EDNAP and 32th 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

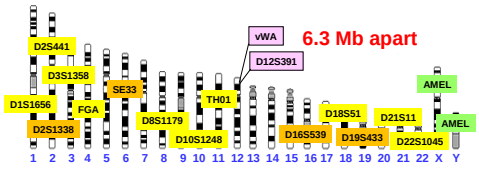
Topics to Address

- Potential linkage with D12S391 and vWA
- Concordance studies with ESX/ESI and NGM kits
- New LT-DNA section of STRBase website
- *Fundamentals* book published *& Advanced Topics* book underway

For additional information on our research activities, see our EDNAP presentation available on STRBase:
http://www.cstl.nist.gov/biotech/strbase/NISTpub.htm

Chromosomal Positions for the European Standard Set

and Other Common STR Markers Used



European Standard Set + D16S539, D2S1338, D19S433, SE33

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)**
  - **AmpFlSTR 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)

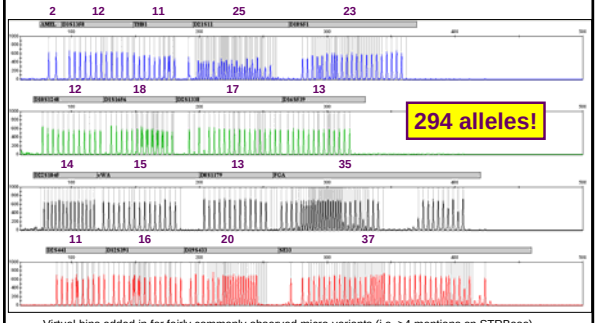
STR Loci Present in Commercial Kits

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
			D12S391	D12S391		
			D1S1656	D1S1656		
			D2S441	D2S441		
			D10S1248	D10S1248		
			D22S1045	D22S1045		
			SE33		SE33	

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

Penta D
Penta E

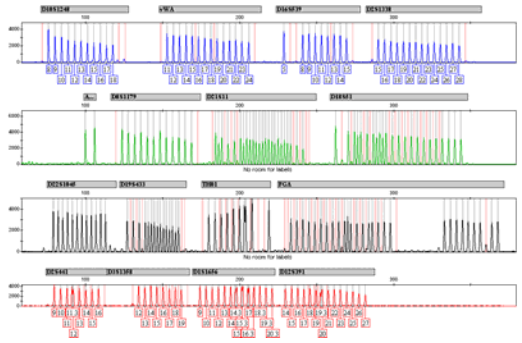
PowerPlex® ESX 17 Allelic Ladders



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

ESX/ESI 17 kits provided by Promega Corporation

AmpFlSTR® NGM Allelic Ladders



NGM kits provided by Applied Biosystems

Summary of NIST Samples Evaluated

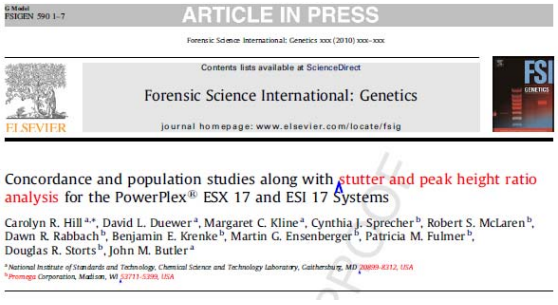
- **U.S. Population Samples** (**657 samples**)
  - Previously studied with Identifiler, MiniFiler, Yfiler, PP16, miniSTRs, and many additional assays *(>200,000 allele calls)*
  - 260 African Americans, 260 Caucasians, 140 Hispanics, and 3 Asians

    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



G Model
FSIGEN 590 1–7

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

ELSEVIER

Concordance and population studies along with stutter and peak height ratio analysis for the PowerPlex® ESX 17 and ESI 17 Systems

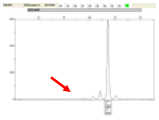
Carolyn R. Hill [a,*], David L. Duewer [a], Margaret C. Kline [a], Cynthia J. Sprecher [b], Robert S. McLaren [b], Dawn R. Rabbach [b], Benjamin E. Krenke [b], Martin G. Ensenberger [b], Patricia M. Fulmer [b], Douglas R. Storts [b], John M. Butler [a]

[a] National Institute of Standards and Technology, Chemical Science and Technology Laboratory, Gaithersburg, MD 20899-8312, USA
[b] Promega Corporation, Madison, WI 53711-5399, USA

D22S1045 Discordance

ESX 17 (prototype) = 17,17

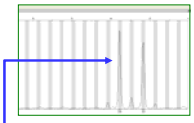
NGM = 17,17



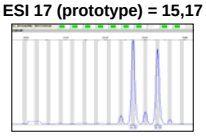
NIST NC01 = 15,17

Destabilized some (but uses lower annealing temperature with fewer amplicons in multiplex)

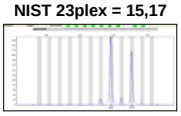
ESX 17 (final) = 15,17



ESI 17 (prototype) = 15,17



NIST 23plex = 15,17



NIST PT84195

G→T 15 bp upstream impacting forward primer binding with ESX17

Promega added additional primer to correct issue

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

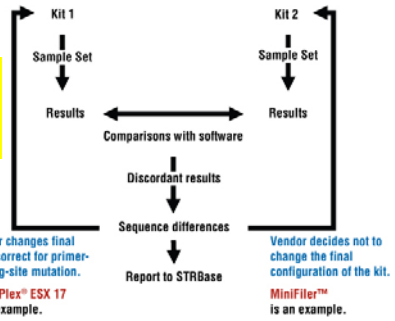
Concordance evaluations are important to conduct to determine if there are any allelic dropout or "null alleles" present in a data set. These studies are performed because there are a variety of commercial short tandem repeat (STR) multiplex kits with different configurations of STR markers available 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 allele 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

Strategies for Concordance Testing

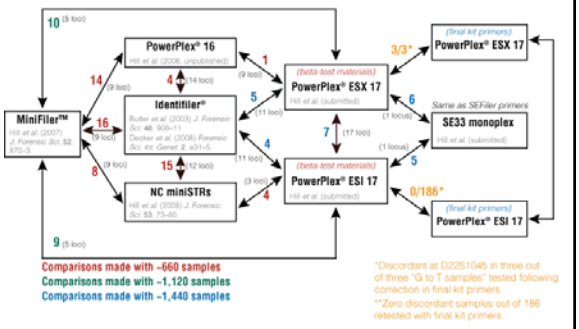
the four S's of concordance studies

Standard samples
Software
Sequencing
STRBase



http://www.promega.com/profiles/1301/1301_08.html

Concordance Comparisons



http://www.promega.com/profiles/1301/1301_08.html

New NIST Software Tools

Developed by Dave Duewer (NIST)

http://www.cstl.nist.gov/biotech/strbase/tools/STR_MatchSamples.xls

STR_MatchSamples

- An Excel-based tool developed to aid comparison of STR genotypes from two or more data sets.

Tools under development (to aid validation studies)

- Peak height ratio
- Inter-locus balance
- Stutter percentages
- Allele frequency

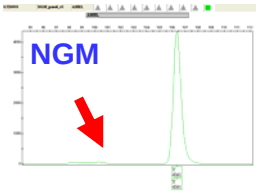
http://www.cstl.nist.gov/biotech/strbase/software.htm

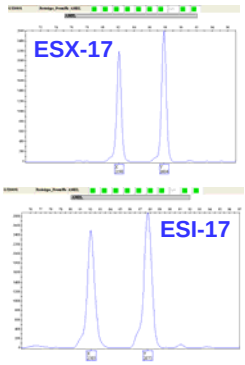
Concordance Testing with NGM Kit

- NGM results compared to ESX/ESI kits with **657 unrelated individuals** (NIST U.S. population set)
- NGM and ESX/ESI 17 are **fully concordant with NIST SRM 2391b** certified values (including D2S441, D10S1248, D22S1045, D1S1656, D12S391)
- **NGM null alleles**:
  - AMEL X (2/657)
  - D18S51 (1/657)
  - D22S1045 (3/657)

Amelogenin X Null Allele

Observed in 2/657 samples tested so far

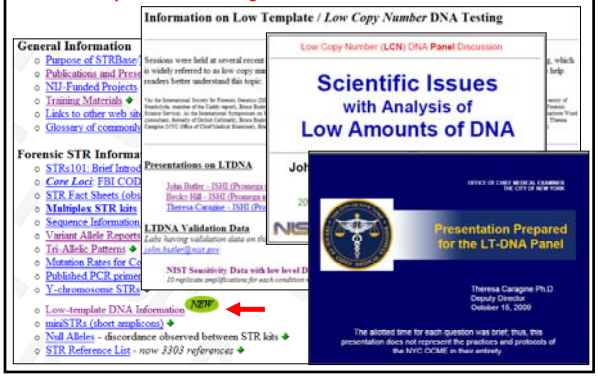




Correct type = X,Y

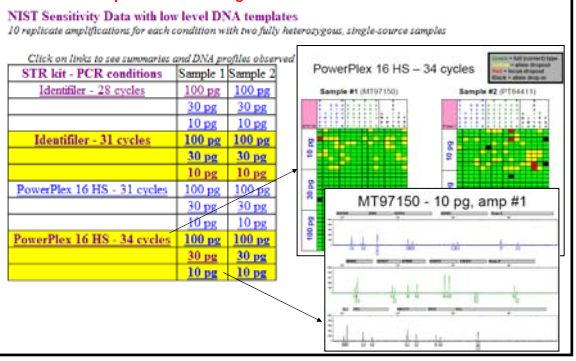
New STRBase Website on LT-DNA (LCN)

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



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

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

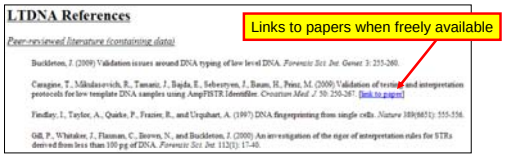


Literature Listing on LT-DNA (LCN)

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

Subdivided into categories

- Peer-reviewed literature *(containing data)*
- Reports *(evaluating the methodology)*
- Review articles *(commenting on other's data)*
- Non-peer reviewed literature *(representing the authors' opinions only)*

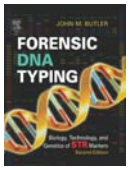


The Expansion of *Forensic DNA Typing*

1st Edition

Jan 2001
335 pp.
17 chapters

2nd Edition

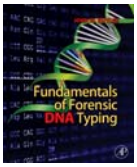


Feb 2005
688 pp.
24 chapters

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

Improved Reference Format

Forensic DNA Typing
(2nd Edition)

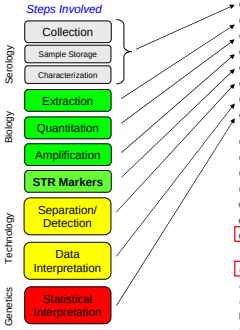
Full list of authors but no article title

Fundamentals
(3rd Edition)

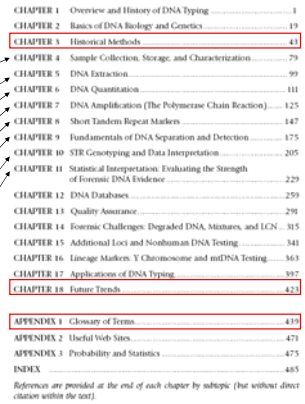
Subdivided by subject with article title provided

>1500 references total
(>600 new since 2nd edition)

Chapters Re-ordered to Reflect DNA Testing Process



Fundamentals of Forensic DNA Typing (2009)

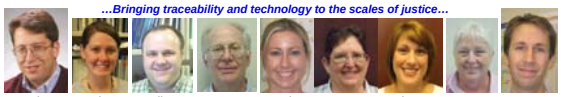


The NIST Human Identity Project Team

(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 — *Project Leader, Forensic DNA*
Erica Butts
Mike Coble
Dave Duewer
Becky Hill
Margaret Kline
Kristen Lewis
Jan Redman
Pete Vallone — *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
STRBase Support

http://www.cstl.nist.gov/biotech/strbase/NISTpub.htm
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001-301-975-4049

Questions?