# QUESTA E' LA VERSIONE ACCETTA DEL SEGUENTE LAVORO:

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## Sexual dimorphism of canine volume: A pilot study

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#### Abstract

Sex assessment is a crucial part of the biological profile in forensic and archaeological context, but it can be hardly performed in cases of commingled and charred human remains where DNA tests often are not applicable. With time literature have analyzed the sexual dimorphism of teeth (and especially canines), but very few articles take into consideration the teeth volume, although with time several technologies have been introduced in order to assess 3D volume (CT-scan, laser scanner, etc.).

This study aims at assessing the sexual dimorphism of dental and pulp chamber volumes of a sample of canines. Cone beam computed tomography analyses were performed by 87 patients (41 males and 46 females, aged between 15 and 83 years) for clinical purposes, and were acquired in order to measure canine volumes. Results show that the dental volume amounted to 0.745 cm3 (SD 0.126 cm3) in males, 0.551 cm3 (SD 0.130 cm3) with a statistically significant difference (p < 0.01). A diagnostic threshold of 0.619 cm3 was stated, which provides a percentage of correct answer of 80.5% in the chosen sample. The novel method was then applied with success to 7 archaeological: where in all the cases the results were concordant with those provided by the assessment of the cranium and pelvis.

The study adds a contribution to the wide analysis of dental sexual dimorphism confirming the statistically significant differences of volume between males and females and providing a method for the diagnosis of sex applicable to forensic cases.

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### 1.Introduction

Sex assessment has a relevant importance in the reconstruction of the biological profile of human remains, and literature provide several methods for reaching a correct diagnosis on the base of different bodily districts, mainly the pubis and cranium [1]. However, both in the forensic practice and archaeological context as well it is not uncommon to recover human remains where these structures are no longer appreciable, for example because of extreme fragmentation or carbonization of the skeleton. In addition, in these cases genetic tests often are not able to produce a reliable profile because of high DNA degradation [2]. From this point of view teeth may provide a relevant contribution, not only to personal identification [3], but also to the diagnosis of sex, thanks to their dimorphism and resistance to taphonomic effects. In literature, canines are known to show the highest degree of dimorphism [4], as also confirmed by several studies performed on different populations [5,6]: in human evolution, canine sexual dimorphism (CSD) has been already known, as it is critical for interpreting the developmental biology, socioecology and phylogenetic position of primate groups, since this phenomenon is absent to weak in extant prosimians [7]. From a forensic point of view, the dimorphism of canine may provide a help to diagnosis of sex, and several authors have analyzed and verified the statistically significant differences of linear metrical indications and ratios between males and females [4,8–10], although the last ones proved to be less reliable than single measurements [4,11,12]. Very few studies deal with the area of teeth, and found that the relative dentin-pulp area of canines and first premolars shows high levels of sexual dimorphism [13]. At the moment, only two studies have so far taken into consideration the sexual dimorphism of teeth volume: the first one was performed on mandibular central incisors and on a sample of 5–6 year oldchildren, where no statistically significant differences were found according to sex [14]. The second one by Tardivo et al. ascertained a sexual dimorphism of mandibular canine volumes and provided a formula for assessing age, given the volume of the tooth in mm3 [15]. However the authors do not provide a formula also for the upper canines. In the last years more and more technologies have been introduced, able to record 3D volume of items and biological material as well: some examples are cone beam CT-scan [16] and laser scanner [17], which have begun in the last years to being applied also to the forensic scenario, although in other fields of research than the diagnosis of sex. Therefore the chances to improve new methods applicable to the forensic context and based on the analysis of teeth volume is worth being taken into consideration. This study aims at confirming the existing data concerning sexual dimorphism of upper canine volumes, analyzing also other indices such as the pulp chamber volume and the ratio between pulp chamber and dental volume, in order to improve a novel method for the diagnosis of sex.

## 2. Materials and methods

A sample of 87 patients (41 males and 46 females, aged between 15 and 83 years) underwent to CBCT (cone beam computerized tomography) for clinical reasons not dependant upon the present study, using a i-Cat Next Generation (Imaging Sciences International, Hatfield, Pa) with the same settings for each subject (voxel size: 0.4 mm; scan time: 8.9 s; scan width: 23.2 cm; scan height: 17 cm). Exclusion criteria were the presence of oral pathologies, facial and oral deformities and systemic diseases. Once the analyses were performed, the images were elaborated by opensource freeware software Osirix in order to estimate the volume of the upper right canine and its pulp chamber. The volumes were evaluated by manually defining the perimeter of each tooth and corresponding pulp chamber on horizontal plane by creating a ROI (region of interest) including the delimited areas every 3–4 slices from the root apex up to the crown. The missing ROI were then created by the function "generate missing ROIs", included in the software. The volume delimited by each group of ROIs was then calculated by the function "ROI volume – compute volume". The volumes of upper right canines were evaluated in cm3, together with the corresponding pulp chamber and ratio between the pulp chamber and total dental volume. In addition, in order to test the novel method 7 canines were sampled from archaeological skeletons dating back to the Roman period and Middle Age recovered from six sites from Lombardia, North Italy (Chiavenna, Lovero, Postalesio, Teglio, Verceia): all the teeth were indicated with the number of tomb (T) and stratigraphic unit (SU). In all the seven cases sex was diagnosed by the assessment of morphological traits of the cranium and pelvis [1]. The canines belong to 2 males and 5 females. In order to acquire the volume of each canine, a 3D scanner (Dental Wings\_) was used: teeth were fixed by plasticine to the basal surface of the scanner and software DWOS\_ elaborated a 3D model which was then exported in a specific software for the elaboration of threedimensional images (VAM\_) for the estimation of the volume parameters (Fig. 1). Sex was then determined by applying the threshold elaborated by the statistical analysis of the 87 subjects.

### 3. Results

Data concerning dental volume and pulp chamber volume are shown in Table 1 and Fig. 2. The mean dental volume amounted to 0.745 cm3 (SD 0.126 cm3) in males, 0.551 cm3 (SD 0.130 cm3) in females; the mean pulp chamber volume was 0.029 cm3 (SD 0.011 cm3) in males, 0.022 cm3 (SD 0.012 cm3) in females. The

mean percentage ratio between pulp chamber and tooth was the same in males and females (3.9%, with a standard deviation of 1.3% in males, 1.7% in females). Among all the measurements, only the dental volume shows a significant difference between males and females (p < 0.01). Therefore, since only the external volume seems to be related with sex, less expensive methods than CBCT can be used, such as a volumeter. In detail, for what concerns the dental volume, a threshold value is estimable in 0.619 cm3: considering individuals with a higher dental volume as males and 73.9% in females (80.5% in mean). For what concerns the test on archaeological samples, the application of 0.619 cm3 threshold gave a result concordant with the results provided by the morphological analysis of cranium and pelvis in all the cases (Table 2).

#### 4. Discussion

Sexual dimorphism of teeth is a well known phenomenon, widely analyzed also from the genetic point of view. Literature report that Y chromosome influences dental growth by promoting both amelogenesis and dentinogenesis by enhancing mitotic activity in tooth germs with a consequent higher dentine thickness in males [18,19]. Amelogenin genes, which is present on both X and Y chromosomes, is implicated in tooth size dimorphism [20], whereas modifications of sex hormones have a more limitedimportance [21]. In addition, literature reports that the dimorphism of the mesiodistal measurements are due to dentin deposition, more pronounced in males than in females, whereas the enamel thickness does not show relevant differences among sexes [22]. This observation is concordant with the existing data concerning the different effects of sexual chromosomes on dental growth: the Y chromosome seems in fact to be active in both dentin and enamel deposition and promoves the mitosis within the dental lamina, whereas the X chromosome is able to influence only the enamel growth [18]. This may explain the differences in size (mainly linear measurements) recorded by literature. From this point of view, the observation of a statistically significant difference also for what concerns dental volume, as observed in the present article, is concordant with the genetic explanations of sexual dimorphism. In detail, the dental volume showed a statistically significant difference between males and females, whereas the same result was not observed for the pulp chamber volume and ratio between pulp chamber and dental volume. The lack of sexual dimorphism pulp chamber measurements and indices is probably due to the influence of age on the pulp size, verified by several authors who highlight also the importance of this phenomenon for age estimation of adults [23–26]. The lack of sexual dimorphism of the pulp chamber can be easily explained by the coexistent modification due to age. The present study provides a contribution to the topic of sex assessment by the adoption of a 0.619 cm3 canine volume threshold, which may give a help to the diagnosis of sex, especially in cases of commingled remains where cranium and pelvis are not available. In addition, the present results highlight the improvement of sex assessment which may derive from the application of modern radiological techniques such as CT-scan, confirmed by the existing literature [27–29]. However, although the best method for assessing canine volume is based on the use of advanced technologies such as laser scanner and cone beam CT scan, the assessment of sex may also be performed through cheaper and more available tools, for example a test tube filled by water and calibrated according to the specific volume threshold. The study of dimorphism of canines may give a contribution also to other fields of application of forensic anthropology, for example enhancing the studies concerning differences of facial soft tissues in males and females, especially for what concerns the oral portion, both from a metrical and morphological point of view [30,31]. Finally the observation of a strong sexual dimorphism incites to extend the analysis in order to verify the same phenomenon also in other types of teeth. In this way, further studies are needed in order to verify if adding similar information from other dental elements may increase the reliability of the method.

#### References

[1] Ubelaker DH. Human skeletal remains: excavation, analysis, interpretation. Washington (DC): Taraxacum; 1999.

[2] Fondevila M, Phillips C, Naveran N, Fernandez L, Cerezo M, Salas A, et al. Case report: identification of skeletal remains using short-amplicon marker analysis of severely degraded DNA extracted from a decomposed and charred femur.Forensic Sci Int Genet 2008;2(3):212–8.

[3] De Angelis D, Cattaneo C, Grandi M. Dental superimposition: a pilot study forstandardizing the method. Int J Legal Med 2007;121(6):501–6.

[4] Ashith BA, Mainali S. Limitations of the mandibular canine index in sexassessment. J Forensic Legal Med 2009;16:67–9.

[5] Yadav S, Nagabhushana D, Rao BB, Mamatha GP. Mandibular canine index inestablishing sex identity. Indian J Dent Res 2002;13(3–4):143–6.

[6] Zorba E, Moraitis K, Manolis SK. Sexual dimorphism in permanent teeth ofmoderns Greeks. Forensic Sci Int 2011;210:74–81.

[7] Schwartz GT, Miller ER, Gunnell GF. Developmental processes and caninedimorphism in primate evolution. J Hum Evol 2005;48:97–103.

[8] Pettenati-Soubayroux I, Signoli M, Dutour O. Sexual dimorphism in teeth:discriminatory effectiveness of permanent lower canine size observed in a XVIIIth century osteological series. Forensic Sci Int 2002;126:227–32.

[9] Punnya VA, Hemani S, Sudeendra P, Ashith BA. Analyses of odontometric sexual dimorphism and sex assessment accuracy on a large sample. J Forensic Legal Med 2013;20:673–7.

[10] Rao NG, Rao NN, Pai ML, Kotian MS. Mandibular canine index: a clue for establishing sex identity. Forensic Sci Int 1989;42:249–54.

[11] Hosmani JV, Nayak RS, Kotrashetti VS, Pradeep S, Babji D. Reliability of mandibular canine as indicators for sexual dichotomy. J Int Oral Health 2013;5(1):1–7.

[12] Acharya AB, Mainali S. Are dental indexes useful in sex assessment? J Forensic

Odontostomatol 2008;27(2):53-9.

[13] Saunders SR, Chan AHW, Kahlon B, Kluge HF, Fitzgerald CM. Sexual dimorphism of the dental tissues in human permanent mandibular canines and third premolars. Am J Phys Anthropol 2007;133:735–40.

[14] Ma JL, Shi SZ, Ide Y, Saka H, Matsunaga S, Agematsu H. Volume measurement of crowns in mandibular primary central incisors by micro-computed tomography. Acta Odontol Scand 2013;71(5):1032–7.

[15] Tardivo D, Sastre J, Ruquet M, Thollon L, Adalian P, Leonetti G, et al. Threedimensional modeling of the various volumes of canines to determine age and sex: a preliminary study. J Forensic Sci 2011;56(3):766–70.

[16] Cappella A, Amadasi A, Gaudio D, Gibelli D, Borgonovo S, Di Giancamillo M, et al. The application of cone-beam CT in the aging of bone calluses: a new perspective? Int J Legal Med 2013;127(6):1139–44.

[17] Sansoni G, Cattaneo C, Trebeschi M, Gibelli D, Porta D, Picozzi M. Feasibility of contactless 3D optical measurement for the analysis of bone and soft tissue lesions: new technologies and perspectives in forensic sciences. J Forensic Sci 2009;54(3):540–5.

[18] Alvesalo L. Sex chromosomes and human growth: a dental approach. Hum Genet 1997;101:1–5.

[19] Alvesalo L, Tammisalo E, Townsend G. Upper central incisor and canine tooth crown size in 47, XXY males. J Dent Res 1991;70:1057–60.

[20] Lau EC, Mohandras TK, Shapiro LJ, Slavkin HC, snead ML. Human and mouse amelogenin gene loci are non the sex chromosomes. Genomics 1989;4:162–8.

[21] Guastelli-Steinberg D, Sciulli PW, Betsinger TK. Dental crown size and sex hormone concentrations: another look at the development of sexual dimorphism. Am J Phys Anthropol 2008;137:324–33.

[22] Stroud JL, Buschang PH, Goaz PW. Sexual dimorphism in mesiodistal dentin and enamel thickness. Dentomaxillofac Radiol 1994;23(3):169–71.

[23] Cameriere R, Ferrante L, Cingolani M. Variations in pulp/tooth area ratio as an indicator of age: a preliminary study. J Forensic Sci 2004;49(2):317–9.

[24] Cameriere R, Ferrante L, Belcastro MG, Bonfiglioli B, Rastrelli E, Cingolani M. Age estimation by pulp/tooth ratio in canines by mesial and vestibular periapical X-rays. J Forensic Sci 2007;52(5):1151–5.

[25] Cattaneo C, De Angelis D, Ruspa M, Gibelli D, Cameriere R, Grandi M. How old am I? Age estimation in living adults: a case report. J Forensic Odontostomatol 2008;26(2):39–43.

[26] Kvaal S, Solheim T. A non-destructive dental method for age estimation. J Forensic Odontostomatol 1994;12(1):6–11.

[27] Ciaffi R, Gibelli D, Cattaneo C. Forensic radiology and personal identification of unidentified bodies: a review. Radiol Med 2011;116:960–8.

[28] Zech WD, Hatch G, Siegenthaler L, Thali MJ, Lösch S. Sex determination from os sacrum by postmortem CT. Forensic Sci Int 2012;221(1–3):39–43.

[29] Bilfeld MF, Dedouit F, Rousseau H, Sans N, Braga J, Rougè D, et al. Human coxalmm bone sexual dimorphism and multislice computed tomography: geometric morphometric analysis of 65 adults. J Forensic Sci 2012;57(3):578–88.

[30] Ritz-Timme S, Gabriel P, Tutkuviene J, Poppa P, Obertovà Z, Gibelli D, et al. Metric and morphological assessment of facial features: a study on three European populations. Forensic Sci Int 2011;207(1–3):239.e1–8.

[31] Ritz-Timme S, Gabriel P, Obertovà Z, Boguslawski M, Mayer F, Drabik A, et al. A new atlas for the evaluation of facial features: advantages, limits, and applicability. Int J Legal Med 2011;125(2):301–6.

## Table 1

Statistical data concerning dental volume and pulp chamber volume in males and females.

	Males	Females
Mean dental volume (SD)	0.745 cm <sup>3</sup>	0.551 cm <sup>3</sup>
	(0.126 cm <sup>3</sup> )*	(0.130 cm <sup>3</sup> )*
Minimum dental volume	0.424 cm <sup>3</sup>	0.308 cm <sup>3</sup>
Maximum dental volume	1.064 cm <sup>3</sup>	0.919 cm <sup>3</sup>
Mean pulp chamber volume (SD)	0.029 cm <sup>3</sup>	0.022 cm <sup>3</sup>
	(0.011 cm <sup>3</sup> )**	(0.012 cm <sup>3</sup> )**
Minimum pulp chamber volume	0.011 cm <sup>3</sup>	0.006 cm <sup>3</sup>
Maximum pulp chamber volume	0.061 cm <sup>3</sup>	0.052 cm <sup>3</sup>
Mean percentage ratio between pulp chamber and tooth (SD)	3.9% (1.3%)	3.9% (1.7%)

SD: standard deviation.

\* Statistically significant difference between males and females (*p* < 0.01).

\*\* Not statistically difference between males and females (p > 0.01).

## Table 2

Results of diagnosis of sex by morphological analysis of cranium pelvis and canine volume applying 0.619 cm<sup>3</sup> threshold.

	Cranium	Pelvis	Canine volume
Chiavenna T3-US43	М	М	М
Lovero, Church of S. Maria Maddalena T1- US147	F	F	F
Lovero, Church of S. Maria Maddalena T1- US147	Μ	М	М
Postalesio, Church of S. Colombano T24	F	F	F
Teglio, Church of S. Pietro US14	F	F	F
Teglio, Church of S. Pietro US28	F	F	F
Verceia, Church of S. Fedele T3	F	F	F

