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DEOXYRIBONUCLEIC ACID AS A PROBLEM-SOLVING TOOL

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ABSTRACT

DNA or Deoxyribonucleic acid is the molecule of life, it is the chemical code specifying our function, appearance and Pedigree and is unique for all individuals except identical twins. An individual's DNA is formed by combination of Deoxyribonucleic acid from his or her parents with half coming from the mother and her from the father. For this reason, Deoxyribonucleic acid testing can be used as evidence of paternity of a child. Deoxyribonucleic acid is found in most cells of the body, including hair roots, Semen, white blood cells and body tissue. Traces of Deoxyribonucleic acid can be detected in body Fluids such as saliva and perspiration. Mitochondrial Deoxyribonucleic acid, which follows the maternal line of an individual, can be extracted from hair and bone samples. This can be used to examine relatedness and common ancestry between individuals and to verify the identity of buried remains. This technique was used in the much-publicized case of the Romanovs. The techniques that are applied in identity testing are Deoxyribonucleic acid profiling, DNA fingerprinting and DNA typing. Although there are some technical differences between these tests. Deoxyribonucleic acid analysis is the key to linking suspects to biological evidence and to identifying individuals in crimes and disasters. An important use of Deoxyribonucleic acid analysis is the establishment of paternity in custody and child support litigation. Deoxyribonucleic acid profiling is also used to diagnose inherited disorders and human diseases. The Present paper is a review on this.

Key Words- Deoxyribonucleic acid, DNA profiling techniques

INTRODUCTION

Forensic science utilizes the properties of deoxyribonucleic acid in several ways. The old adage every contact leaves a trace indicates the importance of a technique able to type trace amounts of genetic material left during the commission of a crime. Saliva or hairs left on a balaclava worn during a robbery, semen located at a rape scene, blood collected from an assault, perspiration on clothing, traces of assailant's skin under a victim's fingernails, can often be deoxyribonucleic acid profiled. This genetic information can then be used to include or exclude suspects as being the source of the genetic material. It is not possible to test the whole of an individual's deoxyribonucleic acid. Forensic analysis involves the testing of regions of an individual's deoxyribonucleic acid. Database have been compiled which list the abundance of a particular fragment of deoxyribonucleic acid in the population. From this information, an estimate of the abundance of combinations of DNA at several regions can be made and compared to the deoxyribonucleic acid of victims or suspects. In this way and individual can be included or excluded as a possible source of DNA found in relation to a criminal investigation.[1,2] Statistical interpretation of information can be made to estimate the possibility of material coming from a particular individual relative to coming from a random member of the population.

The Structure of Deoxyribonucleic acid

The deoxyribonucleic acid molecule is a vast ladder in which the vertical pieces consist of alternating sugar molecules and phosphate group, and the rungs are complementary bases. The four major bases available for the deoxyribonucleotides (A) adenine, (T) thymine, (G)

guanine and (C) cytosine in (figure1). The backbone of the molecule consists of phosphodiester bonds connecting the 3' hydroxyl of the deoxyribonucleotide to the 5' hydroxyl of the next sugar. Two backbone chains running in opposite directions, are paired complementarily by the interaction of a purine (A or G) with a pyrimidine (T or C). The pairing of a purine and pyrimidine results in a regular structure. A can only bond with T and G can only bond with C (figure2) due to steric hindrance, so if the structure of one side of the chain is known, the other can be determined. Deoxyribonucleic acid is a template for reproduction of genetic material and cellular information. The molecule splits along its central axis, providing access to one side of the complementary chain. Enzymes then add the appropriate bases to each chain, giving a new exact copy of the complementary sequence. This ability of deoxyribonucleic acid to repair and replicate itself is exploited in the DNA profiling techniques used in forensic science.

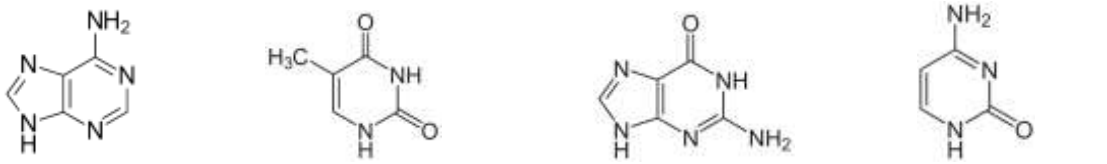
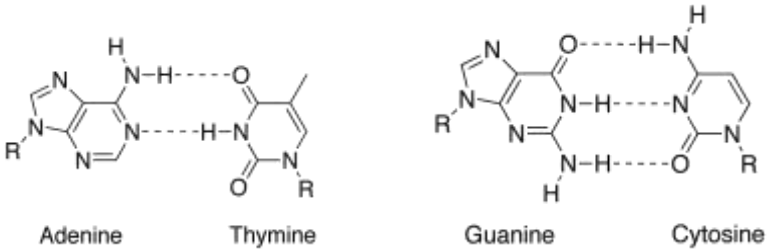


Figure 1



Pairing of a purine and pyrimidine

Figure 2

The Human Genome

In man the nuclear DNA comprises 46 long molecules, each of which forms the genetic backbone of a chromosome. The 46 chromosomes of each cell consist of 23 pairs, one chromosome from each pair having been inherited from the individual's mother the other from the father. This is the basic of Mendel's universal first law of inheritance and means that it is possible by DNA analysis to trace the biological relations within a family, including the segregation of disease genes. One pair of the 23 pairs of chromosomes, the sex chromosomes, differ between the two sexes: two X chromosomes (XX) in the female and X and Y (XY) in the male. Analysis of blood stains and tissue samples for the presence of Y chromosome DNA can therefore be used identify the sex of the donor. The remaining 22 pairs of chromosomes are called autosomes. It is estimated that the human nuclear genome has between 50,000 and 100,000 genes which codes for the many different proteins of the organism. however, only a minor part of the nuclear DNA contains such coding sequence. The non-coding sequences are located within individual genes, forming intervening sequences or introns, as well as between genes. The DNA sequences used in DNA profiling belong to the non-coding sequences. The complexity and variation of genetic material means that each human being, other than monozygous individuals (identical twins, triplets etc.) has a unique genome and thus a personal DNA profile.[3,4]

Extraction of DNA

Before a DNA profile can be prepared from a human sample the DNA must be isolated from other organic and non-organic component of the sample. The type of sample will determine the particular isolation technique used. Isolation techniques may involve the use of enzymes to break down proteins and other cellular material; chelating agents to ensure that non-organic material does not degrade the DNA; and organic solvents to separate the DNA from the other organic and non- organic material where necessary. Once the DNA has been extracted from the sample, a number of different techniques can be used to develop a DNA profile. Techniques for developing profiles are constantly improving or being superseded.

Sources of DNA Evidence president's DNA Initiative (2007)

Evidence	Possible Location of DNA on the Evidence	Source of DNA
baseball bat or similar weapon	Handle, end	sweat, skin, blood, tissue
hat, bandanna, or mask	Inside	sweat, hair, dandruff
Eyeglasses	Nose or ear pieces, lens	sweat, skin
facial tissue, cotton swab	Surface area	mucus, blood, sweat, semen, ear wax
dirty laundry	Surface area	blood, sweat, semen
Toothpick	Tips	saliva
used cigarette	Cigarette butt	saliva
stamp or envelope	Licked area	saliva
tape or ligature	Inside/outside surface	skin, sweat
bottle, can, or glass	Sides, mouthpiece	saliva, sweat
used condom	Inside/outside surface	semen, vaginal or rectal cells
blanket, pillow, sheet	surface area	sweat, hair, semen, urine, saliva
"Through and through" bullet	outside surface	blood, tissue
bite mark	person's skin or clothing	saliva
fingernail, partial fingernail	Scrapings	blood, sweat, tissue

All biological evidence is subject to deterioration. The careful collection and storage of biological evidence will help ensure that this evidence is preserved so that useful information can be obtained from the analysis. Most deoxyribonucleic acid typing methods are robust, and dirt, grease, some dyes in fabrics, and other substances can seriously compromise the deoxyribonucleic acid typing process. Environmental insults will not change DNA allele "A" into allele "B", but they can adversely affect the ability of the scientist to obtain a complete deoxyribonucleic acid profile from the sample[5,6,7,8]

DNA profiling techniques

Restriction Fragment length polymorphism (RFLP)

This technique is outlined double stranded deoxyribonucleic acid is extracted from Blood or semen. The deoxyribonucleic acid is cut into small pieces by a sequence - specific enzyme i.e., an enzyme that cuts the deoxyribonucleic acid wherever a particular sequence of bases occurs. The fragments are then separated out by a process called electrophoresis. The sample is put at one end of a bath of a jelly-like substance called agarose gel and a voltage is applied. The fragment is charged, and the voltage is applied in such a way as to encourage the fragment to migrate to the other end of the gel. Small fragments move much faster than large ones so separation on the basis of molecular weight occurs. After electrophoresis the

gel and fragments are exposed to 0.25M HCl to depurinate the deoxyribonucleic acid and nick the sugar phosphate backbone as this assist in fragment in transfer. The depurinated sites are then cleaved by washing in NaOH/ NaCl. The denatured deoxyribonucleic acid is then transferred to a nylon membrane and the variable minisatellite region of the deoxyribonucleic acid examined by ^{32}P- radiolabelled short pieces of single stranded deoxyribonucleic acid called probes binds to its complementary on the membrane. The radiolabelled membrane is exposed to film produce an autoradiograph. Successive regions of the deoxyribonucleic acid are examined. The distribution of each of the probed regions of deoxyribonucleic acid within the population is estimated from a population database to give an indication of the probability that sample comes from a given suspect.

Short tandem repeat profiling (STR'S)

Short tandem repeat profiling (STR'S) [9,12] is the new generation of DNA profiling. In this technique microsatellite repeating regions are examined. The technique is based on the polymerase chain reaction - the cycle of reactions in which DNA is split replicated then split again for replication. This amplification gives an exponential increase in the number of copies of the original template. The reaction is under Kinetic control, reaching a plateau which is dependent on competition, the ultimate inactivation of the catalytic enzyme and the original number of template molecules, primer and dNTPs deoxynucleotide triphosphates - the building block of DNA. The deoxyribonucleic acid is denatured split at approximately 94^0 centigrade. Short strands of deoxyribonucleic acid, called primers, attached to the target deoxyribonucleic acid at a specific site. Bases are added enzymatically to the end of the primers to form a new complementary strand. Approximately thirty such cycles are carried out to produce many copies the original material. Since the amount of original material is increased; this technique is particularly suited to the analysis of trace amount of deoxyribonucleic acid. The amplified deoxyribonucleic acid is separated by electrophoresis through an ultra-thin denaturing polyacrylamide gel. This technique can be performed manually with repeats visualised using a silver staining or automatically, with multiple loci visualised simultaneously using fluorescent dyes. The number of repeats for a particular individual is determined at several loci. The manual method currently in use examines three loci with an extra male /female sex test. The automated currently examines four loci. Statistical analysis on the abundance of the observed patterns in the population is carried out.

Microchip Technology

Akin to the microchip technology[12,13] which has been the backbone in the area of personal computers, micro - fluidic systems are the very basis of micro electro mechanical system devices in the domin of "Laboratories on a chip." The basic Molecular biology techniques of electrophoresis, thermal cycling and hybridization can be accomplished by microchip formats. microchannel capillary electrophoresis is a miniature version of the currently practiced capillary electrophoresis. Exceedingly small channel lengths are sufficient for detectable separation of DNA fragments. STR determinations have been reliably performed in millisecond electrophoretic runs. Multiple channels can be placed on a single microchip. PCR and capillary electrophoresis can be integrated on a single chip. PE biosystems, Nanogen technologies and caliper, as well as a number of Institutes in USA and Europe such as the whitehead Institute are vigorously pursuing R & D to develop a microchip-based capillary electrophoresis suitable for human identification. forensic analysis is bound to be immensely benefited by the success of these efforts. DNA typing of biological material in the

field will then be a matter of a few minutes and could be performed without any academic expertise in molecular biology.

Applications

DNA profiling is a highly conclusive, informative procedure in identification of biological specimens, genomic diversity in population studies, characterization and tracing antiquity of ancient DNA and for diagnosis of a plethora of diseases. The important forensic applications are in

medico-legal issues

Murder Assailant, victim or the crime scene can be positively identified from very small amount of the tissue left behind with the victim on the scene of crime or on the personal belongings of the accused.[10,11]

Sexual Assault

Rapist can be positively identified from semen on the body or apparel of the victim or from the scene of crime.

Accidents, Mass Disasters

identification of mutilated bodies can be done by comparing the DNA profile of biological remnants with blood relatives.

Parentage Disputes

parentage of a child can be established by comparing the DNA of the child with putative mother and /or father. when one of the parents is not available for the testing, his /her DNA made up can be reconstituted from the DNA of close relatives for identifying potential suspects who DNA May match with the evidence left at crime scenes.

Animal and Plant genomics

Pedigree determination of seed or for livestock breeds. Identification of plant and animal species under in Intellectual Property rights. Seed certification and quarantine programmes.

Anthropology and evolutionary studies

Ancient DNA analysis for addressing the origin of modern humans' migration and evolution. Characterisation of archaic fossil remains for identifying animal and plant species. Understanding genetic structure, variation and affinities of populations.

Wildlife Forensics

identification of protected flora and fauna thus aiding wildlife forensics for conservation and management of endangered species could be used for prosecuting poachers.

Food Technology

Authentication of consumables such as caviar and wine, thus maintaining quality assurance and control.

CONCLUSION

DNA profiling technologies are extensively applied in resolving Civil and criminal cases and in supporting various type of Investigation. As techniques for manipulating and analysing DNA become more efficient and more robust, forensic DNA testing will improve further. An additional advantage of DNA testing is the ability to review previous cases that were decided primarily using classical tests. In these instances, DNA techniques can be used to reanalyse material that may provide previously convicted individuals an opportunity for acquittal. The rapid application of DNA techniques in diverse fields such as medical genetics, wildlife forensics, evolutionary Anthropology, plant and animal genomics has yielded significant inferences with remarkable resolution unseen in earlier systems, thus revolutionizing scientific approaches comprehensively.

REFERENCES:

1. Chisum, W.J., & Turvey, B. "Evidence Dynamics: Locard's Exchange Principle & Crime Reconstruction," Journal of Behavioral Profiling, January, 2000, Vol. 1, No. 1
2. Inman, K., Rudin N., (1997) An Introduction to Forensic DNA Analysis, (New York: CRC Press)
3. Lee, H., Ed., (1995) Physical Evidence, (Enfield, CT: Magnani & McCormick, Inc.,)
4. Thornton, J. I., (Ed.) Kirk, P., (1974) Crime Investigation, 2nd Ed., (New York: Wiley & Sons)
5. Lee HC, Ladd C, Bourke MT, Pagliaro E, Tirnady F. (1994) DNA typing in forensic science. Am J Forensic Med Pathol 15:269-82.

6. Bourke MT, Scherczinger CA, Ladd C, Lee HC. (1999) NaOH Treatment to neutralize inhibitors of Taq polymerase. J Forensic Sci 44:1046-50.
7. Adams DE, Presley LA, Baumstark AL, Hensley KW, Hill AL, Anoe KS, et al. (1991) DNA analysis by restriction frag-ment length polymorphisms of blood and other body fluid stains subjected to contamination and environ-mental insults. J Forensic Sci 36:1284-98.
8. Van Oorschot RA, Gutowski SJ, Robinson SL, Hedley JA, Andrews IR. (1996) humtho1validation studies: effects of substrate, environment, and mixtures. J Forensic Sci 41:142-5.
9. Kashyap VK, Sarkar N, Trivedi R (2002b). Genetic diversity at two pentanucleotide STR and thirteen tetranucleotide STR loci by multiplex PCR in four predominant population groups of central India. Forensic Sci Int, 128: 196-201.
10. Goodwin W, Linacre A, Hadi S. Chichester: John Wiley & Sons; (2011). An Introduction to Forensic Genetics.
11. Kayser M: (2015) Forensic DNA phenotyping: predicting human appearance from crime scene material for investigative purposes. Forensic Sci Int Genet; 18: 33–48.
12. Swaminathan, H., Grgicak, C.M., Medard, M., Lun, D.S. (2015) a computational method to infer the number of contributors to DNA samples analyzed by STR genotyping. Forensic Sci. Int. Genet. 16, 172–180
13. Arfat, Y., Aqib, M., Mehmood, R., Albeshri, A., Katib, I., Albogami, N., Alzahrani, A. (2017) Enabling smarter societies through mobile big data fogs and clouds. Procedia Comput. Sci. 109, 1128–1133