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DNA Testing in Forensic Science

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Abstract

DNA is the hereditary material found in all cells of a human being. The genetic information is passed on through the processes of transcription and translation that result in the formation of proteins. DNA testing is a powerful tool for forensic analysis. DNA can be extracted from any biological material exchanged during a crime scene and analyzed through various techniques. Restriction fragment length polymorphism is one the oldest DNA analysis techniques that produces unique DNA fragment pattern for every individual. However, this technique requires large amount of DNA and was replaced with more efficient methods like polymerase chain reaction that works with even very small amount of sample DNA. New techniques like short tandem repeat analysis have even more discriminating power than simple PCR analysis. Forensic DNA testing has improved a lot over the past few decades and is expected to improve further as the technology is still evolving to provide faster and discriminating results.

Keywords: DNA testing, transcription, translation, RFLP, PCR, STR, forensic science

Introduction

Deoxyribonucleic acid (DNA) is the molecule that contains information required for development, reproduction and survival of an organism. DNA is present in many viruses and all prokaryotic and eukaryotic cells. The chemical nature of DNA was discovered in 1869 by Frederich Miescher while its role in genetic inheritance was found in 1943. Using the discoveries of Rosalind Franklin and Maurice Wilkins, James Watson and Francis Crick revealed the structure of DNA in 1953. They found that is a double stranded helix made up of nucleotides where each nucleotide contained a deoxyribose sugar, one of the four nitrogenous bases and a phosphate group. The nitrogenous bases include pyrimidines i.e. thymine and cytosine and purines i.e. adenine and guanine. The two strands of DNA are held together by the hydrogen bonds between nitrogenous bases (thymine bonds with adenine and guanine bonds with cytosine) and the strands twist around each other to form a stable configuration (Portin, 2014).

DNA is a self-replicating molecule and can from its copies using the replication machinery. During replication, the two strands of DNA separate and one strand guides for the synthesis of a new strand. A segment of the DNA containing several nucleotides that codes for a particular protein is called gene. The process that allows the synthesis of a ribonucleic acid (RNA) from the DNA is called transcription. Transcription takes place in the nucleus of eukaryotic cells. During transcription, the double strand of DNA opens up and RNA polymerase uses one strand of DNA as template to form a new strand of pre-mRNA. The pre-mRNA which is a single stranded copy of the gene undergoes several modifications to form mature mRNA. The process in which a protein is formed from the RNA is called translation (Figure 1). In eukaryotic cells, translation takes place in the cytoplasm. During translation, mature mRNA is used as a template. A group of three bases on mRNA makes a “codon” and each codon codes for a particular amino acid. A ribosome binds the mRNA and makes a strand of amino acids based on codon sequences on the mRNA (Ingloia, 2016).

This chain of amino acids forms a protein that performs its functions in the cell (Figure 1).

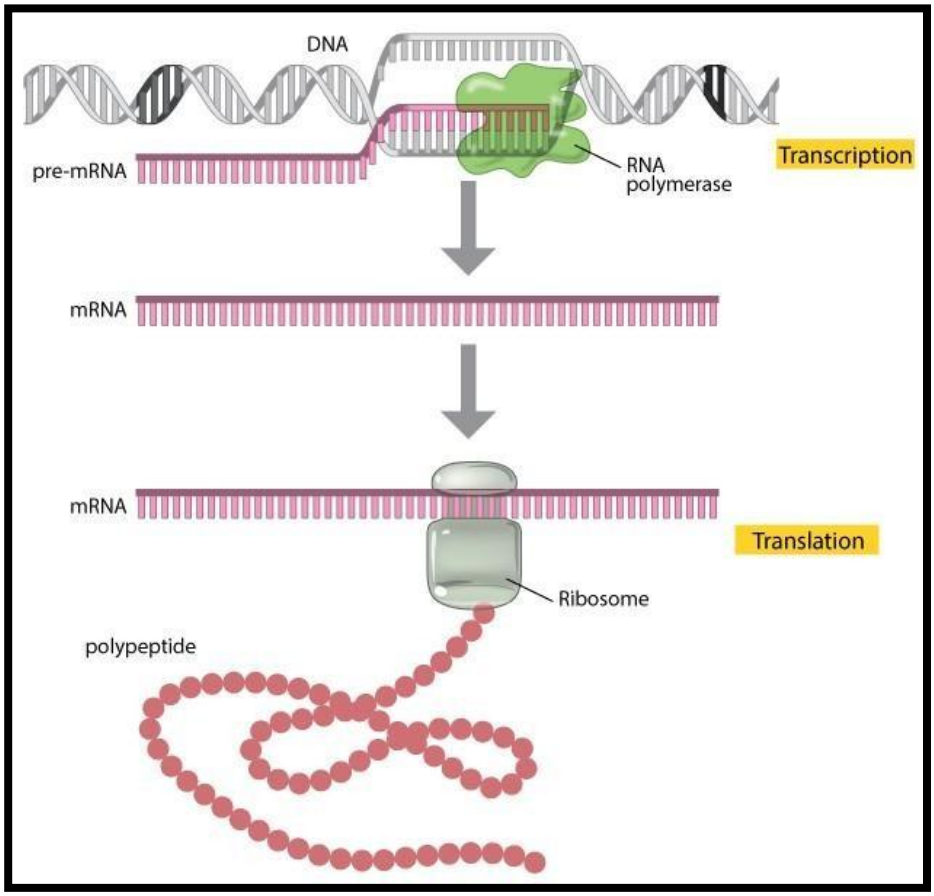

Figure 1. A gene is expressed when mRNA is synthesized from DNA (transcription) followed by synthesis of protein from the mRNA (translation) (Adapted from Clancy and Brown, 2008)

Aim

This review aims to provide brief and simple overview about the importance of DNA testing in forensic science.

DNA Testing in Forensic

DNA can be easily and efficiently isolated from small and complex samples thereby providing criminal justice in many cases. Any biological samples exchanged during a crime scene can be used to identify the DNA and find the culprit. DNA profile of every individual is different from the other except identical twins who needed to be distinguished through further genetic tools. DNA testing was introduced in mid 1980s and plays a significant role in forensic testing. In 1985, an English geneticist Alec Jeffreys discovered DNA typing or DNA fingerprinting when he found that a number of repeated sections in the DNA were different from individual to individual. DNA testing is not only useful for crime scenes where a culprit is involved but also for identifying the disaster victims. For instance, DNA samples of parents can be used to identify an otherwise unidentifiable victim (Butler, 2015).

Techniques used in DNA Testing

The scientific procedure of DNA testing involves collection of samples from crime scenes, disasters or paternity investigation centers. Samples are taken to the laboratories where DNA is extracted and quantified. DNA is then subjected to various methods of DNA analysis followed by determination of sample genotype. The genotypes of question samples are compared with the genotypes of culprit samples and if the genotypes match, profiles are added to population databases and reports are prepared.

DNA sample collection and extraction is only the starting point of forensic DNA testing. The accuracy and success of a DNA test lies in efficient DNA analysis that is performed with various techniques like restriction fragment length polymorphism (RFLP), short tandem repeat (STR) analysis, Y-chromosome analysis, polymerase chain reaction (PCR), mitochondrial DNA (mtDNA) analysis etc. (Sakari et al., 2015).

RFLP Analysis

The first technique used for DNA analysis was RFLP analysis. In this method DNA is extracted from samples (e.g. blood samples) and subjected to restriction digestion with restriction endonucleases. Restriction endonucleases chop the DNA at specific recognition sites and the fragments of DNA are analyzed on the electrophoresis. DNA fragments are then transferred to membrane (Southern blotting) and hybridized with radioactive labeled probes. Extra probe is washed off the membrane and the membrane is sandwiched with X-ray film to see the pattern of DNA fragments. This pattern of test sample is compared with that of suspects to identify the culprit. For instance, in Figure 2, the pattern of sample matched with the pattern of John but not with the pattern of Bill, thereby identifying John as the culprit.

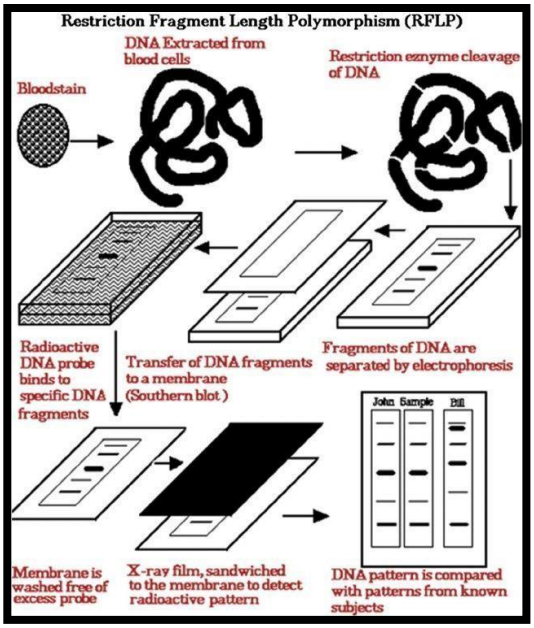

Figure 2. The process of restriction fragment length polymorphism by obtaining DNA sample from blood stain (Adapted from Bluth and Bluth, 2013)

Various modifications, like the use of DNA binding dyes like ethidium bromide to compare the fragment pattern or use of fluorescent probes instead of radioactive probes, have been made to the RFLP technique.

RFLP is a simple method yet it is efficient for forensic testing like crime scene investigation or determining paternity. However, RFLP requires large DNA samples that are often hard to collect from crime scenes. Additionally, this method is laborious, time consuming and hard to automate. RFLP does not work well with degraded DNA as well (Adane, Gebreyohannes and Gebreyohannes, 2016).

PCR Analysis

Polymerase chain reaction allows the amplification of a small amount of DNA from crime scene to create millions of copies that can be compared with the samples of suspects. The DNA samples are mixed with *Taq* polymerase, dNTPs, buffers and suitable primers and subjected to PCR amplification in thermal cycler. The amplified samples are run on electrophoresis gel and test sample is compared with suspects as shown in Figure 3.

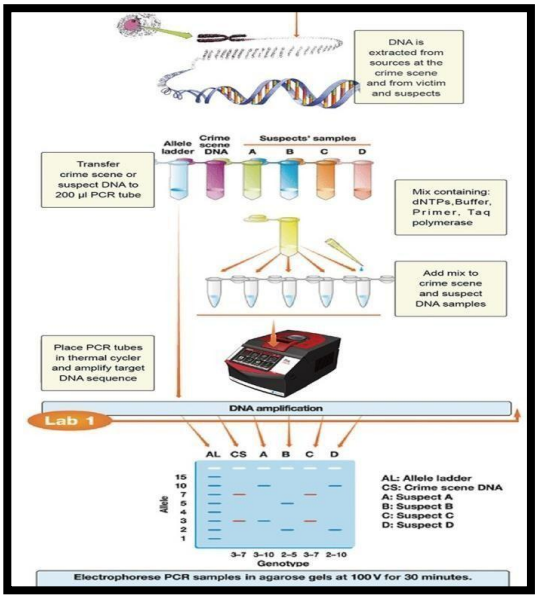

Figure 3. The process of forensic DNA analysis through PCR (Heal Force, 2017)

PCR has an advantage over RFLP that it be conducted with small amount of DNA. Additionally, PCR can be automated and time efficient for a large number of samples. However,

PCR analysis results are not as discriminating as RFLP analysis and it is difficult to analyze mixed or contaminated DNA samples like vaginal swab of the victim after sexual assault. PCR inhibitors or degraded DNA in samples from crime scene can also be a major challenge in PCR analysis. Small target DNA producing small PCR amplicons help to extract information from the degraded samples of DNA (Chaitanya et al. 2015).

STR Analysis

STRs are 2-13 nucleotides on the DNA strand that are repeated several hundreds of time. STR analysis uses STR specific primers to amplify the repeating units through PCR and explores the exact number of these repeating units which are different in every individual. The chance of two people having the same 13 STR regions is almost impossible. STR based DNA testing is commonly used nowadays in various countries. For instance, in UK 17 loci system (DNA-17) is used while in North America, Combined DNA index System (CODIS) is used.

Only 1 ng of DNA is enough to produce good STR results. Therefore, STR analysis is efficient for forensic samples which often contain degraded DNA (Norrgard, 2008). STR analysis can also be automated that reduces the time of analyzing multiple samples. For instance, the cost and time of STR analysis is significantly reduced due to the use of multiplex PCR reactions (Nastainczyk et al. 2009). The actual advantage of STR for forensic DNA testing is its statistical power of discrimination since there is almost one in billion chance that STR results of two people match with each other. However, if the samples are contaminated, it is probable that profile of most common sample will match with the test sample. Therefore, clean equipment, chemicals, buffers and appropriate controls are used for every test to ensure correct results (Cowell et al. 2015). Also, complete profiles are often compared to remove any doubts.

Conclusion

In a nutshell, DNA testing is a powerful tool for solving various forensic cases. Presence of DNA in every eukaryotic cell allows it to be extracted from any biological material exchanged during crime scene. Older techniques like RFLP and PCR have been continuously been improved and replaced with better techniques like STR analysis. Time saving and less laborious techniques for DNA techniques help to analyze large number of samples in cost effective manner. However, contaminated DNA samples may still pose problem for accurate criminal DNA testing. Human genome sequencing projects like next generation sequencing may provide in depth information for detailed analyses. With the advancement of technology, DNA testing in forensic science will further evolve and provide more discriminating results for forensic analysis.

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