Title:

Assessing Size and Strength of the Clavicle for its usefulness for Sex Estimation in a British Medieval Sample.

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Running Title: Sex estimation using the clavicle

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Abstract

The construction of the biological profile from human skeletal remains is the foundation of anthropological examination. However, remains may be fragmentary and the elements usually employed, such as the pelvis and skull, are not available. The clavicle has been successfully used for sex estimation in samples from Iran and Greece. In the present study the aim was to test the suitability of the measurements used in those previous studies on a British Medieval population. In addition, the project tested whether discrimination between sexes was due to size or clavicular strength.

The sample consisted of 23 females and 25 males of pre-determined sex from two medieval collections: Poulton and Gloucester. Six measurements were taken using an osteometric board, sliding callipers and graduate tape. In addition, putty rings and bi-planar radiographs were made and robusticity measures calculated. The resulting variables were used in stepwise discriminant analyses.

The linear measurements allowed correct sex classification in 89.6% of all individuals. This demonstrates the applicability of the clavicle for sex estimation in British populations. The most powerful discriminant factor was maximum clavicular length and the best combination of factors was maximum clavicular length and circumference. This result is similar to that obtained by other studies.

To further investigate the extent of sexual dimorphism of the clavicle, the biomechanical properties of the polar second moment of area J and the ratio of maximum to minimum bending rigidity are included in the analysis. These were found to have little influence when entered into the discriminant function analysis.

Introduction

Sex determination/estimation is of vital importance to biological anthropology, as it is the first assessment made when creating the biological profile of an adult individual. If sex is incorrectly assigned, further assessments such as age and stature estimations will be incorrect as they are most accurate when using sex-specific regressions. For reasons of increased reliability, anthropologists continue to develop new or revise old methods for sex determination/estimation. In this study, sex estimations rather than determinations will be produced as they are not performed on a population with documented sex. (Ousley and Jantz, 2012)

Prior studies have shown that different populations have different identifiable factors for sex estimation (Charisi et al., 2011; Dayal et al., 2008; Frutos, 2005; Králík et al., 2014; Šlaus and Tomičić, 2005). This is the reason why a single, global method cannot be developed. This has led to a multitude of new population-specific techniques to determine biological profiles from different locations (Charisi et al., 2011; Manolis et al., 2009; Mountrakis et al., 2010)

Sexual dimorphism between males and females is what allows sex to be determined in human skeletal remains and is caused by several factors. Firstly, there is natural selection. Because males in the past had to compete with other males in order to increase reproductive success, larger males were more frequently selected for (Armelagos and Van Gerven, 1980). These selective pressures were absent in females, therefore they tend to be smaller than males. Secondly, there was the role of sexual selection. It has been suggested by Puts (2010) that in the past sexual selection for men was in favour of selecting traits such as size, muscularity, strength and aggression. This eventually led to males being larger in general than females. Finally, there are environmental pressures. The ontogenetic and behavioural environment to which males and females are subjected throughout their lives has caused differences in plastic features such as bone robusticity and strength. For example, males traditionally perform much more labour-intensive roles and as a result develop larger muscles, which require stronger and more robust bones to support them. This was investigated by Ruff (1987) who studied the decline of sexual dimorphism from hunter-gatherers to the industrial age and suggested that this decline is due to a steady decrease in the division of labour between the sexes.

[ENTER TABLE 1 HERE]

There are several bones which are commonly used for sex assessment in biological anthropology. Table 1 presents a number of methods used on each bone, with its corresponding accuracy of sex estimation. As expected the pelvis comes out as the most accurate method overall with accuracies up to 95.5% using a variety of techniques (Gonzalez et al., 2009; Patriquin et al., 2005; Steyn and İşcan, 2008). However, metric methods employing long bones such as the femora and clavicles achieved higher percentage accuracies than craniometrics. This is of particular interest as the skull has been traditionally seen as the second best predictor of sex after the pelvis.

The aim of this pilot study is to validate the accuracy of sex estimation of the clavicle using an English Medieval sample. In addition, we aim to determine whether sexual dimorphism in the clavicle is the result of size differences between males and females or if it is also affected by biomechanical factors (here measured as strength). Králík et al. (2014) have called for the need to associate biomechanics and environmental factors to the sex assessment of the clavicle.

Materials and Methods

The skeletal material used in this study comes from two collections, housed in the School of Natural Sciences and Psychology at Liverpool John Moores University, UK. These were the Poulton Project collection from Cheshire and the Gloucester Museums collection from Gloucestershire. The Poulton sample is a farming population dating 1153-1534 AD (Poulton, 2014; Roberts, 1998) and Gloucester is an urban population dating from 1137 AD (Atkin and Garrod, 1990). Although we recognise that pooling both samples will increase the variance due to potential difference in physical activity and diet, we combined the populations in order to ensure the regressions are robust and usable on both urban and rural samples.

Only skeletons with sex estimation based on both pelvic and cranial characteristics were used for the study. The methods used are those most commonly applied by anthropologists, and are outlined in Buikstra and Ubelaker (1994). Skeletons with an evident pathology or with a fragmentary right clavicle were discarded from the sample selection. All skeletons were aged using standard ageing techniques (Buikstra and Ubelaker, 1994) and individuals possessing cranial and postcranial adult sex determination features were included in the sample. This resulted in a sample of young, middle and older adults. Individuals with ambiguous sex characters were excluded from the sample resulting in a sample of twenty-five males and twenty-three females.

[ENTER TABLE 2 HERE]

The linear measurements were taken in mm and are defined in Table 2. Anterior-posterior and superior-inferior x-rays were also made of each clavicle. In addition, the non-destructive method of latex cast moulds, combined with bi-planar radiographs was used to reconstruct cross-sectional geometry at mid-shaft . As discussed by Stock and Shaw (2007) this method is the most robust way in which to examine the cross sectional measurements of J and Imax/Imin. Latex cast moulds of the clavicular cross-section were taken around the circumference of the midpoint. The moulds were removed then scanned into the computer with a flatbed scanner. This is commonly referred to as the latex cast method (O'Neill and Ruff, 2004; Stock, 2002). The cross-section mould scans were then edited in Adobe Photoshop® and the cortical thickness and medullary cavity size were measured by use of ImageJ (Table 2). The software package R was then used to extract the ellipse of the mould and superimpose the image onto the edited mould scan, commonly referred to as the ellipse model method (O'Neill and Ruff, 2004; Stock, 2002). Measures of maximum/minimum bending rigidity ratio (Imax/Imin) and the polar second moment of area J were taken using the Cross-Section R workspace (Sylvester et al., 2010).

Analyses were carried out using IBM SPSS version 21. Intra-observer error was assessed by calculating the percentage difference between three sets of all measurements, taken on three individuals over a two week interval. A t-test was used to check for significant differences between males and females for each measurement. A stepwise discriminant with leave-one-out function analysis was then conducted to determine the most discriminatory factors and also the best combination of factors for successful sex classification.

Results

Intra-observer error rates were low, indicating that there was good repeatability of the measurements. The highest recorded percentage error was 4.6%, which is below the 5% threshold. Descriptive statistics show that overall, mean values for males are higher than those for females (Table 3). However, the mean Imax/Imin measure is only just larger in males than in females by 0.1mm. This is of interest because Imax/Imin is an indicator of rigidity and also the shape of the bone, therefore there is little difference between the cross-sectional shape of male and female clavicles. Student t-tests, show that maximum clavicular length ($t = 6.653$, $df = 46$, $P<0.001$), CVD ($t = 5.446$, $df = 46$, $P<0.001$), CSD ($t = 5.28$, $df = 46$, $P<0.001$) and Circumference ($t = 5.895$, $df = 46$, $P<0.001$) are significantly different between the sexes at the 99% level with equal variances assumed. The polar second moment of area (J) is significantly different between males and females at the 95% level with equal variances assumed ($t = 3.06$, $df = 46$, $P = 0.04$). For the strength factor, it is worth noting that even though there is a significant difference between males and females, there is a large area of overlap due to the high standard deviation for both males (std. dev = 4.742) and females (std. dev = 4.417). Imax/Imin is the only variable that does not show a significant difference between males and females ($t = 0.85$, $df = 46$, $P = 0.4$).

[ENTER TABLE 3]

Using a stepwise discriminant analysis with all variables, only two variables were retained because of their high discriminatory power (Table 4). The two discriminatory measurements were maximum clavicular length and circumference (circumference of the mid-shaft). Adding more variables did not increase statistical significance. The Wilks Lambda value was 0.376 when both variables were entered together (Table 4), which indicates a high predictability of sex from these two factors with $P<0.001$. Table 4 shows that other measurements had much lower canonical values and therefore a weaker discriminatory power.

[ENTER TABLE 4]

The overall classification rate using maximum clavicular length and Circumference combined was 89.6% (Males=84%, Females=95.7%). A cross-validated analysis resulted in successful classification results that were identical to the non-cross-validated results (Table 5).

[ENTER TABLE 5 HERE]

The equation for classifying the sex of an unknown individual from the measurements of maximum clavicular length and Circumference is as follows:

$(\text{Maximum clavicular length} * 0.762) + (\text{Circumference} * 0.675) - 130 = X$

If $X<0$ then the individual is classified as female and if $X>0$ the individual is classified as male.

Discussion

Sex estimation is the starting point of a typical skeletal analysis in biological anthropology. Traditionally, qualitative methods for sex estimation have been used, but this requires years of training and experience to perfect. Anthropologists have therefore a tradition of developing metric methods for sex estimation which allows for easier reproducibility, greater accuracy and increased reliability (Akhlaghi et al., 2012; Gonzalez et al., 2009; Patriquin et al., 2005; Steyn and İşcan, 2008).

According to several studies, certain measurements of the clavicle have been proven to be dimorphic, such as the maximum clavicular length (Akhlaghi et al., 2012; Albanese, 2013; Frutos, 2002; Králík et al., 2014; McCormick et al., 1991; Spradley and Jantz, 2011; Thieme and Schull, 1957; Tise et al., 2013). Due to the high accuracies obtained by these studies, certain measurements are frequently used in sex estimation of the clavicle, such as maximum clavicular length, clavicular vertical diameter, clavicular sagittal diameter and Circumference. Nevertheless, it is necessary to understand the exact nature of these sexual dimorphic features that allow us to use them for identification purposes. Frequently features are size related, or secondary sexual characteristics. However, certain factors (Králík et al., 2014) that could have potential influence on the results such as strength and the maximum/minimum bending rigidity ratio may have been overlooked. By understanding the underlying causes for sexual dimorphism in different regions of the body it is possible to determine their usefulness as cross-population or cross-time period sex estimation traits.

The descriptive statistics show that there are size differences between males and females in many of the measurements (Table 3). The results of this study are consistent with those of others (Akhlaghi et al., 2012; Frutos, 2002; McCormick et al., 1991; Thieme and Schull, 1957) and show that in this medieval British population the measurements of maximum clavicular length, clavicular vertical diameter, clavicular sagittal diameter and Circumference are all larger in males than in females. The values for the polar second moment of area (J) are also significantly higher in males than females which was expected as males tend tobe more physically active. The only variable that is not significantly different between males and females is the Imax/Imin measurement. This suggests that there was little to no difference in the bending rigidity of bones between males and females, regardless of size. It can also be seen that there is a greater variation within sexes for the measure of Imax/Imin than between sexes (Table 3). This is most likely due to the fact that the bending rigidity of bone has no relationship with sex in this sample (Table 3) but this should be explored further with samples of known activity.

Prior to the discriminant analysis, canonical coefficient values were produced (Table 3). It was found that both the polar second moment of area and bending rigidity have the lowest discriminatory powers. Therefore environmental factors such as physical activity and diet of the individual, rather than sexual division of labour are responsible for the reduced sexual dimorphism observed between males and females. It is also worth noting that the effects of biomechanical factors may not have a positive impact on sex estimation, but may actually smooth the differences between sexes, except in cases of extreme sexual division of labour. This is shown by Stock and Pfeiffer (2004) in a population that has known division of labour between the sexes. In this example, males have different levels of bilateral asymmetry in the forelimb reliant on what environment they depended upon. In the same populations females have relatively homogenous robusticity between the environments. This shows that robusticity in a population is highly variable and is determined by tool use and habitual activity.

Stepwise discriminant analysis required two steps to achieve the most effective sex classification from measurements taken from the sample. The analysis did not include any additional factors because they did not significantly increase match probability. Maximum clavicular length had the highest canonical discriminant function of 0.762 (Table 4), identifying it as the single best variable for determining sex in this population. The highest accuracies were achieved when length and circumference measurements were combined. This is consistent with other studies (Kaur et al., 1997; Králík et al., 2014; Murphy, 1994; Thieme and Schull, 1957).

[ENTER FIGURE 1 HERE]

The overall accuracy for correct classification was 89.6% (Figure 1). This is consistent with other studies which have reached accuracies of over 92% of cases classified correctly (Frutos, 2002; Kaur et al., 1997; McCormick et al., 1991; Murphy, 1994; Steel, 1966). The use of clavicular dimensions followed by stepwise discriminant function analysis is highly accurate, allowing for the regression equation to be applied and increase reliability. This makes it invaluable for use in archaeological contexts where there is significant fragmentation of skeletal elements.

As stated previously, the age of individuals was not known but all skeletons in the sample were of an age where sex assessment using conventional techniques such as the Phenice method could be used (Phenice, 1969). However, according to Králík et al. (2014) age has no effect on the discriminant function unless the reference group is older than 61. This is most likely caused by the onset of age-related osteoporosis. Bilateral asymmetry was not considered for these analyses. Králík et al., (2014, p.212) state that 'in the larger proportion of cases, the right clavicle is shorter than the left' and 'Midshaft circumference is significantly larger on the right side in both males and females'. The present study however, only included right clavicles in order to minimise the effect of sidedness.

The main aim of this study was to test whether sex estimation using the clavicle was as accurate for a British Medieval sample as those for the Iranian and Greek populations. Kralik et al. (2014) warned that population affinity has a significant effect on sex determination accuracy when using the clavicle and called for the creation of population specific regression formulae. However, pooling a wide geographical range of samples in order to create a universal sex determination regression may be more useful in an ever increasingly global world. In addition, this study attempted to incorporate other factors such as the polar second moment of area and bending rigidity index to determine whether biomechanical factors influenced the sex assessment of the clavicle. The traditional measurements of maximum clavicular length and circumference performed the best as discriminant functions providing accurate sex estimation in over 89% of cases with use of the regression equation. Measurements of the polar second moment of area and bending rigidity had only weak discriminant power. This suggests size, rather than strength, is the most sexually dimorphic variable. In the future, skeletal samples of known age and sex could be used to further test the clavicle for its usefulness for sex estimation in a broader geographical sample.

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Table 1: Methods used for sex determination and determination accuracies.

Reference	Type of Bone	Method used	Accuracies
Gonzales, et al. 2008	Pelvis	SA with DFA	90.1%-93.4%
Patriquin, et al. 2004	Pelvis	LM with DFA	94%-95.5%
Steyn and Iscan. 2008	Pelvis	Single Os coxae with DFA	79.1%-93.5%
Dayal, et al. 2008	Skull	Non-metric trait analysis with DFA	80%-85%
Steyn and Iscan. 1998	Zygomatic breadth	DFA	80%-86%
Green and Curnoe. 2009	Skull	Morphologika PCA and DFA	86.8%
Kharoshah, et al. 2010	Mandible	CT scans and virtual measurements	83.6%-84.2%
Balci, et al. 2005	Mandible	Seriation of visual landmarks	70.6%-95.6%
Iscan, et al. 1998	Femur	LM with DFA	87%-94%
Frutos. 2005	Humerus	LM with univariate analysis	76.8%-95.5%
Iscan and Shihai. 1995	Femur	SDFA	92.3%
Mall, et al. 2000	Femur	SDFA	91.7%
Akhlagi, et al. 2012	Clavicle	LM with DFA	73.3%-88.3%
Kralik, et al. 2014	Clavicle	LM with DFA	91.62%-92.55%
Papaioannou, et al. 2012	Clavicle	LM with DFA	84.4%-89%
Spradley and Jants. 2011	Post-cranium (clavicle)	SDFA	71.88-94.34% (93.4-93.6%)
Tise, et al. 2013	Post-cranium (clavicle)	SDFA	71.57-89.43 (81.25-93.33%)
Albanese. 2013	Clavicle, Humerus, Radius and Ulna	Logistic regression	97.4-91.9%

SA=Shape analysis, LM = Linear measurements, DFA = discriminant function analysis, PCA = principle component analysis,

SDFA = Stepwise discriminant function analysis

Table 2: Definitions of measurements.

Measurement	Description	Method used
MCL	Maximum clavicular length	Osteometric board
CVD	Vertical diameter at midpoint	Callipers
CSD	Sagittal diameter at midpoint	Callipers
Circumference	Circumference taken from the midpoint	Measuring tape
J	Strength measure ‘J’ (the torsional average bending rigidity) (Ruff, 2008) divided by MCL.	Software analysis
Imax/Imin	The maximum and minimum measures for ‘I’ (bending rigidity) (Ruff, 2008) divided by each other (Imax/Imin) multiplied by 100 to make a percentage.	Software analysis

Table 3: Descriptive statistics and independent samples *t*-test results

	Male				Female							
	N	Mean	S.D.	SE Mean	N	Mean	S.D.	SE Mean	t	CFC	CC	Sig.
MCL	25	148.7	8.457	1.69	23	133.9	6.706	1.40	6.653	0.762	-0.743	*
CVD	25	10.9	1.093	0.22	23	9.2	1.141	0.24	5.446	0.364	-0.614	*
CSD	25	13.5	1.224	0.24	23	11.7	1.187	0.25	5.280	0.584	-0.623	*
Circumference	25	40.9	2.822	0.56	23	35.7	3.313	0.69	5.895	0.675	-0.676	*
Strength	25	12.8	4.742	0.95	23	8.8	4.417	0.92	3.060	0.155	-0.441	**
Imax/Imin	25	1.9	0.614	0.12	23	1.8	0.452	0.09	0.861	-0.42	-0.077	N.S.

MCL: maximum clavicular length, CVD: vertical diameter of the midpoint, CSD: sagittal diameter of the midpoint, Circumference: the circumference of the midpoint, Strength: the strength of the bone calculated by dividing torsional strength (J) by the MCL, Imax/Imin: index of maximum bending rigidity, t is the value assigned to each factor by the T-test, CFC: Canonical function coefficient, CC: Spearman's rho correlation coefficient. The higher the canonical function coefficient, the higher the discriminatory power of the variable in the analysis.The lower the correlation coefficient, the stronger the correlation between sex and the factors. N for all values = 48, df = N-2.

*Significant at P<0.001, **Significant at P<0.005, N.S. not significant

Table 4: Final predictors used in stepwise discriminant analysis.

Step	Variables entered	Wilks Lambda	df	P-value
1	MCL	0.51	1	*
2	MCL Circumference	0.376	2	*

Variables entered are the variables entered together in the stepwise analysis. Wilks Lambda values close to zero imply high predictability while values closer to one imply low predictability. df are the degrees of freedom used in the analysis. P-value is the significance of the results.

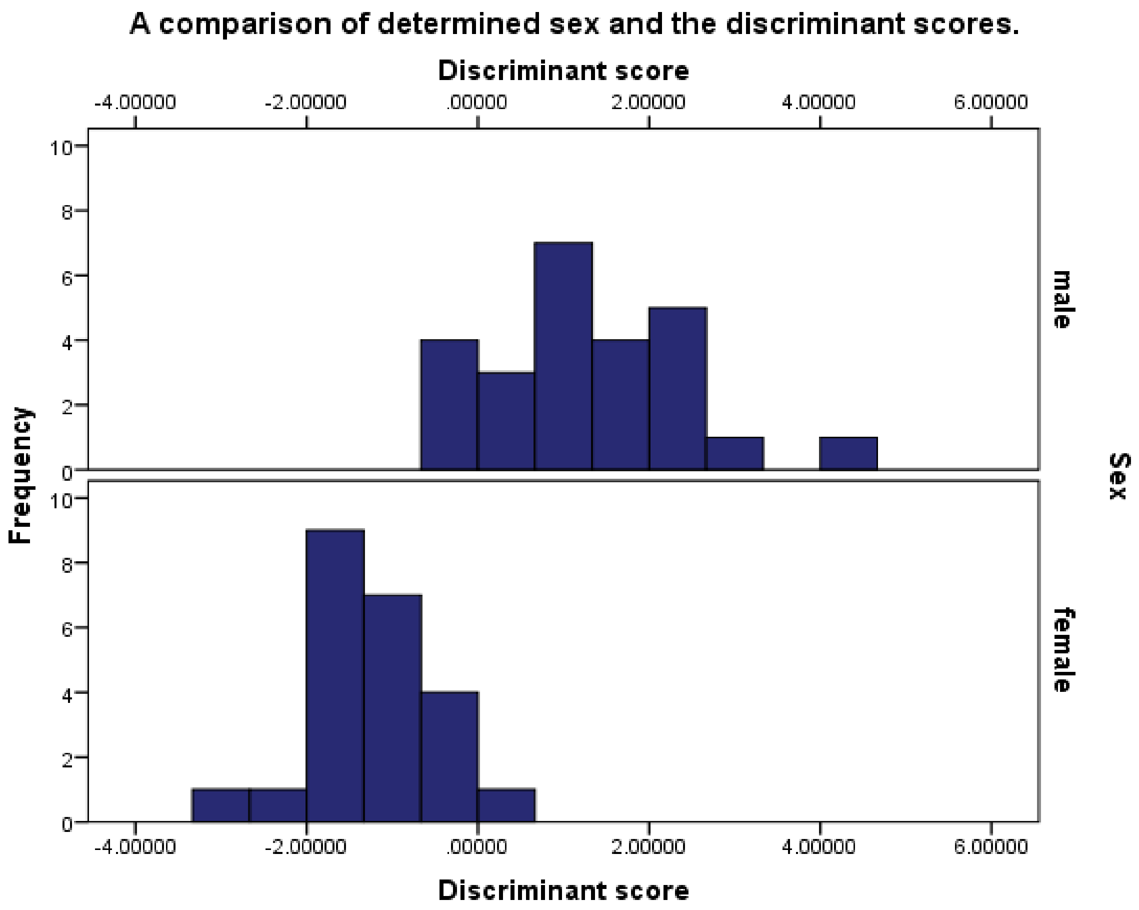
*Significant at $P<0.001$

Table 5: Classification and Cross-validated classification matrix using stepwise discriminant function analysis.

Sample		Predicted group		Percentage Accuracy	
Sex	N	M	F	M	F
M	25	21	4	84%	16%
F	23	1	22	4.3%	95.7%

Overall predictive accuracy is 89.6%. Cross-validated classification values are identical to the original classification matrix.

Figure 1



A comparison of determined sex and the discriminant scores.

A positive discriminant score is assumed male and a negative discriminant score assumes female.

Table and figure list