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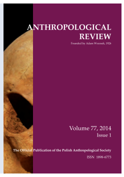

Sex determination from femora in late antique sample from Eastern Adriatic coast (Salona necropolis)

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Abstract: The determination of sex is one of the first steps in anthropological analysis. When skeletal remains are fragmented, the most useful approach is application of osteometric methods. The methods are population specific, and therefore require development of discriminant functions for each population group.

The aim of this study was to test sexual dimorphism of femoral measurements and to calculate discriminant functions applicable for sex determination on fragmented skeletal remains on the late antique sample from the Eastern Adriatic coast (2^{nd}–6^{th} century AD).

214 randomly chosen skeletons from the excavation site Solin-Smiljanovac were analyzed. Sex and age were assessed using standard anthropological methods, and skeletons were examined for pathologic and traumatic changes. In the next step, we selected 27 female and 48 male skeletons free of peri- or post-mortem changes that could affect measurements Eight standard femoral measurements were taken. Sexual dimorphism was initially compared using independent sample t test, after which discriminant functions were computed.

All femoral measurements showed statistically significant sexual dimorphism (p<0.001). Ten discriminant functions for every part of femur were calculated and obtained classification accuracy of 73.1–91.8%.

This study reached relatively high classification, which will improve further analysis of the skeletal remains from the Salona necropolis. Due to similar population structure in the Roman period across the Adriatic coast, the discriminant functions could be applicable for all populations from the same period and area. This study also raised a few methodological questions showing that when creating discriminant functions we should consider not only the accuracy, but also the applicability based on the experience from the anthropology laboratory that considers the state of preservation and frequent pathology.

Key words: discriminant function analysis, osteometric methods, Roman province, Croatia

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Introduction

The determination of sex is fundamental and one of the first steps for recreating the person's biological profile both in forensic and biological anthropology. This stems from the fact that other important elements of an adults biological profile, i.e. age and stature, cannot be properly obtained without data about sex. In forensic anthropology determination of the biological profile eliminates groups of individuals and facilitates identification and final disposition of remains. Furthermore, in biological anthropology it is a variable that enables the study of demography of ancient groups and reconstruction of their life by studying sex specific patterns of burial, violence distribution, mortality, pathology, diet and societal status (Gibbon 2009).

There are three methodological approaches for sex determination on skeletal remains: analysis of DNA, non-metric morphological traits, and metric analysis. The analysis of DNA is the gold standard and indisputably the most accurate method for determination of sex (Gibbon et al. 2009). However, the drawbacks of the method are its high monetary and time costs. Additionally, the results can be affected by insufficient collagen preservation and contamination. Traditionally, the approach that is mostly used is based on the analysis of the skeletal features with pronounced sexual dimorphism, mainly the pelvis and the skull. The method is based on descriptive distinctions of shape and bone configurations that are macroscopically detectable. Although this approach could be appropriate when the examiner is cognizant of population variability, it also relies heavily on the subjective interpretation of the examiner, and it is difficult to assess its accuracy. Moreover, a glaring flaw of this approach is that it cannot be carried out if the skeletal remains, primarily the pelvis and the skull, are not sufficiently preserved. The last method – osteometric analysis is based on various statistical approaches (such as discriminant function analysis, logistic regression, etc.) that classify unknown individuals into one of two categories (female or male) using single or multiple measurements of one or more bones (İşcan and Steyn 2013). The main advantages of osteometric analysis are the reduction of subjectivity and the availability of data on the accuracy of each method. According to new findings a single bone measurement can demonstrate reliability equal or even better than the non-metric morphologic analysis of the skull (Spradley and Jantz 2011). Furthermore, it can also be applicable on fragmented skeletal remains when the pelvis and the skull are not preserved, which is often the case in the analysis of ancient remains and forensic cases. However, osteometric methods are demonstrated to be population specific and therefore require development of standards for each population separately.

So far, nearly all the bones of the skeleton have been analyzed to calculate discriminant functions for numerous archaeological and modern populations. Most of the research has been conducted on long bones like femora, tibiae and humeri, as they are usually the best preserved bones in forensic cases and archaeological findings, and provide high classification accuracy (İşcan and Steyn 2013).

Since the femur is the strongest bone in the human body and is in most of the cases preserved, it is most studied human bone for osteometric sex determination. A great number of studies have been conducted using standard, as well as nonstandard measurements and very accurate classification rates, usually higher than 90%, were obtained (du Jardin et al.; Steyn and Iscan 1997; Šlaus 1997; Šlaus et al. 2003; Albanese et al. 2008; Spradley and Jantz 2011). According to most research, single measurements that performed best were diameter of head and epiphyseal breadth (Steyn and Iscan 1997; Šlaus 1997; Šlaus et al. 2003; Spradley and Jantz 2011). Apart from them, circumference measurements have also performed very well, which is very useful when dealing with fragmented skeletal remains (Black 1978; Safont et al. 2000; Wrobel et al. 2002; Nagaoka and Hirata 2009).

Only a few studies have been published using samples from the area of present-day Croatia. They comprise: medieval femora, tibiae and humeri (Šlaus 1997; Šlaus and Tomičić 2005; Bašić et al. 2013), medieval and contemporary mandible and teeth (Vodanović et al. 2006; Vodanović et al. 2007), and contemporary femora and tibiae (Šlaus et al. 2003; Šlaus et al. 2013). Until now, no such study has been conducted on late antique populations from this area.

Therefore, the aim of this study was to test sexual dimorphism on study sample, and create useful discriminant functions of different measurements, which can be successfully applied even if bones are fragmented.

Materials and methods

Settings

The study sample consisted of 214 randomly chosen skeletal remains from excavation site Solin-Smiljanovac dated to 2nd to 6th century from the sample of more than 1000 skeletons. Solin-Smiljanovac was part of eastern necropolis of antique Salona, the metropolis of the Roman province of Dalmatia. Salona was a coastal stronghold and port of Illyrian Delmats. In that time, in Salona, besides the Illyrians and inhabitants from Greece, a large number of Italics also lived there. After the civil war between Caesar and Pompei in 48 BC, Salona, who sided with Caesar, became a Roman colony with the full name of Colonia Martia Ivlia Salona and became the centre of the Roman province of Illyricum, later a province of Dalmatia. After the Illyrian rebellion was suppressed (during the Baton rebellion that lasted from 6th to 9th year AD), a period of peace and prosperity occurred, mostly seen through its urban development and strong building activity (Anteric et al. 2014).

Anthropological analysis

The skeletons were washed and bone preservation was evaluated.

In the next step the skeletal remains of subadults were excluded from the analysis. In the next step, for further analysis, we selected only adult skeletons with preserved hip bone. Sex of the adult skeletons was determined by the analysis of pelvic morphological traits: sciatic notch, ventral arc, subpubic concavity, ischiopubic ramus, and presence of preauricular sulcus (Phenice 1969; Buikstra and Ubelaker 1994; Walker 2005), and cranial traits: mental eminence, orbital margin, glabellar area, nuchal area, and mastoid process (Buikstra and Ubelaker 1994), when possible.

Skeletons were also examined for pathological changes, dental pathology, and trauma (Aufderheide et al. 1998;

Ortner 2003) and only skeletons free of peri- or post-mortem changes that could affect results were included in the study.

Osteometric measurements

Eight standard osteometric measurements were taken (Moore-Jansen et al. 1994):

1. Maximum Length: distance from the most superior point on the head of the femur to the most inferior point on the distal condyles.
2. Epicondylar Breadth: distance between the two most laterally projecting points on the epicondyles.
3. Maximum Head Diameter: the maximum diameter of the femur head, wherever it occurs.
4. Anterior-Posterior (Sagittal) Subtrochanteric Diameter: distance between anterior and posterior surfaces at the proximal end of the diaphysis, measured perpendicular to the medial-lateral diameter.
5. Medial-Lateral (Transverse) Subtrochanteric Diameter: distance between medial and lateral surfaces of the proximal end of the diaphysis at the point of its greatest lateral expansion below the base of the lesser trochanter.
6. Anterior-Posterior (Sagittal) Midshaft Diameter: distance between anterior and posterior surfaces measured approximately at the midpoint of the diaphysis, at the highest elevation of linea aspera.
7. Medial-Lateral (Transverse) Midshaft Diameter: distance between the medial and lateral surfaces at midshaft, measured perpendicular to the anterior-posterior diameter.
8. Midshaft Circumference: circumference measured at the level of the midshaft diameters.

Length measurement were taken with osteometric board, circumference with metal tape, and the measurements 2–6 were measured with sliding caliper. Each measurement was taken twice by two authors independently, and in cases of discrepancies, measurements were repeated. Left and right bones were measured, and no statistical differences between left and right bone measurements were found (data not shown). Therefore, left bones were used for the analysis, and in cases of missing values, right bone measurements were used.

Statistic methods

We used both univariate and multivariate methods for analysis of sexual dimorphism. Initially, sexual dimorphism was examined with independent samples *t test* and then discriminant functions were calculated. Accuracy of the derived functions was determined by cross-validated classification method, in which each case is classified by the discriminant function calculated from all cases excluding that case. As 80% classification rate is considered useful for sex determination, only discriminant functions that met this criteria were considered relevant for this research (Vance et al. 2011). We also reported eigenvalues, canonical correlations, and Wilk's lambda as indicator of the reliability of the discriminant functions. All analyses were conducted using SPSS (ver 18; SPSS Inc, Chicago, IL, USA), with the significance level set at $p<0.05$.

Results

From the total of 214 examined skeletons, 27 females and 48 males met the selected criteria. Initial comparison showed that males were larger in all femoral dimensions, and that the differences are statistically significant for all measurements (Table 1). The greatest dimorphism exhibited was by maximum head diameter, while maximum length measurements showed to be least dimorphic.

Ten discriminant functions were calculated, eight univariate and two multivariate functions. Among them, five of ten calculated discriminant functions met the criteria of classification rate higher than 80% and enabled reliable sex determination if only part of the bone was preserved, i.e. femoral head, subtrochanteric area or distal epiphysis.

Highest classification rate of univariate functions was obtained with maximum head diameter, while the best multivariate function showed combination of measurements of epicondylar breadth and AP midshaft diameter.

If obtained values are above the sectioning point the skeletal remains are classified as male, if the values are below the sectioning point skeletal remains are classified as female, and if they are equal to the sectioning point sex is considered indeterminate.

Discussion

The results of this study showed that femoral measurements were very useful for sex determination. Since standards for sex determination for antique populations have not been published until now, the main aim of this study was to test their sexual dimorphism and to calculate discriminant functions that can be applied on fragmented skeletal remains. In this paper, the widely used approach for calculating discriminant functions was avoided. More specifically, we produced functions that can be applied if only a part of the bone was preserved, for example only proximal femoral element, and at same time took into consideration various elements that could affect the measurements such as the frequency of osteodegenerative changes on femoral auricular surfaces, bone structure and resistance to deterioration, etc.

In the line with previous studies (Steyn and Iscan 1997; Šlaus 1997; Šlaus et al. 2003; Spradley and Jantz 2011), single measurements that performed best were maximum diameter of head and epiphyseal breadth. Their importance supports the study of Spradley and

Table 1. Initial comparison of femoral measurements (in millimeters).

	Male			Female				
	N	Mean	SD	N	Mean	SD	*t*	*p*
Maximum Length	30	442.50	23.05	21	411.88	27.87	4.283	<0.001
Epicondylar Breadth	29	79.12	4.26	21	70.00	3.02	8.392	<0.001
Maximum Head Diameter	46	47.42	3.36	25	40.64	1.79	11.093	<0.001
Anterior-Posterior (Sagittal) Subtrochanteric Diameter	46	28.25	2.67	26	24.83	2.33	5.441	<0.001
Medial-Lateral (Transverse) Subtrochanteric Diameter	48	33.98	2.80	26	30.27	3.08	5.259	<0.001
Anterior-Posterior (Sagittal) Midshaft Diameter	48	29.15	3.07	27	24.92	2.09	6.368	<0.001
Medial-Lateral (Transverse) Midshaft Diameter	48	28.45	2.57	27	25.62	2.00	4.941	<0.001
Midshaft Circumference	48	92.69	5.86	27	81.96	4.55	8.216	<0.001

Table 2. Discriminant functions and sexing accuracies.

Discriminant functions	Eigen-value	Canonical correlation	Wilks' λ	Cutting point	Accuracy males	%	Accuracy females	%	Overall classification	%
Femur maximum length	0.374	0.522	0.728	434.42	25/30	83.3	13/21	31.9	38/51	73.1
Femur Epicondylar Breadth*	1.467	0.771	0.405	72.69	25/29	86.2	19/21	90.5	44/50	88.0
Femur Max Diam Femur Head*	1.273	0.748	0.44	41.67	40/46	87.0	24/25	96.0	64/71	90.1
Femur AP Sub-Troch Diam	0.423	0.545	0.703	26.03	38/46	82.6	16/26	61.5	54/72	75.0
Femur ML Sub-Troch Diam	0.384	0.527	0.722	32.54	44/48	91.7	14/26	53.8	58/74	78.4
Femur AP Mid-shaft Diam	0.555	0.598	0.643	25.58	41/48	85.4	18/27	66.7	59/75	78.7
Femur ML Mid-shaft Diam	0.334	0.501	0.749	26.18	41/48	85.4	17/27	63.0	58/75	73.3
Femur Midshaft Circumference*	0.925	0.693	0.52	85.36	44/48	91.7	21/27	77.8	65/75	86.7
Epicondylar Breadth x 0.207 + Anterior-Posterior Midshaft Diameter x 0.127 – 20.201*	1.682	0.792	0.373	–0.183	25/28	89.3	20/21	95.2	45/49	91.8
Anterior-Posterior Subtrochanteric Diameter x 0.258 + Medial-Lateral Subtrochanteric Diameter x 0.234 – 14.644*	0.754	0.656	0.57	–0.248	40/46	87.0	19/26	73.1	59/72	81.9

* These functions are recommended to use depending on the preserved part of the bone.

Jantz (2011) that included all standard bone measurements, showing that these femoral measurements were in top four univariate functions, both in white and black modern American population. However, greater weight should be given to the femoral head measurement, as it is more resistant to taphonomic effects in comparison to the distal epiphysis. Additionally, this is a femoral part in which osteodegenerative changes occur later in life (Šlaus 2006; Anderson and Loeser 2010). Although joint surface measurements provide the highest classification rates, they are often insufficiently preserved when dealing with ancient remains. As femoral head and epicondyles are mostly formed of spongy bone, they are easily broken, which prevents obtaining accurate measurements (Safont et al. 2000). For this purpose, two discriminant functions of femoral shaft measurements were calculated and reached classification rates higher than 80%. Midshaft circumference, in comparison to the previous studies gave a surprisingly high classification rate of 86.7%, while prior studies classification rates were between 78 and 85.6% (Black 1978; Safont et al. 2000; Wrobel et al. 2002; Nagaoka and Hirata 2009). Nevertheless, even in this case, there are a few limitations that should be considered. Foremost, if the bone is not sufficiently preserved it is very difficult to pinpoint the middle of the diaphysis, and in that situation, measurements should not be taken. Additionally, this measurement cannot be applied if the skeleton is not sufficiently preserved to show the evidence of the epiphyseal closure. In that case, a combination of subtrochanteric measurements can be applied, regardless of somewhat lower classification rates (81.9%). In contrast to midshaft measurements, it is possible to locate the subtrochanteric landmark, even if the rest of the bone is fragmented. When compared to joint epiphyseal measurements, subtrochanteric area is free of osteoarthritic changes and more durable due to its cortical bone formation. Whilst joint sizes and bone lengths are primary conditioned by intrinsic factors, the diaphyseal dimensions, are, except of differences in bone remodeling in the tubular bones during adolescence (Black 1978), conditioned by functional demands of weight bearing and muscle activity (Šlaus 1997). Thus, in the wake of that fact, population-specific activity patterns and paleopathological data should also be considered when available.

This study reached relatively high classification accuracy for sex determination reaching to 73.1–91.8%, which will certainly enhance further analysis of the skeletal remains from Salona necropolis. Furthermore, due to a somewhat similar Roman period population structure in all of the Adriatic coast area, the studied population can be considered representative of all ancient necropoles from the same period and area, which means that the calculated discriminant functions could be widely applicable. Moreover, it also raised some methodological issues. Namely, the study has pointed out that, when creating discriminant functions, we should take into consideration not only the accuracy, but also the applicability based on the experience from the anthropology lab that considers the state of preservation, frequent pathology, etc. In addition to general paleopathological data, population specific data and activity patterns also should be considered if available, in order to maximize the power of sex determination in all cases. In conclusion, we are hoping for future studies that would test our hypothesis of applicability of newly developed functions on similar populations.

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Authors' contributions

IJ, ŽB, IK, ŠA participated in study design and interpretation of data; IJ, ŽB, and IK conducted anthropometric measurements, performed statistical analyses and drafted the manuscript. IJ, ŽB, IK, ŠA participated in analysis and interpretation of data, revised the manuscript, and gave the final approval of version to be published.

Conflict of interest

The authors declare no conflict of interest.

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