Issues and Challenges with Forensic DNA Analysis

John M. Butler, Ph.D.

NIST Fellow & Special Assistant to the Director for Forensic Science
U.S. National Institute of Standards and Technology
Acknowledgment and Disclaimers

I quote from my recent book entitled “Advanced Topics in Forensic DNA Typing: Interpretation” (Elsevier, 2015). I do not receive any royalties for this book. Completing this book was part of my job at NIST.

Although I chaired the SWGDAM Mixture Committee that produced the 2010 STR Interpretation Guidelines, I cannot speak for or on behalf of the Scientific Working Group on DNA Analysis Methods.

I have been fortunate to have had discussions with numerous scientists on interpretation issues including Mike Coble, Bruce Heidebrecht, Robin Cotton, Charlotte Word, Catherine Grgicak, Peter Gill, Ian Evett …

Points of view are mine and do not necessarily represent the official position or policies of the US Department of Justice or the National Institute of Standards and Technology.

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If you want to be a technician, performing tests on requests, then just focus on the policies and procedures of your laboratory. **If you want to be a scientist and a professional**, learn the policies and procedures, but go much further and learn the philosophy of your profession. **Understand the importance of why things are done** the way they are done, the scientific method, the viewpoint of the critiques, the issues of bias and the importance of ethics.
Overview of My Career

B.S. Chemistry (1992)


Research Conducted at FBI

NIST/NRC Postdoc (Sept 1995 - May 1997)

Some Research at AFDIL

Staff Scientist (May 1997 – Sept 1999)

Research Chemist (Sept 1999 – March 2008)

NIST Fellow & Group Leader (Mar 2008 – Mar 2013)

Special Assistant to NIST Director for Forensic Science (April 2013 - present)

ARMED FORCES DNA IDENTIFICATION LABORATORY

Silicon Valley start-up company doing TOF-MS of DNA

Forensic DNA Project Leader

National Institute of Standards and Technology

GENETRACE

National Institute of Standards and Technology
National Institute of Standards and Technology

• Science agency part of the U.S. Department of Commerce
• Started in 1901 as the National Bureau of Standards
• Name changed in 1988 to the National Institute of Standards and Technology (NIST)
• Forensic science research activities dating back to 1920s
• Partnership since 2013 with U.S. Department of Justice to create the National Commission on Forensic Science (NCFS) and the Organization of Scientific Area Committees (OSAC)

• Primary campus in Gaithersburg, Maryland (near Washington, D.C.)
• >3,400 employees and >3,700 associates
• Supplies >1300 reference materials
• Defines official time for the U.S.
The Best Forensic Scientist You’ve Never Heard Of

Wilmer Souder and the Early History of Forensic Science at the National Bureau of Standards

Kristen M. Frederick-Frost, PhD
Robert M. Thompson, BS
John M. Butler, PhD

LW1: Last Word Society
American Academy of Forensic Sciences
Las Vegas, NV (February 25, 2016)

Slides available on the NIST STRBase website:
What Do I Do in My Job at NIST?

• **Write articles** for scientific journals sharing research results or reviewing efforts in forensic science and DNA
  – Have written >150 articles and 5 textbooks so far

• **Prepare presentations** and speak on forensic science and DNA testing to scientists and lawyers and the general public
  – >300 presentations given in >30 states and 25 countries

• **Participate in meetings** influencing forensic science policy and practice
  – Serve as Vice-Chair of the National Commission on Forensic Science
  – Member of the OSAC Biology/DNA Scientific Area Committee
  – Member of the AAFS Standards Board DNA Consensus Body

• **Visit forensic laboratories** to learn of their challenges and to teach them about potential solutions
Recent Activities of the National Commission on Forensic Science

Written by John M. Butler

In February 2013, the U.S. Department of Justice (DOJ) and the National Institute of Standards and Technology (NIST) announced a partnership that included formation of the National Commission on Forensic Science (NCFS) and what is now the Organization of Scientific Area Committees (OSAC). As a Federal Advisory Committee for DOJ, NCFS involves public meetings, public input on draft documents, and an open website sharing meeting materials and final documents. Video recordings of past meetings are available as well. Co-chaired by Deputy Attorney General Sally Yates and NIST Director Willie May, the Commission meets four times a year and involves energetic discussions on a variety of issues.

The accompanying figure is an attempt to show where the 23 NCFS documents thus far approved impact what can be termed the "forensic science ecosystem," which involves law enforcement, forensic laboratories, scientific (academic) research, medical examiner or coroner's offices, and the legal system. For example, NCFS work product #20 is a recommendation regarding a National Code of Professional Responsibility for Forensic Science and Forensic Medicine Service Providers, which received approval at the March 2016 meeting. The number 20 is shaded in light blue because this recommendation is currently under consideration by DOJ.

At the June 20-21, 2016 NCFS meeting, final drafts for seven work products may be introduced for a vote and approval by the Commission. These documents include recommendations regarding pretrial discovery, a request for NIST to perform developmental validation studies, accreditation of digital and multimedia forensic science service providers, and formation of a national disaster call center. Views documents under consideration cover judicial review of experts, notice and demand provisions, and validation of forensic science methodology.

The Commission's vision is for all forensic evidence to support the equal and impartial application of justice. The NCFS efforts can be framed into three primary goals: (1) foundational—framework and recommendations, (2) implementation—materials, and (3) subject areas—methods and protocols for specific types of forensic science.

First 23 NCFS Adopted Work Products

Note: The recommendations in green below are pending or in the queue to be accepted by DOJ. Other recommendations are being considered (blue) or are outside the purview of DOJ (gray). Views of the Commission are in yellow.

- Questioned sample
- Known sample
- Sample Processing
- Analysis Interpretation
- K Comparison
- Report Issued
- If court, then expert testimony
- Training

Legal Proceedings

Forensic Laboratory

Medical Examiner or Coroner’s Office
Butler Books on Forensic DNA Typing
DNA Capabilities to Aid Forensic Investigations

1. The ability to identify the perpetrator
2. Weight-of-evidence based on established genetic principles and statistics (Hardy-Weinberg 1908)
3. Established characteristics of genetic inheritance enables close biological relatives to be used for reference points using kinship associations
4. Superb sensitivity with PCR amplification (opens the possibility for contamination)
5. Well-established quality assurance measures
6. New technology development aided by genomics

Successful interpretation of DNA (Q-to-K comparison) depends on quality of the crime scene evidence (Q) and availability of suitable reference samples (K)
Forensic DNA Testing in the United States

- We have ~200 public (state and local government) laboratories performing forensic DNA analysis
  - Two large private companies (Bode Cellmark and Sorenson Forensics) and a few smaller ones perform forensic DNA analysis

- Over 15 million DNA profiles in the national DNA database (NDIS: National DNA Index System) run by the FBI Lab
  - Since 1998, the U.S. has included 13 core STR (short tandem repeat) markers; starting in 2017, this number has increased to 20 required STR loci

- Laboratories have many different protocols and in some cases, submitting the same sample to two different laboratories could result in two different results
  - Efforts are underway to improve standardization in the field
Critical Challenges Faced Today

• **Success of DNA testing** → significant growth in sample submissions → sample backlogs
  – Laboratory automation and expert system data review
  – Restrictive case acceptance policies to avoid law enforcement investigator ‘swab-athons’ at crime scenes

• **Greater detection sensitivity** → more complex DNA mixtures and low-template DNA with ‘touch’ evidence
  – Probabilistic genotyping to cope with increase in data interpretation uncertainty
  – Use of a complexity threshold to avoid “skating on thin ice”

Landmark Report Gives DNA Testing a Pass

The U.S. National Research Council of the National Academies issued a major report on forensic science in Feb. 2009.

"With the exception of nuclear DNA analysis, no forensic method has been rigorously shown to have the capacity to consistently, and with a high degree of certainty, demonstrate a connection between evidence and a specific individual or source." (p. 41)

p. 100 mentions limitations with DNA mixtures
Recent Forensic DNA Problems in the News

**Washington DC Crime Lab** problems with **DNA Mixture Interpretation**

Director of D.C.'s embattled DNA lab resigns after suspension of testing

Max M. Houck had been the director since the lab opened in 2012. Auditors found major problems there.

Keith L. Alexander and Julie Zauzmer | Local | Apr 30, 2016

District could spend nearly $1 million for outside lab to test DNA evidence

The District is scrambling to find an alternative after the D.C. lab was ordered to cease DNA testing.

Keith L. Alexander | Crime | Apr 29, 2015

**Texas DNA Mixture Case Review**

August 2015

http://www.fsc.texas.gov/texas-dna-mixture-interpretation-case-review


Austin, Texas lab closed in June 2016

**Broward County Florida DNA Lab**

April 2015


July 2016

The Washington Post
PCAST Report Comments on Forensic DNA

- Supports appropriate use of single-source and simple mixture DNA analysis
- Expresses reservations with complex DNA mixtures (≥3 contributors)

PCAST Co-Chairs

Eric Lander  John Holdren
David Balding: “Low-template DNA cases are coming to court with limited abilities for sound interpretation. ... There are dangers with LTDNA but we know how to handle and manage them. Unfortunately, proper management is not a universal practice.”

Peter Schneider: “If you cannot explain your evidence to someone that is not from the field (like a judge) – and you need a lot of technical excuses to report something – then the result is not good. You should leave it on your desk and not take it to court. This is a very common sense approach to this problem.”
The author’s thoughts and opinions on where the field of forensic DNA testing is headed for the next decade are provided in the context of where the field has come over the past 30 years. Similar to the Olympic motto of ‘faster, higher, stronger’, forensic DNA protocols can be expected to become more rapid and sensitive and provide stronger investigative potential. New short tandem repeat (STR) loci have expanded the core set of genetic markers used for human identification in Europe and the USA. Rapid DNA testing is on the verge of enabling new applications. Next-generation sequencing has the potential to provide greater depth of coverage for information on STR alleles. Familial DNA searching has expanded capabilities of DNA databases in parts of the world where it is allowed. Challenges and opportunities that will impact the future of forensic DNA are explored including the need for education and training to improve interpretation of complex DNA profiles.
## Stages of Forensic DNA Progression

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<thead>
<tr>
<th>Stages</th>
<th>Time Frame</th>
<th>Description</th>
</tr>
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<tbody>
<tr>
<td>Exploration</td>
<td>1985 - 1995</td>
<td>Beginnings, different methods tried (RFLP and early PCR)</td>
</tr>
<tr>
<td>Stabilization</td>
<td>1995 - 2005</td>
<td>Standardization to STRs, selection of core loci, implementation of Quality Assurance Standards</td>
</tr>
<tr>
<td>Growth</td>
<td>2005 - 2015</td>
<td>Rapid growth of DNA databases, extended applications pursued</td>
</tr>
<tr>
<td>Sophistication</td>
<td>2015 to 2025 and beyond</td>
<td>Expanding tools available, confronting privacy concerns</td>
</tr>
</tbody>
</table>

Table 1 from J.M. Butler (2015) The future of forensic DNA analysis. *Phil. Trans. R. Soc. B* 370: 20140252
Current Trends in Forensic DNA

- **Faster results:** Rapid DNA capabilities and new sample-to-answer integrated instruments

- **Higher sensitivity:** New assays lowering the limits of detection, which makes interpretation more challenging

- **Higher information content:** Next-generation sequencing (NGS) for more markers & STR allele information

- **Stronger conclusions:** Mixture interpretation with probabilistic genotyping models

5 Reasons that DNA Results Are Becoming More Challenging to Interpret

1. More sensitive DNA test results
2. More touch evidence samples that are poor-quality, low-template, complex mixtures
3. More options exist for statistical approaches involving probabilistic genotyping software
4. Many laboratories are not prepared to cope with complex mixtures
5. More loci being added because of the large number of samples in DNA databases

Math Analogy to DNA Evidence

\[ 2 + 2 = 4 \]

\[ 2x^2 + x = 10 \]

\[ \int_{x=0}^{\infty} f(x) \, dx \]

**Basic Arithmetic**

**Algebra**

**Calculus**

Single-Source DNA Profile (DNA databasing)

Sexual Assault Evidence (2-person mixture with high-levels of DNA)

Touch Evidence (>2-person, low-level, complex mixtures perhaps involving relatives)

Many laboratories are not prepared to cope with complex mixtures

- Have **appropriate validation studies** been performed to inform proper interpretation protocols? (curriculum & classroom instruction)

- Are **appropriately challenging proficiency tests** being given? (graded homework assignments)

- Would we want to go into a calculus exam only having studied algebra and having completed homework assignments involving basic arithmetic?
Position of Forensic STR Markers on Human Chromosomes

13 Core U.S. STR Loci

D1S1656  D2S1338  D13S1358  D2S441  TPOX

D5S818  D3S1358  FGA

D8S1179  D7S820  CSF1PO

TH01  D10S1248  VWA

D12S391

15 STR loci overlap between U.S. and Europe

1997 (13 loci)

2017 (20 loci)

AMEL  AMEL

Sex-typing

This has required the validation of new DNA testing kits over the past two years
Some Thoughts on Challenges Facing Forensic Science
Important Observations

• The National Research Council 2009 ("NAS Report") called for changes to strengthen forensic science (with 13 recommendations) but these are not really new issues

• The criminal justice system, where forensic science only plays a small part, is not perfect; there have been individuals wrongly convicted for a variety of reasons

• Despite a few well-publicized examples (e.g., Annie Dookhan), forensic scientists generally want to do a good job and are trying to do their best

• Many forces are at play to either change things or to maintain the status quo → which changes are needed?
Culture Clash: Science and Law

Tension exists between science and the law:

• The legal community looks to the past (precedence is desired)

• The scientific community looks to the future (evolving improvement is desired)
Culture Clash: Science and Law

Tension exists between science and the law:

• The legal community **wants finality and absolutes** (guilty or not-guilty court decisions)

• The scientific community **operates without certainty** (rarely with probabilities of 0 or 1)
Nomenclature Challenges

• We often talk past each other (scientists and lawyers or scientists and scientists) because we do not appreciate a subtle or significant difference in the meaning of a word or phrase

• Examples: “validity” or “validation” can mean something very different to lawyers than to scientists and forensic practitioners

• “A reasonable degree of scientific certainty…” (a legal crutch that has no scientific meaning)
What Can You Do to Contribute to Solutions in the Future?

Know the literature
Know the question you are trying to answer
Know the limits of what you can do
Know the Literature

- We must do our homework – and read the literature!

- AAFS 2016 workshop
  - Information Does Exist Beyond the First Page of Your Google® Search!: Tools and Strategies for Forensic Science Literature Searching and Use
  - Search tools and strategies are described
Steps in Forensic DNA Analysis

Gathering the Data

Collection/Storage/Characterization → Extraction/Quantitation → Amplification/Marker Sets → Separation/Detection

Advanced Topics: Methodology

>1300 pages of information with >5000 references cited in these two books

> Understanding Results Obtained & Sharing Them

Data → Stats → Report

Advanced Topics: Interpretation

August 2011

October 2014
Know What Question You Are Trying to Answer

“…Focus on the relevant question. Many misleading statistical approaches [turn] out to be providing valid answers to the wrong questions.”

# Different Calculations

## Answer Different Questions

<table>
<thead>
<tr>
<th>Method used</th>
<th>Questions being answered</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Profile probability</strong> (random match probability, RMP)</td>
<td>What is the rarity of a specific DNA profile given the alleles observed? <strong>What is the chance that a particular profile exists</strong> in a population based on allele frequencies?</td>
</tr>
<tr>
<td><strong>Match probability</strong></td>
<td>Given that a particular profile has been seen (in the crime scene evidence and in the suspect), <strong>what is the chance of it occurring again?</strong></td>
</tr>
<tr>
<td><strong>Database match probability</strong></td>
<td>How often would a DNA profile match the relevant forensic sample in a database of size $N$?</td>
</tr>
</tbody>
</table>

Adapted from Table 11.7, J.M. Butler (2015) *Advanced Topics in Forensic DNA: Interpretation* (Elsevier Academic Press)
“The crucial element that the scientist brings to any case is the *interpretation* of those observations. This is the heart of forensic science: it is where the scientist adds value to the process.”

Know the Limits of What You Can Do


Low-Level DNA and Complex Mixtures

“The limits of each DNA typing procedure should be understood, especially when the DNA sample is small, is a mixture of DNA from multiple sources, or is contaminated with interfering substances.”

NRC I, 1992, p. 8

“For the complex DNA profile, there is no predominant or overarching standard interpretation method.”

Peter Gill (*Gill et al. 2012*, report to the UK Forensic Science Regulator, p. 18)

“The limits of each DNA typing procedure should be understood, especially when the DNA sample is small, is a mixture of DNA from multiple sources…”  (NRC I, 1992, p. 8)
In the fields of observation chance favors only the prepared mind.
Alexander Hamilton

- Men give me some credit for genius, but all of the genius I have lies in this. **When I have a subject in mind, I study it profoundly**, day and night it is before me. I explore it in all its bearings. My mind becomes pervaded with it. The result is what some people call the fruits of genius, whereas **it is in reality the fruits of study and labor** (as quoted in Sterling W. Sill, The Upward Reach, p. 125).
Doug Butler Thoughts on Learning

“You never really learn anything until you have to teach it to someone else.”

My father has written a dozen books covering his field of horseshoeing and started his own school after teaching at three different universities.
National Commission on Forensic Science (NCFS):
www.justice.gov/ncfs

Organization of Scientific Area Committees (OSAC):
www.nist.gov/forensics/osac/index.cfm

www.nist.gov/forensics

+1-301-975-4049  john.butler@nist.gov
International Symposium on Forensic Science Error Management
July 24-28 @NIST, Gaithersburg, MD

Technical Tracks

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- Human Factors
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Or search for “NIST 2017 forensic error management”