

Dimorphism in Dental Tissues: Sex differences in Archaeological Individuals for Multiple Tooth Types

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Abstract

Objectives

Dimorphism in the dentition has been observed in human populations worldwide. However, research has largely focused on traditional linear crown measurements. As imaging systems, such as micro-computed tomography (micro-CT), become increasingly more accessible, new dental measurements such as dental tissue size and proportions can be obtained. This research investigates the variation of dental tissues and proportions by sex in archaeological samples.

Materials and Methods

Upper and lower first incisor to second premolar tooth rows were obtained from 30 individuals (n=300), from 3 archaeological samples. The teeth were micro-CT scanned and surface area and volumetric measurements were obtained from the surface meshes extracted. Dental wear was also recorded and differences between sexes determined.

Results

Enamel and crown measurements were found to be larger in females. Conversely, dentine and root measurements were larger in males.

Discussion

The findings support the potential use of dental tissues to estimate sex of individuals from archaeological samples, whilst also indicating that individuals aged using current dental ageing methods may be under- or over-aged due to sex differences in enamel thickness.

Keywords

Dimorphism, Enamel, Dentine, Proportions, Micro-CT

1. Introduction

The identification of sexual dimorphism in skeletal features has been of longstanding interest in biological anthropology. At its 'simplest' it can be used to aid the estimation of the sex of an individual in paleoanthropological, archaeological and forensic samples. It can also feed into a variety of different bioanthropological conversations involving social, biological and environmental factors. In primates, sexual dimorphism has played a key role in conversations on social structures, such as those regarding breeding systems (monogamy and polygyny), which have then be used to make inferences regarding hominins and other extinct taxa (Larsen, 2003; Kanazawa and Novak, 2005; Plavcan, 2012). Alternatively, skeletal dimorphism has also been used to infer different epigenetic effects. These have included dimorphic patterns in achieved stature linked to nutrition and status (Vercellotti et al., 2011, 2014; Charisi et al., 2016; Dong et al., 2017) and skeletal robusticity linked to differential activity between sexes (Ruff, 1987; Pomeroy and Zakrzewski, 2009; Miller et al., 2018; Hill et al., 2020; Mulder et al., 2020). Teeth, unlike bone, do not remodel. They can therefore provide a unique snapshot regarding an individual. For example, they can give an insight into an individual's evolutionary history or their foetal/early childhood health status.

Dimorphism has been found throughout the human dentition world-wide; males have larger teeth than females when using both crown and cervical MD and BL measurements of the permanent dentition, both varying within and between populations and also between measurements (Moorrees et al., 1957; Garn et al., 1965, 1966; Perzigian, 1976; Garn et al., 1979; Mavroskoufis and Ritchie, 1980; Kieser, 1990; Hillson, 1996; Alt et al., 1998; Lund and Mörnstad, 1999; Harris et al., 2001; Işcan and Kedici, 2003; Kondo and Townsend, 2004; Hanihara and Ishida, 2005; Hillson et al., 2005; Al-Khateeb and Alhaija, 2006; Takahashi et al., 2007; Vodanović et al., 2007; Al-Gunaid et al., 2012; Taturan, 2012; Pilloud et al., 2014; Kerekes-Máthé et al., 2015; Moore et al., 2015). Sexual dimorphism has also been reported in deciduous dentitions, although these differences are often smaller in magnitude than those found in permanent dentitions (Harila et al., 2003; Kondo and Townsend, 2004; Anderson, 2005; Harris and Lease, 2005; Adler and Donlon, 2010; Ribeiro et al., 2013).

There has been a focus on linear MD and BL canine dimensions as these have been noted as particularly dimorphic (Garn et al., 1965; Moss and Moss-Salentijn, 1977; Hillson, 1996; Lund and Mörnstad, 1999; Pettenati-Soubayroux et al., 2002; Işcan and Kedici, 2003; Schwartz and Dean, 2005; Acharya and Mainali, 2007, 2009, Viciano et al., 2011, 2015; Acharya et al., 2011; Ribeiro et al., 2012; Tardivo et al., 2015). Dimorphism has also been observed in premolars and molars (Prabhu

and Acharya, 2009; Viciano et al., 2011, 2013, 2015; Zorba et al., 2011) and occasionally in incisors (Garn et al., 1964; Staka et al., 2016). Sexual dimorphism has also been reported in tooth root number in modern humans (Sert and Bayirli, 2004; Shields, 2005), extant apes, and fossils hominoids and hominins (Abbott, 1984; Shields, 2005; Moore et al., 2015). There has been mixed support for dimorphism of canine and premolar root lengths (Garn et al., 1979; Moore et al., 2015), and less evidence exists for dimorphism for intercuspal distances (Townsend, 1985; Townsend et al., 2003).

The analysis of other dental measurements, such as tissue volumes, has been less common than traditional crown diameters, but dental tissue proportions, tissue volumes and surface areas have also been identified as being sexually dimorphic (Stroud et al., 1994; Harris and Hicks, 1998; Zilberman and Smith, 2001; Schwartz and Dean, 2005; Saunders et al., 2007; Feeney et al., 2010; Tardivo et al., 2015, 2011; Kazzazi and Kranioti, 2017; García-Campos et al., 2018a; b; Sorenti et al., 2019). Despite being used infrequently, the use of dental tissue volumes and surface areas has been recommended for sex determination (García-Campos et al., 2018a).

There is mixed Some evidence exists for sexual dimorphism in enamel thickness (Hall et al., 2007; García-Campos et al., 2018a; b; Sorenti et al., 2019). Overall tooth size, and the sizes of the crown and the root have been found to be larger in males. Enamel volume has been found to be larger in females, and consequently it is thought that the enamel does not significantly contribute to overall dental dimorphism (Stroud et al., 1994; Harris and Hicks, 1998; Feeney et al., 2010; García-Campos et al., 2018b; a). Most studies have focused largely on the posterior dentition although recent studies have analysed canine sexual dimorphism (García-Campos et al., 2018a; b).

The current study investigated sexual dimorphism in dental tissues and proportions. This research is the first to study to concurrently assess this across multiple tooth types. It employed micro-CT imaging to obtain surface area and volumetric measurements from dental tissues and proportions. The potential of using dental tissues for sex estimation in archaeological samples is explored. The aims of this study were twofold: 1) identify sexual dimorphism in dental tissues and proportions and 2) investigate the potential of using them for sex estimation using discriminant function analysis.

2. Materials and Methods

2.1 Materials

The sample studied consisted of permanent teeth, maxillary and mandibular incisors (central and lateral), canines and premolars (central and lateral), with left and right sides pooled for each individual. The sample comprised 300 teeth from 30 individuals (16 Females and 14 Males) from three archaeological samples from the south of the UK. The first sample is derived from the Anglo-Saxon cemetery at Great Chesterford, Essex ($n=10$), dated from 5th – 7th century AD. The second sample is derived from the Early Medieval monastic cemetery at Llandough, South Wales ($n=10$) dated from 7th – 11th century AD. The final sample was obtained from the Late Medieval priory cemetery of St Peter and Paul, Taunton, Somerset ($n=10$) dated from 12th – 15th century AD. Individuals with Molnar (1971) dental wear scores over 4 were excluded, as were those with dental anomalies and pathologies.

Age and sex were estimated according to British guidelines (Buikstra and Ubelaker, 1994). Sex was estimated based upon the dimorphic characteristics of the pelvis and skull, where available (Buikstra and Ubelaker, 1994, pp 15–21). Age estimates were taken from pelvic characteristics of the pubic symphysis (Todd, 1920; Brooks and Suchey, 1990) and auricular surface (Lovejoy et al., 1985), and were then classified by category: young adult, middle adult and old adult (Table 1).

Table 1 Sample composition by period/site (GC- Great Chesterford, LLAN -Llandough and COAS – Taunton), age (Young Adult (YA) and Middle Adult (MA)) and sex.

2.2 Data Acquisition

The teeth were micro-CT scanned using different scanning facilities and parameters. Loose teeth and teeth in small bony fragments were scanned using a SkyScan 1272 at the University of Bristol and a SkyScan 1275 at the Sumitomo Laboratory, Swansea. Loose teeth were scanned at 90 kV and 70 μ A using a 0.5 Al & 0.038 Cu filter, for a target resolution of 17.5 μ m. Teeth in small bony fragments were scanned at 100 kV and 100 μ A using a 1.0 mm Cu filter, for a target resolution of 17.5 μ m. Teeth in large bony fragments and crania were scanned using a Nikon XT H 320 at the National Composite Centre (NCC), Bristol, at 145 kV and 110 μ A using no filter, for a target resolution of 65 μ m (For more details on scan parameters see supporting information).

The scans were reconstructed using Nrecon (Bruker micro-CT, Belgium) and CTPro3D (Nikon Metrology, Herts UK). The data was then segmented using ScanIP (Simpleware, Exeter, UK) based on

thresholding criteria to create individual masks for enamel, dentine, pulp chamber and whole tooth (Figure 1). Cracks were virtually filled in for these masks. For the purposes of this study, cementum was included in the dentine mask as only the external tooth geometry was required. For each threshold range, a surface mesh was generated and exported, resulting in four unique meshes for each tooth: enamel, dentine, pulp and whole tooth.

2.2.1 Measurements

Tooth tissue volumes, surface areas and proportions were obtained for each tooth (Table 2; Figure 1; Figure 2). The root was defined as the tooth present below the CEJ, as obtained in MATLAB (Mathworks, MA, USA). The surfaces were downsampled prior to separation of the crown and root. Volumetric measurements could only be obtained using closed meshes; meshes were closed in ICEM (ANSYS Inc., Canonsburg PA, USA). Degree of wear was recorded qualitatively using Molnar (1971).

Table 2 Definition of dental measurements.

2.2.2 Statistical Analysis

Size, surface area and volumetric measurements were analysed in SigmaPlot 13.0 (Systat Software, San Jose, CA). Data was first checked for normality using a Shapiro-Wilk test and for equal variance using a Brown-Forsythe test. If normality was achieved, a One-way-ANOVA test was performed ($\alpha = 0.05$). Data that failed the normality and/or equal variance tests were analysed using a Whitney Rank Sum test ($\alpha = 0.05$). The effect of dental wear on the results obtained was also tested. A chi-squared test was carried out to determine any association between degree of wear and sex ($\alpha = 0.05$). Finally, an ANCOVA was carried out to control for degree of wear when comparing dental measurements ($\alpha = 0.05$). Degree of wear was also used as a proxy for age, correcting results for any difference in age profile by sex. To visualise variation in tooth measurements, a correlation matrix Principal Component Analysis (PCA) was performed on each tooth type.

The Discriminant Function Analysis (DFA) was carried out on the surface area and volumetric measurements in SPSS 26.0 (IBM Corp, Armonk, NY). A number of multivariate discriminant functions were created. For each tooth, a function was created for the volume (D1), surface area (D2) and both the surface area and volume of each dental tissue (D3). The same was done for dental

tissue proportions (D4, D5, D6). A function was also created for all the dental measurements obtained (D7). Finally, the same functions were created for all teeth pooled together and for the slightly worn teeth only (n=85). Slightly worn teeth defined as having a Molnar (1971) of 2 or below. These were carried out on the original sample as well as using a cross-validation leave-one out procedure.

3. Results

3.1 Wear

Degree of wear was found to significantly differ by sex in all three samples (Table 3), in all cases male crowns were found to have a greater degree of wear than females (Figure 3). When all sites were pooled and each tooth was analysed separately, degree of dental wear was found to significantly differ by sex in upper lateral incisors only (Table 4; Figure 4).

Table 3 Chi-Squared results for sex and tooth wear by site: Great Chesterford (GC), Llandough (LLAN) and Taunton (COAS). Significant results in bold.

Table 4 Chi-Squared results for sex and tooth wear by tooth. Significant results in bold.

3.1 Sex

3.2 Sex Differences

3.1.1 Enamel and Crown

Overall, female enamel and crown measurements were found to be larger than males (Table 5 and 6). In most tooth types studied, EVol was found to be significantly larger in females than males (UI1, UI2, UC, UPM1, UPM2, LPM1, LPM2). Conversely, ESA was not found to be significantly different in any teeth. CVol was found to be significantly larger in females than males in UC, UPM1, UPM2. Whereas CSA was significantly larger in females in UI2, UC and UPM2.

3.1.2 Dentine and Root

DVol was significantly larger in males than females in UPM2, LI2 and LC. However, DSA was only significantly larger in male UPM2 and LCs. The RVol of UPM2 and LC was significantly larger in males. RSA was significantly larger in males than females in most teeth (UPM2, LI1, LI2, LC, LPM1) (Table 5 and 6).

3.1.3 Whole Tooth

Whole tooth measurements (WTSA and WTVol) were found to be significantly larger in male LCs than for females (Table 6).

Table 5 Sex differences in dental measurements for upper teeth. Showing male and female mean measurements with S.D and p values. † Denotes non-parametric test used. Significant results in bold.

Table 6 Sex differences in dental measurements for lower teeth. Showing male and female mean measurements with S.D and p values. † Denotes non-parametric test used. Significant results in bold.

3.2 Wear and Sex

3.2.1 Enamel and Crown

It is possible that the difference detected previously were affected by dental wear. After using an ANCOVA to control for wear, a significant difference was found in enamel and crown measurements of male and female upper canines (Table 7). Upper canine EVol and CVol volume were found to be significantly larger in females than males.

3.2.2 Dentine and Root

After controlling for the degree of wear, numerous dentine and root measurements were found to be significantly larger in males than females in a number of teeth: upper lateral incisor (RSA), upper lateral premolar (DVol and RVol), lower central incisor (RSA), lower lateral incisor (DVol), lower canine (DSA and RVol) and lower central premolar (DSA, RSA and DVol) (Table 7).

3.2.3 Whole Tooth

After constraining for degree of wear, male lower canine WTSA and WTVol was found to be significantly larger than for females (Table 7).

Table 7 ANCOVA results for all dental measurements. Differences between male and female dental measurements after the removal of degree of wear. – indicates that an ANCOVA could not be performed as the data did not pass the normality and/or equal variance test. Significant results in bold.

3.3 Principal Component Analysis

The PCA performed on each tooth type identified the first two PCs accounting for between 77.7% and 87.7% of the variation. Figures 5 and 6 contain the principal component plots of all dental measurements for each tooth type and the corresponding loadings of these measurements. In most

instances, male measurements appear to be more varied than female along both PCs. There is no complete separation of the male and female clusters in any tooth type, however there is a degree of separation in some instances: this is more marked in maxillary teeth. For example, differences are present along PC 2 in upper central incisors, (root volume), and upper canines (crown volume). In upper lateral premolars, separation of male and female clusters occurs along both PC 1 and PC 2, and is related to whole tooth, dentine and root measurements, as well as enamel and crown measurements respectively.

3.4 Discriminant Functions

A summary of the discriminant functions and the accuracy of their classification is given in Tables 8-9 and Supporting Information respectively. Table 7 and 8 contain the Eigenvalue, Canonical Correlation, Wilks' Lambda and Significant value for each discriminant function. The discriminant functions were found to be significant in most instances. DF 1 and 4 were significant in all instances (Table 8 and 9). DF 2 and 3 were mostly significant (Table 9). DF 5 to 7 were found to be less significant than the other discriminant functions (Table 9).

Table 8 Eigenvalue (Eig), Canonical Correlation (CanCor), Wilks' Lambda (Wilks) and Significance value (Sig) for Discriminant 1, 2, and 3 for each tooth type, all teeth and all slightly worn (SW) teeth. Significant results in bold.

Table 9 Eigenvalue (Eig), Canonical Correlation (CanCor), Wilks' Lambda (Wilks) and Significance value (Sig) for Discriminant 4, 5, 6 and 7 for each tooth type, all teeth and all slightly worn teeth. Significant results in bold.

3.4.1 Tooth type

When the discriminant functions were separated by tooth type, classification rate varied between 90% and 66.7%. The accuracy of female classification varied between 100% to 56.25%. The accuracy of male classification varied between 100% to 57.14%. Overall, the female classification rate was usually higher than males. The difference between female and male classification rates was as high as 30.36%.

After cross-validation, the classification rate varied between 87.50% to 35.70%. Female classification varied between 87.50% to 56.35% and male classification varied between 85.71% to 35.71%. The rate of female classification was predominantly higher than males, with the greatest difference at 70% (Supporting Information).

3.4.2 All

When all teeth were pooled together, classification accuracy varied between 72.33% and 67.67%. The rate of female classification ranged between 77.5% and 71.25%. Male classification rate ranged between 70% and 62.14%. The classification of females was greater for all discriminant functions, with the largest difference being 11.79%.

After cross-validation, classification accuracy varied between 71% and 66%. The female classification rate varied between 74.28% and 69.37%, and the male classification varied between 67.14% and 62.14%. The classification rate was greater for females for all discriminant functions, with the largest difference of 10.25% (Supporting Information).

3.4.3 All slightly worn teeth

When only the slightly worn teeth were analysed, the classification accuracy varied between 75.29% and 67.06%. The female classification rate varied between 80.95% and 61.90%, and that of the males ranged between 76.56% and 67.19%. The classification of slightly worn teeth was greater in males in all but one discriminant function, with the greatest difference being 14.66%.

After cross-validation, classification ranged between 71.76% and 67.06%. Female classification varied between 66.67% and 57.14%. Male classification varied between 76.56% and 68.75%. The classification of slightly worn teeth was greater in males in all but one discriminant function, with the greatest difference being 19.42% (Supporting Information).

4. Discussion

4.1 Wear

Wear was found to significantly differ by sex within each sample despite having similar age profiles. This may be indicative of dietary differences between males and females, as diet is intimately connected with social identity. How diet and sex relate is not clear-cut. Isotopic evidence from the sites and others from the period indicate no differential access to dietary resources based on sex (Muldner and Richards, 2007; Mays and Beavan, 2012; Monterrosa Preziosi, 2016; Hemer et al., 2017), and therefore differential access to dietary resources is an unlikely cause of the difference observed.

Differences in dental wear may be indicative of sex differences in enamel thickness, with the female enamel found to be thicker than that of males (Hall et al., 2007; García-Campos et al., 2018a; b;

Sorenti et al., 2019). Consequently, if wear is occurring at the same rate, the thinner enamel in males is likely to wear sufficiently so as to expose the dentine first. If this is the case, an important consideration is raised for age estimation based on dental wear. Current methods that utilise quantity and patterning of dentine exposure to age individuals do not differentiate between males and females (Brothwell, 1981; Molnar, 1971; Smith & Knight, 1984). Failure to consider a difference in enamel thickness may result in the over- or under-estimation of age in males and females respectively.

4.2 Sex Differences

Sexual dimorphism of the permanent dentition has been well established and the results here support this. Previous investigations of dental sexual dimorphism have focused on linear MD and BL canine dimensions (Garn et al., 1965; Hillson, 1996; Lund and Mörnstad, 1999; Schwartz and Dean, 2005; Acharya and Mainali, 2007; Acharya et al., 2011; Viciano et al., 2011, 2015). Dimorphism has also been observed in premolars and molars (Prabhu and Acharya, 2009; Viciano et al., 2011, 2013, 2015; Zorba et al., 2011) and occasionally in incisors (Garn et al., 1964; Staka et al., 2016). Analysis of other dental measurements, such as tissue volumes has been less common. Research has focused on tissue volume and surface areas in canines (De Angelis et al., 2015; García-Campos et al., 2018a; b). De Angelis and colleagues (2015) have advocated the analysis of dimorphism in tissue volumes of other tooth types. The results of this study suggest that this proposition is well-founded, with a significant difference observed in all tooth classes.

The general dimorphic pattern that was identified was a larger surface area and volume of the dentine and the root in males and in the enamel and the crown in females. A similar pattern has been observed in previous studies; males have a greater dentine component and females have thicker enamel (Stroud et al., 1994; Schwartz and Dean, 2005; Smith et al., 2006; Saunders et al., 2007; Feeney et al., 2010; García-Campos et al., 2018a; b). A study of an Iranian archaeological sample found significant sexual dimorphism in the root volume of all teeth (Kazzazi and Kranioti, 2017). In the current study, significant dimorphism in root volume was found for all tooth types (upper central incisor, upper lateral premolar, lower canine and lower lateral premolar). Kazzazi and Kranioti (2017) suggest that their results demonstrate the potential of tooth root volume measurements for sex assessment in archaeological samples. They also recommend the incorporation of more archaeological samples and contemporary populations due to the small sample size of the original study. Both of these statements hold-true here.

Lower canine whole tooth volume was found to be significantly larger in males than females. This is consistent with previous research that has established sexual dimorphism as greatest in canines (Garn et al., 1965; Hillson, 1996; Lund and Mörnstad, 1999; Schwartz and Dean, 2005; Acharya and Mainali, 2007; Acharya et al., 2011; Viciano et al., 2011, 2015). Traditionally such sexual dimorphism has been assessed using MD and BL crown dimensions (Hillson, 1996; Lund and Mörnstad, 1999; Schwartz and Dean, 2005; Acharya and Mainali, 2007; Acharya et al., 2011), later modified to analysis of cervical diameters to avoid effects of dental wear. Recent research has also established this difference in whole tooth volumes (De Angelis et al., 2015; García-Campos et al., 2018a).

Dental wear, in all of its forms, involves the gradual degradation and removal of enamel. Qualitative indices for recording wear typically utilise a grading or scoring system to identify the degree or severity of wear progression (Bardsley, 2008, p 15). Qualitative methods rely predominantly on descriptions of gross wear and are often based on exposed dentine (Bardsley, 2008; D’Incau et al., 2012). After controlling for degree of wear, when this analysis could be performed, a significant difference between male and female enamel and crown measurements was only found in upper canines. Both upper canine enamel volume and crown volume were found to be significantly larger in females.

The specific processes at play are unclear, however it is thought that a combination of genetic and hormonal influences result in dental sexual dimorphism. There is extensive literature, from studying individuals with chromosomal aneuploidies, on the genetic influence of sex-linked genes on the size and shape of the crown (Alvesalo and Portin, 1980; Alvesalo and Varrelä, 1980; Kari et al., 1980; Kirveskari and Alvesalo, 1982; Townsend et al., 1984; Townsend and Alvesalo, 1985; Alvesalo et al., 1987; Midtbø and Halse, 1994a; Nakayama et al., 2005; Lähdesmäki and Alvesalo, 2007), root (Filipsson et al., 1965; Midtbø and Halse, 1994b; Lähdesmäki and Alvesalo, 2004, 2005, 2006, 2007, 2010) and dental tissues (Alvesalo and Tammisalo, 1981). The Y chromosome has been linked to an increase within the activity of dental lamina (Alvesalo, 1997). Conversely, the X chromosome appears to affect enamel deposition (Alvesalo et al., 1991; Lähdesmäki and Alvesalo, 2010).

This research is supported by studies of amelogenin, which plays a crucial role in enamel development, and is specifically responsible for enamel thickness (Gibson, 2011). Amelogenin genes are present on both the X (AMELX) and Y (AMELY) chromosomes. In males, AMELX and AMELY are responsible for 90% and 10% of the amelogenin production respectively (Salido et al., 1992).

Alterations in these genes have shown that differences in their transcriptional products influence proportion of enamel produced (Gibson, 2011; Hu et al., 2012; Cho et al., 2014; Kim et al., 2017; Duan et al., 2019).

The exact contribution of sex hormones to sexual dimorphism is yet to be established (Kondo et al., 2005; Kondo and Townsend, 2006; Guatelli-Steinberg et al., 2008; Ribeiro et al., 2012). Opposite sex twins have been studied to assess the role of intrauterine diffusion of hormones, opposite sex twins have shown greater tooth dimensions than other females (Dempsey et al., 1999; Ribeiro et al., 2012, 2013). Differences in the percentage of dimorphism between the primary and secondary dentition, greater in the permanent dentition, parallels with surges in testosterone (Moorrees et al., 1957; Gingerich, 1974; Kondo and Townsend, 2004; Kondo et al., 2005; Ribeiro et al., 2012). Changes in dentine thickness before and after puberty have been shown to coincide with changing levels of testosterone (Zilberman and Smith, 2001). Growth hormone receptors have also been discovered in dental tissues acting as regulators of growth (Young et al., 1992; Zhang et al., 1997, 2005; Litsas, 2015); they are influenced by oestrogens and others sex hormones (Hietala et al., 1998; Meinhardt and Ho, 2006; Inaba et al., 2013; Houari et al., 2016; Alhodhodi et al., 2017). Research suggests that oestrogen and androgen receptors within the dental pulp play a role in dentinogenesis (Inaba et al., 2013). Evidence, however, has been found against the major role of sex hormones in sexual dimorphism (Alvesalo and Varrel, 1980; Guatelli-Steinberg et al., 2008). More work needs to be done to make clear the exact mechanisms that control dimorphism tissue proportions.

Even though the exact aetiology of the dimorphism of dental tissue volumes and surface areas is unknown, their use for sex determination has been recommended (García-Campos et al., 2018a). The dentine portion of the tooth appears to contribute more to overall size than enamel.

Going forward, further research will help to establish the potential use of dental tissues for sex estimation in humans. In addition to this, the analysis of different hominid samples can help to establish inter-species differences in dimorphism of dental tissues. From this dimorphism in dental tissues, with the advantage of providing a snapshot of early childhood with a limited epigenetic window, may be used in wider conversations related to sexual dimorphism.

5. Conclusions

The general dimorphic pattern identified was a larger surface area and volume of the dentine and the root in males and of the enamel and the crown in females. This corroborates differences found

elsewhere. Dimorphism in dental tissues offers a new potential method of sexing individuals, of value in both forensic science and archaeology. However, these results do bring caveats to future and wider research. Firstly, population comparisons using dental tissue volumes and proportions should only include sexed individuals to avoid skewed results. Secondly, individuals aged using current dental ageing methods may under- or over-aged due to sex differences in enamel thickness.

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Data Availability Statement

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

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Figure Legends

Figure 1 Dental tissue masks from which measurements were taken: whole tooth, enamel, dentine and pulp chamber.

Figure 2 Crown and root surface area and volumetric meshes. Left: Crown surface area (CSA) and root surface area (RSA). Right: Crown volume (CVol), coronal dentine volume (CDVol) and root volume (RVol).

Figure 3 Distribution of degree of wear by sex in each sample. Degree of Wear Score: 1 - unworn, 2 – minimal wear, 3 - slight wear and 4 – wear with minimal dentine showing (Molnar 1971).

Figure 4 Distribution of degree of wear by sex in upper lateral premolars. Degree of wear score: 1 - unworn, 2 – minimal wear, 3 - slight wear and 4 – wear with minimal dentine showing (Molnar 1971).

Figure 5 PCA performed on each upper tooth measurements for females (red) and males (grey). A) PCA plot; B) Loading values showing the measurements associated with PC 1 (top) and PC 2 (bottom).

Figure 6 PCA performed on each lower tooth measurements for females (red) and males (grey). A) PCA plot; B) Loading values showing the measurements associated with PC 1 (top) and PC 2 (bottom).

	GC		LLAN		COAS	
	YA	MA	YA	MA	YA	MA
Male	3	1	4	1	3	2
Female	3	3	5	0	3	2
Total	6	4	9	1	6	4

Measurement	Code	Description	Measurement
Enamel Volume	EVol	Volume of enamel	mm ³
Dentine Volume	DVol	Volume of dentine	mm ³
Pulp Volume	PVol	Pulp volume	mm ³
Enamel Surface Area	ESA	Enamel surface area	mm ²
Dentine Surface Area	DSA	Dentine surface area	mm ²
Whole Tooth Volume	WVol	Volume of whole tooth	mm ³
Whole Tooth Surface Area	WTSA	Surface Area of whole tooth including pulp	mm ²
Crown Volume	CVol	Volume of crown cap including enamel, dentine and pulp.	mm ³
Crown Surface Area	CSA	Surface Area of Crown	mm ²
Root Volume	RVol	Volume of root including dentine and pulp	mm ³
Root Surface Area	RSA	Surface Area of whole tooth including pulp	mm ²
Coronal Dentine Volume	CDVol	Volume of dentine in crown including pulp	mm ³

Site	p
GC	0.012*
LLAN	0.021*
COAS	0.002**

Tooth	p
UI1	0.186
UI2	0.024*
UC	0.258
UPM1	0.343
UPM2	0.240
LI1	0.080
LI2	0.238
LC	0.050
LPM1	0.157
LPM2	0.057

	UI1			UI2			UC			UPM1			UPM2		
	M	F	p	M	F	p	M	F	p	M	F	p	M	F	p
EVol	64.403 (27.577)	83.785 (20.967)	0.037*	43.916 (21.029)	60.555 (14.538)	0.016*	70.331 (25.582)	97.568 (21.740)	0.003**	73.934 (27.957)	96.845 (24.086)	0.022*	72.951 (23.089)	101.216 (25.149)	0.003**
DVol	392.635 (50.017)	361.743 (48.249)	†0.101	251.402 (58.653)	232.019 (28.164)	†0.164	429.104 (107.314)	394.194 (56.717)	0.226	337.581 (62.844)	302.349 (36.365)	0.0726	347.481 (54.081)	295.619 (45.619)	0.008**
PVol	13.216 (5.056)	14.417 (4.841)	0.512	8.903 (2.920)	9.531 (2.899)	0.560	18.889 (5.317)	17.601 (6.377)	0.556	13.273 (5.650)	12.018 (3.149)	†0.983	13.121 (3.622)	11.160 (2.807)	†0.371
ESA	270.175 (78.921)	307.344 (52.943)	0.137	194.424 (57.365)	226.504 (30.726)	†0.059	249.437 (56.088)	277.062 (31.144)	0.101	240.351 (54.348)	262.097 (33.823)	0.193	234.410 (42.252)	260.294 (32.242)	0.068
DSA	418.330 (47.071)	398.834 (42.607)	0.244	322.117 (55.035)	314.548 (36.524)	0.644	464.849 (86.938)	442.993 (53.651)	0.408	419.799 (61.700)	393.722 (44.779)	0.192	416.503 (60.753)	375.094 (42.284)	0.037*
WVol	471.493 (65.404)	460.903 (63.414)	0.656	304.643 (73.333)	302.572 (40.626)	†0.693	519.071 (124.273)	510.450 (71.690)	†0.467	425.475 (75.826)	412.137 (54.574)	0.581	434.511 (54.515)	410.190 (58.803)	0.252
WSA	393.510 (42.103)	385.587 (39.815)	0.601	393.510 (42.103)	385.587 (39.815)	0.920	458.924 (76.910)	423.410 (48.103)	0.813	397.297 (54.322)	383.286 (42.980)	0.437	393.251 (48.373)	367.998 (40.278)	0.130
CVol	193.270 (59.358)	220.873 (37.413)	0.134	125.068 (50.271)	141.191 (26.651)	†0.371	170.952 (54.280)	214.765 (31.565)	0.010*	162.276 (52.837)	197.037 (38.675)	0.047*	156.565 (42.913)	195.386 (34.414)	0.010*
CSA	158.527 (35.613)	180.981 (26.737)	0.059	116.424 (28.364)	134.259 (18.375)	0.048*	143.732 (33.543)	164.220 (17.083)	† 0.036*	149.472 (39.057)	158.545 (20.573)	0.424	138.706 (24.831)	157.831 (18.870)	0.024*

RVol	278.223 (65.856)	240.030 (39.593)	0.060	179.574 (45.227)	161.381 (21.422)	† 0.092	348.120 (105.853)	295.686 (60.918)	0.102	263.199 (70.222)	215.099 (30.645)	† 0.109	277.946 (71.023)	214.804 (35.983)	0.004**
RSA	224.569 (28.927)	208.539 (28.081)	0.135	187.626 (32.032)	170.274 (22.296)	0.092	265.082 (62.569)	251.122 (35.635)	0.452	243.017 (39.921)	220.646 (28.144)	0.084	246.360 (47.985)	207.937 (28.328)	† 0.036*
CDVol	128.867 (40.312)	137.088 (25.274)	0.503	81.152 (36.455)	80.636 (15.338)	† 0.513	100.620 (50.490)	117.196 (25.897)	† 0.513	88.342 (39.973)	100.192 (19.423)	0.301	81.152 (36.455)	80.636 (15.338)	† 0.442

	LI1			LI2			LC			LPM1			LPM2		
	M	F	p	M	F	p	M	F	p	M	F	p	M	F	p
EVol	24.034 (11.135)	30.001 (8.208)	0.103	35.567 (9.686)	40.775 (11.087)	0.070	64.858 (21.775)	69.161 (20.182)	0.579	67.056 (19.741)	82.156 (18.893)	0.041*	69.856 (18.600)	92.779 (28.295)	0.016*
DVol	188.827 (32.366)	167.609 (26.221)	0.0572	231.744 (33.558)	206.742 (31.817)	0.045*	413.750 (70.455)	334.996 (48.136)	0.001**	291.883 (43.412)	266.410 (43.089)	0.119	323.360 (60.132)	292.307 (48.700)	0.137
PVol	5.242 (2.265)	5.585 (1.565)	0.630	7.744 (3.220)	6.790 (1.978)	† 0.519	17.196 (6.700)	14.078 (5.231)	† 0.152	12.692 (3.728)	11.259 (2.784)	† 0.253	13.459 (4.327)	12.103 (2.971)	0.331
ESA	143.555 (40.795)	156.205 (30.847)	0.343	172.513 (31.871)	183.72 (34.219)	† 0.177	240.071 (57.128)	240.559 (44.172)	0.917	221.036 (38.002)	234.289 (32.794)	0.314	221.227 (32.355)	242.783 (44.054)	0.147
DSA	278.136 (41.460)	261.216 (36.443)	0.244	328.215 (38.882)	310.055 (39.946)	† 0.152	463.035 (55.842)	409.659 (53.344)	0.012*	364.219 (46.218)	343.257 (44.425)	0.216	380.850 (54.440)	357.957 (42.557)	0.216
WVol	218.577 (38.715)	203.894 (33.316)	0.274	274.386 (39.774)	255.210 (41.643)	0.209	492.319 (82.327)	419.504 (68.012)	0.013*	372.692 (55.615)	361.024 (56.702)	0.575	407.594 (65.485)	398.281 (71.786)	0.719
WSA	257.914 (35.404)	243.205 (31.701)	0.240	302.013 (31.172)	286.966 (34.769)	† 0.129	422.876 (48.341)	379.220 (47.755)	0.019*	346.439 (38.920)	336.459 (42.752)	0.512	363.591 (43.795)	356.896 (46.587)	0.694
CVol	96.792 (46.701)	81.636 (17.065)	† 0.662	113.649 (64.686)	98.218 (19.716)	† 0.755	158.293 (41.321)	162.289 (38.058)	0.785	162.359 (45.051)	159.020 (30.154)	0.634	157.618 (34.535)	182.031 (42.417)	0.102
CSA	100.254 (33.665)	92.943 (15.684)	† 0.662	112.852 (39.324)	106.153 (16.860)	† 0.493	139.612 (24.772)	141.153 (24.539)	0.886	136.627 (30.487)	140.926 (19.720)	0.646	133.071 (21.334)	149.190 (26.618)	0.084

RVol	121.784 (43.584)	122.257 (19.483)	† 0.884	160.737 (67.249)	156.991 (27.054)	† 0.152	334.025 (66.520)	257.215 (37.302)	† <0.001**	210.333 (36.710)	192.004 (33.456)	0.164	249.976 (57.480)	216.250 (37.267)	0.069
RSA	183.360 (40.719)	151.658 (21.811)	0.011*	202.611 (35.656)	179.547 (21.338)	† 0.040*	269.132 (51.192)	236.509 (26.824)	† 0.044*	229.622 (50.363)	195.178 (28.153)	0.026*	235.416 (43.648)	224.867 (59.935)	† 0.471
CDVol	72.758 (42.284)	51.635 (9.50)	† 0.350	80.082 (58.914)	57.443 (10.653)	† 0.519	93.435 (35.222)	93.128 (19.953)	0.976	95.302 (28.477)	86.863 (14.595)	† 0.852	89.047 (22.116)	89.252 (16.373)	0.977

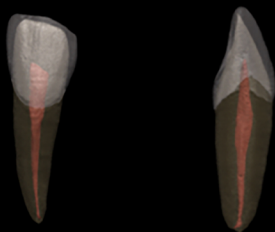
	UI1	UI2	UC	UPM1	UPM2	LI1	LI2	LC	LPM1	LPM2
EVol	0.293	0.535	0.035*	0.146	0.055	0.476	0.141	0.876	0.691	0.168
DVol	-	-	0.879	0.068	0.041 *	0.083	0.048*	<0.001**	0.038*	0.105
PVol	0.609	0.678	0.418	-	-	0.926	0.219	0.124	0.097	0.249
ESA	0.769	-	0.427	0.543	0.535922	0.967	-	-	0.624	0.644
DSA	0.158	0.155	0.278	0.142	0.1093	0.264	-	0.004**	0.045*	0.173
WVol	0.374	-	0.609	0.250	0.205	0.222	0.147	0.005**	0.072	0.161
WSA	0.276	0.194	0.541	0.241	0.187	0.242	0.165	0.006**	0.069	0.181
CVol	0.509	-	0.009*	0.215	0.121	-	-	0.85110	0.245	0.524
CSA	0.337	0.681257	-	0.834	0.177	-	-	0.73887	0.258	0.474
RVol	0.125	-	0.5982	-	0.024*	0.606	0.916	<0.001*	0.121	0.096
RSA	0.255	0.039 *	0.607	0.076	0.061	0.028*	-	-	<0.001*	0.652
CDVol	0.789	-	-	0.509	-	-	-	0.727	-	-

	DF 1				DF 2				DF 3			
	Eig	CanCor	Wilks	Sig	Eig	CanCor	Wilks	Sig	Eig	CanCor	Wilks	Sig
UI1	0.464	0.563	0.683	0.018*	0.295	0.477	0.772	0.031*	1.054	0.716	0.487	0.003**
UI2	0.662	0.619	0.616	0.005**	0.336	0.501	0.749	0.020*	0.631	0.622	0.613	0.029*
UC	0.588	0.609	0.630	0.007**	0.375	0.552	0.727	0.014*	0.644	0.626	0.608	0.027*
UPM1	0.583	0.591	0.650	0.010*	0.259	0.0454	0.794	0.044*	0.583	0.607	0.632	0.039*
UPM2	0.832	0.674	0.546	0.001**	0.364	0.516	0.733	0.015*	1.108	0.725	0.474	0.002**
LI1	0.494	0.575	0.669	0.014*	0.277	0.466	0.783	0.037*	0.533	0.590	0.652	0.053
LI2	0.591	0.610	0.628	0.006**	0.366	0.518	0.732	0.015*	0.592	0.610	0.628	0.037*
LC	0.900	0.688	0.526	0.001**	0.365	0.517	0.733	0.015*	0.971	0.702	0.507	0.004**
LPM1	0.470	0.765	0.680	0.017*	0.247	0.445	0.802	0.051	0.487	0.572	0.672	0.072
LPM2	0.518	0.584	0.659	0.011*	0.285	0.471	0.778	0.034*	0.682	0.637	0.595	0.021
All	0.313	0.488	0.762	<0.001**	0.205	0.412	0.830	<0.001**	0.322	0.494	0.756	<0.001
All SW	0.290	0.474	0.775	<0.001**	0.091	0.289	0.917	0.028*	0.292	0.475	0.774	0.001

	DF 4				DF 5				DF 6				DF 7			
	Eig	CanCor	Wilks	Sig	Eig	CanCor	Wilks	Sig	Eig	CanCor	Wilks	Sig	Eig	CanCor	Wilks	Sig
UI1	0.426	0.547	0.701	0.024*	0.329	0.498	0.752	0.057	0.553	0.597	0.644	0.088	1.269	0.748	0.441	0.042*
UI2	0.557	0.598	0.642	0.008**	0.536	0.591	0.651	0.010*	0.631	0.622	0.613	0.057	0.962	0.700	0.510	0.070
UC	0.824	0.676	0.543	0.001**	0.371	0.520	0.730	0.039*	1.114	0.726	0.473	0.005**	1.391	0.763	0.418	0.015*
UPM1	0.490	0.573	0.671	0.014*	0.157	0.368	0.865	0.278	0.643	0.625	0.609	0.053	1.269	0.748	0.441	0.023*
UPM2	0.686	0.638	0.593	0.003**	0.556	0.598	0.642	0.008**	0.950	0.698	0.513	0.010*	2.379	0.839	0.296	0.002**
LI1	0.596	0.611	0.627	0.006**	0.291	0.475	0.775	0.080	0.627	0.621	0.615	0.058	0.730	0.650	0.578	0.168
LI2	0.914	0.691	0.552	0.001**	0.181	0.392	0.847	0.220	0.991	0.705	0.502	0.009**	1.065	0.718	0.484	0.048*
LC	0.870	0.682	0.535	0.001**	0.575	0.604	0.635	0.007**	0.967	0.701	0.508	0.010*	1.242	0.744	0.446	0.046*
LPM1	0.618	0.618	0.618	0.005**	0.412	0.540	0.708	0.027*	0.894	0.687	0.528	0.014*	0.991	0.705	0.502	0.104
LPM2	0.511	0.582	0.662	0.012*	0.283	0.470	0.779	0.085	0.635	0.623	0.612	0.056	0.719	0.647	0.582	0.175
All	0.308	0.485	0.765	<0.001**	0.158	0.369	0.865	<0.001**	0.321	0.493	0.757	<0.001**	0.344	0.506	0.744	<0.001**
All SW	0.247	0.445	0.802	<0.001**	0.211	0.418	0.825	0.001**	0.344	0.506	0.744	0.001**	0.509	0.581	0.663	<0.001**



Enamel



Whole Tooth



Dentine



Pulp Chamber

Surface Area Meshes

ESA



DSA



Volume Meshes

EVol



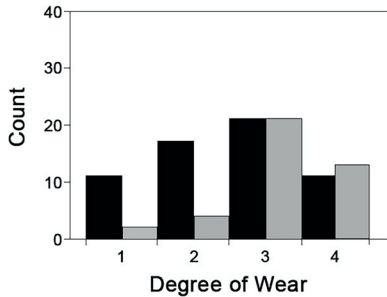
CDVol



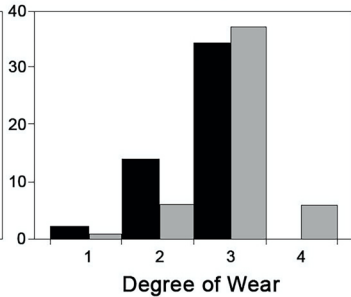
DVol



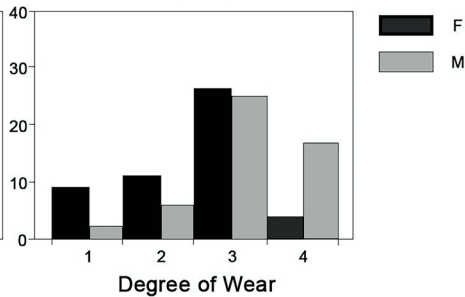
GC



LLAN



COAS



UI2

