# UNIQUENESS OF GROOVE PATTERNS OF MAXILLARY POSTERIOR TEETH IN HUMAN IDENTIFICATION

Samiya Riaz<sup>1</sup>, Mohd Fadhli Khamis<sup>1,\*</sup>, Johari Yap Abdullah<sup>2</sup>, Wan Muhammad Amir Wan Ahmad<sup>3</sup> and Mohammad Khursheed Alam<sup>4</sup>

<sup>1</sup>Unit of Oral Biology, School of Dental Sciences, Universiti Sains Malaysia, 16150 Kota Bharu, Kelantan, Malaysia

<sup>2</sup>Craniofacial Imaging Laboratory, School of Dental Sciences, Universiti Sains Malaysia, 16150 Kota Bharu, Kelantan, Malaysia

<sup>3</sup>Department of Biostatistics, School of Dental Sciences, Universiti Sains Malaysia, 16150 Kota Bharu, Kelantan, Malaysia

<sup>4</sup>Department of Preventive Dentistry, College of Dentistry, Jouf University, 42421 Sakakah, Saudi Arabia

\*fadhli@usm.my

**Abstract**. In forensic odontology, unique features are important concepts for the antemortem and postmortem dental record comparisons. There are studies on the uniqueness of soft and hard tissue structures. However, there is insufficient data specifically on the uniqueness of groove patterns (GP). Therefore, this study was conducted to determine the uniqueness of the maxillary first molar (M1), first premolar (PM1), and second premolar (PM2) using a twodimensional (2D) stereomicroscope for human identification. Ninety dental casts were selected for scanning. Two-dimensional images of M1, PM1 and PM2 were captured with Hirox KH-7700 Digital Microscope System (Japan). The GP tracings followed the central developmental groove and any supplementary groove emerging from it. The GPs were then duplicated into original and duplicate sets of images by examiner-A. Examiner-B decoded the original and duplicate sets of images and made 90 matched and 90 non-matched pairs of GPs of each of M1, PM1, and PM2, which were superimposed by examiner-A using 2D-Hirox. Examiner-B cross-checked to record the correct and incorrect decisions. Examiner-A gave the correct decision for all the pairs of PM1, PM2 and MI, which showed 100 % uniqueness. It was concluded that maxillary M1, PM1, and PM2 showed uniqueness of GP, and hence, may be used for human identification.

**Keywords:** groove pattern, superimposition, maxillary teeth, 2D, uniqueness

## **Article Info**

Received 3<sup>rd</sup> February 2023 Accepted 17<sup>th</sup> April 2023 Published 1<sup>st</sup> May 2023

Copyright Malaysian Journal of Microscopy (2023). All rights reserved.

ISSN: 1823-7010, eISSN: 2600-7444

#### Introduction

Mass disasters, whether they are accidental, criminal, or natural, ended up with many human bodies with a lack of data, resulting in poor identification. Decomposition of the soft tissues leaves us without any traces of fingerprints and face recognition. Furthermore, Deoxyribonucleic acid (DNA) analysis becomes difficult to be performed on such a large scale. Hence, the importance of deceased person identification has been frequently raised for multiple or single causalities.

Studies have shown the uniqueness of palatal rugae and lip prints that may help in gender estimation and positive identification [1,2]. But these techniques rely on degradable soft tissues, hence, limiting their use as a tool for positive identification. Enamel rod pattern analysis is an extensive procedure and requires expertise [3]. Forensic odontologists, then, have to rely on some other biological evidence, which may be present in the healthy teeth.

Alternatively, INTERPOL has approved dental morphological features as one of the most reliable primary sources of human identification [4]. The strong resistance of tooth to force, heat and chemicals allows its availability to the forensic odonatological investigations [5]. There are two primary blocks of dental evidence required for solving the puzzle of true or positive identification. These are the dental remains and accurate antemortem records. True or positive identification demands matching of unique features in antemortem and postmortem records such as any restoration, prostheses, missing or supernumerary teeth, or any structural anomaly [6]. The presence of all healthy and sound teeth makes it difficult to proceed with the comparative identification. In such scenarios, structural anatomy of the maxillary and mandibular tooth surface components can be utilized as a reliable tool in human identification.

Just like the blood types, fingerprints, and DNA, human teeth are also a chronicled biological heritage [7]. Each tooth type has its own morphology, which may contribute to true or positive identification. The occlusal surface of posterior teeth has numerous elongated depressions known as the groove patterns (GPs). The GPs are formed because of the invagination of the enamel organ during the 6<sup>th</sup> intrauterine week [8]. There is scarce data specifically based on the individuality of the GP of maxillary posterior teeth [7]. To date, no previous study has been conducted on the uniqueness of GP of maxillary posterior teeth in the Malay population. This data can be easily obtained using digital photography which has emerged as an easy method to collect, analyse, and store data in a non-invasive method. Several methods of digital photography have been established using multiple equipments. Hirox KH-7700 Digital Microscope System (Japan) is considered a reliable tool with an accuracy of  $0.1 \times 10^{-6}$  [9]. Therefore, the aim of this study was to evaluate the individuality of GP of maxillary first molar (M1), first premolar (PM1), and second premolar (PM2) in a Malay population using 2D-Hirox stereomicroscope.

#### **Materials and Methods**

## Scope

This was a cross-sectional study of retrospective record review that was conducted in the School of Dental Sciences, University Sains Malaysia (USM) on a Malay population. The ethical approval was obtained from the USM Ethics Committee [USM/JEPeM/17100564],

which complies with the Declaration of Helsinki (1964). Ninety dental casts (45 males and 45 females) were retrieved from the Orthodontic Department, retrospectively. Inclusion criteria were Malay origin; age range was from 13-25 years old; maxillary PM1, PM2, and M1 teeth with clearly demarcated GP for easy tracing. Exclusion criteria were the anterior teeth, maxillary 2<sup>nd</sup>, and 3<sup>rd</sup> molars; any tooth with restorations; any dental anomaly that may obstruct the GP tracing; broken or distorted casts or with bubbles on the occlusal surface of the teeth.

## Sample Size Calculation

The sample size was calculated using G\* Power software (Germany), using t-test for matched pairs. Power was 0.80, alpha as 0.05, two-tailed with effect size as 0.5. Although the minimum sample size required was 34, a larger sample size (90 samples) was selected for this study due to data availability.

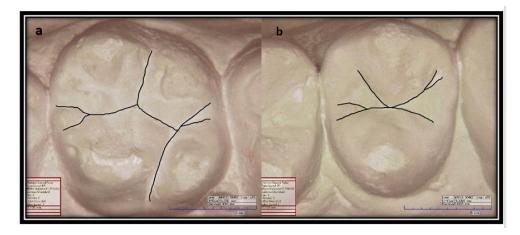
#### Data Collection

Two individuals were directly involved (i) Examiner A, who worked with the dental casts, and (ii) Examiner B, who worked with the images of the GP tracings.

Initially, ninety maxillary dental casts were scanned using Hirox KH-7700 stereomicroscope Digital Microscope System (Japan) to transform the dental casts into 2D digital images by Examiner A. The images were standardized by the auto calibrated system regardless of the distance between the lens and the dental cast. The images of the occlusal surface of each individual tooth (maxillary PM1, PM2 and M1) were captured at 20x magnification with low resolution lens. The images of 300 dpi were saved as jpeg. files on hard drive.

Each file was individually loaded on the Hirox software by Examiner A and subsequently the GP was traced. Tracing was done by marking the developmental groove and the accessory grooves or supplemental grooves that are attached or in continuity with the developmental groove (Figure 1). After tracing, the tooth image from the background was removed and the image of the GP tracing only was again saved as jpeg. file. The images were then duplicated into another set. Finally, we had two sets of images, 90 original GP tracings labelled as original and the other 90 of duplicate GP tracings named as duplicate.

The original and the duplicated images were re-coded by Examiner B while the Examiner A was blinded. The Examiner B made 180 pairs of each tooth GP tracings. The pairs included the original and duplicated images of the same tooth type as 90 matched pairs (Figure 2), and 90 as non-matched pairs (Figure 3). The pairs were made irrespective of the age and gender since the aim of the study was to define a method for 'exact' or 'confirmed' identification of a human body. Examiner B made the matched and non-matched pairs within and between male and female GP tracings.



**Figure 1:** Tracing of the groove pattern of (a) maxillary first molar tooth (M1) and (b) maxillary premolar tooth (PM)

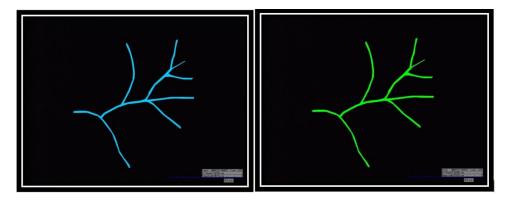


Figure 2: Matched pair of images from original and duplicate image sets

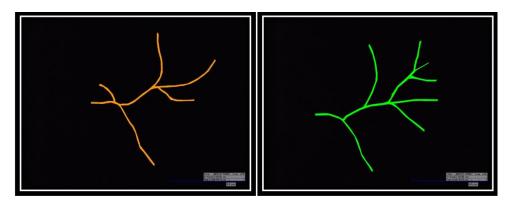
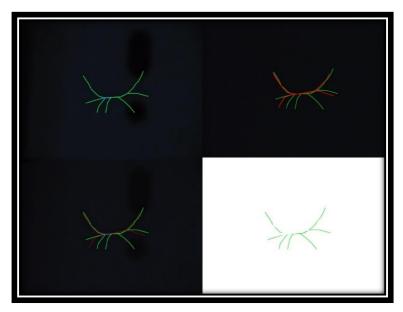


Figure 3: Non-matched pair of images from original and duplicate image sets

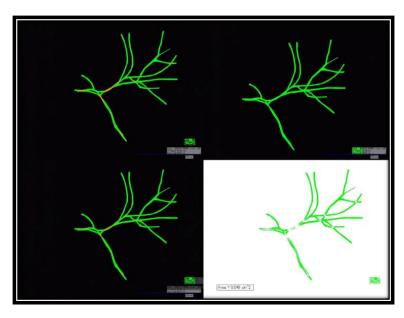
The pairs were then compared by the Examiner A using the superimposition method (compare) in the Hirox software. In this method, 2 images were selected, one from each group of original and duplicate images following the pairs made by the examiner B. Once loaded in the software, box 1 and box 2 showed the GPs selected. The two GPs appeared overlapping in each other in the box 3. The GPs were aligned to overlap the maximum tracing of the developmental groove. The software detected the overlapping GPs. The parts

that remained non-overlapped appeared in box 4 as a result (Figures 4 and 5). This was considered a true negative and marked as unmatched by Examiner A. On the other hand, if the two GP tracings completely overlapped each other, box 4 appeared empty indicating 100 % overlapping (Figure 6). This was considered a true match and marked as "match" by Examiner A. The same procedure was followed for all the pairs of PM1, PM2 and M1 GP tracing pairs and the results were recorded in the excel data sheets. The pairs were recorded according to the same tooth type.

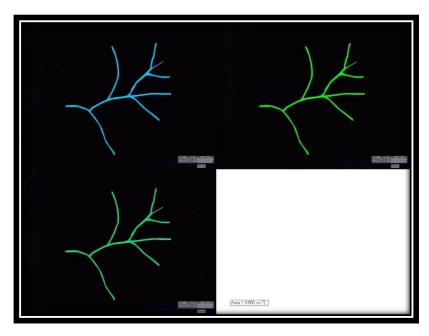
After Examiner A completed all the comparison, Examiner B then observed the results and finally marked them "true" or "false" by comparing the results to the original coding.



**Figure 4:** Incomplete overlapping of the pair of GP tracings of premolar with white box showing part sof tracings that did not overlap.



**Figure 5:** Incomplete overlapping of the pair of GP tracings of molar with white box showing parts of tracings that did not overlap.



**Figure 6:** Complete overlapping of the image pair, white box appeared empty indicating complete overlapping.

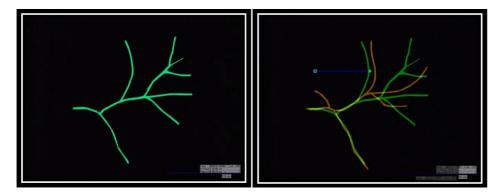


Figure 7: Matched (a) and non matched (b) pairs marked by Examiner A.

# Data Analysis

Data was analysed by Examiner B to determine the decision of Examiner A. If Examiner A made the correct decision, Examiner B marked it as "true". Vice versa, if the decision was wrong, Examiner B marked it as "false".

## **Results and Discussion**

One hundred and eighty pairs of each tooth type (PM1, PM2, and M1) were examined and compared by superimposition. All the pairs for PM1, PM2, and M1 were correctly detected as true positive or true negative by Examiner A. Thus, this demonstrated 100 % uniqueness or individuality of the groove patterns for these teeth.

This study was conducted to determine the uniqueness of teeth as a tool for human identification [7]. Our study focused on the uniqueness of posterior teeth as they are multicusped teeth and have different groove patterns. Dental casts were used for data collection in this study as they are still the common method of data storage. The expensive procedures of 3D scanning restrict most of the hospitals and the clinics to avail that opportunity. In this study, the Examiner B made pairs without gender consideration. The mixed pairing strengthened the study design for use in human identification cases in which the parts of the human remains that could be utilised for gender determination are either degraded or missing.

In this new era of digital dentistry, digital photography has evolved as an easy method to collect, scan, analyse and interpret the data. Storage has also become feasible. Digital photography also carries importance in forensic investigation due to the ease of their use and its non-invasive nature. Hirox digital stereomicroscope Digital Microscope System has been reported to have an accuracy of  $0.1 \times 10^{-6}$  mm [10]. This is the first time Hirox has been used for method establishment in forensic dental investigations using groove pattern tracings. In our study, photographic method of recording the occlusal surface of the teeth was standardised as Hirox had auto calibrated system, which resulted in capturing standardised images regardless of the distance between the lens and the object. Moreover, the high-quality images captured with good resolution made it easy to trace the groove patterns.

Forensic odontology plays a major role in human identification. Tooth has been declared as one of the primary identifiers by the INTERPOL in disaster victim identification. Metric and non-metric features have been studied to analyse their use as a tool in forensic investigations. Groove patterns are the lines present on the occlusal surface of posterior teeth that mark the cusp boundaries. Since tooth has 96 % inorganic content, it is the most mineralized tissue of the human body [11]. This is the prime factor in its ability to withstand high temperatures and disaster intensities; hence, making it readily available for forensic investigations in most of the situations. This is of importance in cases where fingerprints and face recognition become impossible due to the deterioration of the soft tissues. Furthermore, DNA analysis becomes difficult due to high risk of contamination in such scenarios.

In our study, it was noticed that the groove patterns can play a major role in comparative identification. Another study conducted found similar findings using photographs and superimposing them using J image ImageJ software [7]. However, this software does not have auto calibration system. Our studies showed that groove patterns are quite unique in terms of their pattern. Maxillary molar teeth showed 100 % uniqueness. This is because, the groove patterns are not only made up of the central or the developmental groove, but they are accompanied by multiple supplementary grooves giving each groove pattern a unique appearance.

The formation of developmental groove occurs in the early bell stage or the histodifferentiation stage of tooth formation. At this time enamel organ is formed, which causes the deposition of enamel and the dentine matrix [8]. The invagination of this enamel organ later corresponds to the developmental groove. The other small wrinkles that appear along with this invagination manifest as the supplementary grooves [12]. As a consequence, the formation of enamel knots and succeeding epithelial folding in embryonic tooth germs can be closely associated to the ultimate morphology of erupted teeth. The enamel knot is, therefore, considered to be a signalling centre to control the patterning of the tooth cusps. Certain unique signalling molecules, including Shh, BMP2, BMP4, BMP7, FGF4, and FGF9, are expressed in the enamel knot [13]. Hence, tooth shape is determined early as the dentin

and enamel matrices mineralize at the interface between epithelium and mesenchyme and has minimal influence on tooth morphology after the development is complete.

The process of tooth formation is also dependent on various growth factors, which are further interacted with transcription factors [14]. Homeobox 9 gene (PAX9), MSX1, MSX2, DLX3, DLX5, DLX6, DLX7, BARX1, PITX2, LHX6, and LHX7 are the transcription factors involved in the process of tooth development. Signals from the transforming growth factors (TGF-β) and fibroblast growth factor (FGF) families became the first to be explored in developing teeth, shortly followed by signals from the Hedgehog, Wnt, and Notch families [15].

Environmental factors have an effect on the variation in the cusp [16], which may further enhance the uniqueness in groove pattern. The presence or absence of an extra cusp like metaconule may alter the groove pattern since all the cusps are separated by the developmental groove. This means the addition of any cusp, or the absence of any cusp will result in the diversion of a part of groove pattern. Similarly, presence or absence, as well as the degree of expression of carabelli trait effects the size of the protocone cusp and eventually the crown size, especially in maxillary first molars [17], which may result in anatomical variation. Moreover, the hypocone has similar effect, too, based on its degree of expression [18].

Any dental anomaly may also alter the crown shape as well as the occlusal pattern. as Dens evaginatus is a developmental anomaly that results in infolding of enamel and dentine sometimes extending up to pulp and the roots. This infolding results in the formation of tubercles. Tubercles can be present at any location on the tooth occlusal surface of posterior teeth and lingual surface of the anterior teeth [19]. In maxillary molars, paramolar tubercle is expressed on paracone cusp, and in rare occasions on the metacone cusp [20]. It varies in dimensions and pattern from a single pit and groove to prominence like a cusp to surface irregularities. The irregularities cause further anatomical variations.

It has been observed that certain traits run in an ethnicity resulting in similarities within an ethnic group. In our study, it was noted that even within an ethnicity there was uniqueness. This suggests that the groove patterns are distinct from one another.

### **Conclusions**

In conclusions, there is uniqueness in groove patterns of maxillary posterior teeth in the Malay ethnic population. Therefore, groove patterns of posterior teeth can be considered a reliable tool for uniqueness in human identification. Extensive research should be conducted in future on the other ethnic groups and populations to establish the importance of groove pattern uniqueness. Digital image capturing with use of 2D-Hirox Digital Microscope System was easy, cost effective, and non-invasive. Hence, the experts may benefit from this technique and further elaborate the uniqueness of other teeth as well, such as the permanent mandibular posterior teeth and the deciduous teeth.

## Acknowledgements

The financial support for this study was provided by the Ministry of Higher Education Malaysia for Fundamental Research Grant Scheme with Project Code. FRGS/1/2022/SKK11/USM/02/1.

#### **Author Contributions**

Authors contributed toward data analysis, drafting, and critically revising the paper and agree to be accountable for all aspects of the work.

#### **Disclosure of Conflict of Interest**

The authors have no disclosures to declare.

## **Compliance with Ethical Standards**

The work is compliant with ethical standards. The ethical approval was obtained from the USM Ethics Committee [USM/JEPeM/17100564], which complies with the Declaration of Helsinki (1964).

#### References

- [1] Franco, A., Willems, G., Souza, P. H. C., Bekkering, G. E. & Thevissen, P. (2015). The uniqueness of the human dentition as forensic evidence: a systematic review on the technological methodology. *International Journal of Legal Medicine*. 129(6), 1277–1283.
- [2] Dineshshankar, J., Ganapathi, N., Yoithapprabhunath, T. R., Maheswaran, T., Kumar, M. S. & Aravindhan, R. (2013). Lip prints: Role in forensic odontology. *Journal of Pharmacy and Bioallied Sciences*. 5(SUPPL.1), S95-S97.
- [3] Naziya, J., Sunil, S., Jayanthi, R., Rathy, R. & Harish, R. K. (2019). Analysis of enamel rod end pattern for personal identification. *Forensic Corner*. 23(1), 1-5.
- [4] INTERPOL. Disaster victim identification. DVI guide: INTERPOL. (2018). [Online]. [Accessed December 16 2022]. Available from World Wide Web: https://www.interpol.int/INTERPOL-expertise/Forensics/DVI-Page s/DVI-guide
- [5] Peckmann, T. R., Logar, C., Garrido-Varas, C. E., Meek, S. & Pinto, X. T. (2016). Sex determination using the mesio-distal dimension of permanent maxillary incisors and canines in a modern Chilean population. *Science & Justice*. 56(2), 84–89.
- [6] Campobasso, C. P, Dell'Erba, A. S., Belviso, M. & Di Vella, G. (2007). Craniofacial identification by comparison of antemortem and postmortem radiographs. *The American Journal of Forensic Medicine and Pathology*. 28(2), 182–186.

- [7] Roy, J., Rohith, M. M., Nilendu, D. & Johnson, A. (2019). Qualitative assessment of the dental groove pattern and its uniqueness for forensic identification. *Journal of Forensic Dental Sciences*. 11(1), 42-47.
- [8] Nanci, A. (2018). Development of the tooth and its supporting tissues. In *TenCate's Oral Histology: Development, Structure and Function*. 9<sup>th</sup> edition (Elsevier, St. Louis) pp. 68–86.
- [9] Shahid, F., Alam, M. K., Khamis, M. F., Komori, A., Kubo, K. & Maeda, H. (2015). A new method to measure and assess tooth size and tooth size discrepancy via circumferential measurements using stereomicroscope. *Journal of Hard Tissue Biology*. 24(4), 305–310.
- [10] Shahid, F., Alam, M. K., Khamis, M. F., Honda, Y., Sugita, Y. & Maeda, H. (2015). Geomorphometrics of tooth size and arch dimension analysis by conventional digital caliper and digital stereomicroscope to establish standard norms for the Pakistani population. *Journal of Hard Tissue Biology*. 24(2), 155–168.
- [11] Gutiérrez-Salazar, M. del P. & Reyes-Gasga, J. (2003). Microhardness and chemical composition of human tooth. *Materials Research*. 6(3), 367–373.
- [12] Nelson, S. J. & Ash, M. M. (2019). Development and eruption of the teeth. In *Wheeler's Dental Anatomy, Physiology, and Occlusion*. 11<sup>th</sup> edition. (Elsevier, St. Louis, Missouri) pp. 21-41.
- [13] Brook, A. H., Jernvall, J., Smith, R. N., Hughes, T. E. & Townsend, G. C. (2014). The dentition: The outcomes of morphogenesis leading to variations of tooth number, size and shape. *Australian Dental Journal*. 59(SUPPL. 1), 131–142.
- [14] de Sousa-Romero, L. (2016). Growth and transcription factors in tooth development. *International Journal of Oral Craniofacial Sciences*. 2, 15–29.
- [15] Thesleff, I. & Mikkola, M. (2002). The role of growth factors in tooth development. *International Review of Cytology*. 217, 93–135.
- [16] Dempsey, P. J. & Townsend, G. C. (2001). Genetic and environmental contributions to variation in human tooth size. *Heredity* (Edinb). 86(6), 685–693.
- [17] Kondo, S. & Townsend, G. C. (2006). Associations between carabelli trait and cusp areas in human permanent maxillary first molars. *American Journal of Physical Anthropology*. 129(2), 196–203.
- [18] Takahashi, M., Kondo, S., Townsend, G. C. & Kanazawa, E. (2007). Variability in cusp size of human maxillary molars, with particular reference to the hypocone. *Archives of Oral Biology*. 52(12), 1146–1154.
- [19] Levitan, M. E. & Himel, V. T. (2006). Dens evaginatus: literature review, pathophysiology, and comprehensive treatment regimen. *Journal of Endodontics*. 32(1), 1–9.
- [20] Colakoglu, G., Kaya, Buyukbayram, I., Elcin, M. A., Kazak, M. & Sezer, H. (2020). Evaluation of the internal anatomy of paramolar tubercles using cone-beam computed tomography. *Surgical and Radiologic Anatomy*. 42(1), 15–21.