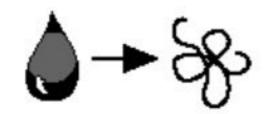


Presented By: Devendra Kumar M.Sc. (Biotechnology)

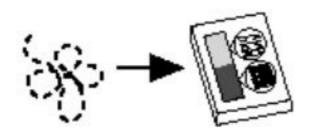
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#### THE PROCESS OF DNAFINGERPRINTING

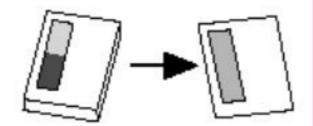
The process begins with a blood or cell sample from which the DNA is extracted.



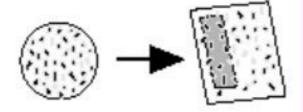
 The DNA is cut into fragments using a restriction enzyme. The fragments are then separated into bands by electrophoresis through an agarose gel.



The DNA band pattern is transferred to a nylon membrane.



4. A radioactive DNA probe is introduced. The DNA probe binds to specific DNA sequences on the nylon membrane.



 The excess probe material is washed awayleaving the unique DNA band plattem.



6. The radioactive DNA pattern is transferred to X-rayfilm by direct exposure. When developed, the resultant visible pattern is the DNAFINGER PRINT.

Figure 3.



# What is DNA Fingerprinting?

- DNA Fingerprinting is a way to identify a certain individual, rather than simply identifying a species or a particular trait.
- A technique used by scientists to distinguish between individuals of the same species using only samples of their DNA

#### Who Invented it?

The process of DNA fingerprinting was invented by Alex Jeffreys in 1985.



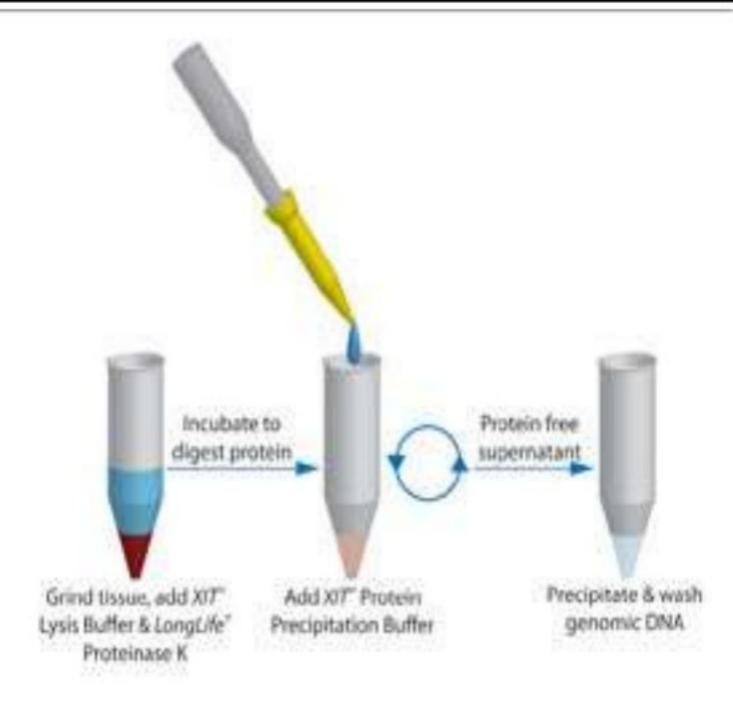
# Biological samples used for DNA fingerprinting

- Blood
- Hair
- Saliva
- Semen
- Body tissue cells
- DNA samples have been obtained from vaginal cells transferred to the outside of a condom during sexual intercourse.





- There are 8 steps for DNA Fingerprinting
- Step 1: Isolation of DNA
  - DNA must be recovered from cells or tissue. Only a small amount of blood, hair, or skin is needed to isolate DNA

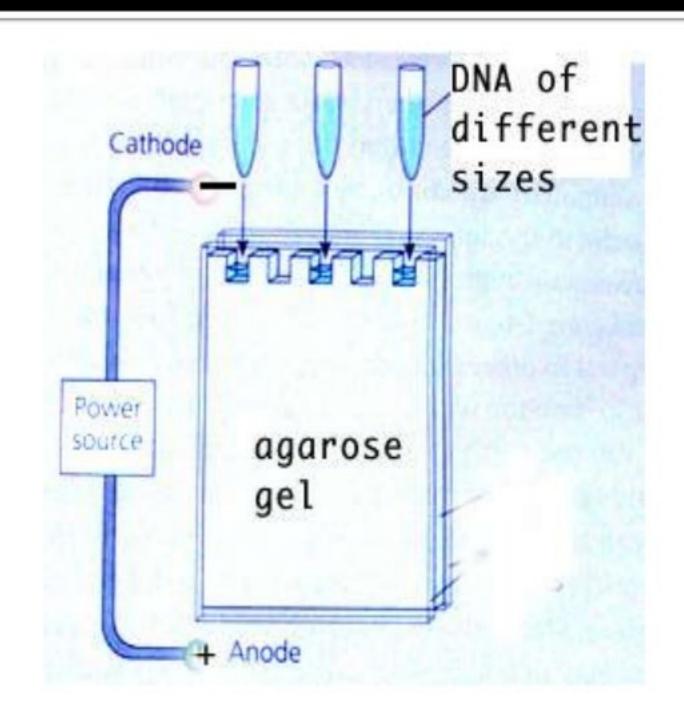


- Step 2:Digestion of DNA by restriction endonucleases
  - The DNA is cut into fragments using restriction enzymes.
  - Each restriction enzyme cuts DNA at a specific base sequence.

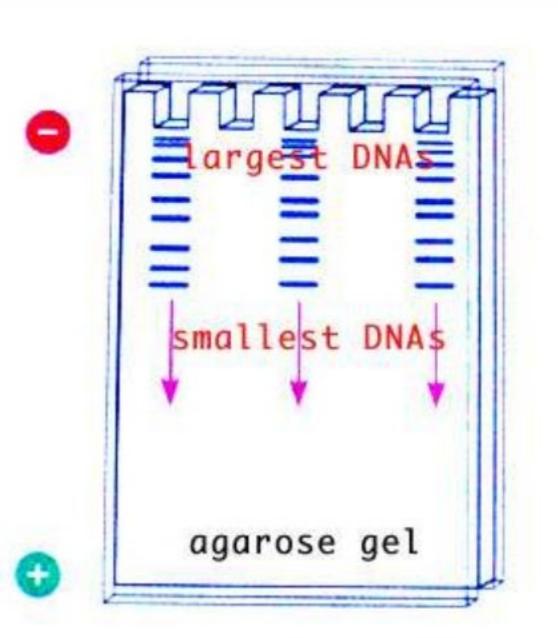


- The sections of DNA that are cut out are called restriction fragments.
- This yields thousands of restriction fragments of all different sizes because the base sequences being cut may be far apart (long fragment) or close together (short fragment).
- Commonly used RE are (hae III, Hinf I, Alu I etc.)
- Reaction mixture is incubated overnight at 37.C

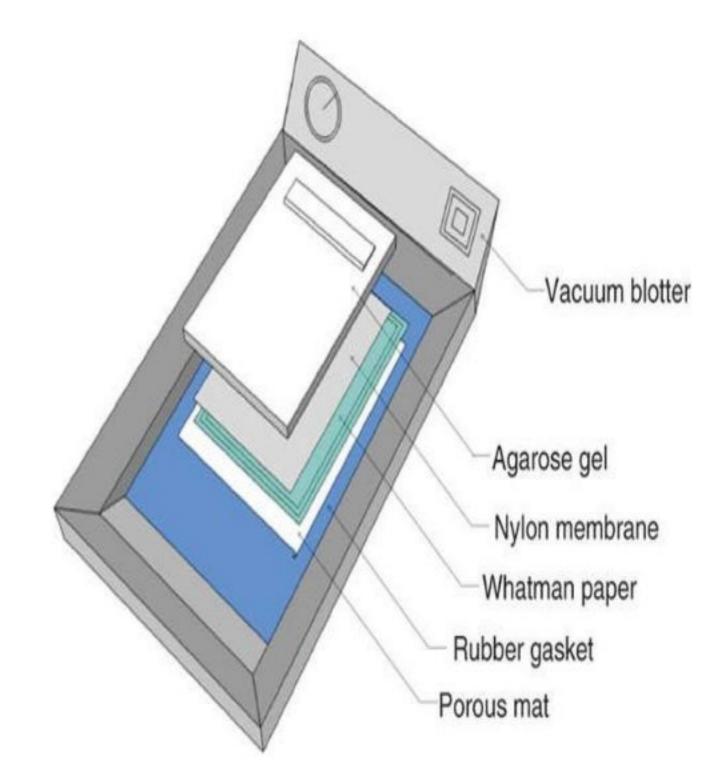
- Step 3: Electrophoretic separation of different fragments
  - Fragments are separated on the basis of size using a process called gel electrophoresis.
  - DNA fragments are injected into wells and an electric current is applied along the gel.

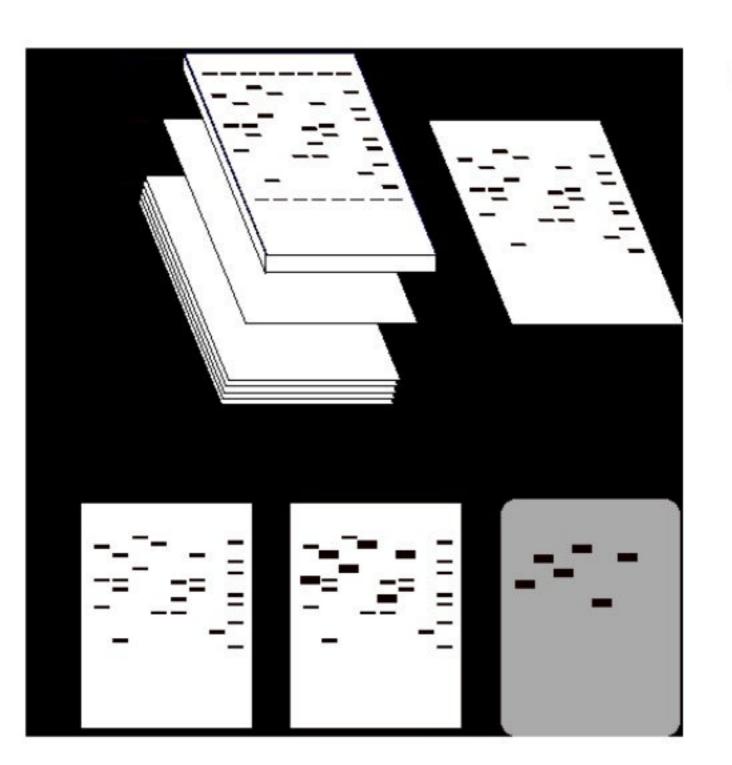


- DNA is negatively charged so it is attracted to the positive end of the gel.
- The shorter DNA fragments move faster than the longer fragments.
- DNA is separated on basis of size.



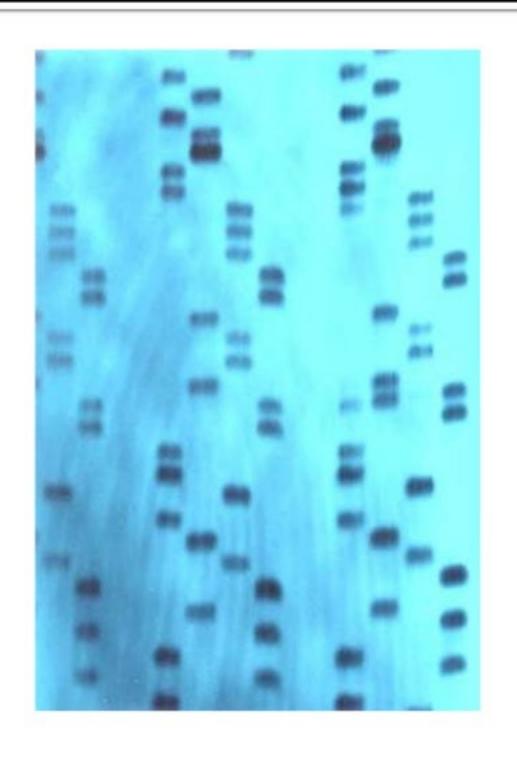
- Step 4: Transfer DNA on Nylon/Nitrocellulose membrane
  - The DNA fragments are transferred to a nylon sheet by placing the sheet on the gel and soaking them overnight by the process southern blot.





- Step5: Probing/probe labeling
  - Adding radioactive or colored probes to the nylon sheet which is complementary to target sequences.
  - Each probe only sticks to one or two specific places on the sheet.

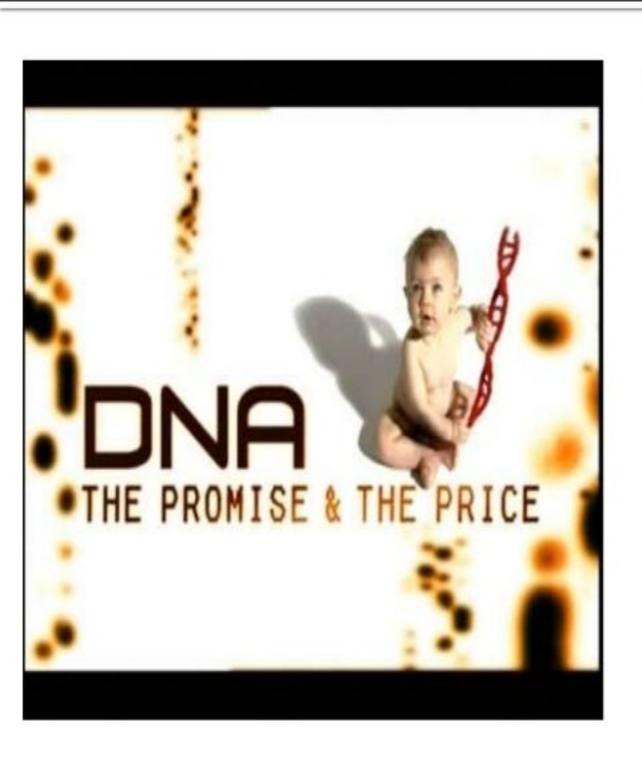
- Step 6: Hybridization
  - labeled probe DNA should be hybridized with the complementary sequences located on nylon membrane for the detection of position of later
  - Membrane is washed to remove non specific binding and clearing of the background



- Step 7: Autoradiography
  - To detect the sequences in genome bound with the hybridized radioactive probe on membrane
  - Technique involves alignment of hybridized membrane with X-ray film
  - The X-ray film alter its development shows multiple no. of bands that looks like bar codes and known as DNA fingerprints.

- Step 8: Interpretation of band patterns
  - Analysis of band patterns of different individuals
  - comparison of position of bans
  - Computer software are also available for for the analysis of DNA fingerprints.

## Uses of DNA Fingerprinting



- Diagnosis of Inherited Disorders
  - Helps diagnose disorders in both prenatal and newborn babies
  - Disorders may include cystic fibrosis, hemophilia, Huntington's disease, familial Alzheimer's, sickle cell anemia, thalassemia, and much more.

### Uses of DNA Fingerprinting

#### Crime

- Forensic science is the use of scientific knowledge in legal situations.
- The DNA profile of each individual is highly specific.
- The chances of two people having <u>exactly</u> the same DNA profile is 30,000 million to 1 (except for identical twins).



#### DNA fingerprinting can solve crimes

- The pattern of the DNA profile is then compared with those of the victim and the suspect.
- If the profile <u>matches</u> the suspect it provides strong evidence that the suspect was present at the crime scene (Note: it does not prove he committed the crime).
- If the profile <u>doesn't</u> match the suspect then that suspect may be eliminated from the enquiry.

#### Example

- A violent murder occurred.
- The forensics team retrieved a blood sample from the crime scene.
- They prepared DNA profiles of the blood sample, the victim and a suspect as follows:

# Was the suspect at the crime scene?

ects Blood s le from cr	 Victims profile

#### Solving Medical Problems

- DNA profiles can be used to determine whether a particular person is the parent of a child.
- A child's paternity (father) and maternity(mother) can be determined.
- This information can be used in
  - Paternity suits
  - Inheritance cases
  - Immigration cases

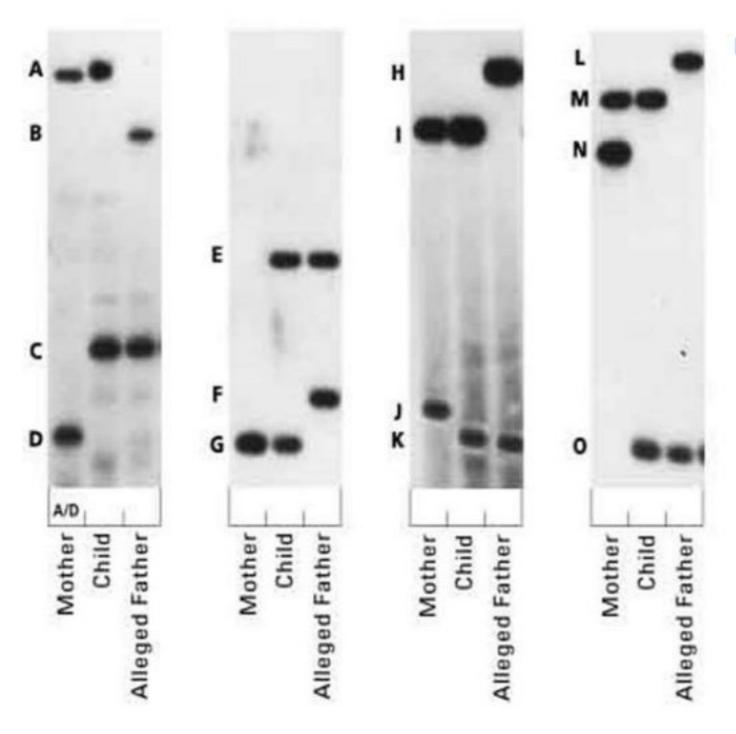
#### Example: A Paternity Test

- By comparing the DNA profile of a mother and her child
- identify DNA fragments in the child which are absent from the mother and must therefore have been inherited from the biological father.

# Is this man the father of the child?

Child	Man
	Child

### Uses of DNA Fingerprinting



- Personal Identification
  - The U.S. armed services are just beginning a program where they collect DNA fingerprints from all personnel for later use, incase they need to identify casualties or missing people.

#### Uses of DNA Fingerprinting

- Some other uses of DNA fingerprinting
  - Identification of carcass of tissues
  - Detection of somatic mutations or cancer
  - Pathogen identification
  - Detection of loci controlling quantitative traits or disease resistance
  - Sex determination
  - Individual identification

#### Famous cases

In 2002 Elizabeth Hurley used DNA Fingerprinting to prove that Steve Bing was the father of her child Damien



#### Famous Cases

- Colin Pitchfork was the first criminal caught based on DNA fingerprinting evidence.
- He was arrested in 1986 for the rape and murder of two girls and was sentenced in 1988.





