

**THREE-DIMENSIONAL SUPERIMPOSITION
ANALYSIS OF OCCLUSAL SURFACE OF UPPER
FIRST PREMOLAR FOR HUMAN
IDENTIFICATION**

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UNIVERSITI SAINS MALAYSIA

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by

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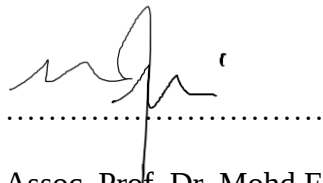
**Thesis submitted in partial fulfilment of the requirements
for the degree of
Bachelor of Science in Forensic Science**

February 2025

CERTIFICATE

This is to certify that this dissertation entitled “Three-Dimensional Superimposition Analysis of Occlusal Surface of Upper First Premolar for Human Identification” is the bona fide record of research work done by Safa Mardiah Binti Mohamed Radzi during the period of October 2024 to February 2025 under my supervision. I have read this dissertation and that in my point of view it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope, and quality, as a dissertation to be submitted in partial fulfilment for the degree of Bachelor of Science in Forensic Science.

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DECLARATION

I hereby declare that this dissertation is the result of my investigation, except where otherwise cited and duly acknowledged. I also declare that it has not been previously or concurrently submitted as a whole for any degrees at Universiti Sains Malaysia or any other institution. I grant Universiti Sains Malaysia the right to use the dissertation for teaching, research, and promotional purposes.

A handwritten signature in black ink, appearing to read 'Safa', is written over a horizontal dotted line.

Safa Mardiah Binti Mohamed Radzi

Date: 26/2/2025

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LIST OF SYMBOLS

$\pm$	Plus or minus
κ	Cohen's Kappa Index
α	Alpha
β	Beta
%	Percentage

LIST OF ABBREVIATIONS

2D	Two-Dimensional
3D	Three-Dimensional
AM	Antemortem
AI	Artificial Intelligence
CBCT	Cone Beam Computed Tomography
DVI	Disaster Victim Identification
DNA	Deoxyribonucleic Acid
ICP	Iterative Closest Point
IOS	Intraoral Scanner
PCA	Principal Component Analysis
PI	Principal Investigator
PM	Postmortem
RMS	Root Mean Square
ROI	Region of Interest
TEM	Technical Error of Measurement
SD	Standard Deviation
STL	Stereolithography
SV	Supervisor
UP1	Right Upper First Premolar
USM	Universiti Sains Malaysia

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KEUNIKAN PERMUKAAN OKLUSAL GERAHAM KECIL PERTAMA MAKSILA UNTUK MENGIDENTIFIKASI MANUSIA

ABSTRAK

Pengenalpastian forensik pergigian menggunakan kaedah perbandingan bergantung kepada ciri unik struktur gigi untuk mengidentifikasi seseorang individu. Kajian ini bertujuan untuk menentukan kebolehpercayaan dan keunikan permukaan oklusal geraham kecil pertama maksila bagi tujuan pengenalpastian manusia. Kajian keratan rentas ini menggunakan model gigi arkib retrospektif daripada individu Melayu Kelantan, yang diperoleh daripada Klinik Ortodontik Hospital USM. Sebanyak 90 pasangan model pergigian 3 dimensi dianalisis, terdiri daripada 45 pasangan sepadan dan 45 pasangan tidak sepadan. Permukaan oklusal diasingkan menggunakan perisian 3-Matic Mimics (Medit Corp, Korea Selatan), dan analisis superimposisi dijalankan menggunakan perisian CloudCompare (CloudCompare, Paris, Perancis). Analisis intra-pemeriksa dan antara pemeriksa ke atas 18 pasang gigi yang sepadan mencapai persetujuan sempurna dengan nilai Cohen's Kappa sebanyak 1.00, sekali gus mengesahkan 100% kebolehpercayaan bagi kedua-dua ujian. Dalam keadaan kajian buta, hanya nilai punca kuasa dua (RMS) digunakan untuk membezakan antara 45 pasang yang sepadan dan 45 pasang yang tidak sepadan. Purata RMS untuk pasangan yang sepadan ialah 0.011 ± 0.005 mm, manakala untuk pasangan yang tidak sepadan ialah 0.219 ± 0.059 mm. Nilai ambang RMS yang ditetapkan pada 0.1 mm berjaya mengenal pasti 100% pasangan yang sepadan dan tidak sepadan tanpa sebarang pertindihan antara kedua-dua kumpulan ($p < 0.05$), sekali gus mengesahkan keunikan permukaan oklusal premolar pertama maksila.

UNIQUENESS OF OCCLUSAL SURFACE OF UPPER FIRST PREMOLAR FOR HUMAN IDENTIFICATION

ABSTRACT

Forensic dental identification using the comparative method relies on the unique features of dental structures to establish an individual's identity. This study aimed to determine the reliability and uniqueness of the occlusal surface of the maxillary first premolar for human identification. A cross-sectional study was conducted using retrospective archived dental casts of Kelantanese Malays obtained from the Orthodontic Clinic of Hospital USM. A total of 90 dental casts were scanned into digital models using the Medit i500 intraoral scanner (Medit Corp, South Korea). The occlusal surfaces were isolated using 3-Matic software (Materialise NV, Belgium), and superimposition analysis was performed using CloudCompare software (CloudCompare, Paris, France). The intra-examiner and inter-examiner analyses on matched 18 pairs achieved a perfect agreement with a Cohen's Kappa value of 1.00, thus confirming 100% reliability for both tests. Under blind study conditions, only the Root Mean Square (RMS) values were used to distinguish between 45 matched and 45 unmatched pairs. The mean RMS for matched pairs was 0.011 ± 0.005 mm, while for unmatched pairs it was 0.219 ± 0.059 mm. The established RMS threshold of 0.1 mm correctly identified 100% of matched and unmatched pairs, with no overlap between the two groups ($p < 0.05$), hence confirming the uniqueness of the occlusal surface of the maxillary first premolar.

CHAPTER 1

INTRODUCTION

1.1 Introduction

Human identification is an important process in criminal investigations and mass disaster responses. Its primary goal is to establish the identity of individuals through the analysis of various biological and physical characteristics. INTERPOL has provided a comprehensive guide for human identification, including three primary and two secondary identification methods. Primary identifiers include DNA analysis, friction ridge analysis of fingerprints, and dental records, which are highly reliable due to their uniqueness and resistance to degradation (Perkins *et al.*, 2024).

Fingerprint analysis is widely used due to the efficiency of digital retrieval and systematic comparison within fingerprint databases (Interpol, 2023). Forensic odontology identifies individuals by comparing antemortem and postmortem dental records, using features such as caries, missing teeth, and dental treatments as markers. Custom prostheses, including implants and dentures, can provide unique identifiers, sometimes with serial numbers that aid in positive identification (Adserias-Garriga *et al.*, 2024). When antemortem records are unavailable, teeth can offer insights into age, diet, and ancestry, which helps in narrowing down the potential matches (Interpol, 2023).

Among these methods, DNA analysis is often considered the gold standard by enabling the matching of a deceased individual's DNA to reference samples (Kayser, 2015). These primary identification methods are often used together to cross-verify evidence and the choice of method depends on the available body structures for examination (Interpol, 2023). In the case of the absence of primary identifiers, a

combination of secondary identifiers can help in the identification process. This includes medical information and evidence such as jewellery or clothing.

DNA analysis is the most reliable method for personal identification, but its high cost, time-consuming, and need for specialised personnel make it impractical for large-scale forensic applications (Putrino *et al.*, 2020). Similarly, although fingerprint analysis is very precise and commonly utilised, its usefulness is greatly reduced when dealing with environmental factors or postmortem changes, especially in cases of decomposed, fragmented, or charred remains, as it becomes difficult or impossible to collect fingerprints.

Among the primary methods, dental identification is particularly significant due to the durability of teeth and their unique morphological features. True identification of teeth is possible either from the comparison of antemortem and postmortem data or by dental profiling (Pretty & Sweet, 2001). Commonly, if the anatomical, morphological or other traits that are natural or acquired through treatment represented in the antemortem and postmortem dental records are the same, it can be identified as the same person (Du *et al.*, 2021). This method of identification is referred to as comparative dental identification, as a presumptive identification exists and an antemortem record is available for confirmation of the hypothesis.

To date, there is a lack of specific publication on the uniqueness of occlusal features of the first maxillary premolar to be used as a feature for human identification. Scientific proof of the uniqueness of this surface is important to prove the fundamental of scientific evidence in the identification process.

1.2 Problem Statement

Dental identification has traditionally relied on unique dental treatments, restorations, and caries to establish positive matches. However, with the improvement in oral hygiene and preventive dental care, the prevalence of such unique identifiers has decreased, posing a significant challenge in cases where antemortem records lack distinctive dental work for comparison. This limitation necessitates the exploration of alternative dental features that remain stable over time and can serve as reliable identifiers. While previous studies have examined dental morphology for forensic applications, the uniqueness of the occlusal surface of maxillary premolars has not been systematically evaluated.

Up to now, there has been a lack of studies discussing the uniqueness of occlusal features of maxillary premolars in the Malay population. Furthermore, the details regarding its uniqueness in 3D dental models for human identification in a primary study have not been well-documented. To comply with the Daubert rule, these techniques and possible forensic significance need to be scientifically validated.

1.3 Justification of the study

The occlusal surface of the maxillary first premolar holds significant potential as a unique identifier in human identification due to its highly individualised morphological features. Unlike other dental identification methods that rely on treatments or restorations, this approach is particularly useful for individuals with healthy intact teeth where traditional markers are absent. The intricate patterns of grooves cusps and fissures on the occlusal surface are naturally distinct and remain consistent over time, making them a reliable source for identification even in challenging conditions such as degraded remains or fragmented dental records.

Establishing the forensic validity of this trait can expand identification options in cases where DNA and fingerprint analysis are unavailable or inconclusive.

The use of 3D superimposition technology further enhances the accuracy and reliability of this method by allowing for precise comparisons between antemortem and postmortem dental records. Current practices in forensic odontology often face limitations when dealing with intact teeth as they lack the unique markers provided by dental work. This can lead to inconclusive results, particularly in cases involving individuals with minimal or no dental interventions. 3D superimposition addresses this issue by enabling detailed and objective analysis of the occlusal surface, reducing subjectivity and improving the overall efficiency of the identification process. This technique complements existing methods and provides a more robust framework for analysing dental morphology. In conclusion, this thesis focused on exploring the occlusal surface of the first upper premolar as a unique identifier and proposing 3D superimposition as a transformative tool to advance the field of human identification.

1.4 Objectives

1.4.1 General

To describe the uniqueness of the occlusal surface of the maxillary first premolar and evaluate its potential as a reliable feature for human identification in forensic science.

1.4.2 Specific

1. To determine the reliability of the superimposition of the occlusal surface of the maxillary first premolar.

2. To determine the uniqueness of the occlusal surface of the maxillary first premolar.

1.5 Research Question(s)

1. Is the superimposition of the occlusal surface of the maxillary first premolar reliable?
2. Is the occlusal surface of the maxillary first premolar unique for human identification?

1.6 Hypotheses

1. The superimposition of the occlusal surface of the maxillary first premolar is reliable.
2. The occlusal surface of the maxillary first premolar is unique for human identification.

CHAPTER 2

LITERATURE REVIEW

2.1 Human Identification Process

Human identification techniques have evolved which involved using both traditional techniques such as fingerprint analysis and dental records and modern techniques such as DNA analysis to ensure accurate and reliable results. Among these methods, dental evidence holds significant importance in terms of uniqueness and resistance under extreme conditions. It becomes the primary means of identification in cases of severely damaged, decomposed, or burnt corpses, especially in challenging scenarios such as mass disasters or criminal activities where visual identification is impossible (Sehrawat *et al.*, 2019).

Teeth are the hardest and most well-protected structures in the human body. They resist decomposition and high temperatures, often remaining intact long after other tissues have degraded. The oral cavity and teeth are also unique to each individual and this provides a reliable basis for identification (Krishan *et al.*, 2015). The process of dental identification typically involves comparing postmortem dental evidence recovered from the scene with antemortem records. Antemortem data refers to data collected before death, such as dental records, images, and dental casts taken during a person's lifetime, while postmortem data refers to data collected after death such as X-rays and descriptive dental records, usually as part of an autopsy or forensic identification (Franco *et al.*, 2019).

High forensic values antemortem records consist of information on the presence and missing teeth, restored teeth, oral diseases, anomalies, and prostheses. This method aligns with INTERPOL standards, which recognise dental analysis alongside scientific methods of DNA and fingerprint as primary identification methods. While the

importance of dental structures in human identification is well-established, there is an ongoing need for more precise and technologically advanced methods of comparison.

The comparative process is significantly hampered if the dentition status is all intact and the unavailability of radiographic images. Thus, there is a need to explore using intact teeth for comparison between antemortem and postmortem records. Nowadays, intraoral digital scanners have been used widely in dental clinics. Therefore, high chance of the availability of 3D images of teeth structures as antemortem evidence.

2.2 Dental Records in Forensic Identification

Dental records play a crucial role in forensic identification, particularly when other methods such as fingerprinting or DNA analysis are unavailable or inconclusive. As defined by Abdul Rahman *et al.* (2024), a dental record is "any item of information specifically related to a patient, regardless of its form or medium, and is created or received by a practitioner, dental office, or any health-related institution, as part of providing care and conducting dental service." These records provide detailed information about an individual's anatomy, including features like tooth presence, morphology, restorations, periodontal tissue characteristics, pathologies, and other anatomical features (Nilceu *et al.*, 2011). These records are important as they can be used to match antemortem dental data with postmortem data in the human identification process. Historically, forensic odontologists have relied on manual approaches to compare dental records by examining various dental features.

The comparison process involving unique dental features has proven effective, with forensic odontology classically identifying about 60% of victims in mass disaster scenarios and contributing to an additional 30% of identifications in collaboration with

other methods (Forrest, 2019). However, the accuracy of identification depends on the availability and quality of the antemortem records. Therefore, forensic odontology has significantly evolved particularly in terms of record-keeping and comparison methodologies. Dental records can be in the form of written treatment records, 2D and 3D images including radiographs, dental casts and other dental items. According to Forrest (2019), while written records have been the traditional standard, they are subjective and prone to error. Two-dimensional images like radiographs and photographs offer more objectivity but are most effective when used in conjunction with written records.

Dental records now can be kept in digital form as they offer several advantages including long-term storage capabilities, easy backup, rapid transmission and preservation of original quality and resolution (Vasilakos *et al.*, 2017). Digital records include high-resolution 2D and 3D images, electronic charts, and scanned data that can be directly compared with postmortem findings. As internet-based dental charts become increasingly prevalent, the forensic identification process, particularly in mass disaster scenarios, is expected to become more accurate and efficient, suggesting the need to use reliable and rapid digital methods (Nagi *et al.*, 2019).

2.3 Comparative Methods in Dental Identification

Dental identification offers two primary approaches for human identification: comparative and reconstructive identification. While reconstructive methods aim to generate a profile from post-mortem data, comparative techniques involve matching antemortem (AM) and postmortem (PM) records. This research focuses on the comparative approach, which is valued for its simplicity, cost-effectiveness, and reliability (Almeida *et al.*, 2015). Traditional comparative methods in dental

identification typically involve visual comparison, dental charting, and radiograph analysis. The process begins with obtaining AM records, often facilitated by presumptive identification through personal effects like wallets or ID cards. Forensic dentists then produce PM records through charting and detailed descriptions of dental structures and radiographs. A methodical and systematic comparison must be applied in examining each tooth and surrounding structures (Pretty & Sweet, 2001).

The comparison process involves noting similarities and discrepancies between AM and PM records. Discrepancies can be categorised as either explainable (e.g., teeth extracted, or restorations placed after AM data collection) or unexplainable (e.g., tooth present in PM but absent in AM records). According to Nagi *et al.* (2019), regardless of the identification method, the comparison of AM and PM data will result in one of the following outcomes:

- I. Positive identification: Sufficient AM and PM data match with no unexplainable discrepancies.
- II. Probable identification: High concordance level, but lack of radiographic support or quality data.
- III. Presumptive identification: Missing information from either source preventing a definitive conclusion.
- IV. Insufficient identification: Insufficient evidence is available to make a scientifically based conclusion.
- V. Exclusion: Discrepancies between the data sets exist and are equally important as positive identification.

There is no minimum number of concordant points required for positive identification that even a single unique tooth can be sufficient for identification (Pretty & Sweet, 2001). This can be illustrated by a case study, where a single and very

distinctive therapeutic identifier on the central incisor contributed to positive human identification in charred remains (Silva *et al.*, 2021).

The comparative method typically involves examining unique dental restorations or implants, such as fillings, crowns, and bridges, as well as malocclusions or dental fractures (Ata-Ali & Ata-Ali, 2014). However, in cases where individuals have healthy teeth with no evidence of decay or restorative treatments, alternative methods of comparison are necessary. Studies like Nomir & Abdel-Mottaleb (2007) emphasise the use of dental morphology, specifically the shape and appearance of teeth, for identification through radiographic comparison.

Unique dental features such as occlusal patterns, groove patterns, and other anatomical features have been increasingly recognised as reliable human identifiers. These anatomical identifiers can be classified as dental, such as the shape and spatial relationship of the teeth and palatal rugae (Gibelli *et al.*, 2018), or non-dental, such as the configuration of the frontal sinus and nasal septum (Saraswathi & Prakash, 2016). Franco *et al.* (2015) highlighted the significance of occlusal patterns in providing individual-specific data through bite mark analysis or detailed examination of dental casts, while Roy *et al.* (2019) and Riaz *et al.* (2023) demonstrated the uniqueness of groove patterns, where no two molar grooves were identical.

Hori *et al.* (2020) also introduced a method for identifying individuals using only molar occlusal surface data by converting 3D STL files into low-frequency 2D images and achieving over 90% accuracy through perceptual Hash and Hamming distance functions. These findings highlight the possibility of using detailed morphological analysis and exploring other possible and less common identifiers for identifying individuals.

2.4 Methodological Variations in Assessing the Uniqueness of Dental Identifiers

Advancements in forensic odontology have introduced diverse methodologies to assess the uniqueness of dental identifiers. These range from traditional 2D imaging techniques to modern 3D imaging and superimposition approaches. These methodological advancements have not only enhanced the accuracy of identifying individuals but also broadened the scope of forensic dental analysis. This section explores the evolution and application of these methodologies in capturing and analysing unique dental features for human identification.

2.4.1 2D Imaging

The applications of 2D imaging techniques are well-established methods for assessing the uniqueness of dental identifiers in forensic identification. These imaging techniques are valued for their ability to capture unique dental identifiers, which are important for distinguishing individuals. These methods primarily involve the use of dental radiographs and clinical photographs. Dental radiographs, such as panoramic X-rays, which capture the overall dentition in a single image, and lateral cephalometric images, which captures a side-view image showing the relationship of facial structure, bone and soft tissues, together with clinical photographs have been used to assess the uniqueness of various dental identifiers, such as the number of molars, missing teeth, and displaced teeth in a study by Angelakopoulos *et al.* (2017). These identifiers were found to be highly unique, with morphological identifiers being particularly significant for human identification purposes.

While 2D imaging techniques have been widely explored, such as matching panoramic dental X-ray images using tooth appearance and geometric similarities, have shown promising results with accuracy rates of 81% to 89% (Oktay, 2018), they also

face several limitations. This includes its inability to convey depth and three-dimensional relationships of dental structures, poor image quality, and variations in imaging angles, which can lead to incorrect matches (Zhong *et al.*, 2013). The 2D feature-based approaches are significantly influenced by the quality of radiographs, which are often blurred due to substantial noise and poor lighting conditions (Ito *et al.*, 2008). The reliance on visual interpretation also introduces a degree of subjectivity, which can affect the consistency of results across different evaluators.

2.4.2 3D Imaging

Recently, 3D imaging, including CT scans and surface scan data, has been recognised as a more reliable and efficient source of AM records than traditional dental charts and odontograms (Forrest, 2019). These advanced imaging techniques can capture distinctive anatomical features from multiple angles and allow for more sophisticated digital analyses, such as automated landmarking and superimposition techniques. The adoption of 3D imaging in routine dental practice also ensures that objective AM data is readily available for rapid comparison and expediting the Disaster Victim Identification (DVI) process (Forrest, 2019). This section discusses two commonly used 3D imaging technologies namely cone-beam computed tomography (CBCT) and 3D scanners.

2.4.2(a) CBCT

Cone Beam Computed Tomography (CBCT) has revolutionised forensic dental identification by enabling precise 3D imaging of dental structures, including teeth, roots, and supporting tissues, from both AM and PM records. It has the ability to produce volumetric datasets through the construction of multiplanar images which serves as a reliable tool for accurate measurement, calculation, superimposition of the

images, and alignment of AM and PM radiographs (Issrani *et al.*, 2022). Recent developments, such as using deep convolutional neural networks (DCNNs) for automated tooth classification, have also improved the accuracy of dental charting based on CBCT images up to 91% and making the process of forensic identification easier without requiring precise tooth segmentation (Miki *et al.*, 2017).

The growing accessibility of CBCT in clinical settings, particularly in maxillofacial and otolaryngology practices, has highlighted its potential for forensic applications in cases involving missing or unidentified individuals. Despite its advantages, CBCT also has limitations, such as higher radiation exposure compared to traditional dental X-rays, the need for specialised training to ensure accurate interpretation, and cost considerations that may restrict its widespread adoption in forensic fields (Alshomrani, 2024). Nevertheless, in assessing the uniqueness of dental identifiers, CBCT is mostly used to evaluate morphological features, particularly those involving root canal structures and alveolar bone wall morphology.

2.4.2(b) 3D Scanner

The assessment of unique dental features increasingly relies on advanced 3D scanning technologies. The fundamental principle behind 3D scanners involves generating point clouds from geometric data collected from object surfaces. These point clouds are then used to reconstruct the object's shape based on the spatial position of each measurement point (Vilborn & Bernitz, 2022). The two primary categories of 3D scanners are contact and non-contact scanners. Contact scanners involve direct physical interaction with the object, while non-contact scanners utilise methods such as lasers or light projections to gather data without touching the surface. Most dental applications favour non-contact scanners due to their ability to quickly and efficiently capture

complex geometries without the limitations associated with contact methods such as their inability to capture surfaces with concavities accurately (Vilborn & Bernitz, 2022).

Among the strengths of non-contact 3D scanning technologies are their speed and versatility. These scanners can acquire data from large areas and complex shapes without the need for extensive setup or environmental controls, making them ideal for dental applications where time and precision are critical (Javaid *et al.*, 2019). For instance, non-contact methods like laser scanning can generate high-density point clouds that facilitate accurate virtual surface reconstructions. However, they often face challenges when scanning shiny or reflective surfaces, which are common in the oral environment due to saliva and varying tissue textures. Deep, undercut, steep, crowded surfaces, along with sharp edges like the incisal edges of anterior incisors, are challenging to scan accurately and often result in less precise point clouds or false representations of tooth morphology (Mizumoto & Yilmaz, 2018; Vilborn & Bernitz, 2022). To overcome this, careful consideration of scanning conditions is necessary to ensure a high point cloud density, which is crucial for accurate virtual surface reconstruction.

Laser scanning, in particular, has gained prominence among other 3D scanners due to its high accuracy and reliability. The process of digital impression often involves triangulation, where laser light is projected onto the surface, and the angle of the reflected light is analysed by the receiver to create precise data and detailed digital representations. It can serve as an effective alternative to traditional plaster models and cone beam computed tomography (CBCT) reconstructions that can reduce costs and space (Camardella *et al.*, 2017; Kim *et al.*, 2014). Furthermore, research by Rajshekar (2017) highlights the applicability of laser scanners in forensic odontology, particularly in bitemark analysis, where they can limit and quantify errors, validate measurements

of landmark features on dental casts, and achieve 100% accuracy in matching of bitemarks to dentitions. Other than bitemark analysis, this 3D scanning has been beneficial in reconstructing skulls and jaws of disaster victims, studying dental anthropology, and advancing biometrics field (Rizwal *et al.*, 2021).

Among the intraoral laser scanners available today, the Medit i500 scanner stands out in this landscape due to its advanced capabilities and proven accuracy. This scanner uses a 3D full-colour streaming capture technology that enhances the level of detail captured during scans. Recent findings indicate that the i500 excels in single-tooth scans and full-arch scans by providing higher reproducibility compared to other intraoral scanners (Selvaraj *et al.*, 2023). A comparative study by Bae & Woo (2022) has also highlighted its effectiveness alongside other intraoral scanners like CS3600 and Trios3, with the i500 being identified as particularly suitable for human identification due to its high reliability and ease of use. In addition, research by Santo *et al.* (2021) on ancestry estimation has also supported that images produced by the i500 have been shown to contain sufficient morphological detail, which is important for assessing the uniqueness of tooth surfaces.

2.4.3 Superimposition Techniques in 3D Analysis

Superimposition is a foundational technique in 3D comparative analysis as it enables alignment and comparison of AM and PM datasets. The process involves overlaying 3D digital models to identify and evaluate similarities or differences in anatomical structures. While this technique has proven effective, its application to 3D imaging is still in its early stages. However, it has long been an accepted method for comparing 2D dental images. For instance, studies by Roy *et al.* (2019) and Riaz *et al.* (2023) demonstrated the uniqueness of occlusal groove patterns using digital superimposition in 2D imaging with 100% correct matching.

3D imaging superimposition often involves algorithms such as the Iterative Closest Point (ICP), which minimises discrepancies between two 3D point clouds by aligning common reference points to facilitate quantitative analysis and use metrics like Root Mean Square (RMS) to measure the alignment accuracy (Kurniawan *et al.*, 2020). Surface-based superimposition remains the most commonly used method, with varied methodologies across studies. The lack of standardisation in software and inconsistent use of Root Mean Square (RMS) values and threshold for alignment assessment has presented challenges.

Segmentation of anatomical structures and alignment techniques are crucial in preparing 3D models for the superimposition process to ensure accurate dental surface assessment. It involves isolating the region of interest (ROI) and removing unnecessary structures that may interfere with analysis. This step involves identifying and extracting specific anatomical features, such as specific teeth and surfaces, to focus comparisons and analysis on relevant areas only. Manual and automated segmentation methods have been discussed in a few studies. For instance, the study by Reesu *et al.* (2020) explored the possibility of adopting an automated segmentation approach using Principal Component Analysis (PCA). PCA ensures consistent positioning of 3D models at the centre of mass during the pre-alignment step and this enabled uniform orientation of models and improved the accuracy of subsequent superimpositions. Automated segmentation, as discussed by Yacout *et al.* (2024), offers efficiency and scalability when processing large datasets. However, manual segmentation ensures higher accuracy, particularly for complex structures like occlusal surfaces, where AI models may overestimate the measurements. Preprocessing steps also include the refinement of 3D polygon meshes, which often contain defects such as holes, overlaps, and

inconsistent orientations that could affect surface matching accuracy (Mizumoto & Yilmaz, 2018).

Alignment is a critical step in the superimposition process, where 3D models are aligned to compare specific anatomical features. Three main types of alignment are commonly used: landmark-based alignment, best-fit alignment, and reference best-fit alignment. Landmark-based alignment involves manually selecting anatomical landmarks, such as cusp tips or incisal edges, to guide the alignment. While this method ensures precision in identifying key structures, it is time-consuming and susceptible to observer bias. Best-fit alignment is a fully automated process where the overall mesh distance errors between two models are minimised using algorithms like the Iterative Closest Point (ICP). Best-fit alignment reduces manual intervention and is particularly suitable for large datasets, as it provides consistent results with high precision (O'Toole *et al.*, 2019).

Reference best-fit alignment, on the other hand, combines the strengths of landmark-based and best-fit alignment. A reference model serves as a baseline to align other models, able to reduce errors and improve reproducibility. O'Toole *et al.* (2019) found this method to have a lower error rate compared to the other two. However, among these methods, best-fit alignment is widely favoured for its efficiency and ability to handle large datasets. Revilla-León *et al.* (2023) demonstrated that best-fit alignment not only achieves higher trueness and precision in virtual cast alignments but also minimises processing time, making it a practical choice for forensic and clinical applications.

The ICP algorithm is a cornerstone of alignment techniques and is widely used in studies to minimise discrepancies between two sets of 3D data points. The algorithm works by iteratively matching points in one dataset to the closest points in another as

illustrated in **Figure 2.1** and improving the alignment to reduce root mean square errors (RMSE). RMSE represents the standard deviation of discrepancies that provides a quantitative measure of alignment accuracy. Kurniawan *et al.* (2020) utilised ICP to superimpose genuine and imposter pairs of 3D datasets from 15 subjects and achieved clear distinctions in RMSE values between the two groups. This study highlighted the effectiveness of ICP in forensic identification, where accurate alignment is crucial for reliable results.

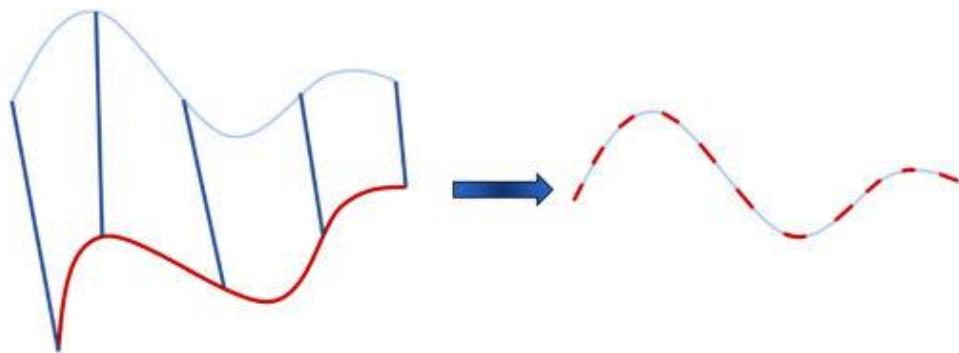


Figure 2.1 A schematic representation of the ICP algorithm (Hu *et al.*, 2021).

Mou *et al.* (2021) further validated the robustness of ICP by demonstrating a 100% match rate for maxillary dentition using RMS analysis. Despite its robustness, limitations arise in cases where full maxillary dental records are unavailable postmortem and this necessitates alternative approaches. Gibelli *et al.* (2019) also used the same iterative closest point (ICP) algorithm and root mean square (RMS) analysis but focused on the entire upper first and second molars. Although this approach was effective and had high precision, it was tested on a small sample size, prompting the need for further research with larger samples. However, ICP performance can be hindered by the lack of preregistration, as noted by Zhou *et al.* (2024).

To enhance accuracy, Zhou *et al.* (2024) proposed a three-stage framework for human identification which involves the extraction of discriminative features, the use of geometric and structural features, and a coarse-to-fine registration process. This framework achieved a Rank-1 recognition rate of 100% for tooth crown contour features and demonstrated the potential for high-precision identification even in cases of partial tooth loss. However, the lack of publicly available intraoral scan datasets highlights the need for more geographically diverse research.

2.5 Maxillary Premolars for Human Identification

Maxillary premolars, particularly the first premolars, hold significant value in forensic odontology for human identification due to their distinct morphological and odontometric features. They are strategically positioned in between the anterior canines and the posterior molars which serve a dual functional role in tearing and chewing food. This tooth features a hexagonal occlusal outline, which is uniquely distinct compared to other teeth and this makes their occlusal surface particularly distinctive. This section provides an overview of the formation, anatomy, and forensic significance of maxillary premolars, with a focus on their occlusal surface morphology and its potential as a reliable identifier in human identification.

2.5.1 Tooth Development

The development of human maxillary premolars progresses through several distinct stages, including the oral epithelium stage, bud stage, cap stage, and both early and late bell stages. Each stage is characterised by specific morphological and histological changes that are crucial for the formation of these teeth.

The development of human upper premolars begins with the oral epithelium stage. This stage marks the very beginning of tooth development, starting around the

sixth week of embryonic development. During the oral epithelium stage, the dental lamina forms from the oral epithelium, which is a critical precursor for tooth development. This stage involves the proliferation of epithelial cells that will eventually give rise to the enamel organ, dental papilla, and dental follicle, setting the foundation for tooth formation (Srbinoska *et al.*, 2023). The interaction between the oral epithelium and underlying mesenchyme is essential, as it influences the differentiation of dental tissues and the overall architecture of the developing tooth (Balic, 2019).

The bud stage occurs around the eighth week of embryonic development. During this stage, the dental lamina forms localised, round swellings called tooth buds. Each bud corresponds to a future tooth and consists of a mass of epithelial cells surrounded by mesenchymal cells. The bud stage is crucial for establishing the basic shape and position of the developing tooth (Rothová *et al.*, 2012). For premolars, this process takes place at specific sites in the dental arches where the premolars will eventually erupt. The tooth bud consists of a group of ectodermal cells that proliferate into the surrounding ectomesenchyme. Multicusped teeth, such as maxillary premolars, develop through the sequential formation of enamel knots. During the bud stage, a primary enamel knot emerges at the tip of the tooth bud, where it expresses various signalling molecules, including fibroblast growth factor (FGF), bone morphogenetic protein (BMP), Wnt family proteins, and sonic hedgehog (Shh) (Jernvall & Thesleff, 2012).

Around the ninth to tenth week, the developing tooth bud progresses to the cap stage. At this point, the tooth bud begins to take on a more defined shape that resembles the shape of a cap or hat. The enamel organ, dental papilla, and dental follicle start to differentiate. The enamel organ forms the outer part of the cap and will eventually produce enamel. The dental papilla, situated beneath the enamel organ, will give rise to

the dentin and pulp. The dental follicle surrounds the enamel organ and dental papilla and will form the supporting structures of the tooth, including the periodontal ligament, cementum, and alveolar bone.

During the early bell stage, which occurs around the fourteenth week, the enamel organ continues to expand and assume a bell shape. The enamel organ continues to develop and differentiate into distinct layers, including the outer enamel epithelium, stellate reticulum, and inner enamel epithelium. The inner enamel epithelium, which lines the inner surface of the enamel organ, begins to differentiate and fold, which is responsible for enamel formation and outlining the future cusp pattern of the premolar. The primary enamel knot acts as a signalling centre, directing the differentiation and proliferation of cells within the enamel organ. This stage is crucial for establishing the precise arrangement of cusps and grooves that characterise the occlusal surface of the premolar.

The late bell stage, occurring around the eighteenth week, involves further refinement of the tooth structure. The inner enamel epithelium cells differentiate into ameloblasts, while cells in the dental papilla differentiate into odontoblasts, which produce dentin. The cusp tips become more pronounced, and the secondary enamel knots emerge, further defining the cusp patterns. Secondary enamel knots appear at the future location of each cusp, with their sequence of formation corresponding to the relative height of individual cusps and the order of their mineralisation (Jernvall & Thesleff, 2012). The tooth continues to develop, with enamel and dentin being deposited in a synchronised manner. The supporting structures, including the periodontal ligament and alveolar bone, also mature during this stage. As the premolar nears eruption, its occlusal surface is fully formed and ready to take on its functional role in the oral cavity.

2.5.2 Occlusal Surface of Maxillary Premolars

The use of dental characteristics in forensic identification is well-established with the occlusal surface of upper premolars presenting a potential but underexplored identifier in human identification. Maxillary premolars, located between the canine and molar teeth, typically feature two cusps: a larger buccal cusp and a smaller palatal cusp that is connected by a transverse ridge. The occlusal surface of maxillary premolars is the topmost area of the teeth that comes into contact with opposing teeth when the mouth is closed. This surface has a complex anatomy designed to assist in the grinding and crushing of food.

The boundaries of the occlusal surface are defined by a curve that connects the marginal ridges, which are the raised borders on the occlusal surfaces, the cusp ridges, which are the sloping edges of the cusps or pointed parts of the tooth, and the cusp peaks, which are the highest points of the cusps. The surface of the first maxillary right premolar is characterised by various grooves and fossae that contribute to its unique morphology as depicted in **Figure 2.2**.

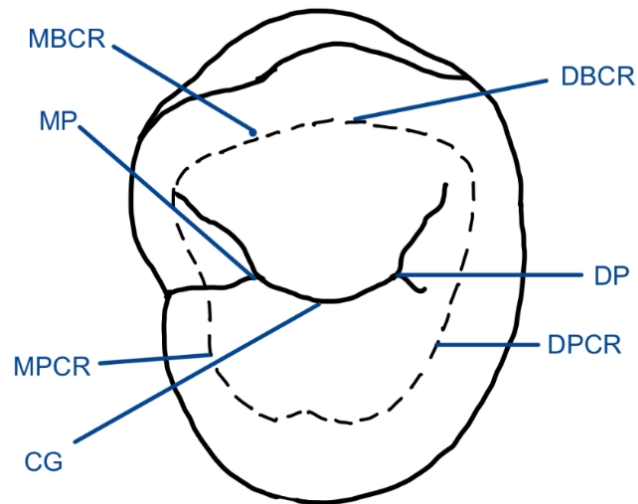


Figure 2.2 Anatomical features of the occlusal surface of the right maxillary first premolar.

MBCR: Mesiobuccal cusp ridge; MP: Mesial pit; MPCR: Mesiopalatal cusp ridge;
CG: Central groove; DBCR: Distobuccal cusp ridge; DP: Distal pit; DPCR:
Distopalatal cusp ridge.

2.5.3 Uniqueness of Occlusal Surface of Maxillary Premolars

Uniqueness refers to the distinct quality of being one of a kind and is characterised by a state or condition that sets something apart as extraordinary or incomparable (Bernitz, 2023). The uniqueness of the occlusal surface of the first maxillary premolar is shaped by a combination of intrinsic and extrinsic factors.

Intrinsic factors are those inherent to the individual's body and genetics. For instance, genetic variations can result in differences in tooth shape, size, and occlusal pattern. Variations in enamel formation and the positioning of cusps and grooves can be influenced by genetics that lead to unique occlusal surface features. Moreover, the development of occlusion is a dynamic process occurring alongside the growth of associated dental structures, such as the nose, maxillary sinuses, facial bones, and muscles (Sabin *et al.*, 2021). Any disruption during this developmental process, such as

the prolonged retention of primary teeth due to impacted permanent teeth or anomalies like ectopic eruptions, can lead to unique changes in the occlusal surface. Early loss of permanent teeth from caries or trauma may also alter the dental arch, further contributing to individuality in occlusion.

Extrinsic factors, on the other hand, are those external influences that affect the teeth over time. These can include dietary habits, which play a significant role in shaping the occlusal surfaces. Consuming hard, citrus, and acidic foods, and a preference for chewing on one side can lead to wear patterns unique to the individual (Elmarsafy *et al.*, 2024). Habits such as teeth grinding (bruxism) or clenching can also create distinctive wear facets on the occlusal surfaces. Environmental factors, such as exposure to certain chemicals or minerals during tooth development and using abrasive toothpastes or hard dental brushes can impact enamel strength and surface characteristics. Furthermore, dental treatments, restorations, and orthodontic interventions can alter the occlusal surface, adding to its uniqueness.

Recent studies have highlighted the significant variability and potential for identification in the occlusal surface morphology of upper premolars. Medhat *et al.* (2023) studied the occlusal morphology in an Egyptian population by analysing groove patterns and occlusal outlines of extracted premolars. They found that the first upper premolar predominantly exhibited a hexagonal outline, while the second upper premolar more commonly showed an oval shape. Variations in crown morphology, although less common compared to root morphology, have also been documented. For instance, Nayak *et al.* (2013) reported a rare presentation of maxillary and mandibular first premolars with a three-cusp pattern. Maxillary premolars also exhibit variations in the presence and expression of accessory cusps, such as the mesial and distal accessory cusps. These non-metric traits have been studied extensively and can provide valuable