Introduction

The identification of individuals and their unique characteristics have been of paramount importance to human society. It is important to identify the deceased to ensure a proper burial and for the satisfactory disposal of legal issues that might arise.¹

Context: Gender determination is central in establishing personal identification from human skeletal remains. The study was conducted to find out the accuracy with which gender can be determined by odontometric methods. Aims: To investigate the mesiodistal (MD) and buccolingual (BL) dimensions of all the teeth of permanent dentition to find new parameters to differentiate between male and female teeth and to assess whether each type of linear measurement can be used independently in odontometric sex differentiation. Materials and Methods: The study was conducted at a dental college on a composite group of 500 individuals comprising 250 males and 250 females. Impressions of upper and lower jaws were made with alginate impression material and casts prepared with dental stone. A digital Vernier calliper was used to measure the BL and MD dimensions of all the upper teeth except the third molars. Statistical Analysis Used: The results were subjected to statistical analysis using univariate analysis and linear stepwise discriminant function analysis to find the variables which discriminate gender significantly. Results: The MD and BL dimensions between males and females were statistically significant. The predicted value for correct classification of gender was also statistically significant. Conclusions: The ability to differentiate gender in the population using stepwise discriminant functions was found to be very high with 99.8% accuracy with males showing statistically larger teeth than females. This is similar to the near 100% success in gender determination using pelvic and skull bones.

Key words: Buccolingual, gender determination, mesiodistal, odontometric

Technically, the positive identification of human remains requires the matching of physical characteristics of the deceased with records of the physical characteristics made before death. Several methods may be used to produce identification with an aggregate high level of reliability. Identification by relatives and friends may not be accurate because of the highly emotional state of the relatives and
also because sometimes, the bodies may be highly mutilated or decomposed.\(^1\)

The most reliable method of identification includes fingerprints and biological methods such as DNA profiling. In some cases, fingerprints may not be available due to trauma and decomposition. DNA profiling exhibits a high degree of dependability, but can be expensive, time-consuming, and may not be feasible in all cases.

Dental examination and comparison between antemortem and postmortem dental records and radiographs produce results with a high degree of reliability and relative simplicity. Teeth are the most durable structures in the body and can resist temperatures of 1600°C without any appreciable loss of microstructure. Hence, teeth form an excellent material for anthropological, genetic, odontologic, and forensic investigations.\(^{1,2}\)

The term “forensic” implies “court of law,” “odontology” means “study of tooth.” Forensic odontology has been defined as the branch of dentistry which, in the interest of justice, deals with the proper handling and examination of dental evidence and with the proper evaluation and presentation of dental findings. Forensic odontology has played a key role in the identification of persons in mass disasters, in crime investigations, and in investigations of decomposed and disfigured bodies.\(^3\)

Various methods are employed in forensic odontology to determine the age and sex of an individual. The methods include rugoscopy, cheiloscopy, bite marks, tooth prints, radiographs, photographic study, and molecular methods.\(^3\)

The determination of gender is central to the process of establishing personal identification from human skeletal remains. An accurate sex diagnosis effectively reduces the number of possible matches by half. It also helps in estimating the stature and age at the death of an individual. Measurement of the long bones may also provide highly accurate sex assessments, but may not be possible in all cases.\(^4\)

In some of the cases, the only available structure for determining gender is the measurement of the permanent dentition. Acharya and Mainali explored the utility of buccolingual (BL) dimension and mesiodistal (MD) dimension in sex differentiation and reported accuracy rates of 62–83% for a Nepalese sample.\(^5\) Ates et al. observed an accuracy of 68–81% for similar measurements in a Turkish sample.

Although teeth are excellent study material, variation in the crown size has been reported among different populations. Numerous factors can contribute to variation in tooth size and may be described broadly as genetic, epigenetic, and environmental influences.\(^6\) Therefore, this study was undertaken to assess the dimorphism of permanent teeth in humans by measuring the BL and MD diameter of the teeth. The results obtained will be subjected to stepwise discriminant functional analysis to enable accurate sex assessment in forensic identification.

**Materials and Methods**

This clinical study was conducted over a period of 4 months on undergraduate and postgraduate students of a dental college. The sample comprised permanent dentitions from 500 individuals of Indian origin, of which 250 were males and 250 females. The sample was the composite of different ethnic groups as participants were from different states of India.

**Inclusion criteria**
1. Healthy state of gingiva and periodontium
2. Caries-free teeth.

**Exclusion criteria**
1. Supernumerary teeth
2. Third molars
3. Developmental abnormalities of teeth
4. Physical and chemical injuries of teeth
5. Teeth with proximal restorations.

**Materials required**

**a.** For examination and selection of study subjects, the following materials are required:
1. Disposable mouth mask
2. Disposable gloves
3. Chlorhexidine mouthwash
4. Cheek retractor
5. Stainless steel kidney tray
6. Cotton holder and cotton
7. Disposable glass tumbler
8. Tweezer
9. Mouth mirror
10. Single-ended straight sharp probe
11. Pro forma and consent form.

**b.** For impression making and obtaining study cast, the following materials are required:
1. Dentulous perforated stock trays for full mouth impressions – upper and lower number 1, 2, 3, 4
2. Rubber bowls
3. Plaster spatulas – straight and curved
4. Alginate impression material – Algitek
5. Type III Gypsum product, dental stone – Denstone
6. Type II Gypsum product, dental plaster – Dentalplast.

**c.** For recording the measurements on the obtained study cast, the following material is required:
1. Digital Vernier calliper calibrated to 0.01 mm.
Method
After obtaining written consent from the participants, impressions of the maxillary and mandibular arches were taken using dental alginate. Casts were poured using dental stone.

Measurements
The MD [Figure 1] and BL [Figure 2] dimensions of all the teeth except third molars were measured on the casts using a digital Vernier calliper calibrated to 0.01 mm (Mitsoyo, Japan). The measurements were noted down on the master chart and later entered in Microsoft excel spreadsheet and were subjected to statistical analysis. The MD measurement was defined as the greatest distance between contact points on the approximate surfaces of the tooth crown and was measured with the caliper beaks perpendicular to the long axis of the tooth. The BL measurement was defined as the greatest distance between the labial/buccal surfaces of the tooth crown measured with the caliper beaks held at the right angles to the MD dimensions.

Results
An observational multivariate discriminate study with 250 males and 250 females was undertaken to determine the significant odontometric variables for discriminating gender.

Statistical analysis
Linear stepwise discriminant function analysis has been performed to find the variables which discriminate the gender significantly. All statistical analyses were performed using statistical software, namely, SAS 9.29 (SAS Institute Inc.), SPSS 15.0 (IBM Corp), Stata 10.1 (Oracle), MedCalc 9.0.1 (Med Calc Software), Systat 12.0 (Systat Software), and R Environment version 12.11.1 (R Foundation for Statistical Computing). Microsoft Word and Excel have been used to generate tables and graphs.

The gender distribution of cases is represented in Table 1. Out of 500 cases, 250 (50%) cases were males and 250 (50%) cases were females.

The descriptive statistics, t-values, and P values of MD measurements of the permanent teeth in males and females are depicted in Table 2. All 28 teeth variables were included in the analysis. Results on continuous measurements are presented on mean ± standard deviation (SD) (Min‑Max) and results on categorical measurements are presented in number (%). Significance is assessed at 5% level of significance. Statistically strongly significant values, with $P \leq 0.01$ were seen in MD values of 13 ($P = 0.001$), 21 ($P = 0.001$), 36 ($P = 0.003$), and 33 ($P = 0.004$). Statistically moderately significant values, with $P > 0.01$ and $P \leq 0.05$ are noted in the MD values of 43 ($P = 0.024$) and 46 ($P = 0.017$), respectively. MD value of 45 with $P = 0.088$ had statistically suggestive significance.

Figure 3 shows the MD odontometrical variables of all teeth in males and females. MD dimensions of male teeth are greater than the MD dimensions of female teeth in 17, 16, 13, 12, 11, 21, 22, 23, 24, 26, 27, 36, 35, 34, 33, 43, 45, and 46.

The descriptive statistics, t-values, and P values of BL measurements of the permanent teeth in males and

| Table 1: Gender distribution |
|-----------------------------|
| Gender | Number of subjects (%) |
| Mesiodistal |
| Male | 250 (50.0) |
| Female | 250 (50.0) |
| Total | 500 (100.0) |
| Buccolingual |
| Male | 250 (50.0) |
| Female | 250 (50.0) |
| Total | 500 (100.0) |

Figure 1: Measurement of mesiodistal width of the study cast

Figure 2: Measurement of buccolingual width of the study cast
females are depicted in Table 3. All 28 teeth variables were included in the analysis. Results on continuous measurements are presented on mean ± SD (Min-Max) and results on categorical measurements are presented in number (%). Significance is assessed at 5% level of significance. Statistically strongly significant values, with \( P \leq 0.01 \) was seen in BL values of 16 (\( P = 0.001 \)), 15 (\( P = 0.002 \)), 14 (\( P = 0.001 \)), 13 (\( P = 0.005 \)), 12 (\( P = 0.001 \)), 11 (\( P = 0.001 \)), 25 (\( P < 0.001 \)), 26 (\( P < 0.001 \)), 27 (\( P = 0.001 \)), 36 (\( P = 0.003 \)), 41 (\( P < 0.001 \)), and 46 (\( P < 0.001 \)). Statistically moderately significant values, with \( P > 0.01 \) and \( P \leq 0.05 \) is noted in the BL value of 17 (\( P = 0.026 \)). BL values of 21 (\( P = 0.096 \)) and 37 (\( P = 0.055 \)) had statistically suggestive significance.

Figure 4 shows the BL odontometrical variables of all teeth in males and females. BL dimensions of male teeth are greater than the MD dimensions of female teeth in 17, 16, 15, 14, 13, 12, 11, 21, 22, 24, 25, 26, 27, 37, 36, 35, 34, 33, 32, 41, 43, 44, 45, 46, and 47.

Table 4 depicts the tooth variables that contributed to the stepwise discriminant analysis for MD dimensions. Wilks' lambda denotes how useful a given variable is in the stepwise analysis and determines the order in which the variables enter the analysis. MD dimension of maxillary right canine entered the discriminant analysis first followed
by maxillary left central incisor, mandibular left first molar, mandibular right lateral incisor, and mandibular right second premolar. The unstandardized coefficients are 0.775 for MD13, +0.691 for MD21, +0.554 for MD36, −0.771 for MD42, and − 0.660 for MD45. Sectioning point is 0.278 for male and − 0.276 for female. Wilks’ lambda is 0.869 with 69.9% predicted value for correct classification and this is statistically significant.

Table 5 depicts the tooth variables that contributed to the stepwise discriminant analysis for BL dimensions. Wilks’ lambda denotes how useful a given variable is in the stepwise analysis and determines the order in which the variables enter the analysis. BL dimension of maxillary right second premolar entered the discriminant analysis first followed by BL dimension of mandibular left central incisor, BL dimension of maxillary right second molar, maxillary right second premolar, maxillary right first premolar, maxillary right lateral incisor, maxillary right central incisor, maxillary left central incisor, maxillary left first premolar, maxillary left second premolar, mandibular left second molar, mandibular left first molar, and mandibular left second premolar. The unstandardized coefficients are −0.207 for MD 21, +0.209 for MD 31, +0.190 for BL 17, +0.484 for BL 15, +0.683 for BL 14, −0.168 for BL 12, −0.219 for BL 11, −0.510 for BL 21, +0.297 for BL 24, +0.214 for BL 25, −0.202 for BL 37, −0.323 for BL 36, and +0.243 for BL 35. Sectioning point is −3.172 for males and 3.159 for females. Wilks’ lambda is 0.090 with 99.8% predicted value for correct classification and this is statistically significant. Hence, this function can predict the new case as male or female with 99.8% accuracy.

**Discussion**

Proper sex assessment of skeletal remains has important ramifications in forensic and bioarchaeological investigations. All the available criteria must be utilized in order to obtain optimal sex prediction. Teeth can resist postmortem insults and are considered one of the strongest tissues in the body. Hence, teeth are of paramount importance when more robust predictors such as the pelvis or long bones are destroyed or fragmented.\[7\]
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Table 5: Stepwise discriminant function analysis (buccolingual)

| Variables | Unstandardized coefficients | Standardized coefficients | Sectioning point       | Wilks' Lambda | P             | Percentage of correct classification |
|-----------|-----------------------------|---------------------------|------------------------|---------------|---------------|-------------------------------------|
| BL15      | 0.469                       | 0.368                     | −3.137 for male        | 0.092         | <0.001**      | 99.8                                |
| BL14      | 0.731                       | 0.441                     | 3.137 for female       |               |               |                                     |
| BL13      | −0.143                      | −0.111                    | 0.092                  |               |               |                                     |
| BL11      | −0.263                      | −0.235                    | 0.082                  |               |               |                                     |
| BL21      | −0.426                      | −0.375                    | 0.082                  |               |               |                                     |
| BL22      | −0.151                      | −0.132                    | 0.082                  |               |               |                                     |
| BL24      | 0.265                       | 0.187                     | 0.082                  |               |               |                                     |
| BL25      | 0.223                       | 0.166                     | 0.082                  |               |               |                                     |
| BL27      | 0.212                       | 0.166                     | 0.082                  |               |               |                                     |
| BL37      | −0.186                      | −0.149                    | 0.082                  |               |               |                                     |
| BL36      | −0.312                      | −0.259                    | 0.082                  |               |               |                                     |
| BL35      | 0.251                       | 0.179                     | 0.082                  |               |               |                                     |

BL: Buccolingual, **: Strongly significant

Table 6: Stepwise discriminant function analysis (mesiodistal + buccolingual)

| Variables | Unstandardized coefficients | Standardized coefficients | Sectioning point       | Wilks' lambda | P             | Percentage of correct classification |
|-----------|-----------------------------|---------------------------|------------------------|---------------|---------------|-------------------------------------|
| MD21      | −0.207                      | −0.138                    | −3.172 for male        | 0.090         | <0.001**      | 99.8                                |
| MD31      | 0.209                       | 0.138                     | 3.159 for female       |               |               |                                     |
| BL17      | 0.190                       | 0.148                     | 0.090                  |               |               |                                     |
| BL15      | 0.484                       | 0.378                     | 0.090                  |               |               |                                     |
| BL14      | 0.683                       | 0.412                     | 0.090                  |               |               |                                     |
| BL12      | −0.168                      | −0.139                    | 0.090                  |               |               |                                     |
| BL11      | −0.219                      | −0.196                    | 0.090                  |               |               |                                     |
| BL21      | −0.510                      | −0.449                    | 0.090                  |               |               |                                     |
| BL24      | 0.297                       | 0.210                     | 0.090                  |               |               |                                     |
| BL25      | 0.214                       | 0.160                     | 0.090                  |               |               |                                     |
| BL37      | −0.202                      | −0.162                    | 0.090                  |               |               |                                     |
| BL36      | −0.323                      | −0.267                    | 0.090                  |               |               |                                     |
| BL35      | 0.243                       | 0.173                     | 0.090                  |               |               |                                     |

MD: Mesiodistal, BL: Buccolingual, **: Strongly significant

Human dentition has been subjected to many studies, and most of the studies have been focused on human dental health, morphology, and odontometric variation. Forensically, teeth have been used mostly for age estimation and sex determination.[8] Out of the two approaches to identify sex, the first is based on a visual assessment of the shape or relative proportions of sexually dimorphic features. The second is a metric approach, which offers advantages over the visual approach as it is inherently more objective, has higher reliability, is less dependent on the previous observer experience, and is more readily amendable to statistical analysis, and thus helps comparisons within the samples as well as with previous studies. The MD and BL diameters of permanent teeth are the two most commonly used and researched features used in the determination of sex on the basis of dental measurements.[9] Most of the odontometric sex determinations have been based on the use of stepwise discriminant function analysis as it calculates the optimal combination of variables and weighs them to reflect their contribution to sex determination.[10]

The aim of the study was to find out the accuracy with which gender can be differentiated by investigating the MD and BL dimensions of all teeth of the permanent dentition. The study was conducted on a composite group of 500 students comprising 250 males and 250 females.

Sexual dimorphism of teeth has been studied by many researchers and it has been reported that permanent teeth are highly dimorphic.[6,11] In our study, both the jaws were utilized as it is known to provide the highest accuracy rate in assessing sex.[12] In all the teeth in which the MD and BL variables showed statistically significant difference, males had larger tooth dimensions when compared to that of females. This finding reinforces the similar conclusions drawn by Vanaki et al., Zorba et al., Pereira et al., and Iscan and Kedici that males exhibit larger teeth than females.[8-11] Zorba et al. have compiled the various reasons put forward by researchers to explain the increased dimensions in male teeth. It includes an increase in jaw size of males, difference in enamel, and the role of Y chromosome in increasing the
mitotic potential of the tooth germ which, in turn, induces dentinogenesis.[11]

Out of the 56 variables, 10 variables exhibited reverse dimorphism, i.e., an increase in the dimensions of female teeth when compared to the dimensions of male teeth, but it did not affect the sex assessment as none of the variables were statistically significant and hence did not contribute to the stepwise discriminant function. The feature of reverse dimorphism has also been reported in a similar study by Prabhu and Acharya. Multifarious interactions between different genetic and environmental factors and the reduced sexual dimorphism and consequent male-female overlap have probably extended to include larger female teeth and result in reverse dimorphism.[12,13]

Of all the permanent teeth, many studies have reported that canines are the most dimorphic teeth.[12,14-16] The present study establishes the existence of a statistically significant difference in the BL measurements of all the teeth in maxillary right quadrant, maxillary left second premolar, first molar and second molar, mandibular right and left first molar, and mandibular right central incisor showed statistically significant difference in the crown size between male and female teeth. Mesiodistally, the maxillary and mandibular canines, mandibular first molar, and maxillary central incisor showed pronounced sexual dimorphism. This is in accordance with the studies conducted by Acharya and Mainali who reported that sexual dimorphism is more in canines, molars, and maxillary central incisors.[9,12] Studies by Garn et al. have also reported a significant sexual dimorphism in canines and molars.[14] The anterior teeth did not contribute in discriminating the sexes as much as the posterior teeth. This was in contrast to the observations of Iscan and Kedici wherein, anterior teeth discriminated the sexes in a better way.[8] Variation in sample size can be considered to be a reason for such a diametrically opposed observation. Maxillary teeth entered the discriminant analysis more than the mandibular teeth just as in the study conducted by Acharya and Mainali.[12]

In similar studies, the BL measurements were found to be more dimorphic than the MD measurements.[6,10-12] Results of our studies have confirmed this finding. The reason for this can be because with time, the MD dimension can be influenced by advanced consumption of special foods and the approximate surfaces could show signs of wear, which, in turn, may give altered dental measurements and impact sex assessment outcomes.[7]

When only the MD variable was used, majority of tooth variables such as 13, 21, 36, and 34, which showed statistically significant univariate dimorphism have contributed to the discriminant analysis. However, the MD dimensions of 33, 43, and 46 that showed univariate differences did not enter the discriminant functions. On the other hand, the MD dimension of 42 did not show significant univariate difference, yet entered the discriminant analysis. Similarly, when only the BL variable was used, teeth with significant univariate dimorphism which contributed to the discriminant analysis were 15, 14, 13, 11, 21, 25, 27, 36, and 35. Whereas, 17, 16, 12, 21, 26, 35, 41, and 46 did not enter the discriminant functions although they showed univariate differences. The BL dimensions of 22, 24, and 27 did not show significant univariate difference, but entered the discriminant function. This is reflected in an earlier work by Acharya and Mainali where a majority of BL dimensions that showed significant univariate differences did not enter the discriminant functions.[12] This suggests that univariate (independent t-test) and multivariate (discriminant) analysis may give different results when used for sexing.

The BL variables were systematically better in sex identification than MD dimensions. The stepwise discriminant analysis for BL variables differentiated the sex with an accuracy of 99.8% while for MD dimensions, the accuracy was 69.9%. The increased accuracy with which BL variables differentiated sex when compared to MD variables is similar to that reported by Prabhu and Acharya and Garn et al.[11,13,17] The latter recommended wider use of BL dimensions. It is plausible that the inability of additional MD variables to enter the stepwise discriminant analysis is responsible for their relatively low accuracy as was noticed in the study conducted by Acharya and Mainali in which there was a low accuracy of BL variables due to the inability of many BL variables to enter the stepwise discriminant analysis.[9]

When the MD and BL variables were combined and used, the posterior teeth discriminated the sexes more than the anterior ones. Although the MD variables entered the stepwise discriminant function first, 11 out of 13 variables that entered the analysis were BL dimensions. About 99.8% accuracy in sexing is seen when the BL and MD variables are used together. This is the highest among similar studies.[5,7-13] Increased sample size in our study can be one reason for this. Another reason can be ethnic mixing which causes changes in dental dimensions and hence gives varying results.[8,12,18]

Conclusions

Discriminant analysis that predicted value for correct classification is as follows:

1. MD variables had 69.9% accuracy in predicting sex with statistically significant value
2. BL variables had 99.8% accuracy in predicting sex with statistically significant value
3. MD and BL variables when combined had 99.8% accuracy for predicting sex with statistically significant value similar to that found when only BL variables are used.
The present study revealed the following findings:
1. Significant dimorphic differences between male and female teeth with males exhibiting larger teeth than females
2. Posterior teeth discriminated the sexes slightly more than the anterior teeth
3. Maxillary teeth entered the discriminant analysis more than the mandibular teeth
4. BL variables, as a unit, provide greater accuracy in gender determination when compared to MD variables
5. The accuracy in predicting gender by BL measurements alone is same as the accuracy obtained when both the MD and BL variables are considered together
6. Few teeth showed reverse dimorphism, but were not statistically significant.

One of the greatest challenges faced by forensic experts is determination of gender using skeletal remains, especially when only fragments of the body are recovered.

The results obtained in this study support the observations made by a plethora of previous studies that male tooth dimensions are statistically larger than females. The ability to differentiate sex in the population using stepwise discriminant functions was found to be very high with 99.8% accuracy. This is similar to the near 100% success in sex assessment using pelvic and skull bones, although the classification accuracy of most functions ranged between 70% and 90%. Consequently, sexing from tooth dimensions has always been considered as an adjunct rather than the sole indicator of gender. Documentation of similar observations with further studies will enable the use of each type of linear measurement independently in odontometric sex differentiation.

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