

Review Research Paper

Application of Genetics and Molecular Biology In Forensic Odontology

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Abstract

Various methods of Forensic Science have been applied to solve many mysteries of world including criminal investigation. Forensic odontology researches are usually associated to the dentist's field of action by using bite marks, dental records and radiographs plays an important role in solving many crimes. These methods help in age determination and sex identification of the people who have lost their identity after death. Nevertheless, since the development of genetics and molecular biology there were an increase in number and quality of solved cases. The present article emphasizes the importance to associate certain forensic biology areas to traditional investigation methods in human identification, especially with forensic odontology. It also shows that in some situations, teeth are an important source to genetic analysis and molecular studies. Finally it was concluded that knowledge of forensic genetics applied to traditional forensic investigations would produce an increase of information to Justice.

Key Words: Forensic Odontology, Genetics, Human Identification

Introduction:

Forensic odontology researches are usually consists of analysis, expertise and evaluation of dentistry related episodes, but it can also be extended to other areas if it is necessary for justice and administration interests. The most common studied area where a dentist acts as a forensic expert is related to oral maxillo-facial trauma caused by different sources such as mechanical, physical and chemical and dental analysis used for human identification. [1]

Development of genetics in the 80's allowed innovations to medical field as well to forensic sciences. This was due to discovery of specific mini satellite regions of human genome which had the so called DNA (deoxyribonucleic acid) "digital impressions". Its analysis led to human individuality information. From that point, genetics research dealing with DNA polymorphism has had a great development

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This human identification methodology relies upon three important steps: DNA isolation or extraction, amplification of specific DNA regions using PCR technique (Polymerase Chain Reaction) and molecule profiles analysis. [2] Human individuality identification tests using DNA are based upon polymorphism of several mini and micro satellite loci. This polymorphism in autosomal chromosomes is considered a great individuality indicator. First, in order to do the amplification phase highly specific probes were developed which allow simultaneous scanning in several loci of the sample. After the material amplification using PCR technique, they are analysed in agarose or polyacrylamide gel or in automatic sequencer. Depending on the quality and quantity of the obtained sample, analysis can be done from nuclear DNA or mitochondrial (mt DNA), both show advantages and limitations. [3]

Computer aid in several laboratory steps associated to biotechnology and forensic mathematics increase the reliability of examinations to determine sex, age estimation, parenthood and human identification. Therefore it became mandatory that forensic experts in several areas of criminal investigation, forensic medicine and dentistry would associate classical investigation techniques to molecular biology analysis and DNA examination in order to achieve more reliable, objective and specific results facing complex cases.

Expertise examination must show a multi disciplinary approach; therefore the

present paper presents a scientific literature review aiming to point out the importance of forensic genetics used along with traditional investigation methods in human identification, mainly with forensic odontology. It also intends to show the tooth forensic relevance as a source of material to genetic and molecular studies.

Molecular Biology Studies:

Nowadays, Forensic Anthropology has molecular biology as a powerful allied, mainly in species, family background, sex and age investigation. Since that, determination of species using bone, tooth, hair or organic fluid samples was done searching for exclusive human molecular indicators. Those indicators are polymorphic and easily detectable in the population. They can refer to a gene, a restricted site or any other DNA sequence which presents different allelic version to that locus. The mechanisms that explain polymorphism can be single changes in nucleotides (substitution) such as SNP (Single Nucleotide Polymorphism), VNTR (variable number of tandem repeats) and STR (short tandem repeats) and the insertion or exclusion of DNA sequences (Indels), such as Alu insertions. It is important to add that some indicators are specific to a determined population, this happens because the difference of frequency between two big populations is more than 50% (such as Europeans and Africans). Those indicators are named ancestor indicators (AIMs). [4]

In order to increase age estimative precision of decomposed bodies such as bones several researches have been developed trying to relate mainly the racemic mixture of aspartic acid (D/L) from tooth tissues with one's chronological age. It is well known that aspartic acid, particularly in its dextrogen form (D), linearly increases along aging. [5] Other biochemical indicators such as gelatinase A and glutamic acid from dentine are also being researched to be used in age estimation. [6]

Human Identification Using DNA:

Genetic material can be obtained from several biological sources such as body fluids, soft tissues and mineralized tissues. When dealing with living suspects in parenthood test (father, mother and son), genetic material is preferably obtained from either blood (leukocyte) or buccal mucosa.

On the other hand, if it is necessary to verify genetic relation involving pot-mortem material, time from death and corpse condition are relevant factors to choose which method of DNA extraction is to be used. In a recent death case blood, viscera and soft tissue are the first

choice of materials, but as time goes by those sources become inappropriate, leading towards the mineralized tissue sources like bones and teeth. Bones are an important source to obtain genetic material since they are located inside the body and are mineralized; also the cortical part protects the medullary part from external factors and micro-organism that may degrade the DNA. [7]

Teeth as Genetic Material Source:

Teeth also are a good source to obtain genetic material. This is true mainly because of their great tissue resistance (enamel, dentin, cementum and pulp) against external injuries. [8] Pulp tissue is a loose connective tissue and it degrades easily when compared to others dental tissues. Dental pulp is protected by tooth structures and therefore can present better condition than others soft tissue for DNA extraction. Lessing et al [9] showed that pulp can be a source of DNA in teeth that had been kept or obtained in different conditions.

Amelogenin can also be studied from dental material that had been through adverse situations. There are several techniques to obtain dental material to extract DNA such as tooth grinding or crushing, tooth horizontal sectioning, pulp extirpation by tooth irrigation and sectioning and nitrogen liquid cryogenic pulverization. [10]

Some legal precautions must be followed when dealing with dental material as source to obtain DNA since the process destroys the material used in the examination. Those precautions involve proper teeth identification, describing all characteristics and if it is possible, taking pictures and radiographs in the original positions when they are removed from dental arch. Those precautions are done to preserve dental characteristics of evidences in order to prove their genuine value when doubts are raised concerning their origin or the results of the person identity.

After the dental recording and filing phase and before the handling of samples, it is necessary to decontaminate dental surface, Sodium Hypochlorite is substance most used, but it is necessary to have an optimization among concentration, time and applying method of this substance. [11]

Multi Disciplinary Approach in Human Identification:

In Forensic dentistry, DNA examination can be used together with traditional techniques showing great results to identify destroyed or advanced decomposed stage corpses. [12] Besides that, DNA analysis obtained from oral

mucosa cells has great importance in dental impressions, also known as bite marks. Normally, the primary investigation approach of this type of evidence is related to the analysis of dental characteristics left in the victim or in the object. Nevertheless, when those marks do not give a conclusive result, biological material collection from the place where bite was applied is extremely important to find the identity of the person who was responsible for the mark. Among the techniques to obtain DNA from human skin there is the double swab technique which consists of applying a swab with distilled water followed by another dry swab on the bite mark. This technique showed good results to obtain biological material to be researched. [13]

In the Borgula et al [14] experimental research was demonstrated that is possible to analyse the genotype of specific bacteria found in the oral cavity (*Streptococcus*) of individuals as an alternative when it is not possible to obtain the DNA of the one who caused the bite mark.

In more complex cases such as presented by Bilge et al [15] a multi field approach investigation was necessary to identify a corpse. Anthropologic techniques, forensic dentistry, computer superposition (face/cranium) and DNA investigation were used. Sex was determined by cranium characteristics and as well by amelogenin analysis. Age was estimated using longitudinal divided crown measurements and computer superposition showed a positive identification between victim facial structures and the found head.

DNA was extracted from dental pulp, bones, muscle tissues and compared to genetic profile of the victim's presumed daughter and wife. Fatherhood indication was verified in 11 examined loci. In another case, Sweet et al [16] presented an identification of human parts from a woman that had been disappeared for 3 years. Investigations showed that the presumed victim had 3 smear cell laminas in the laboratory files. DNA was extracted and compared to genetic profile obtained from the dental sample of the found corpse. The result was positive, showing coincidence in 8 of the 8 examined loci, including amelogenin.

In the identification of charred bodies, the great resistance of mineralized tissues allows the victims to be identified not only by DNA extracted from bone material [17], but also from dental material. Sweet et al [18] showed an identification of a homicide victim that had been

charred with fuel. DNA was obtained from dental pulp extracted from intra osseous third molar.

Facing an overwhelming result presented in the forensic literature and from the even more reliable techniques, DNA examinations and Molecular Biology analysis became an essential tool to help or solve investigation matters that had been considered irresolvable in crime investigation and Forensic medicine. Therefore it is mandatory that those who are in forensic investigations must acquire knowledge about forensic genetics to provide greater justice and benefit the society.

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