RESEARCH ARTICLE

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Determination of Sex in the Domestic Dogs using Femur Morphometric Data*

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Sexual dimorphism is the systematic difference in form between individuals of different sex in the same species. In this study, it was aimed to observe how the femoral osteometric measurements could be useful for determining sexual dimorphism. For this purpose, a total of 67 (33 male, 34 female) canine femurs of known dolichocephalic type were used. A total of 6 osteometric measurements (largest length (GL), biggest width of the proximal end (Bp), the greatest depth of the caput femoris (DC),the smallest width of the diaphysis (SD), the smallest circumference of the diaphysis (CD),the greatest width of the distal tip (Bd) were taken from each femur. Three different indices (proximal index, diaphyse index and distal index) were calculated using these osteometric measurements. Statistical calculations of all osteometric measurements and index values obtained were evalated by the SPSS 21.0 program. The difference between the mean values of femur osteometric measurements of males and females was found to be statistically significant (P<0.01). In conclusion, femur osteometric measurements of dogs did not provide 100% sexual discrimination but, it could make a great contribution to this subject with a predictive coefficient of approximately 85%.

Key Words: *Dog, femur, osteometry, morphometry, sex*

Evcil Köpeklerde Femur Morfometrik Verileri Kullanılarak Cinsiyetin Belirlenmesi

Seksüel dimorfizm, aynı türdeki farklı cinsiyetteki bireyler arasındaki sistematik form farkıdır. Bu çalışmada, femoral osteometrik ölçümlerin seksüel dimorfizmi belirlemede nasıl yararlı olabileceğini gözlemlemek amaçlanmıştır. Bu amaçla dolikosefalik tipi bilinen toplam 67 (33 erkek, 34 dişi) köpek femuru kullanıldı. En büyük uzunluk (GL), proksimal ucun en büyük genişliği (Bp), caput femoris'in en büyük derinliği (DC), diyafizin en küçük genişliği (SD), diyafizin en küçük çevresi (CD), distal ucun en büyük genişliği (Bd) olmak üzere toplam 6 osteometrik ölçüm alındı. Bu osteometrik ölçümler kullanılarak üç farklı indeks (proksimal indeks, diafiz indeksi ve distal indeks) hesaplandı. Tüm osteometrik ölçümlerin ve elde edilen indeks değerlerinin istatistiksel hesaplamaları SPSS 21.0 programı ile değerlendirildi. Erkek ve dişi femur osteometrik ölçümlerinin ortalama değerleri arasındaki fark istatistiksel olarak anlamlı bulundu (P<0.01). Sonuç olarak, köpeklerin femur osteometrik ölçümleri %100 cinsiyet ayrımı sağlamadı ancak, yaklaşık %85 tahmin katsayısı ile bu konuya büyük katkı sağlayabilir.

Anahtar Kelimeler: *Köpek, femur, osteometri, morfometri, cinsiyet*

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Introduction

Sexual dimorphism indicates differences exhibited in shape, size, variation and visual morphological structural differences between males and females of the same species. Sexual shape and size dimorphism is referred to as “Rensch’s rule”. This rule states that variation in male body size is more variable than females (1).

In pets, sexual dimorphism is a common and highly variable subject (2, 3). The difference in body size in both males and females indicates a significant variation known as sexual size dimorphism. Both the degree and direction of sexual size dimorphism vary widely between the individuals belonging to a certain takson (2, 4) and populations within wild species (5), as well as between breeds from the same domesticated species (1, 6, 7). While, sex determination using osteometric measurements has a more widespread use especially in humans (8-11), while it has found a more limited field of study in domestic animals. In particular, the fact that dogs have different sizes and morphological structures from Yorkshire Terrier to Doberman has brought about intraspecific polyformism (12). Therefore, the study of sexual dimorphism in canine anatomy has received little attention, particularly with regard to skeletal elements (13). Yet, this leads to a serious lack of sex determination in zooarchaeological studies. It is very problematic to determine the sex of domestic dogs and other canid species, especially from archaeological sites. It is osteometrically possible to determine the sex of both domestic dogs and wild canid species by comparing body size between males and females of these species (14).

There is a lack of sufficient modern data for sex determination in archaeological remaints (15) and no study has been found, except for two studies (13, 15) on sex

* Present study was produced from the first author’s PhD thesis.

determination in dogs in light of modern data. In one of these studies, sex determination from the humerus was tried with a simple technique, and it was reported that 85% of the male dogs could be predicted from the humeral position on a table (15). A study in German Shepherds suggested that males are larger than females, and femur provides the highest reported score for gender identification from the skeletal element (13). In this study, it was aimed to observe how the femoral osteometric measurements could be useful for determining sexual dimorphism.

Materials and Methods

Research and Publication Ethics: The study was carried out in İstanbul University-Cerrahpaşa, Osteoarchaeology Application and Research Center and approved by IUC Veterinary Faculty Unit Ethics Committee (13.07.2021/139480). A total of 67 adult canine femurs (33 males, 34 females) with dolichocephalic skull type (16) were examined.

Morphometric Measurements: The measurements of the femur were based on Von den Driesch (17) method. For this purpose, a total of 6 osteometric measurements were made on the femur (Figure 1A). A digital caliper was used to take the measurements with 0.1 mm accuracy rate.

Six femur measurements were obtained: largest length (GL), biggest width of the proximal end (Bp), the greatest depth of the caput femoris (DC), the smallest width of the diaphysis (SD), the smallest circumference of the diaphysis (CD), the greatest width of the distal tip (Bd). Three index calculations were made using these osteometric measurements (18, 19): Proximal index: Largest proximal width (Bp) x 100 / Maximum length (GL), Diaphyse index: The smallest width of the diaphysis (SD) x 100 / The largest length (GL), Distal index: The greatest width of the distal tip (Bd) x 100 / The greatest length (GL).

Statistical Analysis: Mean value and standard deviation values were obtained as statistical calculations. Parametric tests were preferred without conducting a normality test because the 'n' numbers were greater than 30 for both groups. Parametric t-test was performed to observe that the difference between the mean the right and left femurs. Correlation analysis of femoral osteometric measurements and indices were performed. For this purpose, SPSS 21.0 (Version 21.0, SPSS Inc., Chicago, IL, USA) program was used.

Results

In this study, osteometric measurements of the 67 adult dogs' femurs were obtained (Table 1). t-test was performed to check whether there was homotypic variation between the right and left bones. As a result, no homotypic variation detected between the right and left femurs. The difference between the mean values of femur osteometric measurements of males and females was found to be statistically significant (P<0.01). Considering GL and CD values, these measurements had a higher value in male subjects. The SD value was appeared to be a smaller value in females and significant at the P<0.01 level for the difference between males and females. A graphical distribution of the SD value versus the GL value in Figure 1B. Femur osteometric measurement indices were presented in Table 2 and were not statistically significant. Of the index values obtained, proximal index (Bp*100)/GL was higher than the distal index of bone (Bd*100)/GL). The SD*100/GL value, which is expressed as the thinness index, gave information about the structural feature of the bone and indicated the strenght of leg bones of the dogs. This value was also relatively lower in females. The index values of the femur had no determinant on sexual dimorphism. It was observed that the differences between the mean index values of female and male individuals were not statistically significant.

Correlation analysis of femur osteometric measurements and indices performed were given in Table 3. Positive high correlations were observed among the osteometric measurements themselves. There were negative correlations between Bp*100/GL and Bd*100/GL indices and osteometric measurements, which was salient. There was a negative correlation between SD*100/GL index and GL and DC values. The statistical significance of the difference between the CD values of female and male individuals and the large difference observed between them are thought to be another factor in the large SD*100/GL index.

There was a significant difference was observed between males and females in GL value (Figure 1C). In males, the highest variation was found in the GL value, and the lowest variation was in the Bd value. The variation in GL and CD values was higher than the other parameters, indicating that dogs with different structural characteristics were used in the study. High variation from female dogs was seen only in middle diaphyseal measurements (SD and CD) compared to males. The presence of relatively short but stocky dogs caused the variation for females to be concentrated in this region. Bp and Bd values showed a more homogeneous distribution than the others (Figure1D and E). Since Bp and Bd values are the most homogeneously distributed osteometric data and the differences between female and male individuals are statistically significant. The Mean ± Standard Deviation values of three measurements (Bp, Bd, CD) were given as; male (n=33): 47.31±7.67, female (n=34): 41.58±5.42, total (n=67): 44.40±7.18 (Table 4) and a simple scatter plot of (Bp+Bd+CD)/3 value versus GL value was created as in Figure1 F.

Table 1. Femur osteometric measurements

Sex	Statistics	GL	Bp	DC	SD	CD	Bd
	Mean	210.96[a]	46.09[a]	23.11[a]	16.70[a]	56.86[a]	38.99[a]
	N	33	33	33	33	33	33
MALE	SD	42.19	7.01	3.69	3.07	10.71	5.76
	Maximum	302.98	62.05	31.11	24.37	84.26	50.18
	Minimum	99.91	26.72	12.88	9.70	34.24	23.29
	Mean	187.43[b]	41.22[b]	20.40[b]	14.36[b]	49.14[b]	34.37[b]
	N	34	34	34	34	34	34
FEMALE	SD	22.87	4.97	2.53	2.25	7.71	4.09
	Maximum	237.05	53.29	26.52	21.24	74.12	45.51
	Minimum	130.24	29.30	14.05	10.35	34.92	23.74

[a,b]: Differences between mean values expressed with different letters in the same column are significant. (P<0.01)

Table 2. Femur index values

Sex	Statistics	Bp*100/GL	SD*100/GL	Bd*100/GL
	Mean	22.1[a]	7.98[a]	18.75[a]
	N	33	33	33
MALE	SD	1.65	0.68	1.78
	Maximum	26.74	9.70	23.67
	Minimum	19.40	6.73	15.81
	Mean	22.03[a]	7.66[a]	18.37a
	N	34	34	34
FEMALE	SD	1.16	0.73	1.06
	Maximum	24.29	9.00	21.25
	Minimum	20.07	6.53	16.45

[a]: Differences between mean values expressed with the same letter in the same column are insignificant

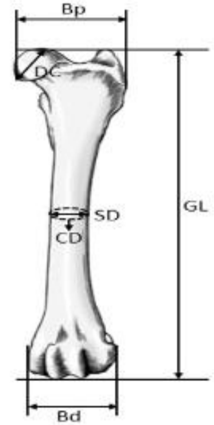
Table 3. Correlation analysis of femur osteometric measurements and indices

	GL	Bp	DC	SD	CD	Bd	Bp*100/GL	SD*100/GL	Bd*100/GL
GL	1	0.946**	0.924**	0.886**	0.878**	0.916**	-0.617**	-0.197	-0.566**
Bp	0.946**	1	0.947**	0.933**	0.926**	0.969**	-0.344**	0.008	-0.344**
DC	0.924**	0.947**	1	0.899**	0.909**	0.964**	-0.390**	-0.010	-0.281*
SD	0.886**	0.933**	0.899**	1	0.984**	0.927**	-0.308*	0.269*	-0.270*
CD	0.878**	0.926**	0.909**	0.984**	1	0.931**	-0.297*	0.254*	-0.238
Bd	0.916**	0.969**	0.964**	0.927**	0.931**	1	-0.326**	0.054	-0.206
Bp*100/GL	-0.617**	-0.344**	-0.390**	-0.308*	-0.297*	-0.326**	1	0.656**	0.867**
SD*100/GL	-0.197	0.008	-0.010	0.269*	0.254*	0.054	0.656**	1	0.629**
Bd*100/GL	-0.566**	-0.344**	-0.281*	-0.270*	-0.238	-0.206	0.867**	0.629**	1

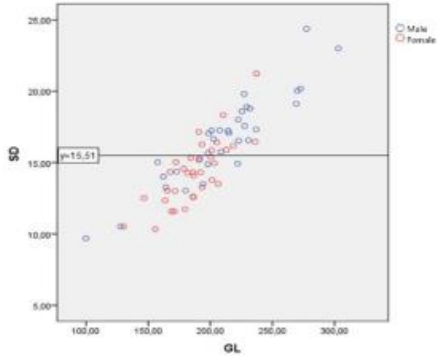
*: P<0.05; **:P<0.01

Table 4. Average values of (Bp+Bd+CD)/3 value

Sex	Mean	N	SD	Maximum	Minimum
Male	47.31	33	7.67	64.04	28.08
Female	41.58	34	5.42	57.64	29.32
Total	44.40	67	7.18	64.04	28.08

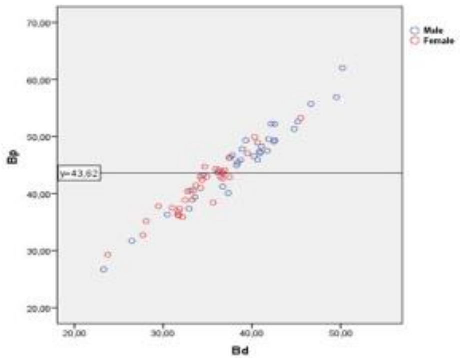


A. The osteometric measurements of femur (General)

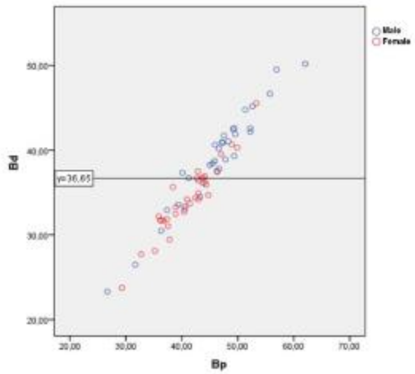


B. Graphical distribution of SD versus GL. Straight line SD (mean) = 15.51

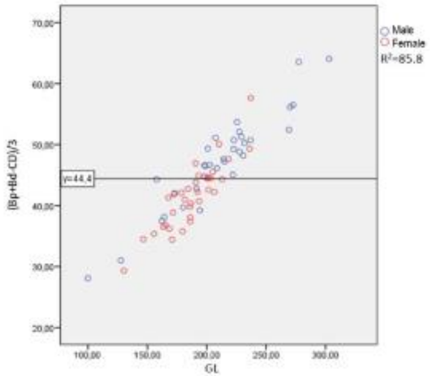
C. Change in coefficient of variation (CV) in femur osteometric measurements.



D. Simple scatter plot of Bp versus Bd. Straight line Bp (mean) = 43.62



E. Simple scatter plot of Bd versus Bp. Straight line Bd (mean) = 36.65



F. Simple scatter plot of (Bp+Bd+CD)/3 versus GL. Straight line (Bp+Bd+CD)/3 (mean)= 44.4

Figure 1. A. The osteometric measurements of femur (General), B. Graphical distribution of SD versus GL. Straight line SD (mean) = 15.51, C. Change in coefficient of variation (CV) in femur osteometric measurements. D. Simple scatter plot of Bp versus Bd. Straight line Bp (mean) = 43.62, E. Simple scatter plot of Bd versus Bp. Straight line Bd (mean) = 36.65, F. Simple scatter plot of (Bp+Bd+CD)/3 versus GL. Straight line (Bp+Bd+CD)/3 (mean)= 44.4

Discussion

The degree of sexual size dimorphism is a factor that varies widely between breeds, whether domesticated (6, 7) or wild species (5). In particular, the fact that dogs have different sizes and morphological structures, from Yorkshire Terrier to Doberman, brought with it intraspecific polyformism (12), which resulted in low interest in the evaluation of sexual dimorphism (13) as dogs have different sizes and visual morphology, they also show different skull typology distribution (16, 20, 21).

From the appendicular skeleton, sexual dimorphism has only been evaluated from the humerus bone of dogs so far, and it has been observed that this has 85% of estimation rate (15). In this study, the role of the femur, which is a bone that is widely used in determining gender in terms of humans (8-11), was investigated on sex determination in dogs with wide intraspecific polyformism (12).

There are many factors (genetics, race, species, etc.) that affect visual morphology and other important factor is sexual dimorphism, which reveals the size difference between male and female individuals (22). This reveals a noticeable difference in height and weight between males and females in many mammals (19). This contributes to the estimation of visual morphology over bones and also to reveal the existence of sexual dimorphism. It has been reported that a comparison between body sizes between males and females is possible with the contribution of osteometric comparisons in determining the sex of dog species (14). Therefore, morphological (15) and osteometric (23, 24) methods are thought to be beneficial for animals. Similar to the widespread use of estimations based on morphometric data in humans (8-11), it has been tried to focus on how it should be used in animals, especially in dogs. The use of dog breeds of different sizes in this study, which are considered dolichocephalic (16), was due to the fact that sexual dimorphism is a factor affecting visual morphology (22). In the population including 22 breed and crossbred dogs, the differences between femur osteometric measurements of female and male individuals were significant at the $P<0.01$ level. This showed the change in osteometric measurements related to gender. The values of male individuals had a higher value than females especially in GL, Bp, DC, SD, CD and Bd values consecutively ; with the mean of 210, 96, 46.09, 23.11, 16.70, 56.86, 38.99 mm in males and 187.43, 41.22, 20.40, 16.36, 49.14, 34.37 mm in females.

Femur GL and diaphysis measurements (SD and CD) are widely used in making predictions such as shoulder height from visual morphological characteristics of dogs (25), body weight (25-27) and thinness index (25). The thought of sexual dimorphism affects visual morphology (22) supports the idea that body sizes differ in both two genders in many animal species. Males in larger species seem to be larger than females and males in smaller species (28), for instance male whippets, irish wolfhounds and standard poodles were found all significantly taller than their female individuals in Sutter et. al's (7) study. In the light of these knowledge the variation observed in the GL value in this study was also due to the presence of dogs in different sizes and GL mean value of both males and females may have been a key point to understand sexual dimorphism (7).

SD and CD values were evaluated together with the GL value, the sexual dimorphism on these values became clearer. The thinness index is an important indicator of the strength of a particular item (23, 25). Compared with the height of the withers, it provides a valuable measure of the structural feature of the legs of the animal in question, which correlates the robustness of the members with the relative weight of the animal, allowing it to be interpreted as stocky or slender (19). In males, the highest variation was found in the GL value, and the lowest variation was in the Bd value. The variation in GL and CD values was higher than the other parameters, indicating that dogs with different structural characteristics were used in the study. The GL value showed large-heavy male dogs, but when evaluated together with the SD value (high value), this definition could be made correctly. Otherwise, tall, slender-legged male dogs could also have a high GL value. Taken together, the graphical distribution of the SD value versus the GL value, supported a sexual dimorphism above the average with 15.51 mm.

Considering the coefficient of variation in femur osteometric measurements in both male and female dogs, it is seen that Bp and Bd values constitute the most reasonable variation and the most homogeneous distribution was seen in Bp and Bd values in terms of sexual dimorphism. When the distribution of Bp value within the overall average is compared to the Bd value, it was observed that females were more clustered below the overall average (Bp=43.62 mm) value, while males were more concentrated above this line. The ones observed below and above the mean line were likely due to the use of different phenotypic dogs in the study. Contrary to this evaluation, when the distribution of Bd value within the general average is compared to the Bp value, a similar situation to that of Bp was seen. It was observed that females clustered below the general mean line (Bd=36.65 mm), while males were over it.

These findings resembles the findings in Belahouse et al's study that showed a relative strong female-biased dimorphism while for the other femoral variables, except length, the dimorphism is male-biased (13).

The scatter graph of (Bp+Bd+CD)/3 value, to which the GL and CD (25-27, 29) values prominent in the prediction of visual morphology were added as addition to the values mentioned above, showed a difference in the distribution density of males and females with R^2= 85.8 a predictive coefficient.

It was also possible to see some extremes in the scatterplots. These were the presence of females in the agglomeration of males or the presence of males in the heap of females. This was due to the presence of dogs with different morphological characteristics, because the morphology of the the bones were affected by size and body weight (28, 30, 31). It was normal for a female St. Bernard and a female Cocker Spaniel to differ slightly in distribution as the predictive rate (R^2) had a value of 85.8%.

On the other hand, there were negative correlations between Bp*100/GL and Bd*100/GL indices and osteometric measurements, which was salient. There was a negative correlation between SD*100/GL index and GL and DC values. Although it had a low correlation coefficient, the increase in bone length had an effect on decreasing this index. A thinness index value with the appearance of long slender legs was emerging. This was probably the case for female individuals. Because the existence of a positive correlation of this index (SD*100/GL) with CD caused the index to increase as body weight increased. This is seen as an effect of the heavy and large-bodied appearance of male individuals (28, 30, 31). The statistical significance of the difference between the CD values of female and male individuals and the large difference observed between them are thought to be another factor in the large SD*100/GL index.

Although the effect of human choice and selection on the emergence of size differences between species was mentioned, according to the present study the argument of this effect was not very valid in determining sex in archaeological remaints by osteometric method (15) was a controversial subject. It is believed that the emergence of dog types with different sizes and morphological structures does not occur on a single gender, but covers both genders. Gender identification through bone morphology in dogs has so far been rather limited (11). Studies have generally focused on os coxae and sacrum (22, 32-34). Although it is claimed that the osteometric method is inadequate in determining gender, the equations created by pelvis morphometry clearly reveal the existence of sexual dimorphism (22, 35). The femoral osteometric measurements used in this study also clearly show the dimorphism between males and females. Obtained osteometric values are directly more decisive parameter than femur index values. The high positive correlations between osteometric values can also explain the higher size increase in males compared to females. An increase in one of the femoral parameters causes the other to increase as well, thus reflecting on the visual morphology. Although the index values of the femur do not have a determining effect on sexual dimorphism, it can provide a clear explanation of what kind of structural feature the limbs have. It is thought that the increase in the osteometric measurements has a negative effect on the indices, due to the fact that the increase in Bp, SD and Bd values is not at the desired level or in the negative direction compared to the rate of increase in the GL value.

On conclusion, although femur osteometric measurements of dogs in a wide morphological range do not provide us with 100% sexual discrimination, they will contribute greatly to the evaluations on this subject with a predictive coefficient of approximately 85%. The measurements in femur could correlate the most with sexual dimorphism in pelvic bones, both skeletal structures being involved in the movements of the hip joint like as pelvic dimensions especially depending on the individual’s breed. (13, 31). Because it is well known that largest breeds show a highly male-biased sexaul dimorphism. And femur may be one of the indicator of sexual dimorphism genetically (30, 31) and measurements of it could be applied to discriminate a male from a female canine in forensic investigations (31).

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