Tooth Morphometry and Pattern of Palatal Rugae Among Monozygotic Twins in Malaysia

Abstract

Introduction: Monozygotic (MZ) twins share the same DNA, placenta, amniotic fluid, and physical features. Genetic factors play a prominent role compared to environmental factors in one’s physical appearance, including dental morphology. Here we studied variation in MZ twin’s tooth morphometry and palatal rugae pattern. Materials and methods: Variation between twins can be a valuable tool in forensics to identify individuals. Ten pairs of MZ twins were selected for this research. The maximum coronal mesiodistal and buccopalatal dimension of the maxillary teeth, excluding the second and third molars, were measured in triplicate using calipers. Palatal rugae patterns of the samples were cast, analyzed, and recorded based on shape and unification. Results: Our results showed a significant tooth dimension correlation between MZ twins. Maxillary central incisors had the least genetic variability, but the Carabelli trait, skeletal pattern, occlusion, and occlusal features exhibited greater correlation in MZ twins. Our findings provide compelling evidence for mirroring of dental features and palatal rugae patterns in MZ twins. This study is the first of its kind reported in Malaysia. Conclusion: Marked similarities in tooth morphometry and other dental physical features were observed between twins, which can be a useful tool in forensics for the identification of individuals.

Keywords: Monozygotic twin, palatal rugae pattern, tooth morphometry

Introduction

Monozygotic (MZ) twins, also known as identical twins, are produced when a single ovum is fertilized by a single sperm, followed by splitting of the embryo into two. The splitting results in two genetically identical twins who share the same amniotic fluid, placenta, and genetic information. Studies comparing physical features in identical twins provide useful insight into the contribution of genetic and environmental factors in dental development.

There are four types of MZ twins, categorized based on the division time of the embryo after conception. [1] Cleavage before the sixth day results in two placentas, two chorions, and two amnions. Cleavage occurring between the sixth- and tenth-day results in one placenta, one chorion, and two amnions (monochorionic diamniotic). Monochorionic diamniotic is the most common form of MZ twins, accounting for about 64% of MZ twins. [2] If the embryo splits between the tenth and the fourteen day, the twins share one placenta, one chorion, and one amnion (monochorionic monoamniotic). This type of twin formation is more uncommon, accounting for only 4% of MZ twins. [1] If cleavage of the embryo occurs after the fourteenth day, there is an increased risk that the division will be incomplete. The resulting twins will be at risk of being conjoined, or what is often called “Siamese” twins. Twins with separate placentas and sets of membranes can be implanted together in the womb that the individual placentas appear to fuse. When observed with the naked eye, only a single placenta seems to be present. This observation happens in 42% of dizygotic (DZ) twin pregnancies and 13% of MZ twin pregnancies, meaning that for approximately 49 out of 100 pairs, an examination of the placenta and fetal membranes will not yield conclusive information about zygosity. [1] [2]

Twin research is essential to understand normal and abnormal dentofacial development. [3] Tooth morphometry is the

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study of the shape and size of teeth, which is determined at about the 18th week of embryonic development of secondary teeth during the morpho differentiation stage of tooth development.\cite{4} It has been reported that normal variation in the size of teeth is the result of multiple inheritance factors, including both genetic and environmental factors.

Forensic makes use of different methods to identify individuals, including methods based on biometric technologies. This method relies on identifying unique characteristics in individuals, from general features such as the face and palm to specific trait patterns such as those present in the retina, iris, and fingerprints. The genetic fingerprint of identical twins is shared; however, biometric verification techniques are not always accurate in identifying individuals. Insight into more biometric traits such as the palatal rugae pattern may provide a way to distinguish between identical twins. More focus on this trait is needed in biometrics technology development and dental forensics. Additionally, analysis of individual palatal rugae patterns can be carried out to identify a person. The palatal rugae refer to the ridges on the anterior part of the palatal mucosa on each side of the median palatal raphe and behind the incisive papilla. Palatoscopy is defined as the study of palatal rugae.\cite{4} Palatal rugae patterns are anatomic landmarks that are unique in each individual, despite there being some similarity patterns between MZ twins.

Another unique and exciting feature in MZ twins is mirror imaging. Mirror imaging is when one of the twin pairs “mirrors” the other for one or more features. It has been reported in about 25% of MZ twins. Despite the increase in twin studies in several countries worldwide, there are no published reports in Malaysia. Therefore, this study was aimed to correlate tooth morphometry, the pattern of palatal rugae, and mirror imaging in identical twins in Malaysia.

Materials and Methods

Ethical approval

Ethical approval for this study (2019-053) was provided by the International Islamic University Malaysia Research Ethics Committee (IERC), Kuantan, Malaysia on 14th February 2019.

Sample size

The study sample consisted of 10 pairs of MZ twins in the 12- to 23-year age group. Informed written consent was obtained from all the participants, including for publication purposes. Zygosity was determined based on physical similarities such as hair color, eye color, nose shape, palm crease pattern, skeletal pattern, and date of birth.

Extra- and intraoral record

Front and profile view extraoral photographs were taken before the intraoral examination. Accurate charting was performed, followed by intraoral photography, and upper and lower arch impression. Quick-setting stone casts were prepared from alginate (Zelgan Plus, Dentsply, Gurgaon, India) and used to prepare impressions of the maxillary and mandibular teeth of each participant.

Dental morphometry

For tooth morphometry analysis, the mesiodistal and buccopalatal dimensions of maxillary molars were measured using calipers. Readings to the nearest 0.1 mm were achieved. The same tooth without caries or restoration of the proximal surfaces or cusps was chosen for measurement in both twins. The measurement was performed as follows: the caliper was held parallel to the incisal or occlusal surfaces of the tooth, with the most pointed beaks of the caliper placed at the tallest points of the cusps. The diameters of the incisor, canine, and molar crown were measured in the same manner. Mesiodistal dimension was recorded as the maximum mesiodistal dimension of the tooth crown parallel to the occlusal and labial surfaces. The buccopalatal dimension was recorded as the maximum crown dimension at right angles to the mesiodistal dimension.

To maintain measurement data validity and accuracy, interoperator validity testing using the Kappa test was performed before carrying out measurements. The measurement of each tooth was also assessed for intraoperator validity on two separate occasions by the same operator. In the case of more than 0.4 mm discrepancy in measurement, the tooth was remeasured.

Rugoscopy record

Rugae shape and unification patterns were analyzed based on the classification.\cite{8} The rugae were first marked on dental casts using a black HB pencil. Then, the number and shape of rugae were assessed. All the rugae patterns from each pair of twins were observed and documented in the same format. Other dental features such as spacing, missing tooth, crossbite, angulation of tooth, and expression of the cusp of Carabelli were carefully examined in all MZ dental casts.

Statistical analysis

All the data collected were analyzed using SPSS version 23 software (IBM SPSS Statistics for Windows, Version 23.0; IBM Corp., Armonk, New York, USA). Parametric data were analyzed using analysis of variance, Pearson’s correlation, Kappa, and intraclass correlation (ICC) test (P-value was set at 0.05).

Results

The final study sample consisted of 10 MZ twin pairs (eight females and two males). All twin pairs were selected from the Malaysian population.

Tooth morphometry

**Rugoscopy**

The differences in the palatal rugae pattern among twins can be appreciated via visual observation. None
of the twin pairs showed a similar shape or number of palatal rugae. The right and left rugae patterns were compared for shape and unification in MZ twin pairs [Figure 1].

The rugae patterns were analyzed based on shape – circular, straight, wavy, curved, unification [Figure 2]. P-value of less than 0.05 in curve and unification indicates mirror imaging of rugae shape between twins [Table 4].

The Pearson correlation coefficients for curve and unification shape of rugae indicate mirror imaging of the rugae in MZ twins. The coefficient values show a strong correlation in the rugae pattern between one side of one twin and the opposite side of the co-twin, both in MZ (0.866 and 0.667). Hence, palatal rugae mirror imaging was observed in the twins.

Other dental features

It was observed that only 1 out of 10 MZ twins had Carabelli cusps (image not shown). High similarities in the pattern of tooth spacing or median diastema were observed in the MZ twins. Additionally, a similar inclination of the central incisor was observed in one pair of twins. In Figure 3, we can observe mirror imaging in the buccally erupted canine between pairs of twins.

Discussion

Determining zygosity is essential medically, genetically, and psychosocially for the individual and family. Zygosity can be determined via various means, including using placental-site ultrasound marker, via physical similarity assessment, or blood type and DNA analysis. This study on identical twins is carried out to discern the influence of genetics and the environment on human dental variation. However, studies on twins are limited in several ways. For example, it is often difficult to obtain a large enough number of twin pairs and ascertain if the twin pairs were exposed to similar environmental factors.

The most reliable diagnosis for zygosity is genotyping; however, this method is expensive and time-consuming. Besides, twin identification kits are not commercially available in Malaysia. Moreover, some participants may not agree to provide biological specimens necessary for DNA extraction, which is sometimes required in zygosity determination methods.
We intended to derive information on MZ twin zygosity from maternal books. However, we found this information to be unavailable. Hence, MZ twin zygosity determination was carried out based on physical appearance (phenotype). Assessing the degree of physical similarity between twins is still a widely used method for diagnosing zygosity. Resemblance can be reported by the twin themselves, their relatives, or spouses. For young twins, this information can be provided by the parents, who can describe the physical differences between the twins and report how often other people confuse the twins for each other. Several physical resemblances that can be assessed include hair type, height, eye color, palm crease patterns, ear shape, nose shape, facial appearance, and skeletal pattern. Confusion by people meeting the twins for the first time and mistaken identity can also be indicators of strong physical resemblance.

In this study, 10 pairs of MZ twins were used to determine zygosity. The 10 pairs of twins had consistently similar phenotypes. Figures 4–6 express the physical similarities between the samples. Table 1 displays the additional similarities in physical features between the twins. The physical similarity data show 100% similarity and support our hypothesis that MZ twins should show 100% similar phenotype. This result also supports the study, which reported 237 identical twins selected for the study had physical similarity results comparable to genotyping analysis results.

Tooth morphometry is primarily controlled by hereditary forces, and the pattern of heritability is distinct for each group of teeth. It has been suggested that normal variation in the size of teeth is the result of multifactorial inheritance, with both genetic and environmental factors being important. Our study analyzed the data using the ICC test to show statistically significant correlation coefficients for tooth morphometry among twins. The permanent maxillary first molars and permanent maxillary canines showed significant P-values [Table 2].

However, we note that the intercuspal difference in the first molar (MB-DB dimension) and diameter of the permanent first central incisors (LL) were not significantly different ($P > 0.05$). Table 3 suggests that common environmental variations may have contributed to the size of this tooth.

Our findings agree with reports stating that frequent environmental contributions to permanent teeth averaged 18% for buccolingual and 10% for mesiodistal diameters. In palatal rugoscopy, we found out that none of our samples showed a similar number and shape of palatal rugae. The differences in palatal rugae patterns were readily appreciated by gross observation without the need for sophisticated tools. This marked difference contrasts with other features assessed between MZ twins in this study that showed 100% similarity. We suggest that palatal rugae patterns are unique and permanent and may help to identify an individual. The result is supported by a study that reported that palatal rugae patterns are not identical between pairs of MZ twins, even when the patterns are related.

Twinning may be linked to mirror imaging. Mirror imaging is observed when one of the twin pairs “mirrors” the other for one or more features. Around 25% of MZ twins display mirror imaging. A study reported significant mirror imaging in MZ and DZ twins, although to a greater extent in MZ twins. In contrast, our study did not show significant mirror imaging between the MZ twins. Only one out of 10 pairs showed mirror imaging of dentition at the buccally erupted canine [Figure 3]. Mirror imaging of curve and unification shape of palatal rugae was observed [Table 4]. Nevertheless, several studies found no association between MZ twinning and mirror imagery.

Other dental traits such as hypomineralization, median diastema, angulation of the teeth, and crowding pattern were observed between twin pairs [Figures 7–9]. This observation suggests that sharing the same DNA may influence dental variation in twin pairs. Oral health professionals in dental practice can use this knowledge to explore differences in orofacial structures in MZ twin patients.

**Conclusion**

This twin study on tooth morphometry and palatal rugae pattern is the first of its kind in Malaysia. We conclude that genetic influence plays a substantial effect in the morphology of permanent teeth. Palatal rugae pattern in twins showed a mirror-image effect; however, the patterns remain unique and distinct enough to be used for individual identification.
Figure 4: Extraoral photograph of MZ twin girls (twin A) and boys (twin B)

Figure 5: Similarities in palm crease patterns in each twin pair
Table 1: Physical features recorded in monozygotic twins for zygosity determination

| Physical features | 1A | 1B | 2A | 2B | 3A | 3B | 4A | 4B | 5A | 5B | 6A | 6B | 7A | 7B | 8A | 8B | 9A | 9B | 10A | 10B |
|------------------|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|
| Hair             | Flat | Flat | Pointed | Pointed | Flat | Flat | Flat | Flat | Flat | Flat | Flat | Flat | Flat | Flat | Flat | Flat | Flat | Flat | Flat | Flat |
| Eye color        | Black | Black | Black | Black | Black | Black | Black | Black | Black | Black | Black | Black | Black | Black | Black | Black | Black | Black | Black | Black |
| Skeletal pattern | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  | CI  |

Table 2: Correlation coefficients obtained from intraclass correlation (ICC) test for tooth morphometry features among twins

| Dental features | $P$-value | $P$-value |
|-----------------|-----------|-----------|
| Intercuspal differences (M1) | | |
| MB-DB           | 0.193     |           |
| DB-DL           | 0.008     |           |
| DL-ML           | 0.000     |           |
| ML-MB           | 0.000     |           |
| Crown diameters (I1) | | |
| MD              | 0.005     |           |
| LL              | 0.371     |           |
| Crown diameters (C) | | |
| MD              | 0.013     |           |
| LL              | 0.010     |           |
| Crown diameters (M1) | | |
| MD              | 0.000     |           |
| BL              | 0.073     |           |

M1, permanent maxillary first molar; I1, permanent maxillary central incisor; C, permanent maxillary canine; MB, mesiobuccal; DB, distobuccal; DL, distolingual; ML, mesiolingual; MD, mesiodistal; LL, labiolingual; BL, buccolingual.

Table 3: Statistically significant correlation coefficients obtained via Kappa test for selected dental features among twins

| Dental features | $P$-value |
|-----------------|-----------|
| Carabelli trait | 0.002     |
| Skeletal pattern| 0.001     |
| Occlusion       |           |
| Incisor occlusion| 0.016   |
| Right molar occlusion | 0.020  |
| Left molar occlusion | 0.005 |
| Occlusal traits | 0.000     |

Table 4: Correlation coefficient values for palatal rugae among twins

| Parameter of rugae analysis | Twin A | Twin B | $R$-value | $P$-value |
|-----------------------------|--------|--------|-----------|-----------|
| Shape                       | Right  | Left   | – (C)     | – (C)     |
| Circular (C)                | 0.278 (S) | 0.437 (S) |           |           |
| Straight (S)                | 0.123 (W) | 0.734 (W) |           |           |
| Wavy (W)                    | 0.866 (c) | 0.001 (c) |           |           |
| Unification (U)             | 0.284 (S) | 0.426 (S) |           |           |
|                            | 0.135 (W) | 0.710 (W) |           |           |
|                            | 0.244 (c) | 0.497 (c) |           |           |
|                            | 0.667 (U) | 0.035 (U) |           |           |
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Conflicts of interest
There are no conflicts of interest.

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