Reliability of Automated Biometrics in the Analysis of Enamel Rod End Patterns- In Vitro Study

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Ameloglyphics is the study of the distinctive patterns found on the enamel surface of teeth, which are different on each tooth and particular to each person. The purpose of this study was to compare the enamel print patterns on the buccal and lingual surfaces of teeth to see if there are any appreciable differences, and to evaluate the sensitivity and reliability of an automated biometrics software. The study involved 62 individuals including 36 females and 26 males, from age range 15-60 years. From each individual one tooth sample was analyzed using a light microscope to capture images of the buccal and lingual surfaces middle third. These images were then analyzed using biometric software called Source AFIS, which processed the images and displayed enamel patterns. Data were entered and analyzed using GPower software (version 3.0). Chi-square test and Goodness of fit tests were done to compare the enamel rod end patterns between the teeth surfaces. These enamel patterns were classified according to the “Manjunath Classification”. The study found that there were no notable distinctions in enamel print patterns when comparing the teeth buccal and lingual surfaces. All 62 samples analyzed showed a high degree of similarity in enamel print patterns on both surfaces. This finding suggests that the same developmental, environmental, and genetic factors likely influence the enamel print patterns on both surfaces of a tooth. The samples' high frequency of comparable sub-patterns on both the buccal and lingual surfaces suggests that they are very similar to one another. This implies that tooth prints may be a valuable tool for forensic personal identification, particularly in the area of forensic dentistry.

Keywords: Automated biometrics; Enamel print patterns; Forensic science; Personal identification.

The increasing use of technology in crimes has made human identification crucial for personal, social, and legal reasons. The most used methods for identification include dental, fingerprint, and DNA analysis. Dental identification is particularly reliable in mass disasters, with a success rate of 75%, as teeth are the most durable and resilient part of the human body. In scenarios such as earthquakes, tsunamis, or car accidents, dental hard tissues have become crucial for personal identification. Due to their ability to endure extreme conditions like high temperatures and acid exposure, as well as postmortem decomposition, teeth have emerged as the preferred method for identifying individuals from badly burned, traumatized, decomposed, or skeletal remains. Forensic odontology employs various methods, including “rugoscopy, cheiloscopy, bite mark analysis, tooth prints, radiography, photographic study, and molecular techniques, to utilize dental evidence in the identification process”.

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“Forensic odontology, which is a branch of forensic science, involves the proper handling, examination, evaluation, and presentation of dental findings.”

It plays a crucial role in detecting crimes and bringing the perpetrators to justice. Due to its reliability, dental evidence is considered a scientifically sound method of identification. Even in the event of identical twins, there are differences between each person’s mouth, according to the fundamental rule of forensic odontology.

Dr. Oscar Amoedo is known as the “Father of Forensic Odontology” because he was able to recognize the victims of a fire disaster in Paris, France, in 1898.

Dental evidence is regarded as a scientifically valid method of identification due to its dependability. Unlike other tissues, the human dentition remains relatively unaffected by physical injury and putrefactive changes. Body identification becomes challenging during mass disasters. While autopsies can be performed, factors such as decomposition and the need for organ histology may limit their usefulness. As a result, forensic odontology has become an essential component of the American judicial system for over a decade.

Forensic odontology encompasses various applications, including rugoscopy, age estimation, ameloglyphics, and PCR, for human identification. However, these techniques may not be suitable for mass disasters where bodies are severely damaged. In such cases, enamel examination, known as “Ameloglyphics,” is the most appropriate method for identification. Enamel prints are unique to each individual, and studies have shown no similarities between different individuals or even within the same person. Enamel prints consist of eight distinct patterns. This research “aims to determine if there are any variations in enamel print patterns between the buccal and lingual surfaces of the same tooth”.

The study’s objective is to analyze and compare enamel rod endings using automated biometric software. If there is no significant difference, enamel prints can be effectively utilized for identification purposes.

MATERIALS AND METHOD

The study involved 62 individuals including 36 females and 26 males, from age range 15-60 years. From each patient, one tooth sample was collected. These are the patients who underwent tooth extraction for orthodontic purposes or due to periodontitis (Figure 1).

The research was carried out at the Department of Oral & Maxillofacial Pathology and Oral Microbiology, Sharda University, Greater Noida. Ethical permission was taken from institutional ethical board.

The Source AFIS biometric software was used to analyze the enamel rod end patterns.
in all the samples, and the resulting patterns were compared. This software employs a technique that involves detecting and matching specific features known as minutiae to create an image. In the case of tooth print analysis, the software reads the enamel prints, which are comprised of a sequence of dot-like patterns known as minutiae, in order to generate an image.

Based on both inclusive and exclusive standards, removed teeth that were physically sound were chosen. The middle third of the buccal and lingual surfaces of the teeth had to be free of cavities, erosion, abrasion, restoration, and fracture in order to meet the inclusive criterion. Carious, corroded, abraded, repaired, and cracked teeth were excluded. The chosen teeth underwent thorough cleaning with distilled water, 10% formalin preservation, open-air drying, and labelling. Using a ruler and a pencil, the incisal, middle, and cervical thirds of each tooth crown were separated into three sections (Figure 2). With the aid of compasses, the dimensions of each sector were equalised. The middle third of the buccal and lingual surfaces of each tooth were chosen for photomicrography as they were least likely to show directional curves and entangling in the rod pattern. The samples were placed in a light microscope using removable adhesive tape, and a digital camera was mounted and connected to the microscope. Photomicrographs of the middle section of the samples were taken, and the images were transferred to a laptop in JPEG format. The processed image or the patterns of enamel print are classified according to Manjunath Classification. (Figure 3). The photomicrographs of the enamel print were analyzed using Source AFIS biometric-based software (Figure 4) which employs a technique that involves detecting and matching specific features known as minutiae to create an image. In the case of tooth print analysis, the software reads the enamel prints, which are comprised of a sequence of dot-like patterns known as minutiae, in order to generate an image. (Figure 5)
RESULTS

The study involved the examination of 62 teeth, with analysis conducted on eight patterns that include Wavy branched, Wavy unbranched, Linear Branched, Linear unbranched, whorl closed, whorl open, loop, and stem-like. Results indicate that while enamel print patterns were collected from the same tooth on both buccal and lingual surfaces, they do not necessarily have identical arrangements. The presence or absence of similar sub-patterns in both surfaces was found to be relatively high.

According to the collected data, all 62 samples showed a combination of at least two of the eight sub-patterns found on both the buccal and lingual surfaces. Some samples even exhibited more than one sub-pattern on the buccal surface. The dominance of enamel print patterns varied among samples, with the wavy-branched pattern

<table>
<thead>
<tr>
<th>A. Wavy-branched</th>
<th>B. Wavy-unbranched</th>
<th>C. Linear-branched</th>
<th>D. Linear unbranched</th>
</tr>
</thead>
<tbody>
<tr>
<td>E. Whorl-open</td>
<td>F. Whorl-closed</td>
<td>G. Loop</td>
<td>H. Stem-like</td>
</tr>
</tbody>
</table>

Fig. 3. “Manjunath classification" of enamel prints

Figure 4a. Enamel print in buccal surface of tooth

Figure 4b. Comparison of enamel print pattern of buccal and lingual surface of the same tooth after being analyzed with Source AFIS biometric - based software
being the most prevalent, followed by linear branched and wavy unbranched in descending order (Figure 6).

Datas were entered and analyzed using GPower software (version 3.0). Chi-square test and Goodness of fit tests were done to compare the enamel rod end patterns between the teeth surfaces (buccal and lingual).

A chi-squared test, also referred to as chi-square or \( \chi^2 \) test, is a statistical test used to examine contingency tables with large sample sizes. It compares observed and expected results to determine whether there is a statistically significant correlation between two variables. The null hypothesis of the test states that there is no significant difference between the observed and expected values.

### DISCUSSION

Naziya et al in 2019 studies that as modern life places increasing importance on human identification, various methods such as passwords, photographs, fingerprints, and DNA analysis have emerged. However, these methods may not be effective when bodies are decomposed, burned, or

#### Table 1. Comparison of the total number of patterns among buccal and lingual surfaces

<table>
<thead>
<tr>
<th>Surface</th>
<th>N</th>
<th>Total no of patterns</th>
<th>Mean</th>
<th>Std. Deviation</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Buccal</td>
<td>62</td>
<td>2.400</td>
<td>.4940</td>
<td>0.05, NS</td>
<td></td>
</tr>
<tr>
<td>Lingual</td>
<td>62</td>
<td>2.250</td>
<td>.4367</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Fig. 5.** Enamel print after being analyzed in the software

**Fig. 6.** Distribution of Most dominant type of pattern among buccal and lingual surfaces
only small fragments of calcified tissues are left. In cases of fire accidents, the intense heat can cause temperatures to vary from 1200°F (649°C), while cremation occurs at temperatures ranging between 871-982°C.52

Temperatures in a fire, influenced by factors like location and combustion details, affect teeth and restorations. Enamel shrinks at temperatures over 1000°C but shows observable prismatic structures. However, identifying cementum with its granular structure becomes impossible at such high temperatures. This is especially valuable in forensic science applications. “Amelloglyphics,” introduced in 2012 by Manjunath et al, is a term similar to dermatoglyphics. Enamel prints, also known as tooth prints, have gained popularity. Ameloblasts, responsible for enamel formation, create unique undulating and interlacing patterns on enamel surfaces, reflected as tooth prints. These prints are individual-specific, but variations exist between individuals, within the same individual, and between genders. Tooth prints are valuable for personal identification in forensic science due to their distinctive characteristics.5

Enamel rod end patterns are unique to each tooth, making them valuable for personal identification alongside other methods. This approach is straightforward, cost-effective, and efficient. It can be used as a supplementary method to maintain dental records for individuals in professions like firefighters, soldiers, jet pilots, and divers. Regular updates of these records are crucial since enamel can wear down with regular use.55

In our study, 62 samples were analysed and pattern were evaluated on both the buccal and lingual surfaces. In all the samples of our study at least 2 patterns were recorded on both buccal and lingual surfaces. In our study, Wavy Branched pattern was the most common found pattern among buccal surfaces. In our study, the linear unbranched pattern was most common found among the lingual surfaces. The result of our study were in consistent with the study of “Manjunath K et al 2009” who also reported a combination of 2 or than 2 different subpatterns in the study.

Our findings were in agreement with “Soo SC et al in 2020” who also had concluded that wavy branched is most dominant subpattern, which also reported in our findings.

Our study specifically examined enamel rod end patterns through automated biometric software. While enamel patterns may differ on different surfaces of the same sample, there is a notable occurrence of similar sub-patterns. Factors such as tooth development, temperature, pressure, and nutrition play a role in shaping these similarities.

As a result, the lingual surface is preferred for enamel print patterns for dental identification in forensic cases due to the high similarity between both surfaces. However, obtaining enamel print patterns is not possible if the tooth surfaces are tainted or have orthodontic appliances and composite restorations. Thus, the lingual surfaces can be a better option in such cases.

Due to its significant resemblance, the lingual surface is preferred for enamel print patterns for dental identification in forensic investigations. However, if the tooth surfaces are contaminated, have orthodontic devices, or have composite restorations, getting enamel print patterns is not possible. The lingual surfaces may therefore be a better choice in these circumstances.

This predominance of wavy branched sub-patterns may be caused by the fact that most enamel rods exhibit a wavy pattern throughout the thickness of the enamel, while many rods follow an undulating course from the dentinoenamel junction to the external tooth surface. The comparable enamel print patterns on the buccal and lingual surfaces of the same tooth are probably the result of several factors working together such as the same placement of the developing tooth bud, temperature, environmental factors, genetics, pressure, and nutrition to the ameloblasts cells in a tooth.57

Our study has demonstrated that tooth prints can be a useful tool for forensic science personal identification since both the buccal and lingual surfaces of the tooth exhibit substantial sub-pattern similarity. This is because of things like heredity, temperature, environmental conditions, pressure, and nourishment for the ameloblast cells in a tooth, as well as the same location of the developing tooth bud. Our findings corroborated those of “Naziya J et al in 2019,” who identified Wavy Branched as the most prevalent pattern in their research.

The result of our study were in partial
agreement with “Joshi PS et al. in 2014” who had reported 3 distinct subpatterns in comparison to 2 distinct subpatterns which was reported in our study.\textsuperscript{58} The undulating and inter-twining path of enamel rods is attributed to the high tensile strength of enamel and the appearance of gnarled enamel and Hunter-Schreger bands. Hence, the enamel rod end pattern should theoretically vary at varying depths of enamel.\textsuperscript{7}

In our study, the arrangement of the patterns was found to be different & unique in every sample which was in similarity to the findings by “Manjunath K et al 2009, Soo SC et al in 2020 and Naziya J et al in 2019”. And the inference could be drawn that biometric analysis of enamel rod end pattern can be used as an adjunct tool for personal identification in forensic odontology.

The use of ameloglyphics in forensic investigations offers several advantages. Firstly, the enamel rod patterns on teeth are unique to each individual, similar to fingerprints, making them a valuable tool for identification when other forms of identification are not available.\textsuperscript{60} Enamel rod patterns are also highly persistent and resistant to environmental factors, such as heat, cold, moisture, and chemical exposure, allowing them to remain intact for long periods of time and making them useful for identifying human remains in forensic investigations.\textsuperscript{5} In addition, enamel rod pattern analysis can complement DNA analysis to provide additional evidence for identification, especially in cases where DNA analysis is not possible or inconclusive. The use of ameloglyphics is also applicable in mass disaster situations, where other forms of identification may not be possible, as teeth are often the most durable part of the human body and can survive disasters such as fires, floods, and explosions.\textsuperscript{61}

The methodology used in this study has many advantages as well. The cellophane tape technique is a popular and non-invasive method that does not require cutting or drilling of the tooth, reducing the risk of damage to the tooth or discomfort to the individual being examined.\textsuperscript{60} This technique is also simple and straightforward, requiring no specialized equipment or training, and can be performed quickly and easily on a variety of tooth surfaces and types, including deciduous teeth, permanent teeth, and teeth with restorations. As it does not require expensive equipment or materials, it is a cost-effective method and a practical option for forensic investigations with limited resources.\textsuperscript{59}

Our research has some drawbacks. Although each person’s tooth print is unique, tooth prints’ reproducibility and permanence are what determine whether they can be used for personal identification in forensic research.\textsuperscript{62} Further research is required to establish if there are any variations in enamel rod end arrangements as depth increases because the enamel rods do not travel the entire length of the enamel in a straight line. For practical purposes, these patterns should be recorded every four years because enamel wear and tear might change it as people age.\textsuperscript{63}

The cellophane tape technique has several advantages, but there are also some disadvantages to consider. One drawback is that obtaining clear and complete impressions of the enamel rod patterns can be challenging.\textsuperscript{64} If the tape is not applied correctly or if there is debris or moisture on the tooth surface, the impression may be incomplete or distorted, making it difficult to compare accurately to a database or the teeth of a known individual. Additionally, teeth with highly irregular surfaces or existing dental work may not provide clear impressions using this technique. Dental X-rays or 3D imaging techniques may be more reliable methods for dental profiling in more complex forensic investigations.\textsuperscript{65}

Various techniques are used to study tooth prints, or ameloglyphics, including acid etching, automated biometrics, and cellophane tape. The easiest and quickest method is using cellophane tape to make a replica of the enamel tooth prints, which can be used for personal identification. This method is commonly used by fingerprint experts.\textsuperscript{6}

However, advances in technology may change the way enamel rod end patterns are analyzed and used in the future. Computer-aided analysis can compare patterns quickly and accurately, making identification faster and more precise. Genetic analysis can also complement enamel rod end pattern analysis to provide additional information to support or confirm a positive identification. Overall, enamel rod end patterns will likely continue to be an essential tool in forensic odontology, and technological advancements will improve their accuracy and usefulness in identifying human remains.\textsuperscript{54}
CONCLUSION

In conclusion, increasing the sample size could improve the accuracy of automated biometric analysis of enamel rod end patterns. Due to the remarkable similarity in sub-patterns between the two surfaces, the study demonstrates that enamel prints on lingual surfaces are favoured for dental identification in forensic instances. Particularly in cases of extreme circumstances where traditional biometric techniques may not be significant, enamel prints may be a lasting and trustworthy biometric tool for personal identification in forensic odontology. These enamel prints’ uniqueness makes it possible to use them as a trustworthy tool for forensic science personal identification.

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Conflict of Interest

Authors declare no conflict of interest.

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