



## REVIEW PAPER

# A review of the significance of oral histology in forensic human identification

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### ABSTRACT

Forensic histopathology is the application of histological techniques to identify the deceased and injuries, specifically in medico-legal cases. The histological aspect of dental tissues is also helpful in forensic human identification. As the tooth can withstand a certain degree of temperature and other environmental factors, oral histology is helpful in various conditions like physical, biological, and chemical injuries, fire, putrefied remains, and archaeological samples. It is also possible for forensic odontologists to know the cause of death, manner of death, and time since death from oral histology, which aids in forensic investigation. In Forensic Dentistry in place of Forensic Science researchers primarily focus on studying oral histological structures, including enamel rod patterns (ameloglyphics), amelogenin (AMEL), perikymata, the incremental line of Retzius, the neonatal line, reparative dentin, sclerotic/transparent dentin, cementum annulation, alveolar bone, ameloblasts, odontoblasts, and blood cells. This article aims to provide a valuable, informative, and critical summary of the importance of oral histology in human identification and biological profiling in forensics.

**Keywords:** Medico-Legal cases; forensic odontology; dental tissues; biological profiling; species identification

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## INTRODUCTION

Oral Histology is the branch of dentistry that deals with a detailed examination of the oral tissues. For a better understanding of the microscopic structures of oral hard and soft tissues, profound knowledge of Oral histology is a prerequisite. Enamel, dentin, cementum, and jaw bone (maxilla, mandible) – are known as dental hard tissues, while pulp, periodontal ligament, gingiva, tongue, soft palate, oral mucosal lining, lip, salivary gland, and muscles of mastication - are known as dental soft

tissues.<sup>1</sup> The histological aspects of these tissues play an important role in forensics. This article will explain the Histological importance of oral tissues in forensic analysis.

### Forensic significance of oral tissues

Histological structures of dental hard and soft tissues have specific forensic significance, like distinguishing human from non-human remains, individual identification, age estimation, gender determination, manner of death, time since death, and geographical identification. The histological study of the

tooth is possible from the sixth week of the intrauterine life of the fetus when deciduous teeth begin to form from the tooth germ.<sup>1</sup> As the tooth is the most resistant tissue to environmental changes, it is also helpful in forensic archaeology and anthropology.<sup>2</sup>

### **Distinguishing human from non-human remains**

When an investigating officer finds any bony skeleton at the scene, there is a need to determine whether the bones are human or non-human. In Medico-Legal investigation, one of the roles of a forensic anthropologist is to identify the human skeleton remains before making a biological profile. There are three primary levels of identification: gross skeletal anatomy, bone macrostructure, and bone microstructure (histology), which distinguish human and non-human bones. The first two levels are non-invasive. Compared to it, the invasive bony histology is more accurate in determining species.<sup>2</sup> The human cortical bone contains a well-organized Haversian system, while non-human cortical presents a Plexiform arrangement. Apart from this feature, osteons' arrangement, density, and diameter can also be measured and quantified. Nor Faridah et al. found statistically significant differences in osteon and Haversian canal parameters, which were relatively more important in humans than Non-human ones.<sup>3</sup> The mandible's thick cortical bone allows histological differentiation between human and non-human bone fragments. Additionally, teeth vary in structure and number across species. David A Crossley found that enamel thickness in dog and cat teeth is 0.1-1mm, while human teeth enamel thickness is more than 1mm. He pointed out the complete examination of dental variation associated with the breed, size, or sex of the animal, which has a primary key role in the identification.<sup>4</sup>

### **Individual identification**

Identification becomes challenging in decomposed bodies, burns, acid attacks, and severe accidental cases where facial features

are lost. Biometric-based identification, such as fingerprint, iris scanning, and facial recognition, is impossible in these cases. The study of enamel rod patterns is known as Amelogyphics (Tooth print). The Tooth print is unique in individuals and reproducible even in burn and acid attack cases; hence, it is helpful in identification.<sup>5</sup>

Developmental defects of enamel, dentin, or oral manifestations of various Systemic diseases like enamel hypoplasia, dentin dysplasia, gingival fibrosis, syphilis-peg lateral, cleft lip and palate, Paget's disease-increase jaw size, Gardner syndrome-supernumerary teeth may be congenital or hereditary. These defects reflect the events in utero and after birth, creating an individual's unique profile. This distinctive profile can help in the identification of victims, specifically in mass disasters, completely charred, putrefied, and mutilated cases.<sup>6</sup>

DNA (Deoxyribonucleic acid) profiling is the most reliable method for individual identification, with teeth and bones being the sole sources of DNA in severely damaged and degraded human remains. Nuclear DNA can be extracted from dental pulp, while mitochondrial DNA can be extracted from dentin and cementum. It is possible to extract DNA from nuclear material even when temperatures reach 300°C or 700°C in mass disasters or burns.<sup>7</sup>

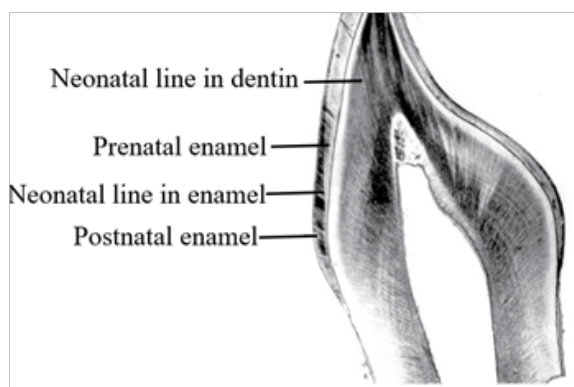
The blood group is genetically determined and remains unchanged for life. ABO blood grouping can be performed from tooth pulp and dentin even after prolonged extraction or in remains.<sup>8</sup> This is a less time-consuming, easy, and cost-effective technique to narrow the search in mass disaster cases. Moreover, blood groups are hereditary and often documented, aiding in individual identification, exceedingly when rare.

### **Age estimation**

Age has its prime role in medico-legal cases as it is directly related to punishment and imprisonment, specifically in minor, juvenile, sexual assaults, and child marriage cases. Dental age estimation has become widely

accepted in the court of law. Since radiographic methods can be performed on living individuals with consent, they can be used in dental age estimation. Histological dental age estimation is only used in the deceased, which helps in the biological profiling of unknown skeletal remains. A histological method is the only option for age estimation of fragmented and single teeth.

The neonatal line, the birth indicator, suggests enamel formation before and after birth, as shown in Fig.1. In forensic science, the birth of a child, whether stillborn or alive, enables the evaluation of the fetus's age. It is possible to distinguish postnatal enamel from prenatal enamel under SEM within one or two days after birth as the rate of enamel formation varies between 2.5-4.5  $\mu$ /day, according to Whittaker and MacDonald.<sup>9</sup> Regressive changes in the tooth, like attrition, formation of secondary dentin, transparent dentin and tertiary dentin, cementum deposition, root resorption, and decreased cellularity, are essential for histological age estimation. Moreover, micrometric measurement by SEM is an advanced technique for histological age estimation. Among all age estimation methods, histological dental age estimation is the most accurate, with the lowest error rate ( $\pm 1.0$  to  $\pm 5.0$ ).<sup>10</sup>



**Figure 1** Neonatal line in enamel<sup>1</sup>

### Gender determination

Amelogenin (AMEL) is a major protein found in the enamel of human teeth. It is produced by ameloblasts (enamel-forming

cells) and helps to determine gender. Dentin offers both nuclear DNA and mtDNA, which can be used to determine gender. The thickness of secondary dentin is higher in males than in females, which can aid in gender determination in forensics.<sup>11</sup>

Females have two identical genes located on the Chromosome, while males have two non-identical genes, one on X-Chromosome and one on Y-Chromosome.<sup>12</sup> In pulp, an active X-chromosome appears as a dense, dark staining spot at the periphery of the nucleus of each female somatic cell, known as the BARR Body. In burn cases up to 200°C, only the BARR Body can be differentiated. Fluorescent staining of the Y-chromosome determines gender reliably in healthy pulp tissue or is limited to enamel or dentin. However, it is applicable only when the pulp is intact. Gender determination is not possible in the degenerated pulp.<sup>12,13</sup>

The drumstick appendages of the polymorphonuclear leukocytes known as Davidson Body are only present in females and are reliable.<sup>14</sup> Real-time PCR can amplify the Gender-determining region Y (SRY) gene, also known as the Sex-determining region Y protein gene, from gingival epithelial cells and scrapings of attached gingiva.

### Manner of death

There are three types of medico-legal death - accidental, suicidal, and homicidal. Suicidal falls involving skull fractures are rare and mainly involve extremities and pelvis bone rather than the skull.<sup>15</sup> Oral manifestations of poison are beneficial in explanation for death, specifically in a suicidal manner. In India, various sources of poison include household, agricultural, horticultural, industrial, and commercial sources, which are readily available. Occupational chronic exposure to heavy metals is associated with epithelial changes in oral tissues.

### Time since death

Different dental tissues like pulp, enamel, dentin, periodontal ligament, and cementum

can be used to estimate PMI (Post-mortem interval). Factors like temperature, humidity, cause of death, burial conditions, and geographic location affect the PMI rate. Hard dental tissue, like enamel, can resist all environmental factors compared to soft dental tissues. Molecular death, succeeded by somatic death, entails the cessation of mitotic cell division in odontoblasts, gradually decreasing their numbers. This phenomenon aids in estimating post-mortem intervals of up to 5 days.<sup>16</sup>

### **Geographical identification**

Race-specific developmental anomalies and cultural traditions are localized to particular regions, which can aid in identifying individual characteristics. The histochemistry of dental plaque provides insights into individual habits, dietary patterns, and lifestyle. It occasionally helps to determine the cause of death, given its link to heightened risks such as endocarditis and heart diseases. Furthermore, the forensic significance of microbiota extends to identifying personal traits, geographical origin, and ethnicity and estimating the time since death. Intentional alteration of tooth shape, known as tooth mutilation, has been documented globally, including in the Pacific, Asian, African, South, and Central American regions. The geographic spread of dental mutilations can serve forensic purposes, helping identify individuals' geographic region, ethnicity, and cultural heritage. Dental mutilations pique medical-legal curiosity, extending beyond mere personal identification.<sup>17</sup>

### **DISCUSSION**

Forensic science primarily concerns evidence, serving as the sole scientific confirmation when identity becomes uncertain. In mass disasters, particularly in burn cases where other evidence may be scarce, teeth offer the primary means for potential identification.<sup>2</sup> Tooth biometrics reveal that enamel rod-end patterns are distinctive for positive individual identification. In the histological section of the tooth, dentin and cementum are more

informative tissues for forensic analysis. Pulp is the only tissue identifying an individual concerning DNA and blood group as it is protected in a pulp cavity and least affected by external environmental factors.<sup>5</sup> The postnatal hard tissue formation amount can be measured microscopically to know the infants' exact survival period.<sup>9</sup> Oral histology is used for age estimation, which includes Gustafson's method, perikymata, neonatal line, amino acid racemization, cemental incremental lines, and dentin translucency.<sup>13</sup> A neonatal line implies live birth, and hence, its absence could be used as evidence of infanticide. Dental pulp can be used for molecular analysis for DNA profiling and to determine age, gender, and blood group antigen. Odontoblasts present in the pulp can be used to assess the age and the time of death. For individual identification and gender determination, buccal swab cells and alveolar bone are also considered in forensics.<sup>16</sup>

### **CONCLUSION**

This article aims to provide a ready source for applying oral histology in forensics. Criminal investigation, mass disasters, and individual identification are crucial aspects of contemporary jurisprudence. The handling, collection, transportation, examination, and comparison of dental evidence of human remains help to provide essential lead in forensic investigation. Dental histology and molecular anatomic structures are finding new ground in the emerging field of forensic odontology. Perhaps this review helps the readers by providing a unique research platform in histology where histological evidence strengthens the principle of forensic science: **"Facts do not lie."**

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