia M. Fulmer<sup>b</sup>, Douglas R. Storts<sup>b</sup>, John M. Butler<sup>a</sup>

<sup>a</sup>National Institute of Standards and Technology, Chemical Science and Technology Laboratory, Gaithersburg, MD 20899-6112, USA  
<sup>b</sup>Promega Corporation, Madison, WI 53711-5299, USA

## D22S1045 Discordance



## D22S1045 Relative PCR Primer Positions



## Profiles in DNA Article Published April 2010

Article Type: Feature

Volume 13 No. 1, April 2010

### Strategies for Concordance Testing

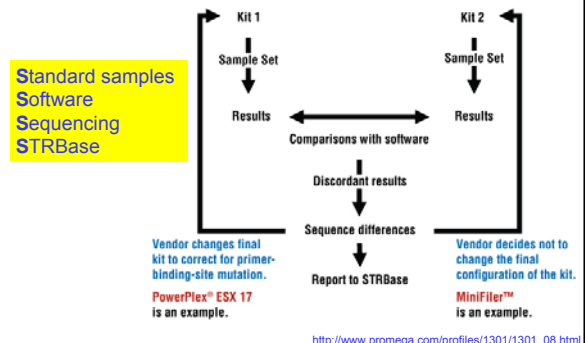
Carolyn R. Hill, Margaret C. Kline, David L. Dueser and John M. Butler  
 National Institute of Standards and Technology, Biochemical Science Division, Gaithersburg, Maryland, USA

Concordance evaluations are important to conduct to determine if there are any allelic dropout or "null alleles" present in a data set. These studies are performed because there are a variety of commercial short tandem repeat (STR) multiplex kits with different configurations of STR markers available to the forensic community. The placement of the markers can vary between kits because the primer sequences were designed to amplify different polymerase chain reaction (PCR) product sizes. When multiple primer sets are used, there is concern that allelic dropout may occur due to primer-binding-site mutations that affect one set of primers but not another.

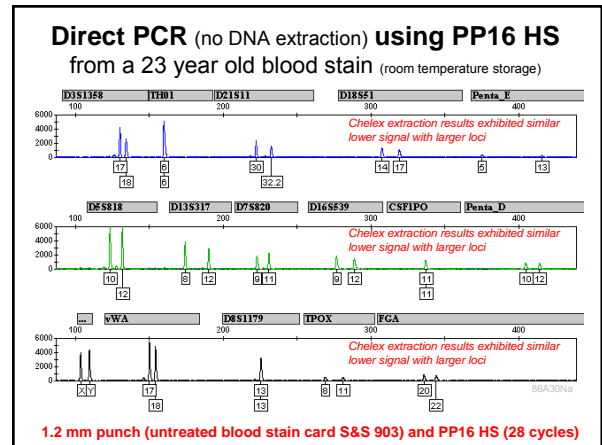
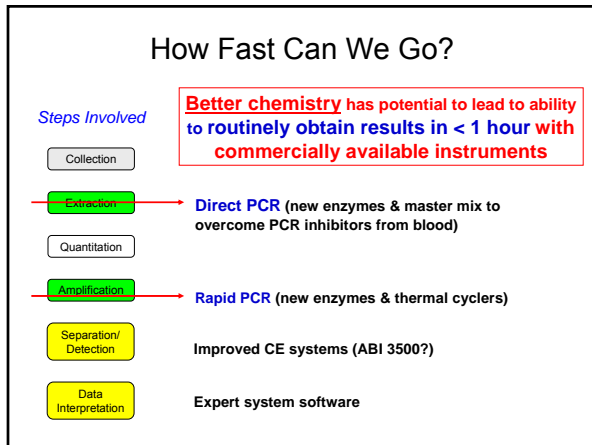
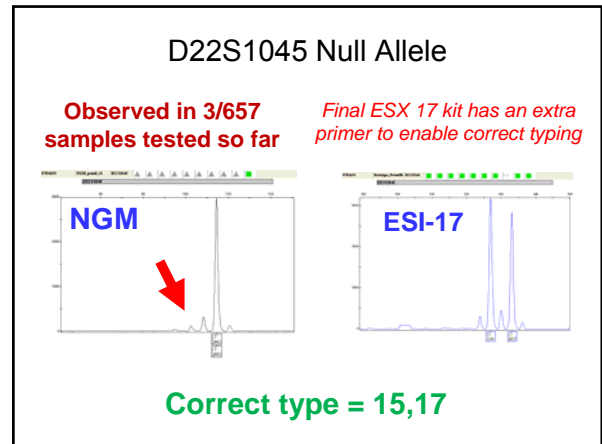
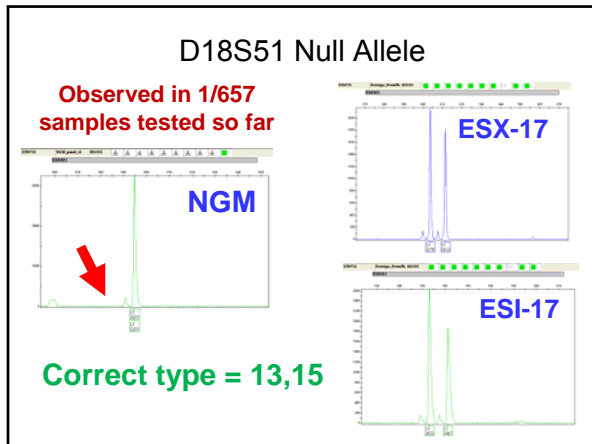
[http://www.promega.com/profiles/1301/1301\\_08.html](http://www.promega.com/profiles/1301/1301_08.html)

## Strategies for Concordance Testing

the four S's of concordance studies







### Rapid PCR work published in *FSI Genetics* (Dec 2008)

**Full STR profiles in 36 minutes (instead of 3 hour PCR)**

Forensic Science International: Genetics 3 (2008) 42-45

Contents lists available at ScienceDirect

Forensic Science International: Genetics

journal homepage: www.elsevier.com/locate/fsig

Short communication

Demonstration of rapid multiplex PCR amplification involving 16 genetic loci<sup>®</sup>

Peter M. Vallone<sup>\*</sup>, Carolyn R. Hill, John M. Butler

National Institute of Standards and Technology, Biochemical Science Division, 350 Bureau Drive, Mail Stop 8311, Gaithersburg, MD 20899-8311, United States

**Complete concordance of STR allele calls (for 60 samples) between the rapid and standard thermal cycling protocols** were observed although there was incomplete adenylation at several of the loci examined and some PCR artifacts were detected. Using less than **750 pg of template DNA and 28 cycles, STR peaks for all loci were above a 150 relative fluorescent unit (RFU) detection threshold** with fully adequate inter-locus balance and heterozygote peak height ratios of greater than 0.84.

### Further Rapid PCR Work

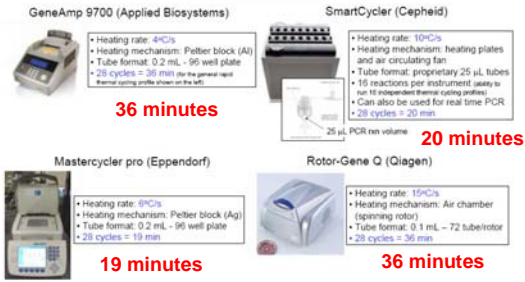
#### Rapid PCR Thermal Cycling Profile

Maximum heating rate of ~4°C/s on a GeneAmp 9700 (Applied Biosystems)

- **Much shorter hold times at each temperature**
- **Faster ramp rates between temperatures**
- **Examination of different enzyme mixes**
  - 0.5 x master mix PyroStart (Fermentas) (\$0.14/rxn)
  - 0.5 x master mix Premix Ex Taq (Takara) (\$0.22/rxn)
  - 0.25 μL = 1.25 units of SpeedStar (Takara) (\$1.09/rxn)
- **Evaluation of additional kits**
  - Identifiler, PP16, Yfiler, MiniFiler and Promega S5
- **Testing thermal cyclers with faster ramp rates**

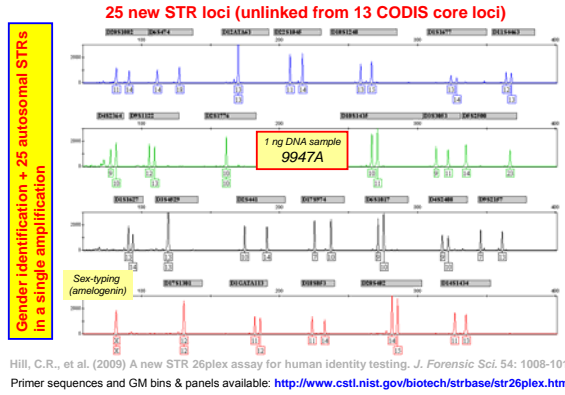
## Four Thermal Cyclers Being Evaluated

How fast can we run 28 cycles?



[http://www.cstl.nist.gov/biotech/strbase/pub\\_pres/VallonePromega2009poster.pdf](http://www.cstl.nist.gov/biotech/strbase/pub_pres/VallonePromega2009poster.pdf)

## NIST 26plex published in *J. Forensic Sci.* (Sept 2009)



## Authentication of Cell Lines Using STRs

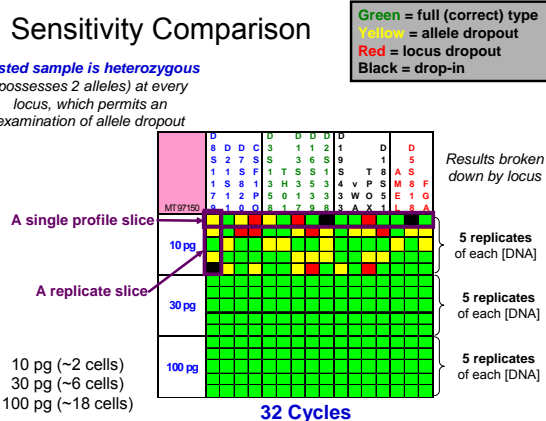
- Working with the ATCC (American Type Culture Collection) to develop standards for testing human cell lines using STR markers
- NCBI (National Center for Biotechnology Information) will soon have a STR database for authenticating human cell lines
- Examining the minimum number of STR markers to separate human cell lines from one another
- Have observed DNA quantitation issues with cell lines particularly Quantifier (hTERT target)

## Experimental Design to Study LT-DNA Issues

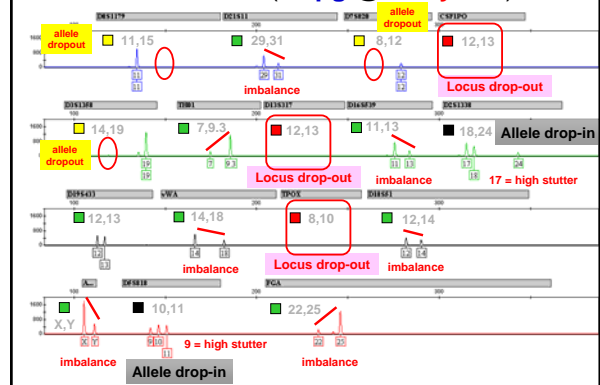
- Pristine DNA Samples
  - 2 single-source samples
  - heterozygous for all loci tested (permits peak height ratio studies)
- Low DNA Template Amounts
  - Dilutions made after DNA quantitation against NIST SRM 2372
  - 100 pg, 30 pg, and 10 pg (1 ng tested for comparison purposes)
- Replicates
  - 5 separate PCR reactions for each sample
- STR Multiplex Kits
  - Identifiler Plus and PowerPlex 16 HS (half-reactions)
- Increased Cycle Number
  - Identifiler Plus (29 cycles and 32 cycles; 28 for 1 ng)
  - PowerPlex 16 HS (31 cycles and 34 cycles; 30 for 1 ng)

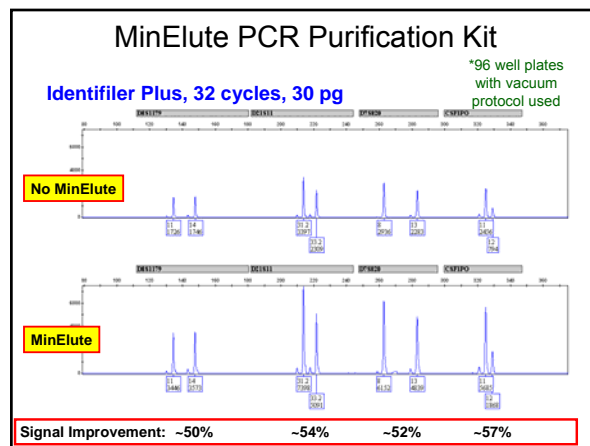
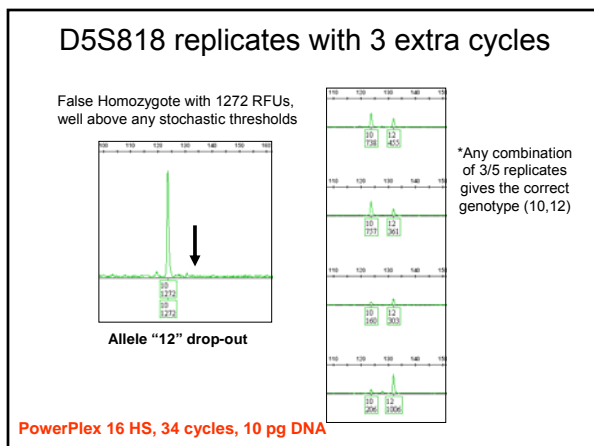
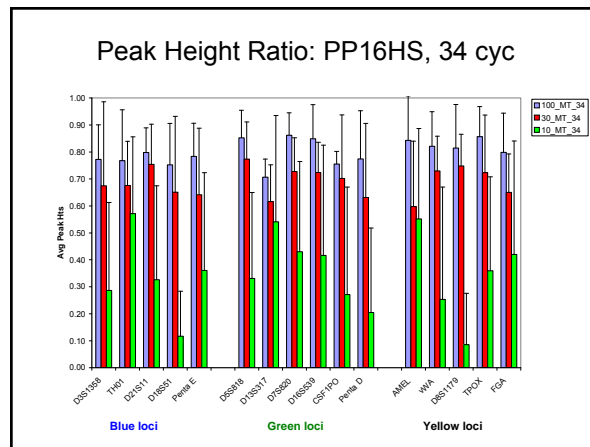
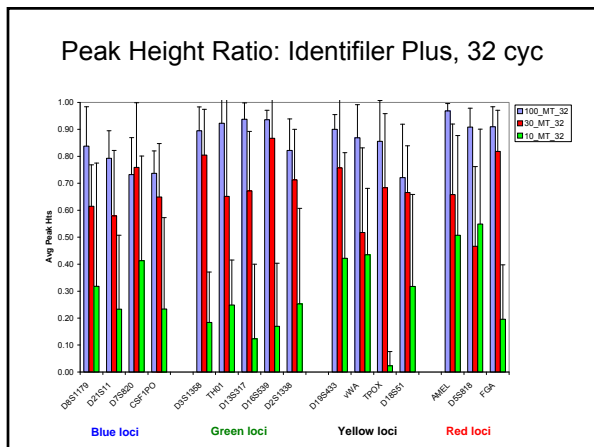
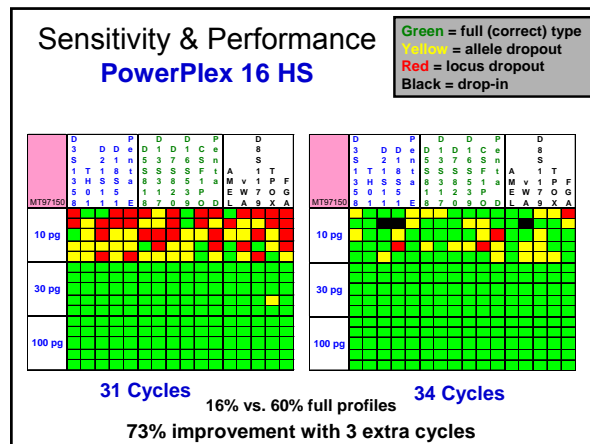
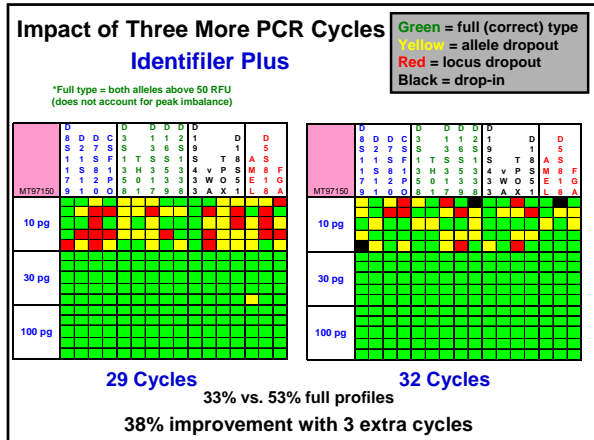
## Sensitivity Comparison

Tested sample is heterozygous (possesses 2 alleles) at every locus, which permits an examination of allele dropout.



## Identifiler Plus (10 pg @ 32 cycles)





## New STRBase Website on LT-DNA (LCN)

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

**Information on Low Template / Low Copy Number DNA Testing**

**General Information**

- o Purpose of STRBase
- o Publications and Presentations
- o NIF-Funded Projects
- o Training Materials
- o Links to other web sites
- o Glossary of common terms

**Forensic STR Information**

- o STRs101: Brief Intro
- o Core Loci: FBI CODIS
- o STR Fact Sheets (jobs)
- o Multiple STR kits
- o Sequence Information
- o Variant Allele Reports
- o Tri-Allelic Patterns
- o Mutation Rates for CODIS
- o Published PCR primers
- o Y-chromosome STRs
- o Low-template DNA Information
- o miniSTRs (short amplicons)
- o Null Alleles - discordance observed between STR kits
- o STR Reference List - now 2302 references

**Low Copy Number (LCN) DNA Panel Discussion**

### Scientific Issues with Analysis of Low Amounts of DNA

Presentation Prepared for the LT-DNA Panel

Theresa Caragine Ph.D.  
Deputy Director  
October 16, 2009

**Presentations on LTDNA**

- John Butler - NIST (Presentor)
- Berke Jibr - NIST (Presentor)
- Thomas Caragine - NIST (Ph.D.)

**LTDNA Validation Data**

NIST Sensitivity Data with low level DNA

10 replicate amplifications for each condition

## Complete Set of NIST Sensitivity Data Available on New LT-DNA Website

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

**NIST Sensitivity Data with low level DNA templates**  
10 replicate amplifications for each condition with two fully heterozygous, single-source samples

*Click on links to see summaries and DNA profiles observed*

STR kit - PCR conditions	Sample 1	Sample 2
Identifier - 28 cycles	100 pg 30 pg 10 pg	100 pg 30 pg 10 pg
Identifier - 31 cycles	100 pg 30 pg 10 pg	100 pg 30 pg 10 pg
PowerPlex 16 HS - 31 cycles	100 pg 30 pg 10 pg	100 pg 30 pg 10 pg
PowerPlex 16 HS - 34 cycles	100 pg 30 pg 10 pg	100 pg 30 pg 10 pg

**PowerPlex 16 HS - 34 cycles**

Sample #1 (MT97150)

Sample #2 (PT04411)

**MT97150 - 10 pg, amp #1**

## LT-DNA Conclusions

- The results with pristine full heterozygous samples demonstrate that replicate testing can produce reliable information with single source samples at low levels of DNA when consensus profiles are created.
- Identifier Plus with 32 cycles and PowerPlex 16 HS with 34 cycles were comparable in performance with low-level DNA analysis.
- With 3 extra cycles, there was better recovery at 10 pg of DNA using both kits including less allelic and full locus drop-out. However, there is a greater potential for allele drop-in or high stutter.
- MinElute PCR Purification Kits were successful in significantly increasing the signal for LT-DNA PCR products and resulted in extra peaks being called at 10 pg DNA samples.

## Profiles in DNA (April 2010)

<http://www.promega.com/profiles/>

**Profiles in DNA**

Each issue provides news and information for researchers and analysts working in the field of genetic identity testing. Topics include forensic genetics, database samples, scientific analysis, legal issues, technical tips, Promega genetic identity product updates, interesting cases and more.

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- Case Reports
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- Legal Focus
- Product News
- Technical Tips

All Products

All Technical Resources

**WELCOME TO THE 20th ANNIVERSARY OF THE LOW COPY NUMBER SESSION AT THE 20TH INTERNATIONAL SYMPOSIUM ON HUMAN IDENTIFICATION**

At the 20th International Symposium on Human Identification, prominent figures in the DNA-typing field shared their view of low copy number (LCN) analysis by responding to a subset of LCN-related questions. Charlotte Ward and John Butler provided introductions to the topic. Links to the introductions, material and individual responses are provided below. Note: All of the responses are not yet available. The other LCN responses will be published in Profiles in DNA as they become available.

A lot of questions can be found here.

**MEETINGS**

- What is LCN?—Definitions and Challenges**  
Charlotte Ward presents a summary of her presentation during the LCN session. She helps us sort through some of the confusion about what constitutes LCN analysis.
- Scientific Issues with Analysis of Low Amounts of DNA**  
John Butler and Carolyn Hill discuss technical issues and challenges that can arise in low template DNA analysis.
- Low Copy Number Analysis From a Legal Perspective**  
Brad Leachman from the Queens County District Attorney's Office shares his view on the biggest challenges with LCN analysis and his advice for forensic scientists working with attorneys on cases that could be considered LCN.

## Our LT-DNA Article in Profiles in DNA

[http://www.promega.com/profiles/03/03/01\\_02.html](http://www.promega.com/profiles/03/03/01_02.html)

Published online April 5, 2010

Article Type: Meetings

### Scientific Issues with Analysis of Low Amounts of DNA

**John M. Butler\* and Carolyn R. Hill**  
National Institute of Standards and Technology, Biomedical Science Division, Gaithersburg, Maryland, USA  
\*Corresponding author: 301-975-6049; john.butler@nist.gov

Faced with limited evidence that yield low amounts of DNA, forensic analysts will continually have to confront the question of how far to push DNA-testing techniques. Low copy number (LCN) analysis, also known as low template DNA (LT-DNA) testing, involves enhancing detection sensitivity usually through increasing the number of PCR cycles. Stochastic effects inherent with analysis of low amounts of DNA yield allele or locus drop-out. Additionally, increasing detection sensitivity can result in a greater potential for contamination or allele drop-in. Validation studies with replicate testing of low amounts of DNA were performed to assess the level of allele and locus drop-out and allele drop-in using 10, 30 and 100 picograms with several commercially available STR-typing kits under both standard and increased number of PCR cycles. The results with pristine, fully heterozygous samples demonstrate that a replicate testing approach can produce reliable information with single-source samples when consensus profiles are created.

\*Based on LT-DNA studies performed in Fall 2009

## The Expansion of Forensic DNA Typing

**1st Edition**

Jan 2001  
335 pp.  
17 chapters

**2nd Edition**

Feb 2005  
688 pp.  
24 chapters

Chinese Translation (2007) Y. Hou, translator

Japanese Translation (2009) Y. Fukuma, translator

**3rd Edition**

Sept 2009

**Fundamentals**  
18 chapters (504 pp.)

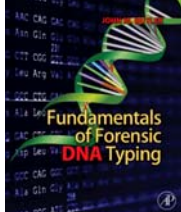
**Advanced Topics**  
25 chapters (~600 pp.)

Planned for Spring 2011



## The New Book...

**3<sup>rd</sup> Edition, Volume 1**



**Sept 2009**  
504 pp.

**Volume 2, Advanced Topics of Forensic DNA Typing, is expected in Spring 2011**

- **Selection of basic information from previous editions** to aid students, lawyers, and scientists understand fundamentals of forensic DNA
- **Updated information** (cites >600 new references since 2<sup>nd</sup> edition was written in 2004)
- **Improved reference format**
  - by topic with title included
- **43 new figures, 17 new tables, 26 new D.N.A. boxes, and numerous new website links**
- **New chapters** on historical methods (Ch. 3) and future trends (Ch. 18)
- **New information** on DNA databases (Ch. 12), quality assurance (Ch. 13), & lineage markers (Ch. 16)
- **New order of chapters** to reflect process of DNA typing (Ch. 4-11)
- **Glossary with >400 key words**

## Improved Reference Format

**Forensic DNA Typing  
(2<sup>nd</sup> Edition)**

*Full list of authors but no article title*

**Fundamentals  
(3<sup>rd</sup> Edition)**

**Subdivided by subject  
with article title provided**

**>1500 references total  
(>600 new since 2<sup>nd</sup> edition)**

### Chapters Re-ordered to Reflect DNA Testing Process

**Steps Involved**

- Forensic Science
  - Collection
  - Sample Storage
  - Characterization
- Biology
  - Extraction
  - Quantitation
  - Amplification
  - STR Markers
- Technology
  - Separation/Detection
  - Data Interpretation
- Genetics
  - Statistical Interpretation

**Fundamentals of Forensic DNA Typing (2009)**

CHAPTER 1	Overview and History of DNA Typing	1
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CHAPTER 8	Short Tandem Repeat Markers	147
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CHAPTER 10	STR Genotyping and Data Interpretation	205
CHAPTER 11	Statistical Interpretation: Evaluating the Strength of Forensic DNA Evidence	229
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*References are provided at the end of each chapter by subject (but without direct citation within the text).*

## Forensic Science Review Article

June 15, 2009 issue of *Analytical Chemistry*

Anal. Chem. 2009, 81, 4695-4711

**Review Contents**

- Forensic DNA Analysis: Collection, Characterization, Preservation, Extraction, and Quantitation of Biological Material
- Short Tandem Repeats
- Single Nucleotide Polymorphisms
- Y-Chromosome and Y-Chromosome Analysis
- Mitochondrial DNA Typing
- Nonhuman DNA Typing Systems
- DNA Databases, Missing Persons, and Disaster Victim Identification
- Interpretation and Statistical Weight of DNA Typing Results
- General Reviews

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**2009 review article covers 160 DNA articles published in 2007-2008**

## Training Workshops Conducted in 2009

**Individual Forensic DNA Laboratories**



**Scientific Conferences**



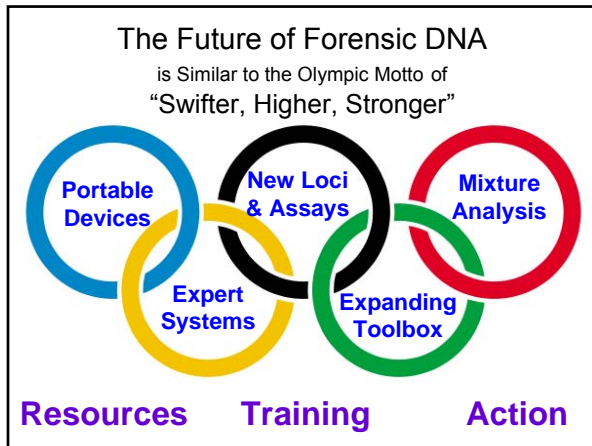
**Universities**



## Overview of NIST Efforts

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- Evaluation of new European loci
- DNA Biometrics (rapid PCR)
- New STR Kits: Analysis and Concordance
- Authentication of Cell Lines
- Projects with Low Template DNA



NIST Publications in 2009 Demonstrating  
"Swifter, Higher, Stronger" DNA Analysis

**Swifter PCR Amplification**

Research article  
Rapid amplification of commercial STR typing kits  
Peter M. Vallone<sup>1\*</sup>, Carolyn R. Hill<sup>1</sup>, Danielle Pablos<sup>1</sup>, John M. Butler<sup>1</sup>

**Higher Levels of Multiplexing**

*J Forensic Sci*, September 2008, Vol. 54, No. 5  
doi: 10.1111/j.1744-4752.2008.01111.x  
Available online at: www.blackwell-synergy.com

Carolyn R. Hill,<sup>1</sup> M.S.; John M. Butler,<sup>1</sup> Ph.D.; and Peter M. Vallone,<sup>1</sup> Ph.D.

**A 26plex Autosomal STR Assay to Aid Human Identity Testing<sup>1†</sup>**

**Stronger Powers of Discrimination**

Research article  
The single most polymorphic STR Locus: SE33 performance in U.S. populations  
John M. Butler<sup>1\*</sup>, Carolyn R. Hill<sup>1</sup>, Margaret C. Kline<sup>1</sup>, David L. Dunson<sup>1</sup>, Cynthia J. Sprecher<sup>1</sup>, Robert S. McLaren<sup>1</sup>, Dawn R. Rabbach<sup>1</sup>, Benjamin E. Kessler<sup>1</sup>, Douglas R. Starks<sup>1</sup>

**The NIST Human Identity Project Team**  
(Forensic DNA & DNA Biometrics)

Funding from the **National Institute of Justice (NIJ)** through the NIST Office of Law Enforcement Standards and the **FBI S&T Branch** through the NIST Information Access Division  
...Bringing traceability and technology to the scales of justice...

John Butler Erica Butts Mike Coble Dave Duewer Becky Hill Margaret Kline Kristen Lewis Jan Redman Pete Vallone

Project Leader, Forensic DNA

Project Leader, DNA Biometrics

Workshops & Textbooks

Mixtures, mtDNA & Y

Concordance & LT-DNA

Kinship Analysis

Rapid PCR & Biometrics

DNA Extraction Efficiency

Software Tools & Data Analysis

Variant alleles & Cell Line ID

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