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LANDMARK BASED SEX DISCRIMINATION ON THE CRANIA OF ARCHAEOLOGICAL GREEK POPULATIONS. A COMPARATIVE STUDY BASED ON THE CRANIAL SEXUAL DIMORPHISM OF A MODERN GREEK POPULATION

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ABSTRACT

The estimation of sex is a fundamental step for physical anthropologists. The present study confirms the presence of sexual dimorphism in cranial traits of a modern Greek population and produces sex predicting logistic regression equations, which are subsequently applied on the crania of archaeological Greek populations. This study uses 24 landmarks and 25 traits based on distance and angle measurements. Equations, were formulated for the cranium in whole, as well as, isolated cranial regions and single traits. The application of these equations on the archaeological Greek populations yielded an accuracy of classification over 70% in the sphenoid region, the maxilla and the cranium in whole and for three single traits. Hence, the results suggest that our equations based on the modern Greek population can successfully be used in sex prediction of archaeological Greek populations.

KEYWORDS: sex determination; 3DGM; logistic regression sex-predicting equations; cranium; Greek populations

1. INTRODUCTION

The utility of sex determination of human skeletal remains is of at most importance, since the skeletal study of archaeological material is the only detailed source of demographical information on ancient populations. A simple example is the demographic analysis of a prehistoric cemetery. Sex discrimination of unknown human remains is an evaluation intense task since sexual dimorphism in humans is not as marked as in other mammals (Hoyme and Iscan, 1989; Schwartz, 1995). Today many unidentified human remains are sexed through DNA analysis (Stone *et al.*, 1996; Tierney and Bird, 2015). However, a number of situations remain in which invasive analysis is not preferred. For instance, in cases of ancient human remains the element may be too degenerated to yield amplifiable DNA.

It is well established from anthropological and anatomical studies (Dwight, 1905; Letterman, 1941; Phenice, 1969; Krogman and Iscan, 1986; France, 1998) that the pelvis is the most reliable indicator of sexual dimorphism followed by skull and humerus. The cranium is of great importance and well-studied, since it is most often retrieved at excavation sites either isolated or along with other skeletal material of less forensic value (Klepinger, 2006).

Standard approach for studying sexual dimorphism include morphological and morphometric methods. Morphometric methods are a trademark for physical anthropology (Olivier, 1969; Buikstra and Ubelaker, 1994; White et al., 2012). Due to the limitations of traditional measurements of Euclidean distances between landmarks, usually measured with traditional tools such as calipers and craniophores, landmark coordinate measurements are used more frequently in the last few decades (Bookstein, 1998; Adams et al., 2004; Slice, 2007). Coordinate data are often collected with instruments such as 3D digitizers and laser scanners. According to Sholts et al. (2010), landmark coordinate measurements from 3D models produced by a desktop laser scanner yield an overall mean SD value of 1.05 mm, whereas landmark coordinate measurements from 3D models produced by a 3D digitizer yield an overall mean SD value of 0.79 mm; precisions sufficient for most craniometric research.

The aim of this study is to demonstrate the presence of sexual dimorphism in cranial traits of a modern Greek population and produce equations for predicting sex using landmark coordinate measurements. Moreover, it tests the application of these equations in crania of archaeological Greek populations. The development of population specific standards relative to sex determination is considered necessary, since a large number of studies have demonstrated that although population genetic composition is the most important contributing factor to sexual dimorphism (Safont et al., 2000; Gustafsson et al., 2007), environmental factors, especially diet (Stinson, 1985; Cowgill, 2007) and mechanical load (Safont et al., 2000; Carlson et al., 2007) affect its expression.

2. MATERIALS AND METHODS

For the purpose of this research 176 crania (94 males, 82 females) of adult individuals from the Athens collection were studied. Regarding the archaeological sample, 7 adult skeletons (3 males, 4 females) from the cemetery Almyros in Corfu, 3 adult individuals (2 females, 1 male) from the Agia Triada Theve collection, 8 adults (4 males, 2 females) from the Edessa collection and 24 crania (9 males, 15 females) of adult individuals from the Ancient Corinth collection were examined. In all cases, individuals without extensive fragmentation of the cranium were selected.

The modern skeletal reference collection (known as the Athens Collection) consists of 225 skeletons. Information on the name and age at death of each individual in the collection is derived from death records (Eliopoulos et al., 2007). The Almyros cemetery in Corfu was dated between the late Archaic Period (7th century B.C.) and the 2nd century A.D (Preka-Alexandri, 1988) and consists of 32 skeletons. The Agia Triada Theve collection consists of 38 skeletons and dates from the 13th to 15th century AD, while the Edessa collection, which dates back to the 3rd century AD consist of 33 skeletons. Finally, the Ancient Corinth collection dates back to the Classical period and consist of 48 skeletons. All studied samples are housed in the Department of Animal and Human Physiology of the Biology Faculty at the University of Athens. Sex determination of the archaeological skeletons was conducted using the morphological criteria of the pelvis (Buikstra and Ubelaker, 1994).

This study uses 24 landmarks on the outer surface of the skull and 25 traits (Table 1; All distances shown in mm), chosen on the basis to adequately illustrate the anatomy of the cranium, but also based on the capacity to obtain the landmarks' coordinates from the 3D models produced with the laser scanner. The landmarks used in the present study are either type I, type II or type III (Bookstein, 1991). The small number of type I landmarks is due to the inherent difficulty to accurately pick point them on the 3D laser scanner models (Sholts et al., 2010). Since the archaeological samples are often fragmented and one of the purposes of the present study is to produce sex-predicting equations, which could also be used in archaeological samples, along with the cranium as a whole, the neurocranium, as well as the frontal, occipital, temporal, sphenoid and maxilla regions were studied separately.

Table 1. List of landmarks and traits.

Landmarks	**Trait**	
	A1	Angle: Maxillonasofrontale Left-Nasion-Maxillonasofrontale Right
Asterion (Left-Right)	A2	Angle: Nasion-Glabella-Bregma
Basion	A3	Angle: Lambda-Opisthocranion-Opisthion
Bregma	A4	Angle: Foraminolaterale Left-Basion-Foraminolaterale Right
Crotaphion (Left-Right)	A5	Angle: Asterion-Entomion-Supramastoid crest
Entomion (Left-Right)	A6	Angle: Frontomalare temporale-Zygomaxillare-Zygotemporale inferior
Foraminolaterale (Left-Righ	D1	Distance: Nasion-Opisthocranion
Frontomalare orbitale (Left-Right)	D2	Distance: Bregma-Basion
Frontomalare temporale (Left-Right)	D3	Distance: Sphenion Left-Right
Glabella	D4	Distance: Frontomalare orbitale Left-Right
Infratemporale (Left-Right)	D5	Distance: Basion-Opisthion
Lambda	D6	Distance: Foraminolaterale Left-Right
Landmark x (Left-Right)	D7	Distance: Maxillonasofrontale (Left-Right)
Mastoidale (Left-Right)	D8	Distance: Staurion-Maxillonasofrontale
Maxillonasofrontale (Left-Right)	D9	Distance: Zygomaxillare Left-Right
Nasion	D10	Distance: Entomion-Mastoidale
Opisthion	D11	Distance: Entomion-Asterion
Opisthocranion	D12	Distance: Crotaphion-Infratemporale
Porion (Left-Right)	D13	Distance: Zygotemporale superior Left-Right
Sphenion (Left-Right)	D14	Distance: Crotaphion-Landmark x
Staurion	D15	Distance: Frontomalare temporale-Frontomalare orbitale
Supramastoid crest - Squamous suture intersection (Left-Right)	R1	Ratio: (Nasion-Opisthocranion)/(Bregma-Basion)
Zygomaxillare (Left-Right)	R2	Ratio: (Staurion-Maxillonasofrontale)/(Zygomaxillare Left-Right)
Zygotemporale inferior (Left-Right)	R3	Ratio: (Entomion-Mastoidale)/(Entomion-Asterion)
Zygotemporale superior (Left-Right)	R4	Ratio: (Crotaphion-Landmark x)/(Crotaphion-Infratemporale)

Regarding the Athens Collection, the coordinates of the landmarks were recorded during a previous research (Chovalopoulou et al., 2013, 2016a,b) using a Microscribe G2X (Immersion Corporation, San Jose, CA, USA) portable digitizer. From the archaeological samples, the data collection was done with a Next-Engine™ HD Desktop 3D scanner and ScanStudio™ HD PRO software (NextEngine™ 2008). After data collection, Meshlab software (Meshlab, Visual Computing Lab – ISTI – CNR, http://meshlab.sourceforge.net/) was used to extract landmarks coordinates.

For calculating the sex-prediction equations, the statistical analysis was performed on the 25 traits of the Athens collection sample. The Shapiro-Wilk test was used in order to assess the normality of the data. The Wilcoxon signed-rank test compared the traits of the right and left sides to ascertain whether any bilateral asymmetry exists. The Mann-Whitney test compared the data between male and female subjects, aiming to identify any statistically significant differences in their mean values. Logistic regression analysis of the sexually dimorphic traits was performed in order to derive the sex prediction equations for males of the general form $z = const. + \sum_{i=1}^{n}(Trait_i \times Coef_i)$, where n is the number of traits. Finally, we applied these equations on the available traits of the archaeological samples and we evaluated their accuracy of correct classification for males, where z>0. All aforementioned statistical analyses were performed using SPSS software (PASW Statistics 23.0) and Octave software (Eaton et al., 2015).

3. RESULTS

Some of the traits used in this research failed the normality test (Table 2). More specifically, for females, the scores of D6, D9, D11 (right side), D13, D15 (both sides), R2 (both sides) and R3 (both sides) traits were significantly different from a normal distribution (p value <0.05).

Table 2. Athens Collections' p-values of the Shapiro–Wilk normality test.

Trait	*Female*		*Male*		*Trait*	*Female*		*Male*	
	N	*p value*	*N*	*p value*		*N*	*p value*	*N*	*p value*
A1	81	.151	94	.037	D10 (Left)	81	.399	92	.407
A2	82	.062	94	.910	D10 (Right)	81	.248	92	.542
A3	81	.442	94	.269	D11 (Left)	81	.179	93	.498
A4	81	.052	94	.082	D11 (Right)	82	.005	93	.269
A5 (Left)	81	.809	93	.059	D12 (Left)	82	.379	92	.130
A5 (Right)	82	.352	93	.036	D12 (Right)	82	.319	91	.847
A6 (Left)	81	.064	92	.048	D13	81	.006	91	.023
A6 (Right)	81	.200	92	.056	D14 (Left)	81	.522	91	.106
D1	82	0.5	94	0.386	D14 (Right)	81	.184	90	.144
D2	81	.793	94	.029	D15 (Left)	81	.000	93	.001
D3	82	.287	91	.074	D15 (Right)	81	.000	93	.000
D4	81	.143	92	.088	R1	81	.329	94	.727
D5	81	.258	94	.084	R2 (Left)	81	.002	91	.124
D6	81	.007	94	.114	R2 (Right)	81	.028	91	.534
D7	81	.072	94	.030	R3 (Left)	81	.000	92	.000
D8 (Left)	81	.084	94	.328	R3 (Right)	81	.000	92	.000
D8 (Right)	81	.251	94	.206	R4 (Left)	81	.601	91	.795
D9	81	.037	91	.078	R4 (Right)	81	.384	90	.766

For males, the corresponding traits are A1, A6 (left side), D2, D7, D13, D15 (both sides) and R3 (both sides). Consequently, non-parametric tests were used in the analysis.

Table 3. Athens Collection's p-values of Wilcoxon signed-rank test for laterality.

Trait	*p value*	*Trait*	*p value*
A5	0.790	D14	0.438
A6	0.484	D15	0.186
D8	0.161	R2	0.250
D10	0.592	R3	0.866
D11	0.816	R4	0.813
D12	0.064		

Table 3 presents the two-tail p value from the Wilcoxon signed-rank test. According to the laterality test results, there are no statistically significant differences in any trait between left and right side. Therefore, the mean values of traits were calculated and used in the subsequent analysis.

Table 4 shows the two-tail p value from the Mann-Whitney test and the demarking points of the sexually dimorphic traits for both sexes. The results demonstrate that not all traits exhibit statistically significant sexual dimorphism. More specifically, most of the angles (A1, A4, A5, A6), the distances D7, D11 and D15 as well as the ratios R3 and R4, are not sexually dimorphic. These traits were not included in the sex prediction equations. Additionally, according to the demarking points, in all traits, apart from the angle formed by landmarks nasion, glabella and bregma (A2) and the R1 trait, males have higher scores than females.

Equations for predicting male individuals are given in Tables 5 and 6. Table 5 presents the equations for each trait separately, while table 6 shows the equations for the cranium as a whole (all traits), for the regions of the cranium as well as the joint traits equation (traits with an accuracy of classification > 70%). Finally, Table 7 presents the classification accuracies of Athens collection and archaeological samples respectively. According to the results regarding the Athens collection, the traits A3, D14, R1 and R2 yield classification accuracies < 60%, while the traits D2, D4, D8 and D13 yield classification accuracies > 70%. As expected, the equation based on all traits (cranium as a whole) yields the highest average classification accuracy, while the equation based on the occipital traits the lowest. Regarding the archaeological samples, the zygotemporale superior left to right distance (D13) yields the highest correct classification rate (75%), while trait D5 yields the lowest (30%).

Table 4. Athens Collection's p-values of Mann–Whitney test for evaluation of sexual dimorphism and demarking points for sexually dimorphic traits.

Sexually dimorphic traits							**Traits that lack sexual dimorphism**			
Trait	**N**		**Demarking point**			**p Value**	**Trait**	**N**		**p Value**
A2	Male	94	Male	<	119.97	0.000	A1	Male	94	0.488
	Female	82	Female	>	123.49			Female	81	
A3	Male	94	Male	>	126.34	0.018	A4	Male	94	0.370
	Female	81	Female	<	124.56			Female	81	

D1	Male	94	Male	>	173.58	0.000	A5	Male	93	0.259
	Female	82	Female	<	171.87			Female	82	
D2	Male	94	Male	>	136.79	0.000	A6	Male	93	0.791
	Female	81	Female	<	133.31			Female	81	
D3	Male	91	Male	>	109.21	0.000	D7	Male	94	0.057
	Female	82	Female	<	107.37			Female	81	
D4	Male	92	Male	>	97.74	0.000	D11	Male	93	0.067
	Female	81	Female	<	94.84			Female	82	
D5	Male	94	Male	>	35.96	0.000	D15	Male	94	0.098
	Female	81	Female	<	35.69			Female	81	
D6	Male	94	Male	>	31.85	0.000	R3	Male	93	0.143
	Female	81	Female	<	31.36			Female	82	
D8	Male	94	Male	>	58.27	0.000	R4	Male	92	0.601
	Female	81	Female	<	57.01			Female	81	
D9	Male	91	Male	>	90.17	0.000				
	Female	81	Female	<	89.93					
D10	Male	93	Male	>	42.68	0.000				
	Female	82	Female	<	41.22					
D12	Male	92	Male	>	42.21	0.000				
	Female	82	Female	<	42.15					
Sexually dimorphic traits										
Trait	**N**		**Demarking point**			**p Value**				
D13	Male	91	Male	>	118.02	0.000				
	Female	81	Female	<	114.34					
D14	Male	92	Male	>	36.18	0.009				
	Female	81	Female	<	35.14					
R1	Male	94	Male	<	1.27	0.012				
	Female	81	Female	>	1.29					
R2	Male	91	Male	>	0.65	0.043				
	Female	81	Female	<	0.63					

The regional traits of the occipital yield the lowest correct classification rate on the archaeological sample, which is consistent with our findings from the athens collection. on the other hand, the joint traits yield the highest correct classification rate, which outperforms its equivalent from the athens collection.

4. DISCUSSION

Former studies related to skull asymmetry proved that it is a characteristic of the human skull (Woo, 1931; Hershkovitz et al., 1992; Seiji et al., 2009). For the particular traits studied in the current research no bilateral asymmetry was found.

However, this does not mean that asymmetry is entirely absent from the crania of the particular individuals.

As observed by the higher values in most distance-related traits of the male group, male individuals are more robust than females. An observation backed by numerous researches on different populations (Kranioti et al., 2008; Zavando et al., 2009). The greater angle, formed by landmarks nasion-glabella-bregma, in the female group (Fig. 1) supports, as expected, that male individuals have a more pronounced supraorbital ridge (White et al., 2012).

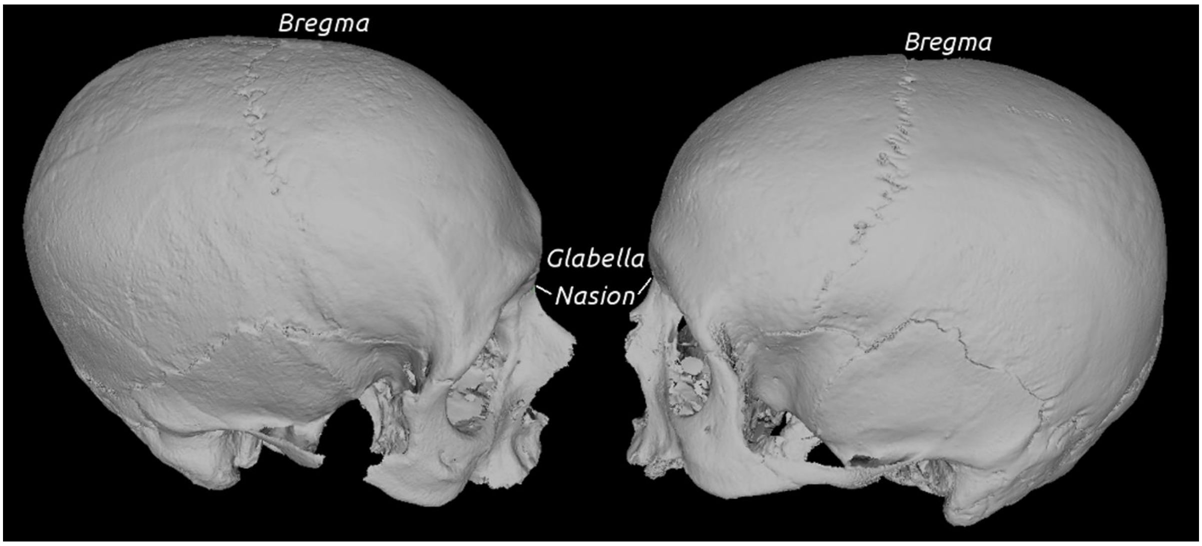

Figure 1. Angle formed by landmarks nasion-glabella-bregma: In the female group (right side) is greater.

Table 5. Logistic regression results and male predicting equations for single traits based on the Athens collection.

Trait	Model x^2	(p)	Nagelkerke R^2	Constant	Coef
A2	49.143	0	0.325	20.163	-0.165
A3	5.078	0.024	0.038	-7.031	0.057
D1	32.601	0	0.226	-24.794	0.144
D2	64.959	0	0.414	-36.92	0.275
D3	36.606	0	0.255	-18.889	0.175
D4	67.006	0	0.429	-33.851	0.353
D5	23.548	0	0.168	-11.173	0.316
D6	28.353	0	0.2	-10.567	0.339
D8	41.809	0	0.284	-19.322	0.338
D9	20.132	0	0.147	-13.516	0.152
D10	35.172	0	0.243	-8.209	0.198
D12	17.965	0	0.131	-6.218	0.15
D13	65.464	0	0.423	-30.588	0.264
D14	7.553	0.006	0.057	-3.142	0.092
R1	6.295	0.012	0.047	8.762	-6.716
R2	4.185	0.041	0.032	-4.76	7.671

According to our results, regarding the R1 trait, in the female skull the length of the base exceeds the sagittal part to a greater extend than in the male. In Germans there is no difference (Dekaban, 1977), while Bartels in 1897 found that in the female skull the sagittal part exceeds the length of the base to a greater extend than in the male. According to Lang (1983), the disparity varies in different populations.

According to our results shown in Table 4, the foramen magnum exhibits size related sexual dimorphism, since the traits D5 and D6 are significantly larger in males rather than females. However, shape related trait A4 yields no sexual dimorphism. These findings are consistent with Kumar et al. (2015), whose studied sample included random collection of 36 adult human dry human skulls (19 males and 17 females) and was carried out in the Human structure and Neurobiology department of Oman Medical College. Analogous results were also reported by Loyal et al., (2013), who studied 202 crania from the Osteology Department of the National Museums of Kenya. The nasal region illustrated with traits A1 and D7 showed no sexual dimorphism, a result in agreement with the study of Schlager and Rudell (2015) on German and Chinese samples, which consisted of 267 individuals each. Concerning the temporal bone, although sexual dimorphism is present in the mastoid process, the squamous part, which articulates with the parietal bone, displayed no sexual dimorphism based on the traits A5 and D11.

Table 6. Logistic regression results and male predicting equations for regions based on the Athens collection.

Region	**Traits**	**(p)**	**Nagelkerke R^2**	**Model x^2**	**constant**	**coef**	**coef**	**coef**
Neurocranium	D1+D2+R1	0	0.455	72.866	-17.683	0.256	0.022	-22.945
Frontal	D3+D4+A2	0	0.539	96.041	-17.841	0.101	0.276	-0.16
Occipital	D5+D6+A3	0	0.257	37.344	-20.068	0.175	0.257	0.046
Maxilla	D8+D9+R2	0	0.326	48.107	-34.519	0.141	0.196	13.853
Temporal	D10+D12+D13	0	0.526	85.199	-39.475	0.164	0.091	0.248
Sphenoid	D3+D12+D14	0	0.287	41.586	-20.377	0.154	0.084	0.009
Joined Traits *	**Model x^2**	**(p)**	**Nagelkerke R^2**	**constant**	**coef**	**coef**	**coef**	**coef**
D2+D4+D8+D13	101.599	0	0.598	-58.538	0.182	0.119	0.14	0.126
Traits	**Model x^2**	**(p)**	**Nagelkerke R^2**	**constant**	**coef D1**	**coef D2**	**coef R1**	**coef D3**
All traits	135.053	0	0.737	-85.213	-0.211	0.428	30.874	-0.004
			coef D4	**coef A3**	**coef D5**	**coef D6**	**coef A4**	**coef D8**
			0.065	-0.193	0.22	0.132	0.095	0.422
			coef D10	**coef D12**	**coef D14**	**coef D9**	**coef R2**	**coef D13**
			0.068	0.082	0.014	-0.182	-28.551	0.189

* Traits with accuracy of classification > 70%

As presented in Table 7, the single traits considered adequately reliable (>70%) for sex determination are D4, D13, D2 and D8. Saini et al. (2011), while studying the biorbital breadth (the straight distance between two ectoconchion) on 112 Indian adult individuals, found an average classification accuracy of 67.9%. On the contrary, in our study, the frontomalare orbitale left to right distance, similar to the biorbital breadth, yields higher correct classification rate (79%). Regarding the correct classification rate of zygotemporale superior left to right distance, our results (76%) are similar to these of Oliveira et al. (2012). Oliveira et al. (2012), examined the presence of sexual dimorphism in 100 Brazilian adult skulls and according to their results, the variable zygion left to right distance, similar to the distance we examined, yielded an average classification accuracy of 75%. Kamath et al. (2015) and Gapert et al. (2008), while determining the sexing potential of the foramen magnum, found that its sagittal diameter yields an average classification accuracy of 69.6% and 60.1% respectively. Regarding the transverse diameter of the foramen magnum, the corresponding results were 66.4% and 65.8%. Kamath et al. (2015), studied 72 skulls (41 male and 31 female) from a South Indian population, while Gapert et al. (2008), examined 158 individuals (82 male and 76 female) from the St. Bride's documented skeletal collection. The results from our study are equivalent. Except the occipital region, all other regions examined in this study are reliable for sex determination. Steyen and Iscan in 1998, took 12 standard cranial and five mandibular measurements from 44 male and 47 female skeletons from the Pretoria and Dart collections and established population specific standards for sex determination. According to their findings, the cranium, based on six measurements, yielded an average classification accuracy of 85.7%, which is almost as good as our results for the whole cranium. Finally, the average classification accuracy of 74% for the neurocranium is similar to the results of Franklin et al. (2005). Franklin et al. (2005), studied 332 (182 male and 150 female) adult crania from the R.A. Dart Collection in order to produce a practical discriminant function for determining the sex of a South African population. According to their results, the vault, based on the variables maximum cranial length, maximum cranial breadth and basi-bregmatic height, yielded a correct classification rate of 75%.

Table 7. Classification accuracies of Athens collection and archaeological samples.

	Results for single traits							
	A2	**A3**	**D1**	**D2**	**D3**	**D4**	**D5**	**D6**
Accuracy of classification *	0.49 (41)	0.41 (37)	0.63 (41)	0.62 (40)	0.73 (33)	0.70 (27)	0.30 (37)	0.58 (36)
Accuracy of classification **	0.69 (176)	0.6 (175)	0.65 (176)	0.74 (175)	0.68 (173)	0.79 (173)	0.63 (175)	0.67 (175)
	D8	**D9**	**D10**	**D12**	**D13**	**D14**	**R1**	**R2**
Accuracy of classification *	0.57 (35)	0.57 (28)	0.65 (40)	0.60 (42)	0.75 (16)	0.56 (25)	0.49 (39)	0.61 (28)
Accuracy of classification **	0.71 (175)	0.68 (172)	0.66 (175)	0.63 (174)	0.76 (172)	0.6 (173)	0.59 (175)	0.56 (172)
	Results for region traits						**Joined traits**	**All traits**
	Neuro-cranium	**Frontal**	**Occipital**	**Maxilla**	**Temporal**	**Sphenoid**		
Accuracy of classification *	0.69 (39)	0.61 (23)	0.51 (35)	0.75 (28)	0.67 (15)	0.71 (21)	0.81 (16)	0.7 (10)
Accuracy of classification **	0.74 (175)	0.81 (170)	0.64 (175)	0.73 (172)	0.78 (170)	0.72 (172)	0.83 (171)	0.88 (168)

* Archaeological samples

** Athens collection

Regarding the archaeological samples, the single traits, which are considered adequately reliable (>70%) for sex determination (also in agreement with the Athens Collection) are D4 and D13. Contrary to the Athens Collection, the sphenion left to right distance, yields higher average classification accuracy (73%). Additionally, the joint traits are the strongest discriminating morphometric variables, the regional traits of the maxilla, the sphenoid and the cranium as a whole are the next best. The rather large difference in the correct sex classification of the cranium as a whole between the modern and archaeological samples, is most likely due to the very small number of individuals of the archaeological sample.

5. CONCLUSION

This study has demostrated that sex predicting equations derived from a modern Greek population can successfully be used in archaeological samples. More specifically, a number of traits and regions (such as D13 or the maxilla) yield consistent correct classification between modern and archaeological populations. While proving that direct applicability can produce useful and accurate results on sex determination of ancient crania, further research on larger archaeological sample is needed to more accurately estimate the correct classification rates that our sex-predicting equations yield on the archaeological population.

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