

Department of Medical Laboratories Techniques Human genetic Lab.8: DNA Fingerprinting M.Sc. Mazin E. Hadi



DNA FINGERPRINTING

Introduction :

Every cell of our body contains DNA and approximately 99.9% of DNA is similar between two humans. Only 0.1% difference is responsible to make someone unique (except identical twins) and this 0.1% of DNA plays an important role in DNA Fingerprinting.

As we know, only 3 % of our genome are coded and act for protein synthesis

i.e. called *gene* and rest of 97% are non-coding, repetitive and junk; this junk DNA are utilized to perform DNA fingerprinting. The sequence structure and number of repeats varies between individuals and organisms. On this basis, the DNA print can be prepared.

DNA fingerprinting technique was first developed by British Professor Sir Alec Jeffrey in 1984. He realized that we can detect variations in human DNA, in the form of minisatellites. Jeffrey created the first DNA profile using the restriction digestion length polymorphism.

Definition -

"It is a molecular method to identify an individual or any living organism from their DNA sample by looking at unique patterns in their DNA."

Satellites DNA :-

The satellites DNA are non-coding, repetitive DNA regions, located on telomeres and centromeres. It helps in proper replication, whereas mutation in those sequences causes replication errors.

It is of two types present in human genome based on their repeat sequence nature :-

- 1. Minisatellite
- 2. Microsatellites

Third stage



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Minisatellite DNA sequences are 10 to 60 base pair long and repeated 5 to 50 times in a genome. They are highly variable (polymorphic), unique, and GC-rich sequences. It is generally present in telomeric regions.

Microsatellites are smaller than minisatellites. It contains repeated DNA sequence of 1-6 base pair and 5 to 10 times in a genome. These regions are also hypervariable, non-coding and telomeric.

Process of DNA Fingerprinting :-

Restriction fragment length polymorphism (RFLP) and polymerase chain reaction (PCR) amplification of short tandem repeats (STRs) are two main DNA tests widely used for DNA fingerprinting.

- 1- The first step of DNA fingerprinting was to extract DNA from a sample of human material, usually blood.
- 2- Molecular 'scissors', called restriction enzymes, were used to cut the DNA. This resulted in thousands of pieces of DNA with a variety of different lengths.
- 3- These pieces of DNA were then separated according to size by a process called gel electrophoresis.
- 4- The shorter pieces of DNA moved through the gel easiest and therefore fastest.It is more difficult for the longer pieces of DNA to move through the gel so they travelled slower.
- 5- As a result, by the time the electric current was switch off, the DNA pieces had been separated in order of size. The smallest DNA molecules were furthest away from where the original sample was loaded on to the gel.

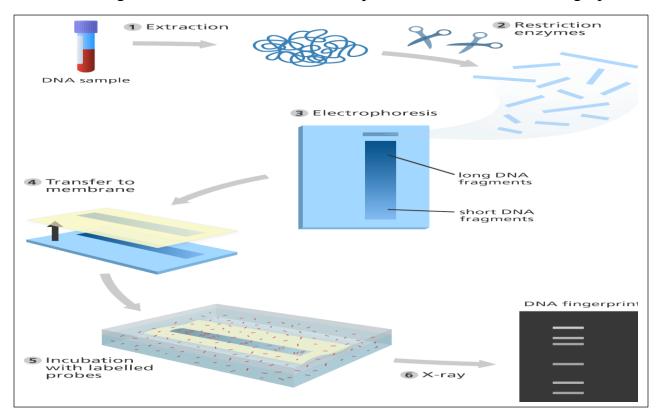


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RFLP analysis :-

RFLP is the first method used for DNA fingerprinting. It has been performed with restriction enzymes, which is used to cut the DNA. This resulted in thousands of pieces of DNA with a variety of different lengths. With the help of agarose gel electrophoresis these DNA has been separated on the basis of their size. Further the piece of DNA has been transferred to nylon membrane. The nylon membrane was incubated with radioactive probes (Probes are small fragments of minisatellite DNA tagged with radioactive phosphorous). The probes only attach to the complementary pieces of DNA and here, they attached to the minisatellites in the genome. The minisatellites attached with probes were then visualized by exposing the nylon membrane to X-ray film. When exposed to radioactivity a pattern of dark bands appeared on the film at the sight of the labelled DNA. This pattern is called the DNA fingerprint.



Third stage

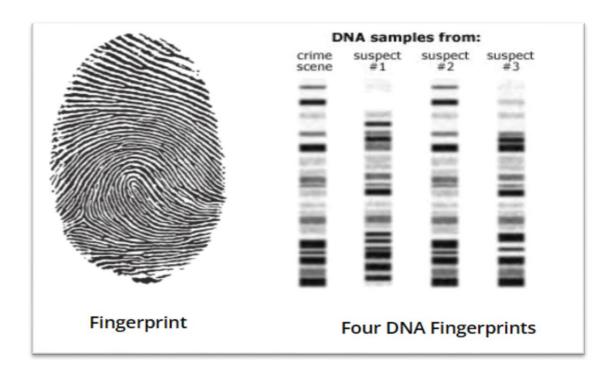




Applications of DNA fingerprinting

DNA fingerprinting finds its applications in various medical tests, investigations, forensic laboratory and many more.

- DNA fingerprinting is widely used to find out the real biological parent or offspring in paternity-maternity disputes.
- DNA fingerprinting is a sure and best method to identify the real culprit involved in crimes, such as murder, rape etc.
- DNA fingerprinting is also used to find to the original background and historical migration of a particular racial group.
- DNA also fingerprinting its application in gene therapy as it helps to provide information regarding some specific alleles.



Third stage