



RESEARCH ARTICLE

Geometric Morphometric Analysis of the Condylus Occipitalis and Foramen Magnum in Sheep and Goat

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Koyun ve Keçilerde Condylus occipitalis ve Foramen Magnum'un Geometrik Morfometrik Analizi

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Öz

Amaç: Yapılan çalışmada, foramen magnum ve condylus occipitalis' in türler arasındaki şekil analizi yapılarak dimorfik yapılarının ortaya konulması ve koyun-keçi arasındaki değişkenliklerinin belirlenmesi amaçlandı.

Gereç ve Yöntem: Çalışmada toplam 81 (46 koyun, 35 keçi) kafatasından alınan veriler kullanıldı. Foramen magnum'un çerçeve şeklini belirlemek ve condylus occipitalis varyasyonunu anlamak için tip I (anatomik) ve tip III (semilandmark) landmarklardan faydalanıldı.

Bulgular: Buna göre toplam şekil varyasyonunun PC1, PC2 ve PC3'ün sırasıyla %30.76, 14.94 ve 14.07'sini açıkladığı, PC1'e göre şekil varyasyonundan birincil derecede condylus occipitalis'i, ikincil derecede ise foramen magnum'un sorumlu olduğu belirlendi. PC2'ye göre sağ condylus occipitalis şekil varyasyonunu büyük oranda açıkladığı, PC3'te ise sağ condylus occipitalis'in tüm, sol condylus occipitalis'in en lateral köşesi ile foramen magnum'un sol kenarı şekil varyasyonunun açıklanmasına neden olduğu belirlendi. Diskriminant fonksiyon analizi sonucunda Procrustes ve Mahalanobis distance sırasıyla 0.12293879 (p<0.0001) ve 67.7482 (p<0.0044) olarak tespit edildi.

Öneri: Sonuç olarak geometrik morfometri yöntemi, türler arası kafatası şeklindeki farklılıkları tespit etmek için kullanılabilir bir araç olduğu ve bu nedenle taksonomik, arkeolojik ve adli amaçlar için başarıyla kullanılacağı düşünülmektedir.

Anahtar kelimeler: Craniometri, Geometrik morfometri, Şekil analizi, Temel bileşenler analizi

Abstract

Aim: The aim of this study was to reveal the dimorphic structures of the foramen magnum and condyle occipitalis through an interspecies shape analysis and to determine the variability between sheep and goats.

Materials and Methods: The study includes data from 81 skulls (46 sheep and 35 goat) for this aim. The foramen magnum frame shape and the condyle occipitalis variation were determined using type I (anatomical) and type III (semilandmarks).

Results: Accordingly, was determined 30.76, 14.94 and 14.07 of the total shape variation of PC1, PC2 and PC3, respectively. It was determined that condylus occipitalis was primarily responsible for the shape variation according to PC1, and foramen magnum was responsible for the secondarily. It was found to explain the shape variation of the right condyle occipitalis to a great extent compared with PC2, while in PC3, it caused the entire right condyle occipitalis to explain the shape variation of the extreme lateral corner of the left condyle occipitalis and the left edge of the foramen magnum. The discriminant function analysis determined the Procrustes and Mahalanobis distances to be 0.12293879 (p < 0.0001) and 67.7482 (p < 0.0044), respectively.

Conclusion: As a result, the geometric morphometry method is regarded to be a useful tool for detecting changes in skull shape between species and can thus be used successfully for taxonomic, archaeological, and forensic research.

Keywords: Craniometry, Geometric morphometry, Principal component analysis, Shape analysis.



Introduction

In classifying species that are taxonomically close to each other, differences in the skeletal system are used as a reference. The intraspecific and interspecific data revealed by using these differences are of great importance not only to the science of taxonomy, but also to the archeological and forensic sciences (Tecirlioğlu 1983). In the skeletal system, the bones that are the most commonly used to distinguish sex, species, and race are the skull and pelvis (Scheuer 2002, Bärman et al 2013). Species identification based on skull morphology is very difficult because it shows high intraspecific diversity (Bärman et al 2013). Classical morphometry alone is also usually insufficient for differentiation. For this reason, geometric morphometry has been increasingly preferred in recent years (Bernal 2007, Aytek 2017, Demircioğlu et al 2021).

Geometric morphometry is a method that determines the shape differences of objects based on landmark coordinates (LM) and indicates the degree of shape change (Viscosi and Cardini 2011, Zelditch et al 2012). By analysing the orientation of the coordinates of LMs identified on the Cartesian coordinate plane, the intra-group and inter-group differences and similarities of the revealed structure are revealed. LMs are identified as points common to all samples and located in the same positions (Slice 2007, Bigoni et al 2010). They are divided into three types based on their anatomical location. Type I LMs are the group that is the most suitable for geometric morphometry and easiest to replicate. They are points with positions and definitions that are clear and easy to identify. Type I LMs are the group best suited for geometric morphometry and are the easiest to replicate. They are points whose positions and definitions are clear and easy to identify. Type II LMs are points positioned at the most extreme or distinct parts of anatomical structures (e.g., columns and appendages). Type III LMs (semi-landmarks) are points placed on the base of other LMs (Aytek 2017). The method of geometric morphometry, which is applied in many fields, has been intensively studied for some time, especially in connection with sheep and goat breeds, which show a high intraspecific polymorphism (Parés Casanova 2014, Parés Casanova and Bravi 2014, Demircioğlu et al 2021, Gündemir et al 2023, Yaprak et al 2023).

The occipital bone is one of the bones of the neurocranium that shape the caudal part of the cranium. It consists of the basilar part, the two lateral parts and the squamous part. At the junction of these three parts, there is the foramen magnum (FM) that constitutes the transition between the cavum cranii and canalis vertebralis. The occipital condyles is articulated with the atlas, which is found in the lateral partes region of the occipital bone. It also marks the lateral boundaries of FM (Bahadır and Yıldız 2008, Demiraslan and Dayan 2021). The size and shape of FM and the occipital

condyles show dimorphism based on sex and breed and provide information about cranio-vertebral biomechanics (Murshed et al 2003, Naderi et al 2005, El-Barrany et al 2016).

The aim of this study was to perform a morphological analysis of FM and the occipital condyle between species, to visualise dimorphic structures, and to identify variations between two species.

Material and Methods

Material

Research Samples

Data obtained from the crania of a total of 81 animals (46 sheep and 35 goats) were used. The materials were samples that were being used for educational purposes at the laboratories of the Anatomy Departments of the Veterinary Medicine Faculties at Harran University, Burdur University, and Bingol University. Therefore, no animals needed to be euthanized for the study. There was no pathology in the samples. In addition to this issue, it was ensured that the laboratory records of the included samples did not have conditions (e.g., orthopedic or neurological conditions) that could affect the results. Based on dental examinations, all samples came from adult animals. While the analyses in this study were carried out only based on the species factor, the breed and sex information of the samples is presented in Table 1.

Table 1. Distributions of the samples according to breed and sex, n

Material	Sheep		Goat	
	Akkaraman	Morkaraman	Kıl	Honamlı
Female	10	14	10	9
Male	10	12	9	7

Methods

Photography and digitization

For 2D analyses, the samples were photographed (Canon 650D) from a 30 cm distance with a focus on the center of FM. Care was taken to ensure that the transverse axis of FM and the lens of the camera were in parallel with each other. The photographs were saved on a computer as JPG files. Type I (anatomical) and type III (semi-landmark) LMs were utilized to determine the outline of the shape of FM and understand the variations of the occipital condyles. For this, first of all, tps file was created in the tpsUtil (version 1.79) program (Rohlf 2019). On this file, using the tpsDIG2 (version 2.31) (Rohlf 2018) program, 40 LMs in total (9 type I, 31 type III) were marked (Figure 1). In this process, the x and y Cartesian



coordinates of the LMs, which are the most fundamental requirements for measuring morphological variations, were identified. LM1 represented the dorsal median point of FM (superimposed with LM32 in the figures), LM33 represented the dorsomedial corner of the left occipital condyle, LM34 represented the lateralmost corner of the left occipital condyle, LM35 represented the ventromedial corner of the left occipital condyle, and LM36 represented the medial junction point of the dorsal and ventral articular surface parts of the left occipital condyle. LM37, LM38, LM39, and LM40 respectively corresponded to the LMs on the right side contralateral to the ones on the left.

To determine morphological differences, a generalized Procrustes analysis (GPA) of the coordinate values of the LMs that were marked in the study was carried out. This way, by eliminating differences in the photographs such as those in size, position, and direction, (Aytek 2017) Procrustes coordinates were obtained. Using these new values, to reduce dimensionality and demonstrate the variations in the principal components, a principal component analysis (PCA) was conducted (Zelditch et al 2012, Villalobos-Leiva and Benítez 2020).

The LMs around which morphological differences were gathered, the presence of an allometric effect (multivariate regression on Procrustes coordinates), and the clustering characteristics of the samples (Discriminant Function Analysis-DFA) were analyzed. All these analyses were performed using the MorphoJ program (Klingenberg 2011).

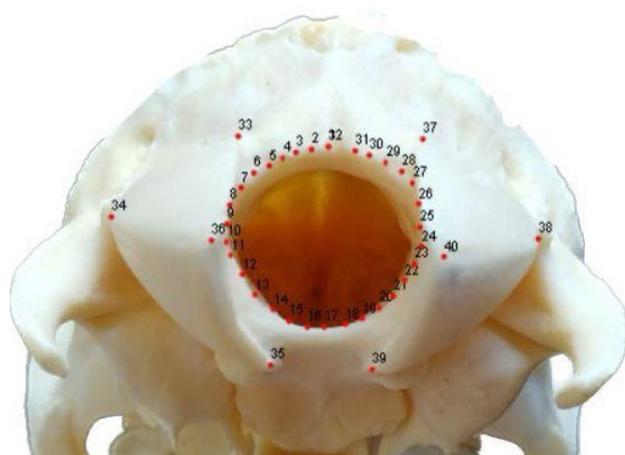


Figure 1. Landmarks

Results

In this study, a small allometric effect (2.6%) of the centroid side on the data was identified. Despite this, the allometric effect was significant in the 10000-round permutation test ($p=0.0283$). Based on the results of the regression analysis conducted to determine the effects of the allometry on the

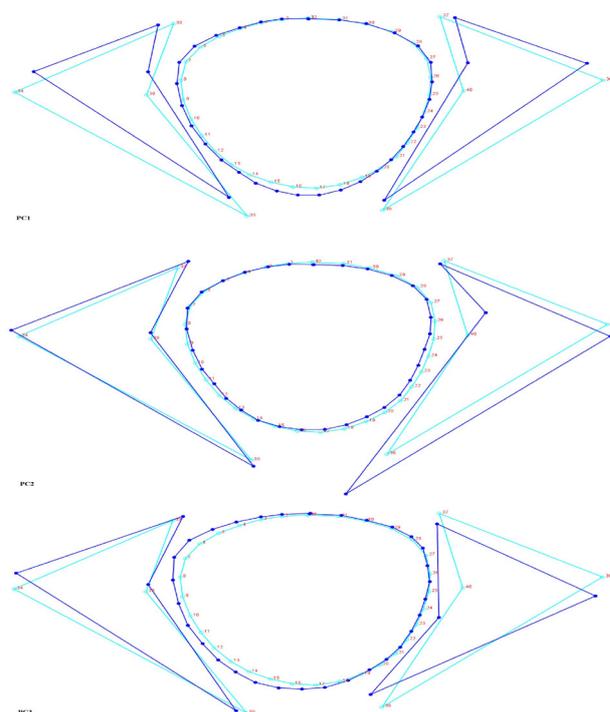


Figure 2. Wireframe morphological change plots according to PC1 (30.76%), PC2 (14.94%), and PC3 (14.07%)

principal components, 9.64% of the morphology according to PC1 ($p=0.005$) and 0.40% of it according to

PC2 ($p=0.569$) was estimable by dimension. Accordingly, it was seen that in the comparisons of the individuals based on the species factor, morphological variations were dimension-independent.

In the PCA, 76 PCs were calculated. It was determined that PC1, PC2, and PC3 explained the total variance in morphology by 30.76%, 14.94%, and 14.07%, respectively. According to PC1, the occipital condyle was the primary factor for the variation in morphology, whereas the upper-left corner and ventromedial side of FM were the secondary factors (Figure 2). According to PC2, the right occipital condyle explained the variation in morphology to a large extent (Figure 2). In the case of PC3, the variation in morphology was explained by the entire right occipital condyle, the lateralmost corner of the left occipital condyle, and the left side of FM (Figure 2).

The scatterplot of the individuals that was obtained as a result of the PCA is presented in Figure 3. According to this scatterplot, the individuals were noticeably distinguished from each other. The results of the DFA that was performed to observe the relationship between the groups more clearly showed that the Procrustes and Mahalanobis distances were consecutively 0.12293879 ($p<0.0001$) and 67.74 ($p<0.0044$). According to the cross-validation results, the goats were grouped with 83% accuracy (29:6), while the sheep were grouped with 82% accuracy (37:9) (Figure 4). The results



on the morphological variations between the groups were compatible with the PCA results. The DFA results (Figure 4) revealed larger occipital condyles and more basal localization of the ventral parts of these condyles in reference to FM in the goats compared to the sheep. The goats also had a more dorsal placement of the ventral articular part of the occipital condyle in reference to FM. In the sheep, FM was broader along the ventromedial and left lateral lines in comparison to the goats. The mean morphologies of the regions that were analyzed in the sheep and goats are shown in Figure 5. Based on these results, the ventromedial edge of FM in the sheep was more conical compared to that in the goats. The FM of the goats had an elliptical appearance.

Discussion

In areas where visual morphology can fall short in terms of interspecies classification, morphometry, which reveals the variety and differences of morphologies with metrics, is utilized (Rohlf and Marcus 1993). Classical morphometry alone is also inadequate in terms of the comprehensive analysis of the shapes of structures (Zeder 2005). Although there are different studies in which the cranial morphologies of sheep and goat breeds have been investigated from dorsal, ventral, and lateral directions (Parés Casanova 2014, Parés Casanova and Bravi 2014, Demircioğlu et al 2021, Parés-Casanova and Domènech-Domènech 2021, Yaprak et al 2022, Yaprak et al 2023), morphological analyses carried out

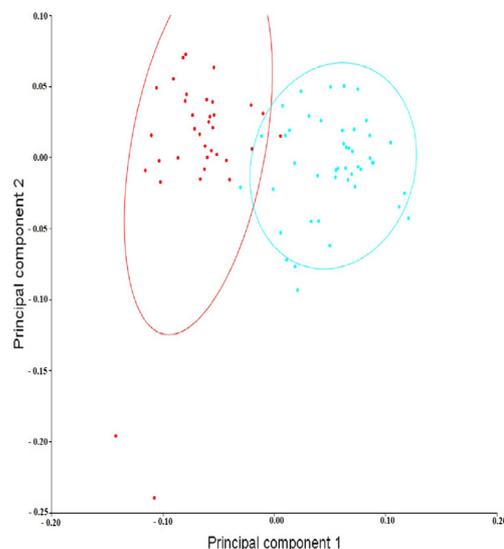


Figure 3. Scatterplot of goats and sheep according to PCA (G: Goat: Red, S: Sheep: Blue)

from the caudal aspect of the cranium are highly limited, and this dearth in the literature constitutes the most significant limitation of this study.

The size and shape allometries of the cranium provide important clues in the revelation of evolutionary and developmental changes (Parés Casanova and Sabaté

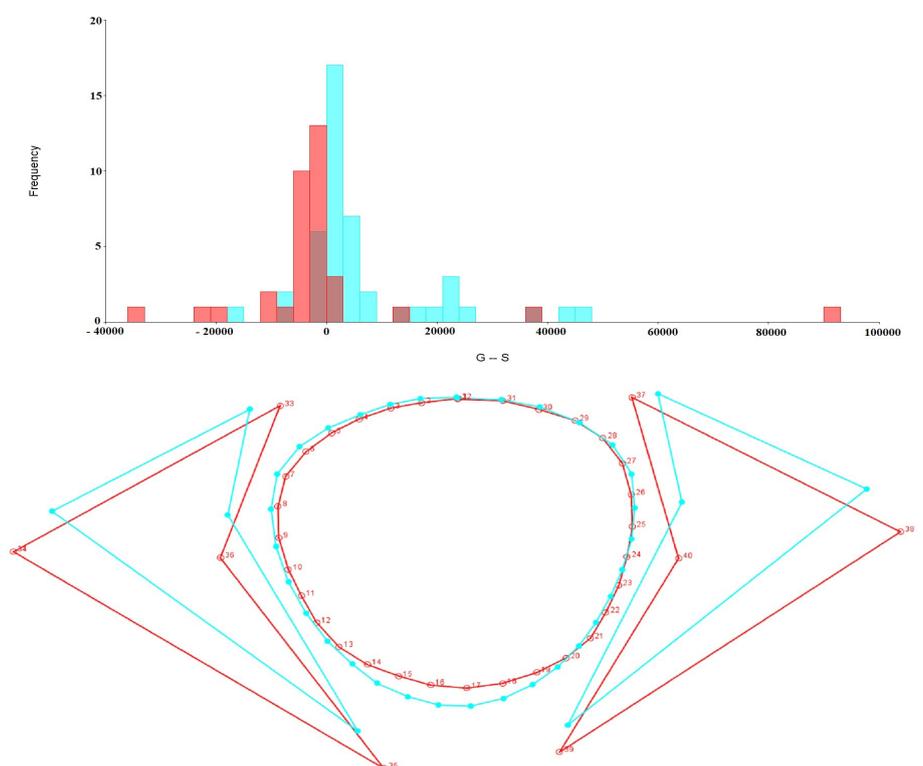


Figure 4. Cross-validation and intergroup morphological variation plots according to DFA (G: Goat: Red, S: Sheep: Blue)

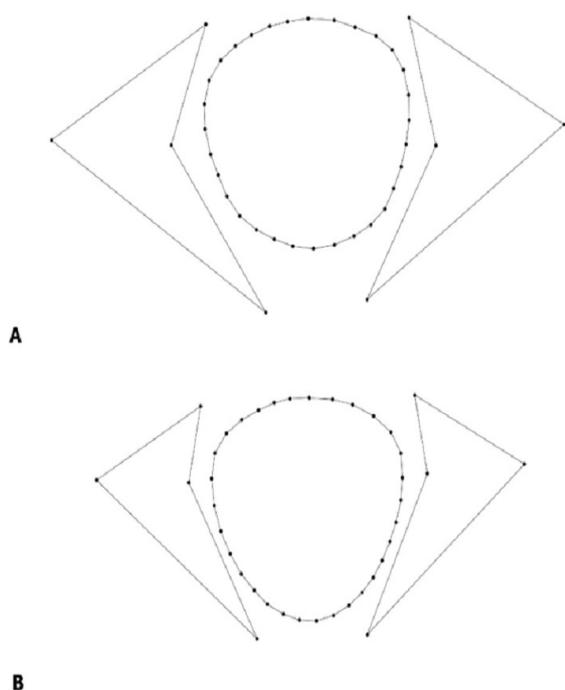


Figure 5. Mean morphologies of regions that were analyzed in sheep and goats (G: Goat, S: Sheep)

2013). It was reported that the Awassi and Hamdani sheep breeds, which have similar conditions in the geographical areas where they are bred, had morphologically different crania, and these intergroup morphological differences are significant (Demircioğlu et al 2022). In another study (Demircioğlu et al 2021) noticeable sexual dimorphism was found from the lateral to the dorsal in the crania of Awassi sheep. In their morphological analyses of the os sphenoidale of domestic sheep and goats, Parés-Casanova and Domènech-Domènech (2021) showed that the two species had morphological differences, and they stated that the first three components in their PCA (PC1, PC2, and PC3) explained these differences at a rate of 71.456%. In this study, the rate of the total variance in morphological differences explained by the first three components in the PCA (PC1, PC2, and PC3) was found as 59.776%. Therefore, it is seen that in the cranial morphology analyses of sheep and goats, the sphenoidal bone shows more allometric variation compared to the occipital condyle and FM.

The occipital bone is the most mobile part of the vertebral column by which the head and neck movements in the craniocervical junction (CCJ) constituted by the atlas and the axis are performed. The rotation, extension, and flexion movements of the cranium are associated with the harmony of the bones constituting this compound structure with each other (White III and Panjabi 1978, Bellabarba et al 2006). Goats usually graze at rockfaces and highlands, while sheep graze in tablelands and foothills. The chins of sheep stay close to the ground during grazing, and they are suitable for grazing close to the soil. On the other hand, when they can

stand on their hind limbs, goats can feed on sprouts, buds, and leaves that are found on trees in higher areas (Shackleton and Shank 1984, Altın 2005, Garip 2013). According to the DFA results of our study, it was seen that compared to the sheep, the goats had larger occipital condyles, the ventral ends of their occipital condyles were localized in a more basal direction in reference to FM, and their ventral articular parts were more dorsally positioned in reference to FM. The sheep, on the other hand, had a broader FM along the ventromedial and left lateral lines in comparison to the goats. The mean morphologies of the regions that were analyzed in the goats and sheep revealed that the ventromedial side of the FM of the sheep was more conical compared to that in the goats, and the FM of the goats had a more elliptic appearance. It is believed that these data demonstrated in our study resulted from changes in the biomechanics of CCJ originating from differences in grazing behaviors. Furthermore, in our study, it was found that according to PC1, the occipital condyle was the primary factor for the variation in morphology, whereas the upper-left corner and ventromedial side of FM were the secondary factors. According to PC2, the right occipital condyle explained the variation in morphology to a large extent. Based on PC3, the variation in morphology was explained by the entire right occipital condyle, the lateralmost corner of the left occipital condyle, and the left side of FM. These asymmetries suggested that there may be a dominance on one side of the body originating from the development of the associated parts of the brain.

Factors such as nutrition, breeding style, and climate conditions can result in some variations, even among individuals of the same breed. This is why various metric measurements are needed to identify not only interspecies but also intraspecies dimorphisms.

Conclusion

In conclusion, it is believed that with this study, data that will contribute to several different disciplines are provided by presenting interspecies similarities and differences by conducting the morphological analyses of the foramen magnum and the occipital condyle, which participate in the formation of the caudal part of the cranium in sheep and goat breeds, which have existed in the history of humanity for millennia.

Conflict of Interest

The authors declare no conflict of interest.

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Author Contributions

Motivation/Concept: ID, YD; Design: ID, YD, IG; Control/Supervision: ID, FAK; Data Collection and/or Processing: ID, YD, FAK, OO; Analysis and/or Interpretation: ID, YD; Literature Review: ID, YD, FAK, IG, OO; Writing the Article: ID; Critical Review: ID, YD, FAK, IG, OO.

Ethical Approval

Bingöl University Experimental Research and Application Center, Animal Experiments Ethics Committee 09.05.2023, 2023/E.106395 Number Ethics Committee Decision

