Review Article

Truth from Untruth: Dental Pulp and Its Role in Forensic Odontology – A Retrospective Review

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Abstract

Forensic identification by its nature is a multi-disciplinary approach relying on positive identification methodology. This branch dealing with the identification of the deceased has many maxims, the best known of which, is that every contact leaves its trace. The identification of dental remains are of primary importance when the deceased person is skeletonized, decomposed, burned, or dismembered. A google literature search was done on various studies done using dental pulp in forensic odontology. Based on the available data, the details were analysed and reviewed. Pulp plays a pivital role in forensic odontology. Pulpal tissue can be used for molecular analysis to determine Age, Sex and Blood group antigen. Apart from these, the extracted DNA from Pulp can be used for Personal Identification. Odontoblasts present in pulp can be used to assess age as well as the days of death. To conclude Dental pulp has a high potential value in forensic odontology.

Key Words: *Age estimation, blood group antigen, dental pulp, forensic odontology, sex determination*

Introduction

Over the past 100 years, dental and orofacial findings play a vital role in the identification of deceased individuals through the comparison of antemortem and postmortem records.[1] Dr. Oscar Amoedo is regarded as the father of forensic dentistry.[2] The Federation Dentaire Internationale defines forensic odontology as "that branch of dentistry which, in the interest of justice, deals with the proper handling and examination of dental evidence and the proper evaluation and presentation of dental findings."[3] Dental tissues have the ability to withstand major assaults and have the significant feature of retaining some of its original structure.[4]

Historical View

As we all know, the use of teeth as evidence in the court of law is not recent. The history of forensic odontology backdates to 49 A.D., where Nero's mistress, Sabina, could be identified as Nero's wife by her black anterior tooth. On May 4, 1897, the victims of the Bazar de la Chariti fire were identified using forensic odontology which occurred in Rue Jean-Goujon, Paris. When an ether-oxygen film projector ignited a rapidly fire, about one hundred and twenty-six members of the Parisian aristocracy were perished. The victims were identified visually or by personal effects, mainly jewelry, on the day of fire except thirty of them.[5] Failure of identification of remains occurs during postmortem changes, traumatic tissue injury, or lack of a fingerprint record invalidates the use of visual or fingerprint methods. In such incidents, dental identification attributes a primary role in the identification of remains. It also plays a significant role when the deceased person is skeletonized, decomposed, burned, or dismembered. Like any other hard tissues in the body, which is often preserved after death, teeth can also be preserved. The combination of decayed, missing, filled teeth as well as the status of a person's teeth which changes throughout life can be measured and compared at a fixed point of time. Tooth can withstand the temperature of 1600° without the loss of microstructure and is the most durable part in the body.[6]

After all other soft tissues and skeletal tissues have been destroyed by decay or incineration, teeth have the capacity to subsist its structural integrity. Pulpal tissue is one of the most protected oral tissues being surrounded from all sides by the dental hard tissues.[7]

Histology of Pulp

Dental pulp can be defined as a richly vascularized and innervated connective tissue of mesodermal origin enclosed by dentin with communications to the periodontal ligament. The pulp consists of blood vessels and nerve trunks in the pulp core and at the periphery, it is composed of three zones [Figure 1]: (1) Odontoblasts, (2) cell-free zone, and (3) cell-rich zone.[8]

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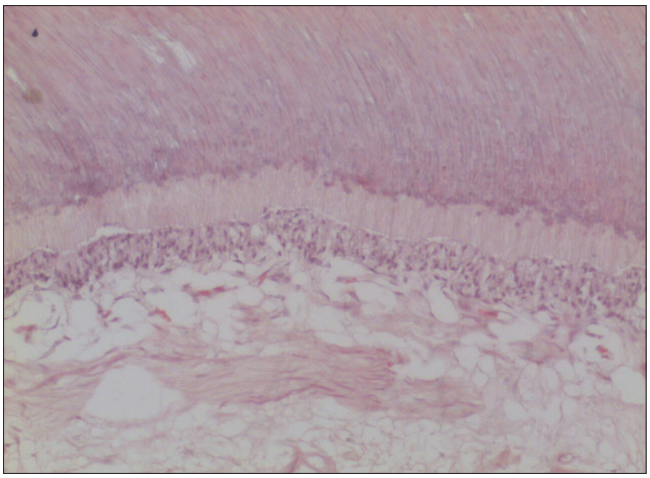

Figure 1: Photomicrograph of pulp showing the zones of pulp (H and E, ×4)

The odontoblasts present in the odontogenic zone vary in size, shape, and arrangement. The cytoplasmic process extending from the apical cytoplasm is usually devoid of organelles and extends to about 2/3rd of the lengths of the dentinal tubules. The cell-free zone contains subodontoblastic plexus of nerves and vessels. Pulp consists of fibroblasts, defense cells such as histiocytes, plasma cells, and pluripotent undifferentiated mesenchymal cells and stem cells.[8]

The apical foramen serves as a passage for blood vessels, lymph vessels and nerves. There are two types of nerves present in pulp that is unmyelinated parasympathetic nerves and myelinated nerves and somatic nerves which lose their myelin sheath before they branch and form plexus in the cell-free zone. This plexus is termed as plexus of Raschkow.[8]

Some of the physiological changes occur during advancing age. Decreased cellularity, increase in fibres and degeneration of nerves and calcifications. The calcifications are either diffuse or organised into masses called pulp stones.[8]

Age Estimation

Age estimation is a crucial concern in establishing the distinctiveness of an individual as the development of human dentition follows a consistent developmental sequence of teeth starting from 4 months *in utero* until the emergence of third molars, i.e. second to third decade of life.[9]

Assessing tissue alterations such as reduction in the size of pulp chamber, formation of pulp stones, dystrophic calcifications, increasing dentinal thickness and reduced cell populations, all of these guides in estimation of age. Neural function also declines along with decreased blood flow.[8,10]

The quantitative evaluation of pulp cavity and emergence of third molar are two essential criteria to assess the age of an individual. As age advances, increased deposition of secondary dentin occurs thereby reducing the dimension of the pulp cavity that can be determined by radiography which may be considered as a guideline for age estimation in an individual.[11] Evaluation of coronal pulp tooth ratio in forensic perspective acts as an indicator in age estimation in surviving individuals of unknown data.[12]

Minimal changes in pulp tissue may be noted at 6 hours after removal of tooth, but still pulp tissue can be maintained for three days in disparity to other body tissues wherein destruction and degeneration occurs within a single itself.[13]

Odontoblasts in Pulp

Odontoblasts and subodontogenic cells undergo apoptic cell death by apoptic cell markers such as bcl-2.[8] As age advances decrease in length, cytoplasmic organelles with reduced capacity of synthetic and secretion are observed in odontoblasts. Occurrence of apoptosis and degeneration reduces the number of odontoblasts. The durability of odontoblasts regarding its survival is greater compared to other body tissues.[14] The morphology of odontoblasts vary with advancing age transforming from loosely packed tall columnar to densely packed short ovoid cells,[15] with diminished reparative capacity. The mass of the odontoblasts may also be used as a guide for age estimation, wherein the number of odontoblasts depends on vitality of tooth. The postmortem change in relation to odontoblasts number is time dependent. Up to 5 days postmortem the days of death may be estimated by observing the reduction of odontoblasts number.[16]

Sex Determination and DNA from Pulp

In this era of molecular analysis, the evaluation of tooth size and morphology provides inadequate characteristics for forensic identification.[17] Several external factors influence the morphology of tooth; the most accepted method for gender identification is DNA molecular analysis.[5,18] Numerous protocols are available for the extraction of DNA from unknown person which can be later compared with antemortem DNA samples.[18] Pulp is a good source for extraction of DNA. Restriction fragment length polymorphism, polymerase chain reaction (PCR), and microarrays are various methods to analyze DNA.[18]

Barr bodies

Barr bodies are intensely stained chromatin material present in nucleus of female somatic cells which plays a pivotal role in gender identification in individuals.[19] These Barr bodies are profoundly basophilic structures measuring 0.8 μ ×1.1 μ depicting various morphologies such as spherical, rectangular, plano-convex, biconvex, and triangular in light microscopy. In electron microscopy, Barr bodies resemble alphabets such as V, W, S, or X. In terms of forensic context to a period of 4 weeks, gender identification can be done by X and Y chromosomal study. Simple stain such as Papanicolaou stain can demonstrate Barr bodies.[20] The study of Barr bodies provides valuable information and sufficient evidence in remains of burnt and mummified bodies. Barr bodies test is not affected by burial conditions in terms of pH and salinity. Under burial conditions, Barr body test can be used to determine gender with 98.9% certainty.[21]

F-bodies

F-bodies are present in Y chromosomes and can be used in sex determination. Numerous studies have been carried out to identify F-bodies from pulpal tissue. The most efficient and reliable method to determine gender is by fluorescent staining of Y chromosome in healthy pulps.[18]

Sex-determining region "Y" gene

The sex-determining region Y (SRY) gene extracted from pulp DNA can be used for gender determination in forensic samples.[22] For further development as male, these SRY genes are accountable. 2 X chromosomes (46 XX) and 1 X and 1 Y chromosome (46 XY) are present in females and males, respectively. The short (p) arm of the Y chromosomes at position 11.3 harbors SRY gene.[18]

Amelogenin gene

During development, the enamel consists of protein (30%); among this, 90% are amelogenins (AMELs). Amelogenesis occurs with the involvement of AMEL. AMEL is formed with the help of AMEL gene. DNA consists of AMEL X gene and AMEL Y gene. Hence, the female has similar AMEL genes, and the male has two different AMEL genes. These genes may assist in determination of sex in minute samples of DNA.[5,18]

As dental pulp is unaffected by external assaults, teeth may be considered as an efficient source of DNA. The concern of DNA profiling should include identifying victims, associated body parts, and identifying criminals.[22] The extracted DNA from the teeth of an unidentified person can be compared with the antemortem DNA samples. The different types of DNA are nuclear DNA and mitochondrial DNA (mtDNA). DNA can be analyzed by various methods such as restriction fragment length polymorphism and PCR.[11]

Mitochondrion of embryo is derived from mother's egg whereas genomic DNA is obtained from father's sperm. Hence, every child has same mtDNA as its mother. Therefore, it can be used as a significant marker to identify a missing individual from unidentified remains with that of maternal relative available.[23]

Telomere Shortening

Dental pulp DNA can also be used to estimate the age of an individual. Reduction in the length of chromosomes can be seen during aging process. The end of human chromosomes and senescence-associated distension of satellites are formed by telomeres. During the aging processes of many cells and tissues, shortening of telomere occurs. At each cell division, shortening of cell division occurs, due to nonreplication of DNA polymerases at the end of linear molecules. Thus, estimating telomere shortening of extracted DNA from pulp is a valuable method in determining age at the time of death.[24]

Blood Group Determination

As pulp is highly vascularized, most undoubtedly, blood group antigens are present in the pulp. Aswath *et al*. conducted a study to determine ABO blood groups and Rhesus factor in dental pulp tissue for identification of deceased individual of 60 samples using slide agglutination, wherein out of 60 samples, blood group of 57 samples was matched. The authors concluded that the dental pulp tissue harbors stable blood group antigen and can be used for forensic analysis.[25]

Identification in Mass Disasters

Forensic dentistry not only plays an important role in mass disasters (terrorist attacks, earthquakes, and tsunamis), child/elder/spouse abuse, bite mark analysis, criminal/natural deaths and injuries, bioterrorism, etc., but also helps in identification of decomposed and charred bodies like that of drowned persons, burns, and victims of motor vehicle accidents.[11]

The identification of a large number of casualties in mass disasters is complex and fraught with hazards, both physically and emotionally. The role of forensic anthropologist plays a critical role in forensic excavation of human remnants, festered, burned remains identified by crime investigative officers, or members of community.[4]

Antemortem and postmortem reports play an important role in the identification of mass disaster victims. In case where antemortem reports of the victims are unavailable, DNA profiling becomes the lone and unswerving methodology for identification.[4] Since the dental pulp in encased inside the tooth, it provides excellent source of DNA which is crucial in identifying the deceased, providing closure for family members as wells as legal issues.[23]

Conclusion

Forensic odontology essays a fundamental task in establishing and identification of an individual where other available sources fail. The use of features unique to the human dentition as an aid to personal identification is widely accepted within the forensic field. The dental pulp can be used as an indicator to determine age and gender. Molecular analysis of DNA from extracted pulp may be analyzed to determine sex. Furthermore, the dental pulp is a warehouse for blood group antigen, thus facilitates in identifying a deceased individual. Table 1 summarizes the pulpal tissue in younger and older age group.[8]

However, further research should aim in establishing the accuracy of application used in dental pulp with regard to its forensic discipline with a large diversified sample size. Although there are various methodologies to determine the identification of age, gender, days of death, blood group, etc., accuracy relies on the selected methodology and its concerned specialties. Furthermore, it requires sophisticated and well-equipped clinical and biochemical laboratories coupled with studies focusing on molecular biology aspect in the identification of human remains that should augment DNA extraction with minimal sources available and under gradually hostile conditions.

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Table 1: Normal pulp in younger and older age[8]

Pulp	Younger age	Older age
Cells in pulp core	Numerous	Reduced
Blood vessels	Numerous	Reduced
Fibroblasts	Numerous	Reduced
Collagen	Less	Dense
Odontoblasts	Numerous	Reduced
Nerve fibers	Numerous	Reduced
Pulp volume	Large	Smaller

Conflicts of interest

There are no conflicts of interest.

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