


**Overview of Rapid DNA Testing**  
2009 Biometric Consortium Conference  
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Dr. Peter M. Vallone  
Biochemical Science Division  
National Institute of Standards and Technology

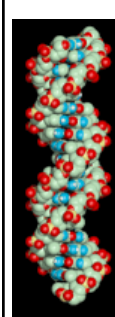
### Outline

- Basics of DNA Typing
- Rapid PCR



### Basics of Forensic DNA Testing

### General Characteristics of Genomic DNA



- Each person has a unique DNA profile (except identical twins)
- Each person's DNA is the same in every cell (DNA from skin cells will match DNA from blood cells)
- An individual's DNA profile remains the same throughout life
- Half of your DNA comes from your mother and half from your father

### Forensic DNA Testing

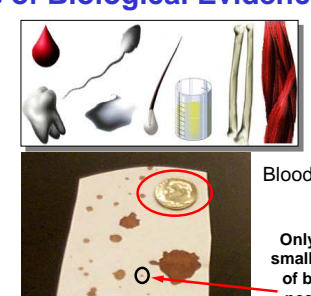
Probe subsets of genetic variation in order to differentiate between individuals

DNA typing must be done efficiently and reproducibly (information must hold up in court)

Typically, we are not looking at genes – little/no information about race, predisposal to disease, or phenotypical information (eye color, height, hair color) is obtained

### Sources of Biological Evidence

- Blood
- Semen
- Saliva
- Urine
- Hair
- Teeth
- Bone
- Tissue



Blood Sample

Only a very small amount of blood is needed to recover DNA

best results with >100 cells, but DNA profiles can be recovered from as little as a single cell

## Applications

- Forensic cases: matching suspect with evidence
- Paternity testing: identifying father
- Missing persons investigations
- Military DNA “dog tag”
- Convicted felon DNA databases
- Mass fatalities: putting pieces back together
- Historical investigations
- Genetic genealogy
- **DNA as a biometric tool**

## DNA Testing Requires a Reference Sample

A DNA profile by itself is fairly useless because it has no context...

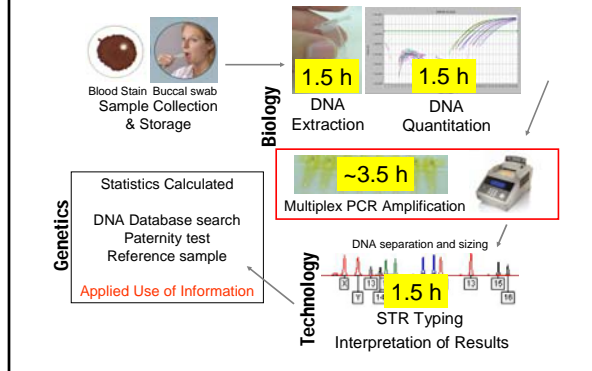


DNA analysis for identity only works by comparison – **you need a reference sample**

**Crime Scene Evidence** compared to **Suspect(s)** (Forensic Case)  
**Child** compared to **Alleged Father** (Paternity Case)  
**Victim's Remains** compared to **Biological Relative** (Mass Disaster ID)  
**Soldier's Remains** compared to **Direct Reference Sample** (Armed Forces ID)

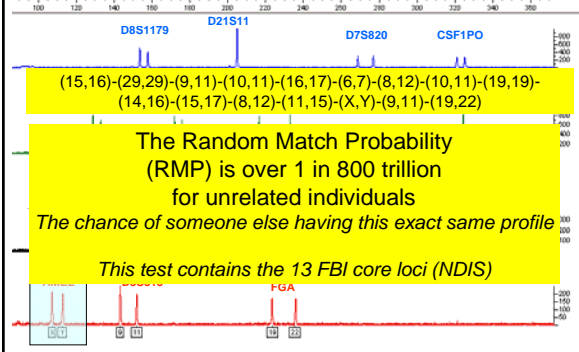
## Steps in Forensic DNA Analysis

Usually 1-2 day process (a minimum of ~8 hours)



## Identifiler (Applied Biosystems) 15 STR Loci Kit

Information is tied together with multiplex PCR and data analysis



## Recent Work with Rapid PCR

At NIST we are working on new PCR methods to reduce the time for PCR down to 20 minutes

Polymerase Chain Reaction (PCR)

Is a means to create billions of exact copies of the human genome – necessary/essential for DNA typing

~3.5 h → ~20 min?

Multiplex PCR Amplification



## Why go Faster? Applications for Rapid PCR

- **Integrated devices** ('Lab on a Chip')
- **Screening** at a point of interest (airport, border, crime scene, intelligence community)
- **Rapid STR typing 'in the field'**
  - Potential for situations/cases when a quick result is needed
  - Provide initial screening information
- Decrease overall time required for STR typing

**DNA as a Biometric tool**

### Current Efforts Towards Portable/Mobile DNA Devices

- Network Biosystems (Woburn, MA)  
<http://www.netbio.com>
- MicroLab Diagnostics and Lockheed Martin (Charlottesville, VA)  
<http://www.microlabdiagnostics.com>
- Microchip Biotech (Dublin, CA)  
<http://www.microchipbiotech.com>

### Goals for Rapid DNA Typing Platforms

- Create an **integrated system** capable of taking a swab and perform DNA testing in **approximately 1 hour**
- Little user interaction (or experience)
- Rugged
- Robust **Swab in...answer out**
- Simple data interpretation

### Typical STR Typing Workflow

*Can the time required for PCR thermal cycling be reduced?*


Sample Extraction ~2 h	Quantitation ~1.5 h	PCR ~3 h	CE Run ~1.5 h
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Alter thermal cycling parameters  
Evaluate faster polymerases  
Evaluate faster thermal cyclers  
Test commercial STR typing kits

Data Review ?


**Goal: cycling in less than 40 minutes**  
Trying simple things first...

### Thermal Cyclers




Cepheid SmartCycler  
Ramp rate = 10°C/s

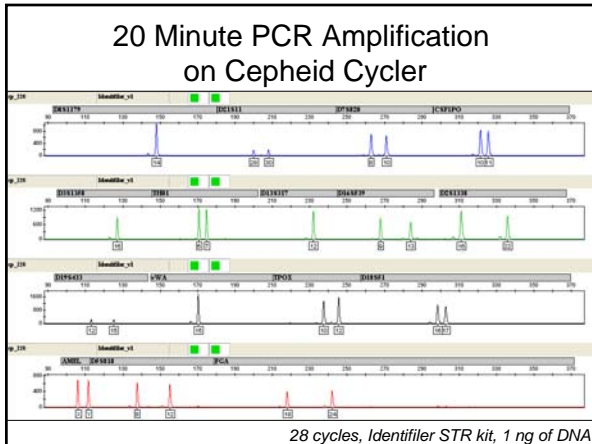
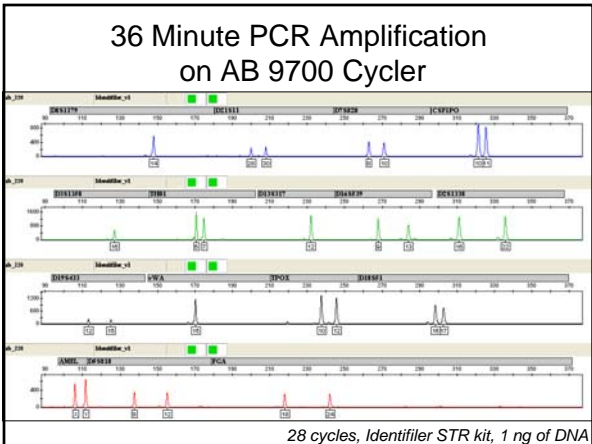
Purchased with FBI funding April 2009

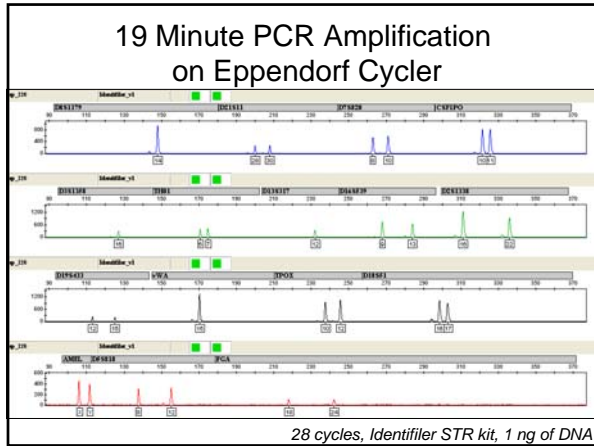


Eppendorf Mastercycler pro  
Ramp rate = 6°C/sec



Applied Biosystems 9700  
Ramp rate = 4°C/s





### Rapid PCR Article

Vallone, P.M., Hill, C.R., Butler, J.M. (2008) Demonstration of rapid multiplex PCR amplification involving 16 genetic loci. *FSI Genetics* 3(1): 42-45.

Rapid Amplification of Commercial STR Typing Kits  
Presented at the International Society of Forensic Genetics (ISFG)  
meeting in Buenos Aires Argentina (September 16, 2009)  
*(Voted Best Poster Presentation)*

- ### Rapid PCR Summary
- Rapid multiplex PCR amplification is possible
    - Compatible with commercial STR typing kits
    - Provides same genotypes as standard cycling
  - **Fast (optimized) polymerases are needed**
  - Further work
    - Applying techniques to integrated platforms
    - Formal validation of technique
    - [Sharing results with PCR community](#)
    - Understanding the kinetics of PCR

### Thank you for your attention!

Questions?

Peter.Vallone@nist.gov  
301-975-4872

Outside funding agencies:  
[FBI - Evaluation of Forensic DNA Typing as a Biometric Tool](#)  
[NIJ – Interagency Agreement with the Office of Law Enforcement Standards](#)