

RESEARCH ARTICLE

RADIOMETRIC AND GEOMETRIC MORPHOMETRIC ANALYSIS OF THE CARPAL JOINT AREA IN 2-YEAR-OLD THOROUGHBRED HORSES

Ozan Gündemir^{1*}, Nedžad Hadžiomerović², Gülsün Pazvant¹, Dilek Olğun Erdikmen³

¹Department of Anatomy, Faculty of Veterinary Medicine, Istanbul University-Cerrahpasa, Istanbul, Turkey

²Department of Anatomy and Histology with Embryology, Veterinary Faculty, University of Sarajevo, Bosnia and Herzegovina

³Department of Surgery, Faculty of Veterinary Medicine, Istanbul University-Cerrahpasa, Istanbul, Turkey

***Corresponding author:**

Dr. sc. Ozan Gündemir

Department of Anatomy, Faculty of Veterinary Medicine, Istanbul University-Cerrahpasa, Istanbul, Turkey

Phone: +90 539 661 04 44

ORCID: 0000-0002-3637-8166

Email: ozan_gundemir@hotmail.com

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ABSTRACT

This study aimed to examine radiographs of Thoroughbred horse and compare radiometric and geometrical features between male and female individuals. To this end, dorso-palmar images of the carpal joint of 34 (17 female, 17 male) clinically healthy two-year-old Thoroughbred horses were analyzed. In total, 15 lengths and two angles were analyzed radiometrically, while geometric morphometrics was applied on 20 landmarks. In 7 radiometric measurements, the difference between female and male individuals was found to be statistically significant. All length measurements of males were found to be higher than of females. The statistically most important difference between the sexes was the measurement of the greatest breadth of proximal end of basis metacarpalia II-III-IV ($p < 0.001$). There was no difference between the sexes for angle values. It was seen that the geometric morphometric analysis made with the Landmarks used in the study was not determinative between males and females. Thanks to the developing technology, the number of three-dimensional modeling technique and radiometric measurement studies is also increasing. In this study, the differences between the sexes were revealed using radiographs of horses of the same age, and obtained values will be the references for future radiometric and clinical studies.

Keywords: Equidae, principal components, radiographs, sex determination

INTRODUCTION

The carpal joint is a compound joint located in the front leg. It includes four different levels of articulation: *articulatio antebrachiocarpea*, *articulationes intercarpeae*, *articulatio mediocarpea* and *articulationes carpometacarpeae* (Konig and Liebich, 2004). This region is significant for lameness in horses (Moore and Schneider, 1995). In addition to physical examinations for the purpose of the diagnosis of lameness in the area, imaging systems have helped to detect abnormalities of the area (Nagy and Dyson, 2012). In recent years, the advantages of imaging systems have been used to detect anatomical shape differences or differences between genders. Studies in humans have also tried to reveal the differences between the sexes as a result of radiometric measurements of bones (Hudson et al., 2016). Radiometric measurements were also taken on canine teeth, and the difference between females and males in humans was revealed (Foad et al., 2012). In animals, imaging systems are generally used in modeling studies. Samples were taken from gazelles, and models were created electronically with these samples (Demircioglu et al., 2020). In addition, modeling was performed using the imaging systems in dog, cat and New Zealand rabbit, and it was possible to take the measurements from that (Atalar et al., 2017; Özkadif et al., 2014; Yılmaz et al., 2020). Geometric analysis is a type of morphometric analysis performed on a 2 or 3-dimensional plane, revealing the shape differences rather than the size differences of the samples (Corruccini, 1987). Two-dimensional photos can be obtained using x-rays, and 3-dimensional images can be obtained using computed tomography. Landmarks are used on these images and then the positions of those Landmarks relative to each other are evaluated in the x-y coordinate system (Zelditch et al., 2012). In

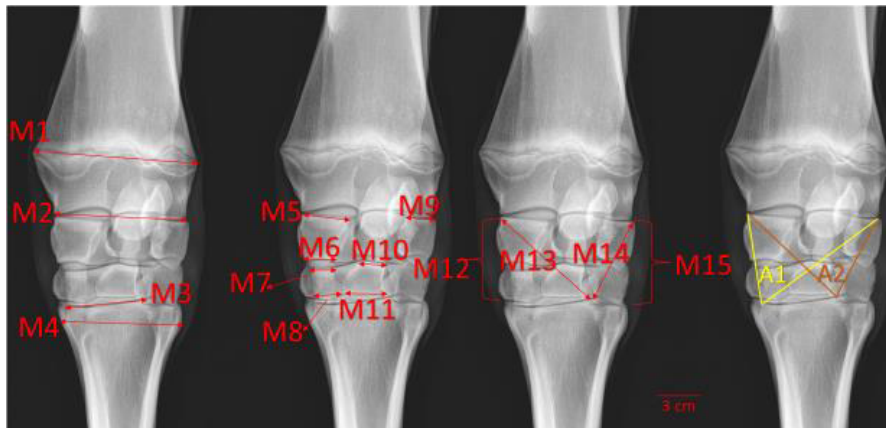
the veterinary field, there are cross-species studies using geometric morphometric analysis (Gürbüz et al., 2020; Haruda, 2017). In addition, in the study using photographs, the differences between the sex were tried to be revealed geometrically (Gündemir et al., 2020).

In addition to clinical research, using radiometric measurements of the carpal area in healthy animals the differences between species and sex can be revealed. Oheida et al. (2019) tried to statistically reveal the differences between the two horse species by taking different radiographic parameters from the carpal joint in Thoroughbred and Standardbred racehorses. This study reported that differences between the two species were statistically significant in 4 parameters. However, studies on this subject are limited in the reference information. This study aims to examine whether there is a sex difference in Thoroughbred horses by taking radiometric measurements of the carpal region. In addition, by using the dorso-palmar image of the carpal joint examined was whether there was a difference in shape as a result of geometric analysis.

MATERIAL AND METHODS

Animals and Radiographs

In the study, 34 (17 females, 17 males) healthy 2-year-old Thoroughbred horses were used. Radiographs were taken from horses of Jockey Club of Turkey. The analysis was applied on dorso-palmar image of the left carpal joint. No pathological condition was observed in the images. Beside the carpal joint bone, the distal end of the radius and the proximal end of the metapodium bones were also visualized. Gierth X-ray was used to capture the images. The study was approved by the Local Ethics Committee of Faculty

**Figure 1**

Dorso-palmar radiographs of the left carpus (Carpal measurements)

of Veterinary Medicine, İstanbul University-Cerrahpaşa (2019/44).

Radiometric measurement

The measurements of the parameters were taken using the Radiant DICOM Viewer (version 2020.2.2). In total, 15 lengths and 2 angle values were evaluated. Centimeter was used as the measurement unit. Measurement points taken from the image (Figure 1):

- M1:** Greatest breadth of distal end of the radius
- M2:** Greatest breadth of the facies articularis carpea
- M3:** Breadth of facies articularis of metacarpalis III
- M4:** Greatest breadth of proximal end of basis metacarpalia II-III-IV
- M5:** Length of facies articularis of radial carpal bone (proximal)
- M6:** Length of facies articularis of radial carpal bone (distal)
- M7:** Length of facies articularis of second carpal bone (proximal)
- M8:** Length of facies articularis of second carpal bone (distal)
- M9:** Length of ulnar carpal bone (proximal)
- M10:** Length of facies articularis of third carpal bone (proximal)

M11: Length of facies articularis of third carpal bone (distal)

M12: Medial carpal height (from the most proximal level to the most distal level of the carpus)

M13: Distance between the most medial point of facies articularis carpea of radius to most lateral point of facies articularis of metacarpalis III

M14: Distance between the most lateral point of facies articularis carpea of radius to most lateral point of facies articularis of metacarpalis III

M15: Lateral carpal height (from the most proximal level to the most distal level of the carpus)

A1: The angle between a line drawn from medial point of facies articularis carpea of radius to most medial point of facies articularis of metacarpalis III, and from that point to most lateral point of facies articularis carpea of radius

A2: The angle between a line drawn from medial point of facies articularis carpea of radius to most lateral point of facies articularis of metacarpalis III, and from that point to most lateral point of facies articularis carpea of radius

Geometric Morphometric Analysis

For geometric morphometric analysis, radiographs of images of 34 animals were first converted into the picture files (png) and then into “tps” files by means of the tpsUtil (Version 1.74) program. Then, as shown in Figure 2, 20 Landmarks were placed using the tpsDig (Version 2) program. The obtained files were converted into “text” files. Utilizing the morphoJ (Version 1.07a) program, the differences between male and female are graphically shown in Figure 2.

Statistical analysis

SPSS 22 program was used for statistical analysis. ANOVA was performed to test the statistical difference between the parameters. The homogeneity of variance between the groups is shown in mean value and standard deviation, as displayed in the Table. Mean value and standard deviations were determined and shown in the Table. In order to reveal the statistical differences, p values for each parameter were examined for male and female individuals. Past (Version 2.17c) was used for geometric morphometric analysis. Principal component (PC) analysis was performed. Each PC variation value between female and male was examined.

RESULTS

The average values and standard deviations of 15 parameters are given in Table 1. It was observed that the variances between the groups were homogeneous. According to the data obtained, all male values in terms of length were found to be higher than of females. While there was no statistically significant difference in the measurement of the greatest breadth of distal end of the radius between male and female, the measurement of the greatest breadth of proximal end of basis metacarpalia II-III-IV was statistically significant between the sexes ($p < 0.001$). The difference between the greatest breadth of the facies articularis carpea was also statistically significant ($p < 0.05$).

Seven measurements (M5, M6, M7, M8, M9, M10 and M11) were taken of the carpal bones. In these measurement values, a significant difference between female and male individuals was found in the measurement of the length of facies articularis of radial carpal bone (distal) and the measurement of the length of facies articularis of second carpal bone (distal) ($p < 0.05$). Four measurements were taken for the total length of the carpal joint (M12, M13, M14 and M15). Among these measurements, the measurements of medial carpal height (M12) and the measurements from the most lateral point of facies articularis carpea of radius to the most lateral point of facies articularis of metacarpalis III (14) were found to be statistically different between genders ($p < 0.01$).

Table 1 Means, standard deviations and p values of measurement points

Measurement	Sex	N	Mean (cm)	SD	Minimum (cm)	Maximum (cm)	P Value
M1	Female	17	9.90	0.47	9.01	10.52	.151
	Male	17	10.16	0.55	9.22	10.90	
M2	Female	17	7.93	0.31	7.41	8.42	.026
	Male	17	8.22	0.41	7.48	8.87	
M3	Female	17	4.74	0.25	4.30	5.17	.045
	Male	17	4.91	0.25	4.34	5.29	
M4	Female	17	7.46	0.33	6.96	8.17	.000
	Male	17	7.92	0.34	7.25	8.74	
M5	Female	17	2.80	0.16	2.59	3.21	.194
	Male	17	2.90	0.27	2.36	3.41	
M6	Female	17	1.76	0.20	1.33	2.03	.023
	Male	17	1.92	0.18	1.49	2.19	
M7	Female	17	1.51	0.19	1.22	1.86	.203
	Male	17	1.61	0.27	.99	2.01	
M8	Female	17	1.38	0.21	1.07	1.76	.014
	Male	17	1.59	0.26	1.05	2.02	
M9	Female	17	1.57	0.15	1.28	1.76	.088
	Male	17	1.66	0.12	1.45	1.85	
M10	Female	17	1.42	0.17	1.06	1.69	.687
	Male	17	1.45	0.24	.91	1.73	
M11	Female	17	2.38	0.13	2.14	2.61	.084
	Male	17	2.48	0.19	2.06	2.79	
M12	Female	17	5.95	0.29	5.47	6.36	.001
	Male	17	6.31	0.29	5.84	6.77	
M13	Female	17	7.62	0.35	7.05	8.26	.051
	Male	17	7.89	0.42	7.24	8.69	
M14	Female	17	5.64	0.33	5.10	6.24	.009
	Male	17	5.97	0.37	5.25	6.57	
M15	Female	17	5.52	0.29	5.02	6.04	.287
	Male	17	5.65	0.40	4.75	6.26	
A1	Female	17	58.99	1.55	56.6	62.2	.103
	Male	17	58.13	1.42	55.2	61.2	
A2	Female	17	70.84	2.07	67.2	74.0	.941
	Male	17	70.89	2.04	67.8	74.7	

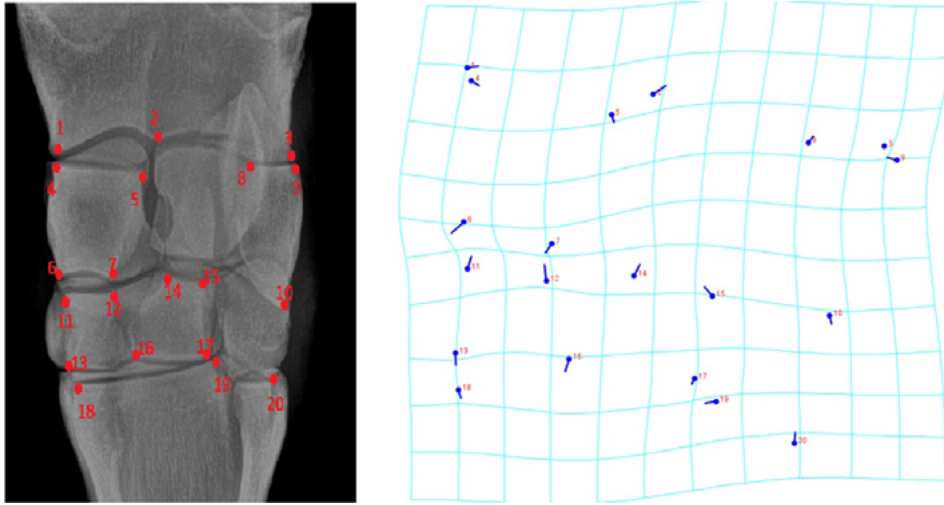


Figure 2 Geometric morphometric analysis. Left picture: locations of Landmarks used. Right picture: Amount and direction of change between male and female markers (morphoj) (the dots represent the female. The extensions show how different the male individuals are and in which direction this difference is)

Twenty Landmarks were used and 33 principal components were obtained. It was observed that PC1, which had the highest value, explained 28.44% of the total variance. The distinction between males and females was not obtained in geometric morphometric analysis. The p value was statistically insignificant in the discriminant analysis between the sexes (P value: 0.9279). Figure 2 shows differentiation between female and male individuals and the direction of that differentiation. In the distal Landmarks of the radial carpal bone and the Landmarks applied to the second carpal bone and the third carpal bone more changes have been identified than in the other regions (LM6, LM7, LM11, LM12, LM13, LM14, LM15, LM16). It was observed that the joint space between this region was narrower in shape in male individuals (LM6, LM7, LM11, LM12). Also, there were differences between male and female in the direction of change of the other regions.

DISCUSSION AND CONCLUSION

In the study, sex determination was made using radiographs of 34 horses. Radiometric measurements and geometric morphometric analysis measurements were applied on the images. The values of geometric morphometric analysis using x-ray images were found to be statistically insignificant for sex differentiation. However, the shape changes between male and female could be interpreted by showing them through Landmarks. In radiometric measurements, seven measurements were found to be statistically significant between females and males. The most significant difference between the sexes was the greatest breadth of proximal end of basis of metacarpalia II-III-IV ($p < 0.001$). There was no difference between sexes for angle values.

The alignment of the bones in the carpal joint area and their related angles are particularly

important in racehorses. The direct effects of the movements of those bones on adjacent tissues were examined and statistically significant changes were identified (Olusa et al., 2020). Radiometric studies that identify the differences of this region between the species are quite limited. Oheida et al. (2019) tried to reveal the differences between Thoroughbred horses and Standardbred horses using radio-palmar images of this region. In this study, the angles were used and they said that the angle differences between the two breeds were also statistically significant. In this study, different length measurements were used, and the differences between male and female were determined with carpal radiometric images of the same species. The greatest breadth of proximal end of basis of metacarpalia II-III-IV measurement was found to be the most determinant measurement point in terms of sex in the carpal radiometric picture ($p < 0.001$).

In veterinary medicine, photographs are taken of the bones and geometric analyzes are made, and in this way, the differences between species and sexes have been tried to be determined (Haruda, 2017; Gündemir et al., 2020; Gürbüz et al., 2020). In this study, on the other hand, geometric analysis was performed on x-ray images to determine the differences between genders. We think that geometric analysis made from X-ray images is more advantageous. More material can be obtained with X-ray images. It is also possible to

measure from live animals and apply geometric analysis. Also, thanks to the x-ray images, samples taken from live animals can be repeated, and the differences in change over time can be determined geometrically with this method.

It has also been reported that the carpal conformation varies between different age groups (McIlwraith et al., 2003). It was thought that the different ages of the horses used for gender studies could directly affect the results. For this reason, the present study was conveyed on individuals of the same age group.

Various studies on traditional measurement techniques (osteometric, morphometric, etc.) used in veterinary anatomy and sex and race discrimination were carried out. However, thanks to the developing technology, the number of three-dimensional modeling technique and radiometric measurement studies is also increasing. In this study, the differences between the sexes were tried to be revealed by using radiographs of horses of the same age. Reference values were obtained. These values are thought to be a reference for future radiometric and clinical studies.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

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RADIOMETRIJSKA I GEOMETRIJSKA MORFOMETRIJSKA ANALIZA PODRUČJA KARPALNOG ZGLOBA KOD DVOGODIŠNJIH PUNOKRVNJAKA

SAŽETAK

Cilj ovog istraživanja je pregledati rendgenske snimke punokrvnih konja i usporediti radiometrijske i geometrijske karakteristike između mužjaka i ženki. U ovu svrhu su analizirane dorzopalmarne snimke karpalnih zglobova 34 klinički zdrava dvogodišnja punokrvna konja (17 ženki, 17 mužjaka). Radiometrijski je analizirano ukupno 15 dužina i dva ugla, dok je za geometrijsku morfometriju korišteno 20 tačaka. Kod 7 radiometrijskih mjerenja je uočena statistički signifikantna razlika između ženki i mužjaka. Sva mjerenja dužine kod mužjaka su pokazala veću dužinu u odnosu na ženke. Statistički najvažnija razlika među spolovima je bila u mjerenju najveće širine proksimalnog kraja baze metakarpalnih kostiju II-III-IV ($p < 0.001$). Nije bilo razlike među spolovima za vrijednosti uglova. Uočeno je da geometrijska morfometrijska analiza sa tačkama korištenim u istraživanju nije bila određujuća za mužjake i ženke. Zahvaljući razvoju tehnologije, broj studija o trodimenzionalnim modelirajućim tehnikama i radiometrijskim mjerenjima se povećava. U ovom istraživanju, razlike između spolova su određene korištenjem rendgenskih snimaka konja iste starosti, a dobivene vrijednosti će se koristiti kao referentne za buduće radiometrijske i kliničke studije.

Ključne riječi: Ekvide, glavne komponente, određivanje spola, rendgenski snimci