

The Investigation of Sexual and Species Dimorphism in the Foot of Chukar Partridge (*Alectoris chukar*) and Gray Partridge (*Perdix perdix*) Using Geometric Morphometric and Symmetric Analyses

Kınalı Keklik (*Alectoris chukar*) ve Çil Keklik (*Perdix perdix*) Ayaklarındaki Cinsiyet ve Tür Dimorfizminin Geometrik Morfometrik ve Simetrik Analizler Kullanılarak İncelenmesi

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ABSTRACT

Morphological differences can provide insights into species' ecological adaptations and evolutionary processes. This study focuses on examining the effects of sex and species dimorphism on foot morphology in two different partridge species, the Chukar Partridge (*Alectoris chukar*) and the Gray Partridge (*Perdix perdix*). A total of 68-foot samples, including both right and left foot, were analyzed from 34 partridges collected in Sivas province. The analysis revealed that, regardless of sex, the first toe of Chukar Partridges was longer than that of Gray Partridges. When comparing species, the angle between the toes of female Chukar Partridges was wider than that of female Gray Partridges, while the angle between the toes of male Gray Partridges was wider than that of male Chukar Partridges. In terms of sex differences, the angle between the toes of male Gray Partridges was wider than that of female Gray Partridges, while the angle between the toes of female Chukar Partridges was wider than that of male Chukar Partridges. The contribution of directional asymmetry to variation was found to be lower than that of fluctuating asymmetry in both shape and size, suggesting that the asymmetry may result from developmental differences rather than lateral bias. Overall, the width of the toe angle was linked to habitat preferences and ecological adaptations. These findings suggest that the foot morphology of partridges may be shaped by factors such as sex, species, and habitat, and that these adaptations may help birds cope with environmental challenges.

Keywords: Chukar partridge, gray partridge, geometric morphometry, morphology, symmetry analysis

ÖZ

Şekil farklılıkları, türlerin ekolojik adaptasyonları ve evrimsel süreçleri hakkında bilgi sağlayabilir. Çalışmanın odak noktası, iki farklı keklik türü olan Kınalı Keklik (*Alectoris chukar*) ve Çil Kekliklerin (*Perdix perdix*) cinsiyet ve ırk dimorfizminin ayak morfolojisine etkilerini incelemektir. Sivas ilinden toplanan 34 keklikten sağ ve sol olmak üzere toplam 68 ayak örneği analiz edildi. Analiz sonuçlarına göre cinsiyet fark etmeksizin kınalı kekliklerin baş parmağı çil kekliklerden uzundu. İrklar kıyaslandığında dişi kınalı kekliğin parmakları arasındaki açısı dişi çil keklikten genişti. Erkek çil kekliğin parmakları arasındaki açısı ise erkek kınalı keklikten daha genişti. Cinsiyetler kıyaslandığında ise erkek çil kekliklerin parmakları arasındaki açısı dişi çil kekliklerden, dişi kınalı kekliklerin parmakları arasındaki açısı erkek kınalı kekliklerden daha genişti. Directional asimetrisinin varyasyona katkısı, şekil ve boyut üzerindeki fluctuating asimetriden daha düşüktü. Bu da asimetrisinin, taraf farkından ziyade gelişimsel farklılıklardan oluştuğunu göstermektedir. Sonuç olarak parmaklar arasındaki açının genişliği habitat tercihleri ve ekolojik adaptasyonlarla ilişkilendirildi. Kekliklerin ayak morfolojisinin cinsiyet, ırk ve yaşam alanı gibi faktörlerle şekillenebileceğini ve bu adaptasyonların kuşların çevresel zorluklarla başa çıkmasına yardım ettiği düşünülmektedir.

Anahtar Kelimeler: Çil keklik, kınalı keklik, geometrik morfometri, morfoloji, simetri analizi

INTRODUCTION

When examining evolutionary processes, morphological traits undergo variations in shape and size due to the strong influence of biological factors, and these variations play a significant role in species identification. While the dimensions of morphometric traits may be similar, species can often be distinguished by their shapes.¹⁻³ In recent years, there has been a noticeable increase in the use of geometric morphometric analyses in studies of bird species.^{4-6,25-30} Morphometric analysis focuses on the dimensions of materials, whereas geometric morphometric analysis concentrates on shape. The use of geometric morphometric analysis is advantageous over traditional size analyses, as shape is a more decisive factor and tends to exhibit greater variation within species groups than size.^{7,8,31-34} Since form and function are closely linked to species' ecology, body traits can provide insight into how animals forage for food or evade predators.^{9,10}

The fundamental principle of geometric morphometric analyses is the examination of shapes using Cartesian coordinates based on anatomical landmarks.¹¹ Various ecological and biological processes can lead to shape differences between species and within-species groups, which may also result from factors such as disease, geographic adaptation, ontogenetic development, or evolutionary divergence.¹² The Chukar Partridge (*Alectoris chukar*), a ground-dwelling bird specialized in running and walking, matures early. Its locomotor performance is influenced by factors such as age, sex, species, and breeds.^{13,14} The Gray Partridge (*Perdix perdix*), on the other hand, nests on the ground and inhabits open agricultural fields and grasslands. Once common in agricultural landscapes, the Gray Partridge has experienced a significant population decline over the past fifty years, necessitating its classification for conservation purposes.¹⁵

Partridges have strong, short, and sturdy feet, which help them move easily in steep terrains. Being anisodactyl, they have a total of four toes, three facing forward and one facing backward. The forward-facing toes are separated and strong, enhancing grip on the ground. The hind toe (hallux) is usually small and makes minimal or no contact with the ground. The toes end in strong claws, which are used for digging, gripping, and defense.³⁶

In this study, we aim to investigate the effects of sex and species dimorphism on the foot morphology of two wild partridge species, the Chukar Partridge and the Gray Partridge, using geometric morphometric analysis. Additionally, we aim to analyse symmetry-asymmetry patterns to assess differences between the left and right foot.

MATERIALS AND METHODS

Animal

In the province of Sivas, a total of 34 partridges were utilized in this study, comprising 22 Chukar Partridges (*Alectoris chukar*) (15 males and 7 females) and 12 Gray Partridges (*Perdix perdix*) (6 males and 6 females) harvested by hunters from the Gürlevik and Yılanlı General Hunting Grounds in Hafik District, the Gaziköy and Uçuk General Hunting Grounds in Şarkışla, and the Aşağı Boğazkesen General Hunting Ground in Sivas Center. Partridges are all similar-aged wild birds that have hatched in the wild. The average weight of male Chukar Partridges was 582 g, while females weighed 458 g; for Gray Partridges, males weighed 358 g and females 311 g. The foot of the partridges taken from the hunters were immediately photographed without any processing. For the geometric morphometric analysis, the left foot of each partridge was used, while the right and left foot were utilized for symmetry-asymmetry analysis. The foot was photographed from a dorsal view at a 90-degree angle from the Os tarsometatarsus, positioned at 31 cm for analysis. Ethical approval for the study was obtained from the Ethics Committee of Selçuk University Faculty of Veterinary Medicine (Date: 05/09/2024, Approval No: 2024/139).

Geometric Morphometric and Symmetric Analysis

To facilitate the analysis of the photographs, it was necessary to convert them into Tps format. This conversion was performed using TpsUtil (version 1.74). Landmark coordinates were established by placing landmarks on the photographs converted to Tps format using the tpsDig (version 2.30) program. A total of nine landmarks were utilized (Figure 1). The dataset containing the coordinates was uploaded to Morphoj (version 1.07a) for geometric morphometric analysis and symmetry-asymmetry analysis. The landmarks used are as follows:²⁰

- Lm1: Lateral of the articulatio metatarsophalangea I
- Lm2: Phalanx terminalis digiti I
- Lm3: Medial of the articulatio metatarsophalangea I
- Lm4: Phalanx terminalis digiti II
- Lm5: Tela interdigitalis intermedia
- Lm6: Phalanx terminalis digiti III
- Lm7: Tela interdigitalis lateralis
- Lm8: Phalanx terminalis digiti IV
- Lm9: Lateral of the articulatio metatarsophalangea IV

Statistical Analysis

For geometric morphometric analysis, regression analysis and Principal Component Analysis (PCA) were conducted using the MorphoJ software. The PCA analysis determined

the direction and magnitude of change in the principal components relative to the total shape variation from the mean shape. Discriminant Function Analysis (DFA) was performed to identify group characteristics based on sex and species.²⁰ Additionally, symmetry-asymmetry analysis was conducted in MorphoJ to determine the symmetric and asymmetric characteristics of the right and left foot.³⁵

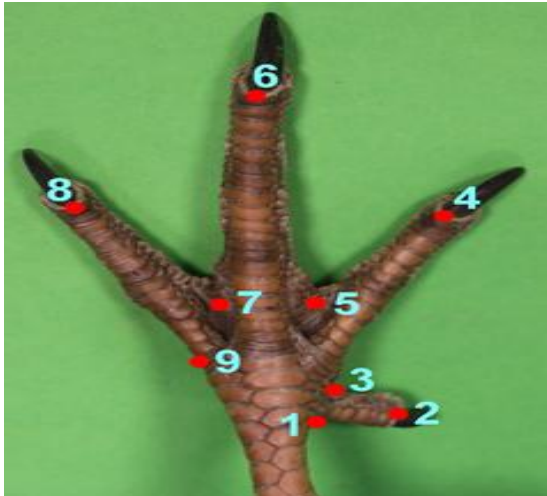


Figure 1. Representation of landmarks; Lm1: Lateral of the articulatio metatarsophalangea I, Lm2: Phalanx terminalis digiti I, Lm3: Medial of the articulatio metatarsophalangea I, Lm4: Phalanx terminalis digiti II, Lm5: Tela interdigitalis intermedia, Lm6: Phalanx terminalis digiti III, Lm7: Tela interdigitalis lateralis, Lm8: Phalanx terminalis digiti IV, Lm9: Lateral of the articulatio metatarsophalangea IV.

RESULTS

Prior to conducting Principal Component Analysis (PCA), regression analysis was performed. The regression analysis conducted on the shape centroid size (PCs) indicated that species accounted for 4.09% of the shape variation ($P = .2900$). Based on this finding, it was determined that the foot shape variations concerning the species factor addressed in this study were not size-dependent. Consequently, no statistically significant allometric component was identified.

According to the results of the Principal Component Analysis, the first principal component (PC1) explained 31.40% of the total shape variation, while the cumulative contribution of the first two principal components (PC1 + PC2) reached 60.19% of the total shape variation. A significant inflection point among the principal components was particularly observed between PC2 and PC3.

Figure 2 presents wireframe graphics obtained from PCA, illustrating the direction and extent of variation related to PC1 and PC2 in terms of total shape variation. The regions where shape differences associated with PC1 and PC2 are

concentrated include the terminal phalanges of the first (Lm2), second (Lm4), third (Lm6), and fourth (Lm8) toes. The most pronounced changes in the mean shape according to PC1 are a reduction in the angle between the third and fourth toes and an increase in the angle of the first toe. In contrast, the most significant changes observed in PC2 involve an increase in the angles between the second and third toes, as well as between the third and fourth toes.

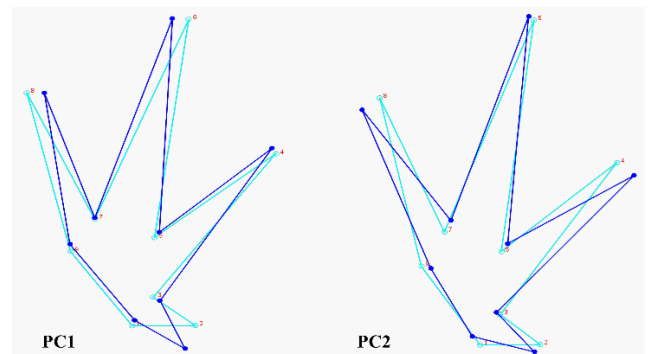


Figure 2. Wireframe graphical representation of shape differences concerning PC1 and PC2. Dark blue represents the positive bounds of principal component scores.

The discriminant function analysis (DFA) compared the foot morphology of the partridges in relation to gender and species factors. Figure 3A depicts the comparison of the foot morphology of the chukar partridge, while Figure 3B illustrates that of the gray partridge, with a focus on the gender factor. According to the analysis, the angle between the second and third toes of female gray partridges is narrower compared to males. Conversely, the angle between the second and third toes of female chukar partridges is wider than that of males.

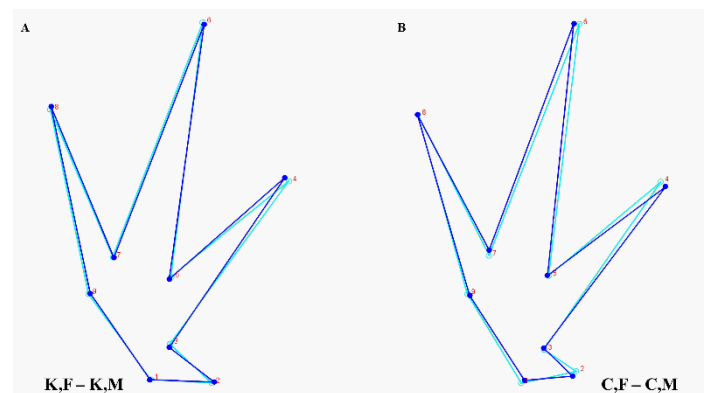


Figure 3. Wireframe graphics of the foot according to sex in Discriminant Function Analysis (K: Chukar partridge, C: Gray partridge, F: Female, M: Male). In Figure 3A, the light blue color represents the female Chukar partridge, while the dark blue color represents the male Chukar partridge; in Figure 3B, the light blue color represents the female Gray partridge, and the dark blue color represents the male Gray partridge.

The DFA graphs presented in Figure 4 illustrate the comparison of partridge foot morphology based on species factors. In Figure 4A, the angle between the second and third toes of female gray partridges is narrower than that of female chukar partridges, while the angle between the third and fourth toes is wider in female gray partridges compared to females of the chukar species. Additionally, the hallux of female chukar partridges is longer than that of female gray partridges. Figure 4B shows that the angles between the second and third, as well as the third and fourth toes of male gray partridges, are wider compared to those of male chukar partridges. Furthermore, the hallux of male chukar partridges is longer than that of male gray partridges.

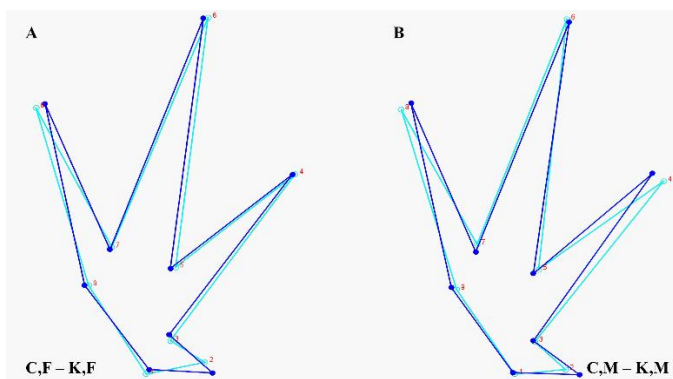


Figure 4. Wireframe graphics of the foot according to breed in Discriminant Function Analysis (K: Chukar partridge, C: Gray partridge, F: Female, M: Male). In Figure 4A, the light blue color represents the female Gray partridge, while the dark blue color represents the female Chukar partridge; in Figure 4B, the light blue color represents the male Gray partridge, and the dark blue color represents the male Chukar partridge.

The Procrustes ANOVA analysis indicated that measurement error was not significant. Variation among individuals in terms of "Shape" and "Size" was statistically

significant ($P < .0001$). While fluctuating asymmetry for "Size" was statistically significant ($P < .0001$), directional asymmetry was not significant ($P = .9057$). The symmetric component was statistically significant ($P < .0001$). Regarding "Shape," both symmetric ($P < .0001$) and asymmetric components (fluctuating ($P < .0001$) and directional ($P = .0005$)) were statistically significant. The variation rates of differences among individuals were 97.42% for "Size" and 51.87% for "Shape" (Table 1). The contribution of directional asymmetry (DA) to variation was lower than that of fluctuating asymmetry regarding shape and size, suggesting that asymmetry is primarily due to developmental differences rather than side differences. Nevertheless, it was observed that the partridge foot included in the dataset exhibited a tendency in usage, resulting in a statistically significant value for directional asymmetry in terms of shape (Figure 5).

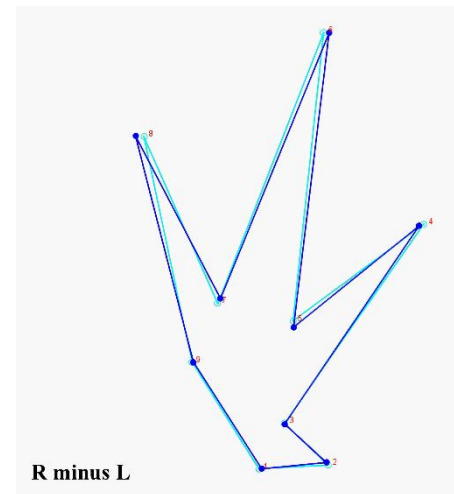


Figure 5. Symmetry-asymmetry graph. Dark blue represents the left foot.

Table 1. Morphological variation in foot size and shape calculated by Procrustes ANOVA.

Size	% Variance Explained	SS	MS	dF	F	P
Individual	97.425	343212.290249	16343.442393	21	45.91	<.0001
Side	0.001	5.112705	5.112705	1	0.01	.9057
Ind*Side	2.121	7475.255787	355.964561	21	16.83	<.0001
Error 1 (Imaging)	0.264	930.610306	21.150234	44	1.41	.0864
Error 2 (Digitizing)	0.187	660.234013	15.005318	44	0.83	.7491
Shape	% Variance Explained	SS	MS	dF	F	P
Individual	51.873	0.51334933	0.0017460862	294	1.66	<.0001
Side	4.252	0.04208750	0.0030062498	14	2.86	.0005
Ind*Side	31.179	0.30855610	0.0010495105	294	9.89	<.0001
Error 1 (Imaging)	6.605	0.06537211	0.0001061236	616	1.06	.1817
Error 2 (Digitizing)	6.200	0.06135782	0.0000996069	616	0.97	.6780

SS: sums of squares; MS: mean squares; dF: degrees of freedom, F: statistics and parametric p values are provided for each effect.

DISCUSSION

Natural habitats, foraging behaviour, ecological niche, and factors such as genetic variation significantly influence the development of foot morphology.¹⁶ This study aimed to investigate whether the factors of species and sex contribute to shape variation in the foot of two distinct partridge species, based on the hypothesis that “the variation in the foot of species inhabiting similar geographical regions with comparable foraging conditions of the same sex would be limited.” The findings indicate that the different environmental conditions experienced by male and female chukar partridges inhabiting rocky, steep, and rugged terrains, as well as male and female gray partridges residing in fields, flat landscapes, and pastures, result in phenotypic variation arising from adaptations influenced by these distinct living conditions and sexual dimorphism.

Geometric morphometric analyses revealed that, regardless of sex, the hallux of the gray partridge is shorter than that of the chukar partridge. Chukar partridges inhabit rocky and rugged terrains,¹⁷ which necessitates a longer hallux for improved grip and stability. A longer hallux enables these partridges to effectively navigate steep slopes and rocky surfaces while foraging. Additionally, it enhances their ability to probe the ground for seeds and insects, thereby increasing their chances of survival. Conversely, gray partridges, typically found in more open and flat fields,¹⁸ possess a shorter hallux that suffices for food searching in these relatively flat environments, minimizing competition with grasses. Furthermore, an increase in toe length provides a broader surface area for weight distribution, enhancing maneuverability in steep and uneven terrains. In summary, toe length may reflect various ecological adaptations and habitat preferences. Similarly, the angle between the toes is largely related to these factors. In our study, the angle between the toes of female gray partridges was found to be narrower than that of female chukar partridges. A wider toe angle enhances stability and grip, which is a critical adaptation for chukar partridges living in rocky and uneven environments. This adaptation facilitates a more balanced distribution of weight on steep inclines and challenging terrains, thereby improving stability and gripping ability. Moreover, the angle between the toes influences the bird's maneuverability while foraging, allowing for quick directional changes necessary for evading predators. A narrower toe angle in gray partridges renders them more adept at running and making rapid maneuvers in flat and pasture-like environments. Consequently, it is hypothesized that the toe angle may serve as an adaptable trait that can be developed in response to varying

environmental contexts and locomotion requirements.

Another notable finding from our study is that the angle between the toes of female gray partridges is narrower than that of male gray partridges. Male partridges possess greater body weight and longer, stronger legs compared to females. The balanced distribution of body weight across the foot and toes is critical for stability and grip. It is posited that a wider angle between the toes may facilitate this weight distribution. One of the adaptations associated with sexual dimorphism in partridges may be the differences in toe angles. Interestingly, it was found that the angle between the toes of female chukar partridges is wider than that of male chukar partridges. Furthermore, male gray partridges exhibit a greater angle between their toes compared to male chukar partridges. The relatively shorter femur and more streamlined body structure of gray partridges, as well as the rounder body shape characteristic of chukar partridges adapted to rugged terrains, suggest that the influence of species may play a role in these sexual adaptations. These morphological variations may be a response to the ecological demands and habitat preferences specific to each species and sex.

When reviewing previous studies on this topic, Lombardo et al.¹⁹ examined the ratio of the lengths of the second and fourth toes in some male and female birds. In their study, they found no statistically significant differences in the ratios of the lengths of the second and fourth toes on the left foot of house sparrows, tree swallows, budgerigars, and chickens. Similarly, in Japanese quail, which also underwent analysis regarding this ratio, no statistically significant differences were found between the sexes. In the same study, no sexual dimorphism was observed in the ratios of the lengths of the second-third and third-fourth toes; however, it was noted that the second toe in male quail was more curved inward compared to females.²⁰ Additionally, Ruuskanen et al.²¹ found that in their study of wild birds, the ratios of the lengths of the second-third and second-fourth toes were higher in males than in females. The differences in toe lengths between the sexes may be related to variations in hormone levels in males and females.^{22,23} In studies comparing the right and left foot, a hypothesis was proposed suggesting that one leg may be used more than the other.²⁴ These findings contribute to a broader understanding of the factors influencing morphological variations among avian species and highlight the complexity of sexual dimorphism and its ecological implications.

Although there have been studies on birds concerning their skulls, beaks, and wings, there is a scarcity of geometric morphometric research focused specifically on their foot.

In the present study, we employed geometric morphometric methods to elucidate the phylogenetic relationships of the foot structures in partridges. A detailed shape analysis was conducted on the foot of chukar partridges and gray partridges, which are distinguished by their differing habitats. The obtained data indicate that the overall shape of partridge foot is significantly influenced by factors such as species, sex, and geographical region. Consequently, the findings from this study are expected to contribute to the classification of partridges, enhance our understanding of the adaptations they have undergone, and be utilized for insights into the identification of fauna and evolutionary processes. The low population density of chukar partridges in the study area is a limitation of this research. The scarcity of individuals may have affected the sample size, potentially limiting the statistical power and generalizability of the findings. Despite this limitation, the current results provide valuable insights into the morphological characteristics of the species and contribute to the existing literature.

As a result of, regardless of sex, the thumb length of chukar partridges was longer than that of grey partridges. When comparing species, the angle between the toes of female chukar partridges was wider than that of female grey partridges. Similarly, the toe angle of male grey partridges was wider than that of male chukar partridges. In terms of sex differences, the toe angle in male grey partridges was wider than in female grey partridges, while the toe angle in female chukar partridges exceeded that of male chukar partridges. The contribution of directional asymmetry to variation was lower than that of fluctuating asymmetry in both shape and size. This indicates that the observed asymmetry arises primarily from developmental differences rather than consistent lateralization.

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