



NIST Historical Perspective: Rapid DNA Identification

Erica Romsos

Rapid DNA Technology Forum

Alexandria, VA

August 16, 2017



Disclaimer

We will mention commercial STR kit and instrument names, but we are in no way attempting to endorse any specific products.

NIST Disclaimer: Certain commercial equipment, instruments and materials are identified in order to specify experimental procedures as completely as possible. In no case does such identification imply a recommendation or it imply that any of the materials, instruments or equipment identified are necessarily the best available for the purpose.

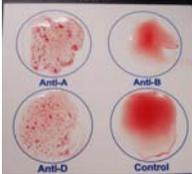
Information presented does not necessarily represent the official position of the National Institute of Standards and Technology or the U.S. Department of Commerce.

Rapid Overview

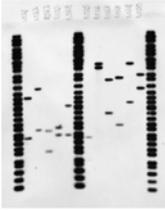
- Before there was Rapid DNA
- Rapid DNA Advancements
- NIST's role in Rapid DNA
- Future of Rapid DNA



Before there was Rapid DNA



ABO Blood Typing
(~1900-1985)



RFLP
(~1985-mid 1990s)



DQ alpha & Dot Blot Hybridization
(Early-mid 1990s)

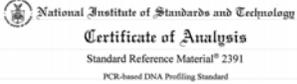
42 U.S. Code Part A - DNA Identification

- § 14131 - Quality assurance and proficiency testing standards
- § 14132 - Order to facilitate law enforcement exchange of DNA identification information

The DNA Identification Act of 1994

Then in 1995...

SRM 2391 PCR-based DNA Profiling Standard Released



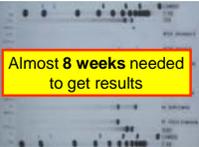
DNA typing presented for the first time in a major case

Questions of evidence handling
Validity of DNA typing process not questioned

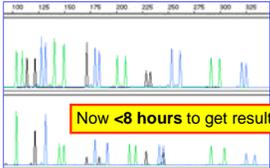


Since 1995...

Forensic DNA typing moved from RFLP to commercial multiplex STR kits



RFLP discontinued US-wide 2000



- 1996 first multiplex STR Kits available
- 2000-2001 single coamplification kit of CODIS STRs





DNA Core Loci Expanding
Thu, Dec 15 2016 by Seth Augenstein
Expanded January 2017

From 2000s to Present Day

The forensic science world is on the brink of a revolution



Human Genome Sequenced

Commercial Multiplex STR Kits >20 Loci

ABI 3100 Genetic Analyzer introduced

ABI 3500 Genetic Analyzer introduced

NDIS exceeds 12 million offender profiles

NEWS

What If All Law Enforcement Agencies Could Do Instant DNA Analysis?

Forensic News
Thu, Oct 4 2012

No human intervention

Minimal operator education

Small liquid volumes

Faster PCR

Expert analysis software

Rugged & Portable

Single instrument

Faster Separation & Detection

No liquid handling steps

Stable reagents

Microfluidics

DNA Typing Process

Extraction

Quantitation

Multiplex PCR

Separation & Detection

Interpretation of Results

HOW IT WORKS LIFESAVING TECH

THE LAB THAT FITS IN YOUR HAND

Entire DNA typing process in less than 2 hours

Rapid Advancements



- Many efforts made to reduce time within the DNA typing process
- Predominant focus area in reduction: **PCR**

Forensic Science International: Genetics 18 (2015) 96-99

Contents lists available at ScienceDirect

Forensic Science International: Genetics

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

Review

Rapid PCR of STR markers: Applications to human identification

Erica L. Romson^a, Peter M. Vallone

^aNational Institute of Standards and Technology, 800 Burns Drive, MS 8519, Gaithersburg, MD 20899-6106, USA

Alternative DNA Polymerases

- Allow for a higher processivity than AmpliTaq Gold
- Higher resistance to inhibitors
- 16-32 fold increase in efficacy with shorter extension times
 - Allows for reduction in PCR thermal cycling time

In vitro
 A novel strategy to engineer DNA polymerases for enhanced processivity and improved performance
 Yan Wang, Dennis E. Prosen, Li Mei, John C. Sullivan, Michael Finney and Peter B. Vander Horn

Anatomy of a Polymerase - How Structure Effects Function

bioRxiv preprint doi: <https://doi.org/10.1101/184007>; this version posted October 1, 2017. The copyright holder for this preprint (which was not certified by peer review) is the author/funder, who has granted bioRxiv a license to display the preprint in perpetuity. It is made available under aCC-BY-NC-ND 4.0 International license.



Rapid PCR Protocols Standard PCR Cycling Time: ~3 Hours

2009: Identifiler Optimized - 36 minutes

2012: Identifiler 2-step PCR - 26 minutes

2013: MP7 direct PCR - 16 minutes

2014: Identifiler of 3-step and 2-step PCR across six thermal cyclers - 14 minutes with Streck Philisa

Rapid direct PCR for forensic genotyping in under 25 min

Rapid PCR protocols for forensic DNA typing on six thermal cycling platforms



Rapid PCR on a Chip

- Reduction of PCR volume
- Miniaturization and integration with microfluidic

Table 3
 Summary of studies performing PCR of STR markers on a chip. E.L. Roman, P.M. Valore / Forensic Science International: Genetics 18 (2015) 90-99

Assay/Primers	Polymerase	Thermal cycling	Time (min)	Reference number
AmpliTaq Gold	AmpliTaq Gold	Miniature analytical thermal cycler instrument	46	[66]
Proteinase Plus	SpeedSTAR	Custom thermal cycler	12.5	[70]
PowerPlex ESX 17	Hot Report	Custom hydrogen psPAM beads	10	[71]
Identifiler	Taq Cold	Non-contact infrared	90	[76]
Identifiler	SpeedSTAR	Non-contact infrared	45	[76]
Identifiler			77	[77]
Identifiler			78	[78]

TECHNICAL NOTE

Heidi Giese,¹ Ph.D.; Roger Lam,¹ M.Sc.; Richard Selden,¹ M.D., Ph.D.; and Eugene Tan,¹ Ph.D.

Fast Multiplexed Polymerase Chain Reaction for Conventional and Microfluidic Short Tandem Repeat Analysis
 Devices: A Review

lic devices



Forensic Integration - Microfluidics

Integrated Microfluidic Systems for DNA Analysis

Review of microfluidic systems which were composed of 2+ microdevices

Samuel K. Njoroge, Hui-Wen Chen, Małgorzata A. Witek, and Steven A. Soper

Integration of liquid extraction with a pre-existing microfluidic PCR platform and μCE

An integrated sample-in-answer-out microfluidic chip for rapid human identification by STR analysis†

Delphine Le Roux,¹ Brian E. Root,² Jeffrey A. Hickey,³ Orion N. Scott,⁴ Anchi Tsai,⁵ Jingqi Liu,⁶ David J. Saul,¹ Luc Chassignac,⁶ James P. Landers¹ and Philippe de Mazancourt^{1*}

Anal. Chem. 2016, 88, 6991-6999

Integrated Microfluidic System for Rapid Forensic DNA Analysis: Sample Collection to DNA Profile

First demonstrated fully integrated device without any manual intervention

Andrew J. Hapwood,^{1*} Cedric Burth,¹ Jianing Yang,¹ Zhi Gu,¹ Nina Moran,¹ John G. Lee-Edgill,¹ Alan Nordquist,¹ Paul Longhi,¹ Matthew D. Eades,¹ John P. Halsey,¹ Gotta R. McMillen,¹ Xiaojie Chen,¹ Carter Brooks,¹ Stan Smith,¹ Keith Elliott,¹ Floris Koenig,¹ Frederic Zentgraf,^{1*} and Gillian Tully¹

Run time: 4 hours

Research and Development, Forensic Science Service, Tipton Court 2000 South Parkway, Birmingham Business Park, Birmingham UK B37 7YU, and Center for Applied Neurobiology and Medicine, The University of Arizona College of Medicine, 425 N. Fifth Street, Phoenix, Arizona 85024

Commercial RDNA Instruments

IntegenX

- RapidHIT 200
 - PowerPlex 16HS
 - Globalfiler
- RapidHIT ID
 - Globalfiler





NetBio/ANDE

- ANDE/DNAScan
 - PowerPlex 16
- ANDE
 - FlexPlex (27)





Rapid Advancements: NIST Edition

How does NIST fit into Rapid DNA Identification?

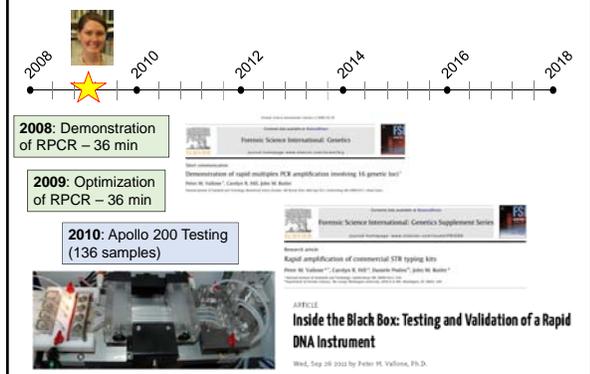
Who are we?

- NIST falls within the Department of Commerce
 - Mission: to promote innovation and industrial competitiveness by advancing measurement science, standards, and technology

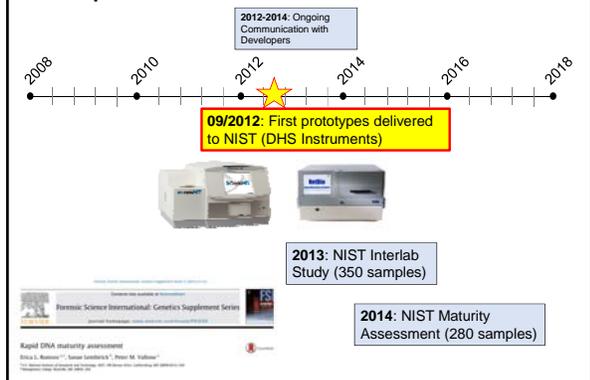
- Our focus is on making measurements
 - Robust examinations of technology
 - Collaboration with other federal/state/local users
 - Collaboration with industry

- Interagency collaboration with the FBI and DHS

Rapid Advancements: NIST Edition



Rapid Advancements: NIST Edition



NIST: 2012 to 2014

- Many developmental changes and upgrades during 2012-2013 timeframe
 - Software, hardware, reagents, consumables, etc
- Over 700 samples run between both platforms
- NIST participation in the Rapid-DNA committee within the Scientific Working Group on DNA Analysis Methods (SWGDM)

NEWS
FBI DNA Quality Assurance Standards Now Include Rapid DNA Analysis

Mon, Dec 8 2014, by rwaters

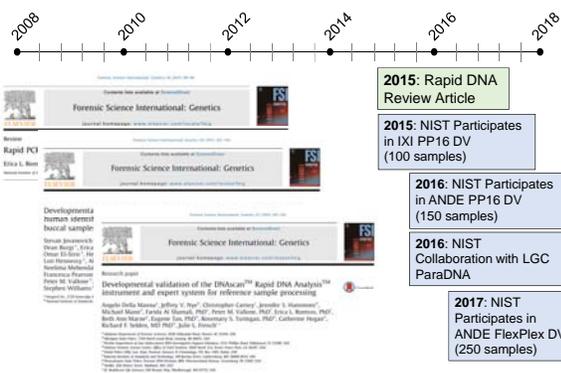
NIST: Interlabs and Maturity Assessments

- Collection and distribution of samples to all participating laboratories
 - IL: 3 laboratories, 350 samples total
 - MA: 7 laboratories, 280 samples total
- Coordination of all testing sites to include return of all data to NIST for analysis and review
- Analysis and compilation of all data
- Summary of results presented across multiple meetings within the forensic and biometric communities

Rapid DNA Maturity Assessment
 Erica L. Romagnolo¹, Sarae Lemberick², and Peter M. Vallone¹
1 U.S. National Institute of Standards and Technology, 100 Bureau Drive, Gaithersburg, MD 20899-8314, USA
 2 Montgomery College, Rockville, MD 20850, USA



Rapid Advancements: NIST Edition

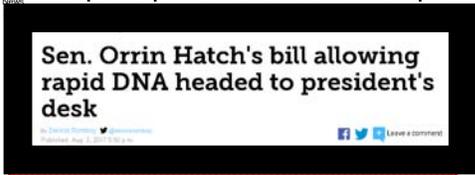


NIST: 2017 and Beyond

- Continuing to provide data in support of discussion within the SWGDAM R-DNA committee
- Subject matter experts for R-DNA for DHS
- Continued support to the R-DNA community and developers

Future of Rapid DNA

NEWS
Legal Hurdles Threaten to Slow FBI's 'Rapid DNA'
 Revolution Op-ed: Update law so labs can use Rapid
 Forensic News Thu, Sep 14, 2017 10:30 AM



Section 1, Short title

This Act may be cited as the "Rapid DNA Act of 2017".

Hatch's Rapid DNA Act will make communities safer

FRIDAY, JULY 27, 2017, 4:38 AM

Future of Rapid DNA

- FBI integration of commercial Rapid DNA profiles into CODIS with search against NDIS
 - Rapid DNA Instrumentation within Police Booking Stations
- DHS continual efforts to employ Rapid DNA typing for immigration, refugee status, and mass fatality response operations for kinship testing



<p>ANDE/MIT-LL Eugene Tan Melissa May Julie French Richard Selden Catherine Hogan Paula Pomianowski Collins Marcus Lines Themis Parodos Anthony Lapadula Martha Petrovick Rosemary Turingan John Johnson</p>	<p>IntegenX/IQT Stevan Jovanovich Jacklyn Buscaino Omar El-Sissi Helen Franklin Stafanie Gangano Lori Hennessy David King Charles Park Robert Schueren Jason Werking Timothy Look Robert Courter Roy Swiger Chris Contonis Jason VanSice Stephanie Rogers Richard Brooks Benny Wicks Paul Kotturi Joseph DiZinno Bob Barrett Keith Elliott Kevin O'Connell Stephen Williams</p>	<p>Federal Past and Current Members of SWGDAM RDNA Committee Karen Olson Brigid O'Brien Ken Kroupa Jeff Salyards William Towns Michael Kessler Roland Castillo Daniel Hoffhines Robert Driscoll Colin Steven Jennifer Wendel Elisabeth Johnson Shahram Orandi Melanie Glass Bob Zimmerman Michael Mann Julie Kidd Jerry Sellers James Loudermilk Lilliana Moreno Stephen Cargo Travis Hite Yonas Nebiyeloul</p>	<p>Industry Steve Kelly Chris Connelly Michael van Waes Richard Healey Jeff Hickey Abby Mackness Alice Chung Kathy Webb Yvette Crandall Amanda Sozer Stephanie DeDore</p>
<p>Lockheed/UVA Joan Bienvenue Delphine Le Roux Brian Root James Landers John Mears</p>	<p>LGC/ParaDNA Simon Wells Mark Dearden Eloise Busby Paul Rendell Michael Winter Jim Huggett</p>	<p>State/Local/Academic Cecelia Crouse Bill Hudlow Mavis DateChong Christopher Carney Bruce McCord Daniele Podini Bobby LaRue Bruce Budowle Susan Greenspoon Theresa Caragine Amy Mundorff</p>	

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