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Source / Izvornik: Croatian Medical Journal, 2019, 60, 221 - 226

Journal article, Published version Rad u časopisu, Objavljena verzija rada (izdavačev PDF)

https://doi.org/10.3325/cmj.2019.60.221

Permanent link / Trajna poveznica: https://urn.nsk.hr/um:nbn:hr:105:250140

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Croat Med J. 2019;60:221-6 https://doi.org/10.3325/cmj.2019.60.221

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Aim To determine the sexual dimorphism of acetabular measurements in contemporary Croatian population and to provide a discriminant function equation for sex estimation.

Methods The sample consisted of 200 adult pelvic bones (100 male and 100 female) from positively identified victims of Croatian War of Independence. In total, 96 left (48 male and 48 female) and 104 right (52 male and 52 female) acetabula were measured. One author measured two acetabular parameters using a sliding caliper: acetabular diameter (AD) and transverse acetabular diameter (TAD). Another author re-measured the acetabula of 40 randomly selected individuals to determine the inter-observer error.

Results Both measured variables showed significant sexual dimorphism. Men had significantly higher values for AD and TAD than women. Receiver operating characteristic curve analysis showed that the cut-off point for prediction of male sex when using acetabular diameter was higher than 54 mm. For transverse acetabular diameter it was higher than 52 mm. The discriminant function was generated by using both acetabular variables, with 88% of accuracy in sex estimation. Inter-observer error was not significant.

Conclusion The acetabular measurements can be used for sex estimation in contemporary Croatian population with high accuracy.

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Received: November 9, 2018 Accepted: March 13, 2019

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Sex estimation of human skeletal remains is of crucial importance in medicolegal context, as it is usually the first step in the identification of remains (1-3). Two methods of sex estimation are commonly used - morphological and osteometric (4,5). As the morphological method is based on the visual evaluation of sexually dimorphic morphological bone features, it is prone to subjective judgment and depends on the examiner's experience. The need to find more reliable and objective methods led to the development of osteometric methods. These are based on statistical analyses of measurements of different skeleton parts, meaning that formulae are generated from discriminant function analysis (6-10). One of the main problems of osteometric method is inter-observer difference. Namely, it is hard to precisely define the starting and finishing point of each measurement, which leads to different readings of the same measurement by two observers (11). In addition, bone fragmentation and damage resulting from body decay decreases the number of measurements that can be obtained. Another problem is that formulae generated from discriminant function analysis are population-specific, as various populations differ with regard to general body size and degree of sexual dimorphism, and are not as reliable outside the population they were based on (6,7,12,13). However, some recent studies indicate that some formulae are possibly less population-specific (14,15).

The aim of this study was to develop standards for sex estimation of Croatian population by using two acetabular parameters. The acetabulum was chosen as one of the most diagnostic pelvic variables for sex assessment and one of the body parts that are less vulnerable to post mortem decay and damage. To our knowledge this is the first study in Croatia and the surrounding region to use pelvic bone parameters in developing population-specific standards for sex assessment.

MATERIALS AND METHODS

The study sample were the skeletal remains of the contemporary Croatian population, precisely, the victims of Croatian War of Independence (1991-1995). The sample consisted of pelvic bones from 100 male and 100 female victims who had already been positively identified.

Victims had been identified at our Institute using various methods, including anthropological examination, dental records, antemortem x-ray, and DNA analysis. The sample includes individuals from all socioeconomic categories and Croatian regions, with the mean age of 51 years

(mean age for men 45.3 years, range 20-76; mean age for women 57.1 years, range 34-80). Only complete, undamaged acetabula were measured. If both acetabula were complete, only the better preserved one was measured. Consequently, the measurements were performed on 96 left (48 male and 48 female) and 104 right acetabula (52 male and 52 female). All of the measurements were made in period from 2014 to 2017.

By using a sliding caliper, the first author measured two acetabular parameters (Figure 1):

- 1) acetabular diameter (AD): maximum diameter of the acetabulum measured in a superior to inferior direction diameter of the acetabulum along the axis of the body of the ischium (16).
- 2) transverse acetabular diameter (TAD): maximum acetabular diameter from the pubic eminence on the acetabular rim (17).

Both dimensions were measured to the nearest millimeter.

Normality of distribution was tested using Kolmogorov-Smirnov test. Both AD and TAD variables showed normal distribution on the left and right side in both sexes. The

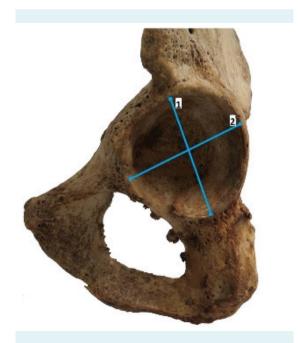


FIGURE 1. Measured parameters of the acetabulum: acetabular diameter – 1 and transverse acetabular diameter – 2.



independent t test was used to compare measurements between men and women, and between the right and left acetabula of different individuals. After the original data set was collected, we determined the repeatability of measurement by the inter-observer test. The second author selected 40 acetabula from the original data set using True Random Number Generator (Random.org) and re-measured them. The two sets of values for these 40 individuals were compared with Pearson correlation coefficient r and dependent t test. Sexual dimorphism data are presented using standard descriptive statistics. Receiver operating characteristic (ROC) curve analysis was performed to determine the cut-off value for male sex prediction. Discriminant function analysis was performed by using both measured variables. The data were analyzed with SPSS, version 25.0 (IBM Corp, Armonk, NY, USA).

RESULTS

There were no significant differences between the left and right acetabula in both sexes (independent t test, P = 0.157). Both measured variables showed significant sexual dimorphism. AD and TAD values were significantly higher in men

than in women (P<0.001). Men exhibited greater variation than women for TAD, but not for AD. These results suggest that the measured variables are useful in evaluating morphological differences between the sexes (Table 1).

The cut-off value for prediction of male sex for AD was higher than 54 mm, and for TAD was higher than 52 mm. Therefore, the highest accuracy for AD was observed at specificity of 90% and sensitivity of 84%, and for TAD at specificity and sensitivity of 88%. Since the sample consisted of an equal number of men and women, the overall accuracy of this method was 87% for AD and 88% for TAD (Table 2).

The area under the curve was large for both parameters and almost equal. This means that both parameters predicted male sex with similar accuracy (Figure 2).

The standardized coefficients indicate the relative contribution of each variable to the function. We used non-standardized coefficients to calculate discriminant function score from the raw data (Table 3). A discriminant score was obtained by multiplying each variable with its

TABLE 1. Descriptive statistical analysis of acetabular diameter and transverse acetabular diameter for both sexes

		Arithmetic Standard mean deviation Minimum		25th Maximum centile Median			75th centile	- 3		Р	
Acetabular diameter	female								-14.2	198	<0.001
		51.95	2.59	47.00	59.00	50.00	52.00	54.00			
	male	57.12	2.56	52.00	63.00	55.00	57.50	59.00	17.2		
Transverse acetabular diameter	female	50.72	2.05	46.00	56.00	50.00	51.00	52.00	-14.8	100	<0.001
	male	55.76	2.73	50.00	62.00	54.00	56.00	58.00	-14.8	198	

TABLE 2. Cut-off point values of acetabular diameter and transverse acetabular diameter in the prediction of male sex obtained by receiver-operating characteristic curve analysis

	Criterion	Sensitivity (%)	Specificity (%)	Area under the curve	Р
Acetabular diameter	>54 mm	84.00	90.00	0.92	< 0.001
Transverse acetabular diameter	>52 mm	88.00	88.00	0.93	< 0.001

TABLE 3. Standardized and non-standardized discriminant function coefficients, structure matrix, and classification for acetabular diameter and transverse acetabular diameter

Model	Structure matrix	Standardized canonical discriminant function coefficients	Non-standardized canonica discriminant function coefficients	l Wilks' Lambda	n P	Average accuracy		
Acetabular diameter	0.967	0.431	0.168	0.495	< 0.001			
Transverse acetabular diameter	0.932	0.618	0.256	0.476	< 0.001			
Constant			-22.778			88% correctly		
Group centroids for females			-1.079			classified (88% cross-validated)		
Group centroids for males			1.079			CiOss-validated		
Sectioning point			0.000					

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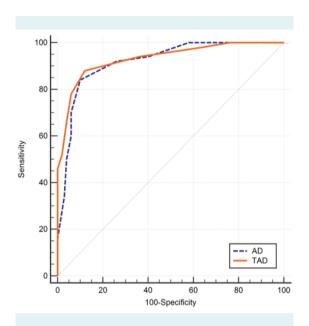


FIGURE 2. Receiver-operating characteristic curve for acetabular diameter and transverse acetabular diameter.

non-standardized coefficient, and adding them together along with the constant. The constant has no inherent value, and only serves to calibrate the sectioning point to zero when the number of men and women is equal. The discriminant function formula employing AD and TAD was the following:

$$Df = -22.8 + 0.17 \times AD + 0.26 \times TAD$$

All values higher than 0.00 indicate male sex, while all values lower than 0.00 indicate female sex. The accuracy of sex estimation using this discriminant function was 88%.

Inter-observer error for measurements performed by two different authors was not significant (P=0.858 for AD, P=0.126 for TAD). There was a strong correlation between measurements obtained by two observers for both AD (r=0.963) and TAD (r=0.937). This indicates that both measurements were repeatable and could be measured with a high degree of accuracy.

DISCUSSION

The 87% accuracy of sex prediction with AD and 88% with TAD obtained in our study show that the acetabulum is a very good indicator for sex estimation in contemporary Croatian population. The cut-off points for AD

correctly classified 90% of women ("specificity") and 84% of men ("sensitivity"), while the cut-off points for TAD correctly classified 88% of women ("specificity") and 88% of men ("sensitivity"). Employing both variables in sex discrimination function formulae leads to the accuracy of sex estimation in 88% of cases. Hence, with these two simple-to-use variables, we achieved a high level of accuracy necessary for working with skeletal remains in forensic context (6) and court procedures (18,19). Interestingly, the discriminant function formula using both AD and TAD can achieve hardly better accuracy than AD and TAD separately. This implies that sexual dimorphism in our specimen is almost entirely size-based, with only a minor influence of the shape of the acetabula.

While conducting the forensic analysis of a high number of skeletal remains from the Croatian War of Independence (over 4000 examined skeletal remains), we have often found the acetabulum to be well preserved, even after a long postmortem period. We measured either the left or right acetabulum of one individual and obtained an almost equal number of measurements for both sides. This is why we believe that the results can be used on both sides, especially because no significant difference was found between the groups of the left and right acetabula.

Our results are in accordance with the previous research in contemporary Croatian population, which showed that the maximum femoral head diameter (complementary part of the hip) can be used to discriminate the sex with 94% accuracy (20).

Acetabulum sex dimorphism has been studied in various populations. Our results can be compared to those obtained by Goméz-Valdéz et al (21), who have shown that TAD was one of the important sex indicators in the contemporary Mexican population, with the accuracy of almost 87%. A slightly higher accuracy obtained when using AD in Croatian population can suggest some sort of population specificity. Murphy (22) analyzed a sample of Polynesian innominate bones, using a single variable - maximum diameter of the acetabulum, which yielded 86% accuracy of sex determination. Stayn and Iscan (23) have established that the use of maximum acetabulum diameter, in a superior-inferior direction, in modern Greek population achieves the accuracy of 84%. Benazzi et al (11) achieved the accuracy of 96.4% based on the planar image of the acetabulum and related metric data measuring area, perimeter, longitudinal, and transverse maximum width.



The mentioned research indicates inter-population variability. We believe that the standards elaborated in this study are not suitable for use in forensic analysis of skeletal remains of populations other than Croatian. If our measurements are compared to the those obtained by Goméz-Valdéz et al (21) (mean AD and TAD for Croatian vs Mexican population for women are 51.9 vs 50.7 and 48.8 vs 46.3 respectively; and for men 57.1 vs 54.5 and 55.7 vs 52.2 mm respectively), the need for population specific results becomes obvious - Croatians of both sexes have on average larger AD and TAD than Mexicans. We believe, however, that our results could be applied to similar populations; primarily to other contemporary populations in Southeastern Europe. We also believe that these results can be used in other populations with a similar or the same sample mean and demarking points.

In conclusion, our results clearly show that acetabular measurements can be used for sex estimation in contemporary Croatian population. They are particularly important in cases of fragmented and incomplete skeletal remains when the acetabular region is preserved. The need for this is obvious since in Croatia there are still over 1500 missing and unidentified war victims (24). These findings will also be of special interest in routine medicolegal investigations. The results from this study encourage the development of new osteometric standards for sex determination based on other parts of the skeleton in contemporary Croats.

Funding None.

Ethical approval The permission to use skeletal remains from the Croatian War of Independence was given by the Ministry of Croatian Veterans' Affairs.

Declaration of authorship PB and DM conceived and designed the study; PB and MB acquired the data; PB, MB, VP, and DM analyzed and interpreted the data; PB and MT drafted the manuscript; PB, MB, MT, and DM critically revised the manuscript for important intellectual content; PB, MB, VP, and DM gave approval of the version to be submitted.

Competing interests All authors have completed the Unified Competing Interest form at www.icmje.org/coi_disclosure.pdf (available on request from the corresponding author) and declare: no support from any organization for the submitted work; no financial relationships with any organizations that might have an interest in the submitted work in the previous 3 years; no other relationships or activities that could appear to have influenced the submitted work.

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