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Contemporary Applications of Next Generation Sequencing in Forensic Science: A Brief Note

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Abbreviations: NGS: Next-Generation Sequencing; STRS: Short Tandem Repeats; RFLP: Restriction Fragment Length Polymorphism; NGS: Sequencing Techniques; AIMS: Ancestry Informative Markers;

Summary

Over the past few decades, investigations for medicinal and therapeutic purposes have looked into the potential of microbial diversity. However, its application in forensics is expanding because of its efficacy in situations when conventional procedures fall short of generating a solid conclusion or are unable to do so [1]. The use of the human microbiome may be used to identify the kinds of stains in salivary and bodily secretions as well as to link certain stains to specific people. Additionally, because microbes affect the decomposition process, that might be utilized to estimate the time since death. This is like how the microbiome composition of a soil sample can be used to determine geographic origin or link people, animals, or objects with a particular location. Next-generation sequencing (NGS) technology, which appeared ten years before, had completely changed the field of genetic research [2]. These contributions have shown how new opportunities for forensic genomic case work are provided by NGS. Markers such as short tandem repeats (STRs), Single Nucleotide Polymorphisms (SNPs), restriction fragment length polymorphism (RFLP), insertion/deletions, mRNA are the combinations for analyzing, which cannot be evaluated concurrently with the current conventional PCR-CE procedures that could allow the scientists to learn more from distinct samples in a single experiment [3]. In this brief note, we will succinctly describe all sequencing techniques NGS, especially shotgun metagenomic sequencing, and address the potential uses of NGS in forensic genetics.

Development of sequencing technique's

Forensic scientists now have access to more pertinent data to support probability assignments, as well as knowledge of the transmission, persistence, prevalence, and recovery of DNA, which is important for helping with investigations into suspected criminal activity and guiding factfinder's [4]. Forensic genetics has a long history with DNA sequencing. In the late 20th century, mitochondrial DNA (mt DNA) was assessed in sequencing technique and utilized for an investigation during a period when RFLP examination became a circumstance for human identity and years prior to the first STR assays were created. With the Sanger di-deoxynucleotide (dd NTP) chain termination technique, sequencing was done by stopping a developing DNA chain from being extended by the DNA polymerase [5].

In 1996, pyrosequencing was proposed as a Sanger sequencing substitute for real-time sequencing. A sequence of enzymatic processes that involves 3 enzymes (Apyrase, Luciferase, and ATP Sulfurylase,) and utilizes the pyrophosphate that is produced to create light during the DNA synthesis process, which involves sequential additions of nucleotides. CCD camera was utilized to detect the light in real-time in which the output of the sequencing process does not need to be electrophoresed. Pyrosequencing was used for mt DNA sequencing because it was less expensive and quicker than Sanger sequencing [6]. Even though the first pyrosequencing apparatus never really took off in science, the real-time sequencing idea and the pyrosequencing technology ultimately served as the foundation for the present advancement in DNA sequencing.

Pyrosequencing was used by the genome sequencer 20 from 454 Life Sciences, the first commercialized high-throughput sequencing system [7]. In this report, we will just provide a brief introduction to high throughput sequencing and concentrate on

potential uses of NGS in forensic genetics. Some NGS systems have so many features that their main purpose is to perform sequencing tests that can identify every dsDNA molecule present in the sample material [8]. Shotgun sequencing, often referred to as sequencing without targeting, involves shattering µg of DNA into minuscule fragments of 50–500 bp either mechanically, by enzymatic absorption, or by randomly adding transposons. Shotgun sequencing or RNA sequencing is the best technique to analyze cDNA containing samples, may produce a gene expression outline of the sample [9,10].

NGS role in forensic science

Full genome sequencing appears exorbitant in many forensic genetic situations, where the main objectives are to identify the individual who provided the sample, may be predict any inherited physical characteristics of the person, or determine the specific cell types in the sample. This just involves a limited indicators, and an acquisition depended method will be far further affordable and needed fewer samples [11]. X chromosome STRs, Y chromosome STRs, Autosomal STRs, mt DNA SNPs, autosomal SNPs, phenotypic markers, ancestry informative markers (AIMs), Y chromosome SNPs, mRNA, insertion, and deletion other markers are now tested separately (Figure 1). Aside from the essential forensic markers, which are categorized by PCR-CE. NGS takes two to three days on average, whereas PCR-CE could be accomplished in a one-time work. However, if an acquire for the appropriate genomic region can be designed, all (or the majority) of the PCR-CE analysis might be integrated into a single NGS analysis.

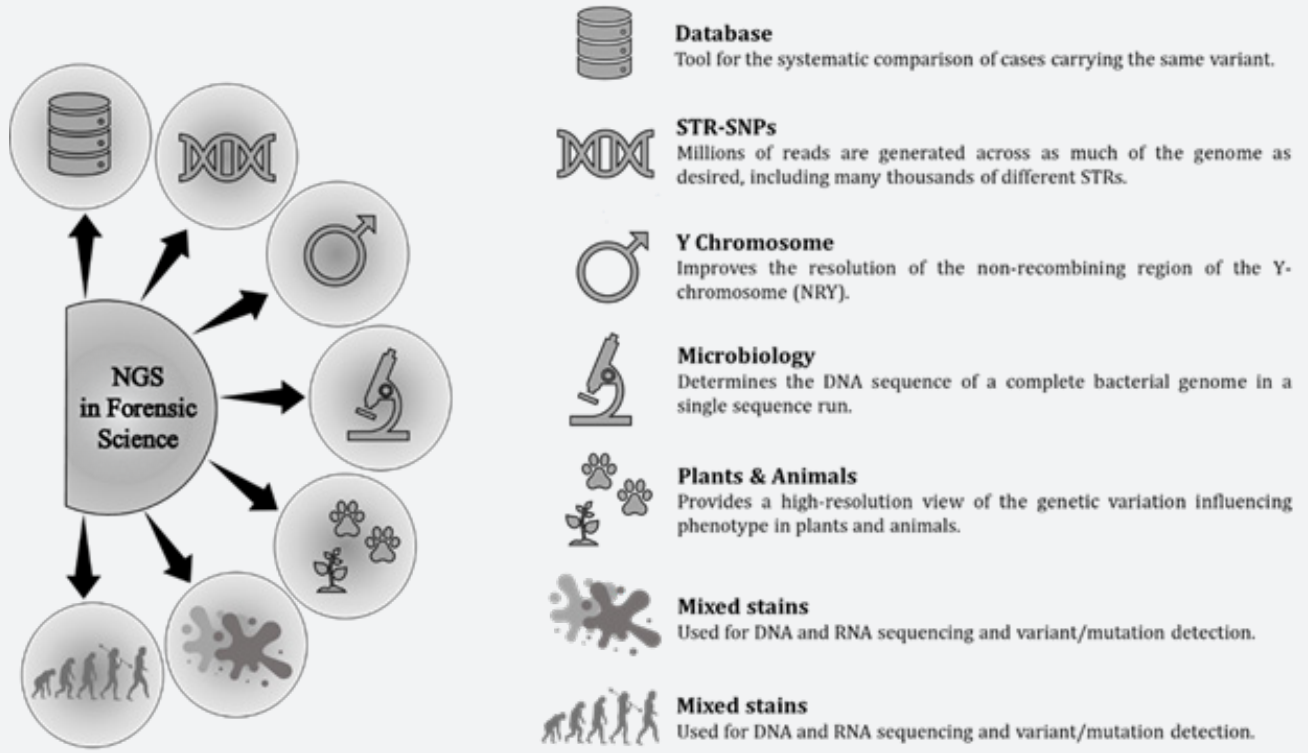

Figure 1: VARIOUS APPLICATIONS OF NGS IN FORENSIC SCIENCE.

In situations when additional studies are required, one NGS test with several distinct markers will save time and shorten the total period as evidence is evaluated in the research laboratory [12,13]. These comments are fascinating, however forensic genetics faces numerous difficulties and must consult in which major issues, such as how many and which genomic region to examine, what to communicate, and if it is sensible to ignore sequence information. There will also be many ethical and legal issues to consider when introducing NGS in forensic genetics. Hence, techniques are inevitable in the field of forensic science to analyze evidence such as DNA, phenotypic studies, identical twin's studies, species identification, plant and microbiology research.

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