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Role of Dental Pulp in Human Identification

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Abstract **Review Article**

Identity-related issues are a major focus of forensic science. For society, it has always been crucial to identify an unidentified person. Positive identification is necessary not only to guarantee the deceased's correct burial but also to address other matters, including criminal investigations, settlements, and military procedures. But if the deceased is burnt or decomposed it will be difficult to identify due to lack of soft and hard tissues for forensic and medicolegal examination. In those situations, medicolegal examination can be conducted by utilizing the dental pulp. As dental pulp is well protected by hard and resilient dental tissue like enamel and dentin, it would serve as a great source for human identification.

Keywords: Dental pulp, Sex, Blood, Forensics, Age.

INTRODUCTION

One of the most taxing fields that humans are tackling till now is human identification (Nayar AK *et al*., 2017). According to Parikh (1999) Forensic medicine is defined as 'the application of medical and paramedical scientific knowledge to certain branches of law, both civil and criminal'. The Federation Dentaire Internationale (FDI) characterizes forensic odontology as "that subdivision of dentistry that within the interest of justice, deals with the proper handling and examination of dental evidence and the best possible analysis and presentation of dental findings" (Sachdeva, A *et al*., 2021). Enamel is the hardest part in human body which forms the outer layer of tooth and protects the inner layers (dentin, pulp) of tooth even in adverse environment. Dental pulp is the most reliable tool in forensic odontology as it is well protected, munificently vascularized and is great source of DNA. This plays an important role when the deceased person is decomposed, skeletonized or burned and in mass calamities (Sachdeva, A *et al*., 2021). Dental pulp will provide information like age sex and blood group which will help to build a profile of that deceased human.

DENTAL PULP

Dental pulp is a highly specialized mesenchymal tissue which is encased by dentin and liaises with periodontal ligaments. It is intruded by blood vessels, lymph vessels, nerve fibers through apical foramen (Sachdeva, A *et al*., 2021). It consists of cells like odontoblasts, fibroblasts, dendritic cells, macrophages, mast cells, lymphocytes and undifferentiated mesenchymal stem cells. As the dental pulp is well vascularized it serves as a good source for blood grouping. Dental pulp undergoes age related changes like fibrosis, degeneration, calcifications, atrophy decrease in cellularity and size of pulp chamber (Cohenca, N *et al*., 2012). Along with dental pulp odontoblastic process, fibers of periodontal ligament, cellular cementum serves as a source of DNA with which a person`s sex can be determined (Bansal, S. P *et al*., 2021).

METHODS OF RETRIVAL OF PULP TISSUE

CRUSHING ENTIRE TOOTH: It is the simplest method, but the major disadvantage is that it destroys the tooth completely making it impossible to do other

investigations like radiographic, anatomic, biochemical studies. The other disadvantage is that there is a high chance of contamination of bacterial DNA endonucleases during DNA extraction (Bansal, S. P *et al*., 2021).

CONVENTIONAL ENDODONTIC ACCESS: This method involves preparation of access cavity and excavating pulp using hand instruments. The complexity of this method relies on pulp chamber morphology and size of access cavity. The drawbacks are destruction of occlusal surface and restoration make it unfeasible for further radiographic analysis and assurance of complete debridement of pulp chamber (Sharma *et al*., 2017).

VERTICAL SPLIT: This method involves sectioning of tooth along its long axis followed by excavation of pulp. This method provides access to the entire length of pulp in single rooted teeth, but it limits access in multi rooted teeth. This method lowers the risk of contamination of pulp (Sharma *et al*., 2017).

HORIZONTAL SECTION: This method involves sectioning of teeth in cervical area below the cementoenamel junction. This method provides accessibility to both radicular and coronal pulp without hindering the restorations (Bansal, S. P *et al*., 2021).

ORTHOGRADE ENTRANCE TECHNIQUE: This technique suggests entrance through enamel surface instead of root apex which reduces the risk of tooth damage (Bansal, S. P *et al*., 2021).

REVERSE ROOT CANAL: This technique suggests entrance from root apex followed by filling the interior of tooth. The tooth will be fragile, so this method has high risk of crushing the tooth (Bansal, S. P *et al*., 2021).

SEX DETERMINATION

Sex, a biological trait that encompasses anatomical and physiological features like chromosomes, gene expressions, reproductive anatomy and hormone levels. Sex is determined at birth and is categorised as male (XY) or female (XX). In forensic medicine determining sex is the most crucial step in identifying a person. As there are various methods to determine sex like metric and nonmetric methods which includes odontometrics, incisor index, mandibular canine index, analysing radiographs, dental morphology and anomalies, biochemical method possess the greatest accuracy and reliability. Pulp is a good source of DNA as it is well preserved in teeth for very long period. It consists of fibroblasts, odontoblasts, endothelial cells, peripheral nerve, undifferentiated mesenchymal cells. and nucleated components of blood which are the source of DNA. There are various methods like PCR amplification, amelogenin protein analysis, sex determining region Y protein (SRY) analysis (Heng, D *et al*., 2022).

BARR BODIES

Barr bodies were 1st discovered by Barr and Bertram in 1949. They are inactive X chromosomes that are found in cell with more than one X chromosome present (Heng, D *et al.*, 2022). They are strongly stained chromatin material present in females, one per cell nucleus. Simple stains like Papanicolaou and eosin and haematoxylin dyes can be used to view Barr bodies (Heng, D *et al*., 2022, Sachdeva, A *et al*., 2021). They are basophilic structures measuring 0.7x1.2 micrometres (Sreeja, C *et al*., 2015). They demonstrate various shapes spherical, rectangular, triangular, plano-convex and biconvex. In electroscopy, they look like alphabetical letter sets like V, W, X or S (Sachdeva, A *et al*., 2021).

AMELOGENINS

These are primary extracellular matrix protein found in enamel (Heng, D *et al*., 2022). AMEL gene is responsible for formation of amelogenin which is responsible for amelogenesis (Sreeja, C. *et al*., 2015). This gene is found in both X and Y chromosomes so sex can be determined by identifying size difference between these chromosomes (Heng, D. *et al*., 2022). Sex can be determined by either running a known primer against sample or mass spectrometry (Heng, D *et al*., 2022). The AMEL X and AMEL Y genes are in the DNA at 106 and 112 bps, respectively (Sreeja, C *et al*., 2015). Male will have two different AMEL genes whereas females have two similar AMEL genes with which sex can be identified (Sreeja, C. *et al*., 2015). In recent times the efficacy of AMEL gene in sex determination is questioned because of false outcomes (Heng, D *et al*., 2022).

Y CHROMOSOME

The genetic difference between male and female is determined by presence or absence of Y chromosome. So, sex determining region of the Y chromosome (SRY) is used as a sex marker in forensic science (Heng, D *et al.*, 2022). Females have 2X chromosomes [46XX] and males have 1X and 1Y chromosome [46XY]. It is located on the short arm of Y chromosome (Sreeja, C *et al*., 2015). Polymerase chain reaction (PCR) or real time PCR for SRY amplification (Prasad, P *et al*., 2021). Certain cases like maternal-foetal micro chimerism, syndromes like Klinefelter syndrome (46, XXY), Turner syndrome (46, X0) and persons who have undergone organ transplantation or blood transfusion there may be false positive results (Prasad, P *et al*., 2021).

F-BODIES

These are found in Y chromosome which can be used to identify sex of one person (Sachdeva, A *et al*., 2021). Fluorescent staining of Y chromosome is the effective way to determine the sex for which quinacrine mustard can be used which gets accumulated in guanine portion of DNA (Sreeja, C *et al*., 2015). But these

techniques only reliable if the pulp is healthy (Sreeja, C *et al.*, 2015).

BLOOD GROUP DETERMINATION

Blood groups are determined by presence or absence of antigens on RBC surface and rhesus factor protein. Blood group is determined by the genes that is inherited from the parents. There are 8 main blood group types, which are combination of 4 blood groups and 2 Rh factors which are A, B, O, AB and the 2 Rh factors are + and -. Identifying a person`s blood group will be very useful in forensics because it cannot be affected by drugs, disease or living conditions. Blood typing can also be used to identify one`s paternity (Harbison *et al*., 2016). As dental pulp has numerous blood vessels, antigens will be most likely to present (Aswath, N *et al.*, 2012).

ABSORPTION-ELUTION TECHNIQUE

The Absorption-Elution method functions by first enabling the absorption of blood-group-specific agglutinin by a substance containing blood-group agglutinogens. After the absorbed antibody has been eluted at a high temperature, blood cells containing the matching antigens are agglutinated (Rao, K. A *et al*., 2024).

This technique least alters the nature of the substrata and is incredibly sensitive and specific. Additionally, it has been discovered that the material can be reused with nearly minimal loss of its antigenic properties after it has been utilized. It is possible that recent tooth specimens will offer reliable sources for blood group determination. Nonetheless, variance may result from autolysis, dehydration, pulp antigen loss, or a high frequency of mistakes brought on by bacteria-borne foreign antigen in carious teeth [Ramnarayan, B *et al*., 2013]. As the duration increases, antigenicity decreases, and findings tended to be unfavourable. Dental pulp can provide up to six months of data on Rh factors and ABO blood types (Das, M *et al*., 2023). Particularly in the elder age group, erroneous results may occur because of regressive changes in pulp (Aswath, N *et al*., 2012).

ABSORPTION-INHIBITION TECHNIQUE

The Absorption-Inhibition method is mostly used to determine blood group from dry blood samples. Other than blood the antigens are also secreted in body fluids. So, using this method blood group can also be determined from body fluids like saliva. This method works by reducing the strength of antigens (Kumaraswamy, J *et al.*, 2017). This technique is less significant than absorption-elution technique because it gave more negative results than positive results. As the duration increases or due to adverse environmental conditions this technique will be very less sensitive.

AGE DETERMINATION

Estimating an individual's age is essential for determining their uniqueness since the human dentition develops in a predictable order, with teeth appearing 4 months after birth and emerging in the second or third decade of life, after that. Age is estimated by evaluating changes to the tissue, such as shrinkage of the pulp chamber, pulp stone production, dystrophic calcifications, increased dentinal thickness, and decreased cell population (Metgud, R *et al*., 2016).

ODONTOBLASTS

Odontoblasts are cells that are responsible for formation and maintenance of dentin. Apoptic cell death is induced in odontoblasts and sub odontogenic cells by apoptic cell markers such as bcl-2. Cytoplasmic organelles in odontoblasts exhibit a decreased potential for synthesis and secretion with advancing age. The quantity of odontoblasts decreases when apoptosis and degeneration take place. When it comes to survival, odontoblasts are more resilient than other bodily tissues. The morphology of odontoblasts evolves with age, from short, ovoid cells with poor reparative capabilities to densely packed, tall, columnar cells. The mass of odontoblasts can also serve as a reference for predicting age, since the number of odontoblasts is depending on the health of the tooth (Metgud, R *et al.*, 2016). Odontoblast density will be highest in young ages than older ones (Baker, A *et al*., 2019). After death, the number of odontoblasts varies and is time dependent. The number of odontoblasts can be used to estimate the days of death up to five days after death (Metgud, R *et al*., 2016).

TELOMERE

Telomeres are proteins that are found at the end of each chromosome. It protects the ends of DNA from binding to itself or each other and its length serves as a biological clock that determines the life span of a cell. If its length reaches a particular limit the cell undergoes apoptosis or senescence (Shammas, M. A, 2010). Therefore, telomere length will get shortened as the age increases. As dental pulp is a great source of DNA, after its extraction quantitative polymerase chain reaction can be done to assess the length of telomere. It will be very useful in cases where morphological remnants are not available (Tejasvi, M. A *et al*., 2021).

PULP SIZE

As one ages, secondary dentin deposition causes the dental pulp chamber's dimensions to shrink both vertically and horizontally. Secondary dentine deposition is the most significant age-related alteration along internal tooth surfaces and is typically thought to be well preserved from various challenges. Analysing the pulp/tooth length ratio is one indirect method of measuring secondary dentine deposition.

Age groupings differ significantly from one another. As age increases, there will be a progressive decline in pulp size in relation to cervical pulp width and total pulp length, and this has a strong association with the chronological age. Age estimates among adults therefore become much more varied and precise because of age-related changes.

Evaluation of these radiographic data can therefore be used as a tool for estimating age. Therefore, using standard intraoral periapical radiographs, one can estimate an adult's chronological age based on the size of their dental pulp. Without using radiographs using only digital vernier callipers and k file pulp tooth ratio can be calculated, and age can be determined. The advantage of employing radiography to determine a person's chronological age from dental pulp is that it is a non-invasive method (Shetty, U. A *et al*., 2017).

CONCLUSION

In situations where a person's body is purposefully dismembered or highly decomposed to conceal their identity, as well as in man-made disasters like bomb blasts, terrorism, mass murder, and landslides, floods, and earthquakes, personal identification becomes necessary. Dental tissues are a good source for forensic examination since they are resistant to environmental assaults like burning and mutilation. In the human body, the tooth pulp is the most robust and durable biological material when compared to other sections. It is resilient to adverse environmental circumstances and provides excellent protection for the pulpal tissue found inside teeth. The cells in the tooth pulp so stay stable for a very long period. Thus, the dental pulp has a high potential value. Hence, it can be recommended that the uniqueness of pulp can be used as a powerful tool in victim identification.

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