

Morphological Variations in the Skulls of Male and Female Persian Fallow Deer (*Dama dama mesopotamica*)

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Abstract

BACKGROUND: Persian fallow deer (PFD), *Dama dama mesopotamica*, is one of the rarest members of the Cervidae family currently listed as endangered by the International Union for Conservation of Nature.

OBJECTIVES: Morphological variations in the skulls of male and female PFDs were evaluated in this investigation.

METHODS: In cooperation with the Department of Environment, skulls and mandibles were obtained from five male and four female animals. After the usual practices of bone cleaning, the specimens were evaluated for morphological differences. Next, 29 parameters were measured on the skulls and mandibles by a digital Vernier caliper for morphometric studies. In addition, ten, six, and nine landmark points were defined on the left lateral photos of mandibles, dorsal, and left lateral photos of skulls, respectively. The points were digitized on two-dimensional images using the TpsDig2 software. The shape differences between the two genders were analyzed using discriminate function analysis in the MorphoJ software.

RESULTS: The interfrontal ridge was more prominent in male PFDs than in female animals. Moreover, there were some significant differences in the measured parameters, mostly in the mandible. The geometric morphometric evaluations showed no significant differences between the two genders.

CONCLUSIONS: The findings of the present investigation revealed some morphological differences between the skulls of male and female PFDs.

KEYWORDS: Dimorphism, Head, Mandible, Persian fallow deer, Skull

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Introduction

Persian fallow deer (PFD) with the scientific name *Dama dama mesopotamica*, as one of the

rarest deer species in the world, was previously abundant throughout western Asia. Nowadays,

they are inhabiting only a small habitat in Khuzestan, Southern Iran, two rather small protected areas in Mazandaran Province (Northern Iran), and some other small areas in the Near and Middle East. This member of the subfamily Cervinae belonging to the family Cervidae is currently listed as endangered by the International Union for Conservation of Nature (Jantschke, 1990; Rabiei and Saltz, 2011; Berger-Tal *et al.*, 2012; Ekrami *et al.*, 2016; Vigne *et al.*, 2016).

Due to the importance of skull anatomy in diverse fields of science, including clinical and surgical practices, wildlife and evolutionary sciences, and sexual dimorphism. Therefore, many studies have been carried out on distinct animals, such as various species of dogs, equines, felines, porcine, sheep, talpas, and rodents (Kieser and Groeneveld, 1992; Evans and McGreevy, 2006; Carreira and Ferreira, 2016; Pitakarnnop *et al.*, 2017; Kyllar *et al.*, 2016; Choudhary and Singh, 2016; de la Barra *et al.*, 2020; Selçuk *et al.*, 2019; Selçuk *et al.*, 2018). Marzban Abbasabadi *et al.* (2018) evaluated the morphometric differences of the skull between male and female Zell sheep. They found significant in sexual dimorphism between males and females. Parés-Casanova (2015) analyzed the skulls of adult White Rasquera goat breed to explore sexual dimorphism based on geometric morphometric in this local goat breed. Their results demonstrated that the differences in the skull of the two genders may be attributed to the extensive management styles of the animals as under a low anthropogenic influence they tend to reinforce their natural sexual size dimorphism.

In this regard, some studies have evaluated skull anatomy in various species of the Cervidae family. Markov (2014) analyzed the extent of sexual dimorphism in the skull features of red deer (*Cervus elaphus L.*) in Bulgaria and reviewed the population morphometric variations in the skull of red deer from mountainous and lowland habitats. In addition, there are many

studies on skull anatomy in European roe deer (And and Reig, 1993; Sabalinkiene *et al.*, 2017). However, information about PFD skull anatomy is limited. Therefore, the current study aimed to evaluate morphological differences in the skulls of male and female PFDs.

Materials and Methods

Samples Preparation

In cooperation with the Department of Environment of Mazandaran province, Iran, a total of nine PFD skulls or heads (five adult male and four adult female) were obtained from the museum of the Department of Environment and the dead deer of Dasht-e Naz Wildlife Refuge, Sari, Iran protected area during the past two years. Only the skulls of adult PFDs with completely formed dentition were used (Selçuk *et al.*, 2018). The heads and skulls were examined for any skeletal damages or deformities and then the heads were prepared by boiling method. Next, the mandibles were disarticulated from the temporomandibular joint. The samples were numbered and photographed (Canon EOS 4000D, Tokyo, Japan) from dorsal, lateral, ventral, and caudal views. During photography, the camera and the samples were fixed in place to keep the distance and angles of the shots constant.

Morphological Observations

At the first step, the male and female PFD skulls and mandibles were assessed to find remarkable morphological differences. As the horns or cornual process are different between sexes, the other components of the skull and mandible were searched. Afterwards, the accuracy of the found hallmarks was examined.

Morphometric Geometric Studies

As demonstrated in [Figures 1](#) and [2](#), ten, six, and nine landmark points were defined on the left lateral photos of mandibles and the dorsal and left lateral photos of skulls, respectively.

These points were digitized on two-dimensional (2D) images using the TpsDig2 software version 2.16 (Rohlf, 2010).

The adequacy of tangent shape for statistical analysis was investigated utilizing the TpsSmall (Rohlf, 2003). The non-shape information was removed from landmark configurations applying General Procrustes Analysis

The covariance matrices were generated and the shape differences between the two sexes were analyzed by the discriminant function analysis (DFA) in the MorphoJ version 1.02j (Klingenberg, 2011). The patterns of skull and mandible shape differences were illustrated in the wireframe relative to each other for quantification and visualization purposes.

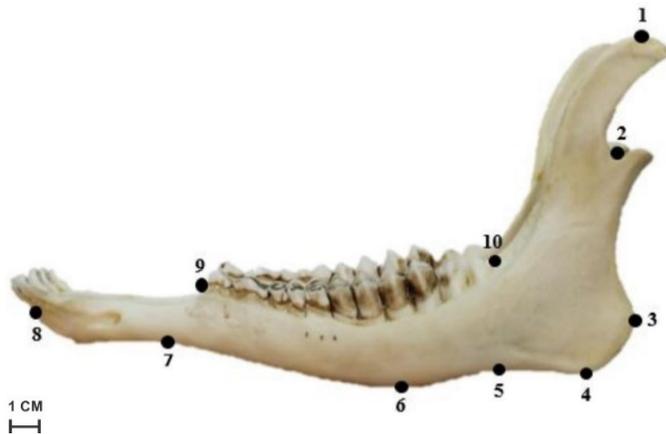


Figure 1. Location of the used landmarks of the left lateral view of Persian fallow deer (*Dama Dama Mesopotamica*) mandible: 1. The highest point of coronoid process; 2. The deepest point of mandibular notch; 3. Caudalmost point of angle of mandible; 4. Ventralmost point of angle of mandible; 5. The most concave point of mandible at the junction of body and angle of mandible; 6. The ventralmost point of body; 7. The most concave point of mandible at the junction of molar part and incisive part of the body; 8. The anteriormost point of mandible; 9. The anteriormost point of premolar alveoli; 10. The posteriormost point of premolar alveoli.

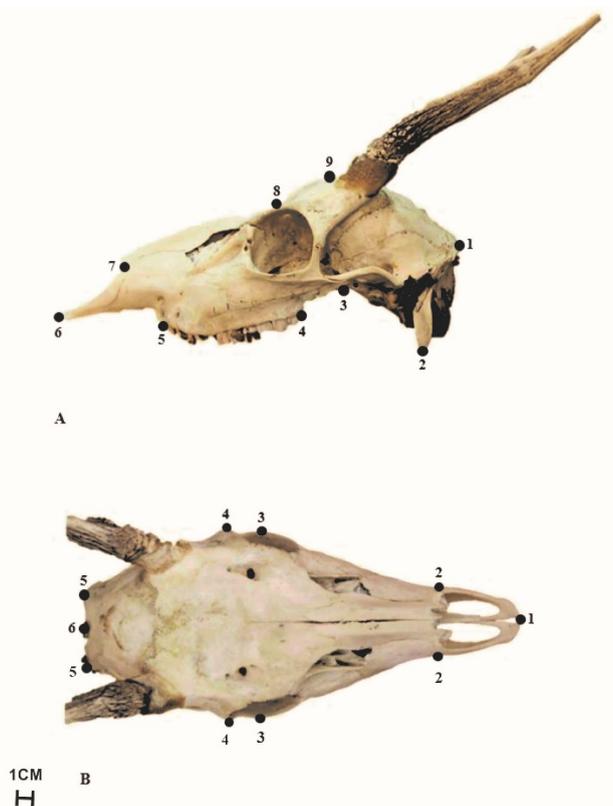


Figure 2. (A) Location of the used landmarks of the left lateral view of Persian fallow deer (*Dama Dama Mesopotamica*) skull: 1. Posteriormost point of occipital bone; 2. Ventralmost point of jugular process; 3. The most concave part of sphenoid bone; 4. The most caudal part of alveolar process of maxillae bone; 5. The most cranial point of alveolar process of maxillae bone; 6. The anteriormost point of incisive bone; 7. The Posteriormost point of nasoincisive notch; 8. The most concave part of frontal bone; 9. The highest point of frontal bone (B) location of the used landmarks of the left lateral view of Persian fallow deer: 1. The anteriormost point of incisive bone; 2. The junction of incisive and maxillae bone; 3. The most concave point of orbit; 4. The most prominent part of zygomatic process of frontal bone; 5. The most caudal part of occipital bone; 6. The most concave point of nuchal crest.

Morphometric Measurements

For morphometric measurements, 11 parameters in the mandibles and 18 parameters in the skulls selected based on the previous studies (Figures 3 and 4, Tables 1 and 2) were calculated utilizing digital Vernier caliper (Digimatic Caliper, Japan) with an accuracy of 0.01 mm.

All measurements and observations were blinded (Pitakarnnop et al., 2017; Onuk et al., 2013). Data were analyzed by the independent samples t-test using the SPSS software version 16 (SPSS Inc., Chicago, USA). The level of significance was considered as P-value<0.05. The Data are presented as mean ± SE.

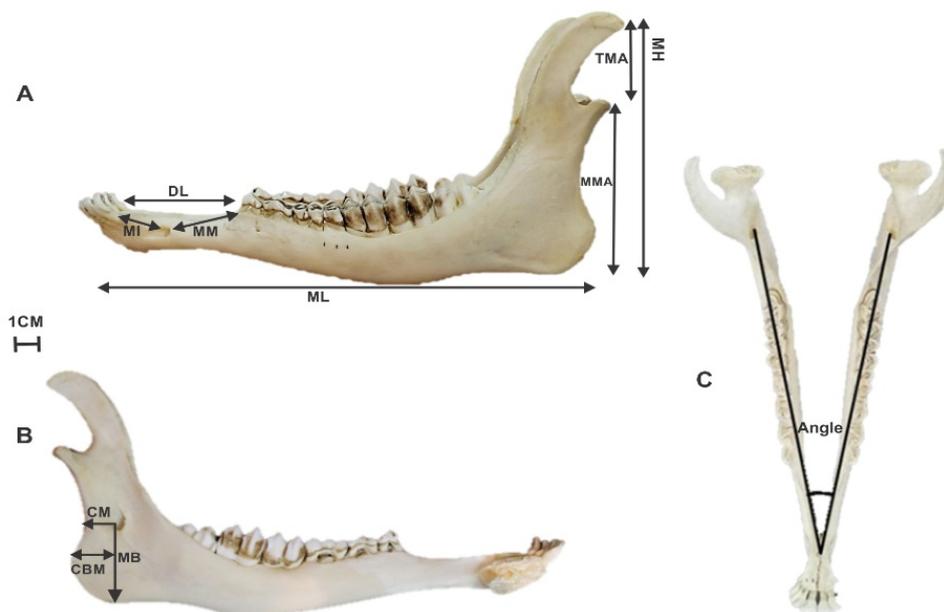


Figure 3. Measured parameters in lateral view (A), medial view (B) and dorsal view (C) of the mandible of Persian fallow deer (*Dama Dama Mesopotamica*): (A) Condylod fossa to height of mandible (TMA), Condylod fossa to base of mandible (MMA), Maximum height of mandible (MH), Total length of mandible (ML), Mental foramen to first premolar (MM), Lateral alveolar root to mental foramen (MI), Length of diastema (DL). (B), Caudal border of mandible to beneath of mandibular foramen (CBM), Mandibular foramen to the caudal border of mandible (CM), Mandibular foramen to base of mandible (MB). (C), The interior angle between the bodies of the mandible (Angle)

Table 1. Description of 11 measurements taken from the mandible (see details in Figure 3).

Mandible	
TMA	Temporalis muscle moment arm; measured from the posterior end of the condyle to the apex of the coronoid process.
DL	Length of diastema; measured from the posterior end of I4 to the anterior end of P1
ML	Length of the mandible; measured from the anterior limit of the dentary bone between I1 to the posterior end of the mandibular condyle.
MI	Mental foramen to the I4, measured from the posterior end of the alveolus of the I1 to the mental foramen.

Mandible	
MM	Mental foramen to the P1, measured from the anterior end of the alveolus of the P1 to the mental foramen
MMA	Masseteric moment arm; measured from the dorsal surface of the condyle to the ventral border of the angle of the mandible.
CBM	Mandibular foramen to the caudal border of the angle mandible.
CM	Mandibular foramen to the caudal border of ramus.
MB	Mandibular foramen to the ventral border of the angle mandible.
MH	Total height of mandible; measured from the highest point of the coronoid process to the ventral border of the angle of mandible.
Angle	The interior angle between the bodies of the mandible

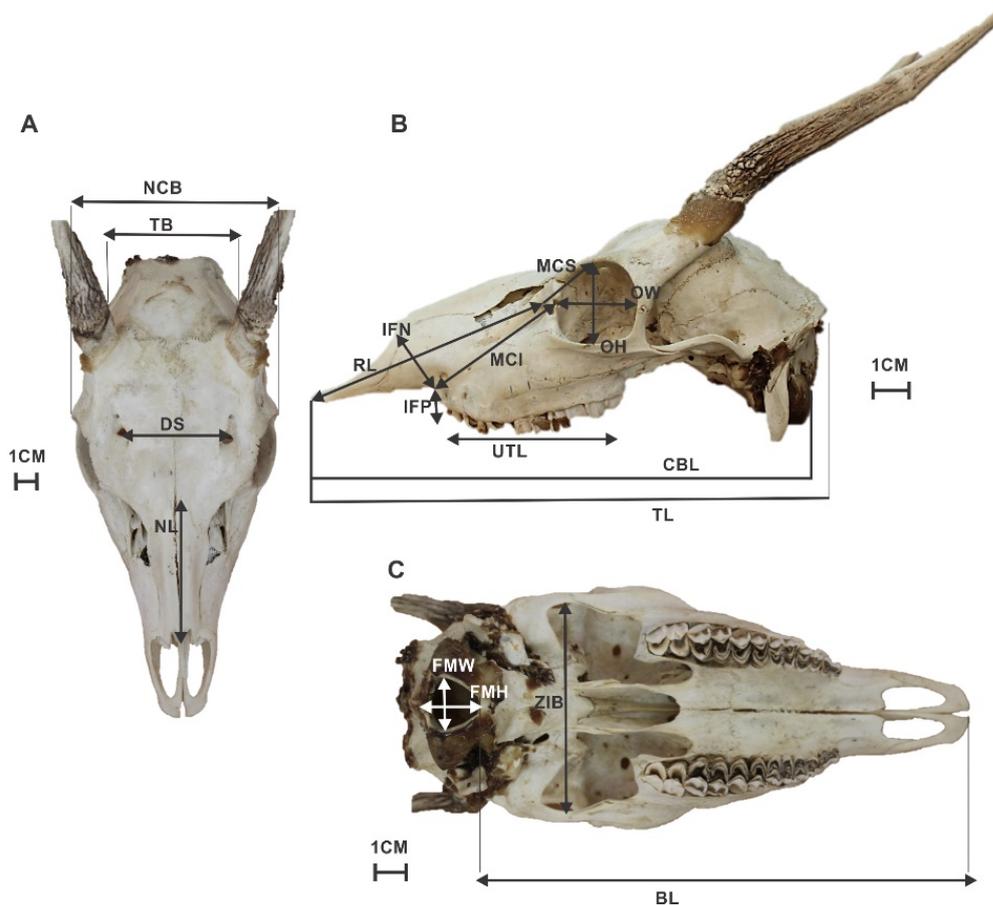


Figure 4. Measured parameters in dorsal view (A), lateral view (B) and ventral view (C) of the skull of Persian fallow deer (*Dama Dama Mesopotamica*): (A), NCB. Neurocranium breadth; TB. Total breadth; DS. distance between two supra-orbital foramen; NL. Nasal length. (B), TL. Total length; CBL. Condylobasal length; UTL. Upper tooth row length; MCI. Medial canthus to infraorbital foramen; RL. Rostrum length; MCS. Medial canthus to supraorbital foramen; OH. Orbital height; OW. Orbital width. (C), ZIB. Zygomatic breadth; BL. Basal length; FMH. Foramen magnum height; FMW. Foramen magnum width.

Table 2. Description of 18 measurements taken from the skull (see details in Figure 4).

Skull	
TL	Total length; measured from the anterior edge of the incisive bone to the posteriormost part of the occipital bone.
NL	Nasal Length; measured from the most cranial end of nasal bone to the posteriormost part of it.
RL	Rostrum length; measured from the medial cantus of the orbit to the anterior limit of the Incisive bone.
MCS	Medial canthus to supraorbital foramen; measured from the Medial cantus of the orbit to the supra orbital foramen.
MCI	Medial canthus to infraorbital foramen; measured from the Medial cantus of the orbit to the infra orbital foramen.
CBL	Condylbasal length; measured from the anterior edge of the incisive bone to the posteriormost projection of the occipital condyle.
ZIB	Zygomatic arches internal breadth; the greatest distance between the inner margins of the zygomatic arches, with two anterior measurements.
NCB	Neurocranium breadth; The widest point of the braincase across parietals.
TB	Total breadth; the greatest width of the skull, including the mastoids.
BL	Basal length; measured from the anterior end of incisive bone to the posterior end of basioccipital bone
UTL	Upper tooth row length; measured from the anterior end of P1 to the posterior end of M3.
FMH	Foramen magnum height; the maximum height of the foramen magnum.
FMW	Foramen magnum width; the greatest width of the foramen magnum.
OH	Orbital height; the maximum height of the orbit.
OW	Orbital width; the greatest width of the orbit.
DS	Supraorbital foramen distance; measured the interior distance between two supraorbital foramens.
IFN	Infraorbital foramen to the nasoincisive notch; measured the distance between the infra orbital foramen to the nasoincisive notch.
IFP	Infraorbital foramen to the nasoincisive notch; measured the distance between the infraorbital foramen to P1.

Results

Descriptive Anatomy of Male and Female Persian Fallow Deer

As our observations revealed, the interfrontal ridge was prominent in male PFD and flat in female PFD (accuracy rate: 90.47%). In addition, the dorsal surface of the frontal bone was

concave in females and flat and more extended in males (accuracy rate: 85.71%) ([Figure 5](#)).

Morphometric Geometric Results

According to the DFA performed in this study, there was no significant sexual dimorphism in the skull of PFD. However, in some

defined landmarks, some differences were observed between sexes, which were somewhat consistent with the findings of morphometric studies (Figure 6).

Morphometric Measurements

Results of the descriptive analysis are presented in Tables 3 and 4. The results showed significant differences in some measured parameters of the skull and mandible of male and

female PFDs. The total breadth of the skull, upper tooth row length, temporalis muscle moment arm, diastema length, mental foramen distance to P1 and I4, and mandible total height were significantly larger in male PFDs, compared to female animals. However, the distance between the mandibular foramen and the caudal border of mandible angle was significantly higher in female PFDs than in male cases (Tables 3 and 4).

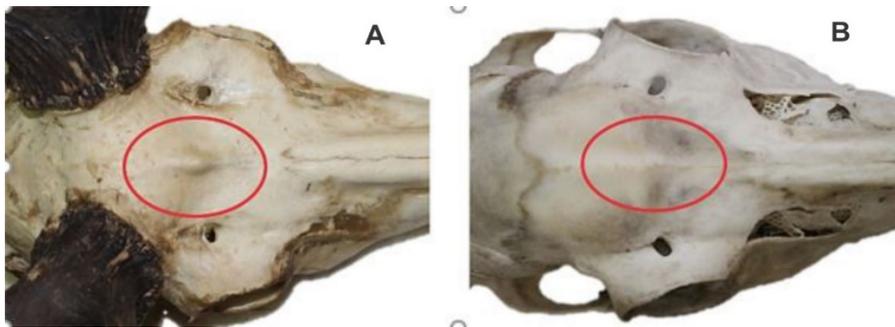


Figure 5. Dorsal view of the skulls of the male (A) and female (B) Persian fallow deer (*Dama Dama Mesopotamica*), showing the more prominent interfrontal ridge in male PFD (red oval) (A) (accuracy rate= 90.47%).

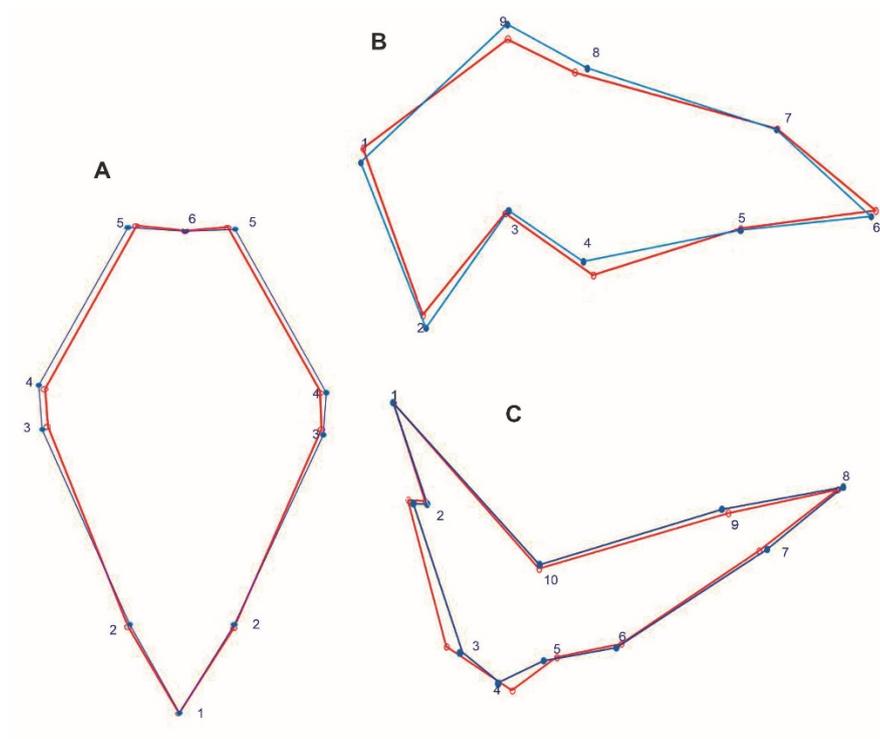


Figure 6. Visualization of the relative shape differences (by Morpho J software) among species based on wireframes between sexes in Persian fallow deer skull (dorsal view (A) and lateral view (B)), and mandible (lateral view (C)); Red lines: female, Blue lines: male.

Table 3. Morphometric data of Persian fallow deer (PFD) skull in mm (M±SE).

Parameters	Male PFD (N= 4)	Female PFD (N= 3)	P-value
TL	300.52 ±0.51	276.73 ±0.41	0.411
NL	79.92 ±4.77	93.84 ±1.43	0.163
RL	149.43 ±11.12	149.69 ±3.02	0.993
MCS	35.05 ±2.44	29.06 ±3.85	0.241
MCI	67.57 ±3.08	74.35 ±4.46	0.276
CBL	251.19 ±20.72	271.12 ±3.45	0.569
ZIB	100.08 ±0.68	102.12 ±0.98	0.292
NCB	99.59 ±15.36	86.86 ±3.19	0.557
TB	130.67 ±8.18	94.06 ±2.02	0.006 ^a
BL	211.14 ±19.22	265.87 ±12.13	0.118
UTL	97.08 ±3.18	81.82±0.01	0.006 ^b
FMH	25.53 ±0.19	25.55 ±0.005	0.340
FMW	23.49 ±0.35	26.25 ±2.15	0.417
OH	47.11 ±2.33	44.28 ±0.91	0.534
OW	50.09 ±3.39	44.56 ±1.36	0.181
DS	64.46 ±3.19	63.95 ±0.37	0.338
IFN	37.76 ±2.68	36.65±0.04	0.796
IFP	10.587 ±1.00	9.63±0.13	0.576

^{a,b} Values within a row with different superscripts differ significantly at $P<0.05$

Table 4. Morphometric data of Persian fallow deer (PFD) mandible in mm (M±SE).

Parameters	Male PFD (N= 4)	Female PFD (N= 3)	P-value
TMA	38.642 ±1.32	29.224 ±1.69	0.007 ^a
DL	64.12 ±1.53	52.39 ±0.28	0.004 ^b
ML	222.31 ±2.95	208.42 ±5.85	0.071
MI	28.39 ±0.88	23.54 ±1.94	0.024 ^c
MM	35.36 ±1.163	29.65 ±0.29	0.002 ^d
MMA	88.736 ±2.09	83.18 ±3.80	0.227
CBM	22.02 ±0.42	24.98 ±0.003	0.026 ^e
CM	15.14 ±0.53	16.81 ±0.01	0.093

Parameters	Male PFD (N= 4)	Female PFD (N= 3)	P-value
MB	42.38 ±1.64	42.45 ±0.02	0.968
MH	129.14 ±0.29	113.65 ±0.56	0.000 ^f
ANGLE	30.29° ±0.43	28.59° ±0.27	0.064

^{a,b,c,d,e,f} Values within a row with different superscripts differ significantly at $P < 0.05$

Discussion

Due to the importance of skull anatomy (Al-safy *et al.*, 2014; de la Barra *et al.*, 2014; Farhadinia *et al.*, 2014; Samuel *et al.*, 2016; Parés-Casanova and Fabre, 2013; Mohammadpour, 2011), morphological variations in the skulls of male and female PFDs were evaluated in this investigation. Descriptive morphological examinations of male and female skulls demonstrated more prominent interfrontal ridge and flatter frontal bone in males, compared to female subjects. There is no published study revealing hallmarks between male and female PFDs. However, it was expected to find differences in the bony structures of this area between sexes because of the pressure of horn growth on the frontal zone (Pételis and Brazaitis, 2003).

According to the morphometric-geometric results of this study, male and female PFDs were not significantly different in terms of the defined landmarks of the skulls and mandibles. However, some differences were observed in the caudal curvature of the mandible and some parameters defined in the facial part of the skull lateral view. The latter differences were somewhat consistent with the morphometric results. Based on the morphometric results, five parameters of the mandible, including the distance between mental foramen to the first premolar and forth incisor tooth (MM and MI), length of diastema (DL), Total height of mandible (MH), and the length of temporalis muscle moment arm (TMA), as well as two parameters in the skull, namely the total breadth (TB) and upper tooth row length (UTL), were significantly

larger in males than females. Furthermore, the CBM was significantly larger in female PFDs than the male cases.

It was shown that the mandible is a more accurate bone for distinguishing the skulls of male and female PFDs. Reig (1993) compared the craniometrics variability between two roe deer populations in Spain. They reported the mandible to be the most variable trait with significant differences between populations. Marzban Abbasabadi *et al.* (2018) surveyed morphometric differences of the skull in male and female Zell sheep and observed the only significant difference in mandible. Their findings indicated that the distance of lateral alveolar root to mental foramen was significantly higher in male Zell sheep than in females. In addition, in studies on skull dimorphism on other animals, such as the Feline (*Felis catus*) species, the only significant morphometric difference of skull was observed in the mandible where the masseteric moment arm (MMA) was higher in males than female animals (Pitakarnnop *et al.*, 2017).

In the Cervidae family, some studies have used skull or other parts of the skeleton to find the distribution of the subspecies, differentiate between populations, and assess the colorations with age, body mass, and some other variables. Most such studies have been performed on roe deer to examine morphometric differences. In some of these investigations, the diversity of the craniometrics parameters was large enough to divide the population into different groups or ecotypes. Blagojević and Milošević-Zlatanović

(2011) evaluated sexual shape dimorphism in Serbian roe deer (*Capreolus capreolus* L.) by combining the multi-variate statistical procedures with geometric morphometrics and visualization techniques. These authors reported distinct patterns of shape variability in male and female animals as the cranial base was broader in males, while elongated and slenderer-shape with narrower basicranium in female skulls.

Sabalinkiene *et al.* (2017) revealed that geographical location has a significant effect on the antler morphometry traits and skull size of male roe deer. Furthermore, they reported the impact of gender on the skull morphology traits at juvenile age. Based on their results for the zygomatic arch internal breadth (ZIB), the influence of gender was equally important in all age classes, whereas for ML, UTL, NL, the gender effect was stronger at the juvenile stage than at the mature stage. Concerning DL, age had a relatively stronger effect at the mature stage. They revealed that males showed higher measurement values in almost all measured traits in all age classes.

Kim *et al.* (2013) detected the sexual dimorphism of the Korean water deer in cranial trait by the size of TL, CBL, and BL, which were similar to our results. However, they reported that the skull size of female subjects was larger than that of male animals, which is a unique pattern among mammals. According to the previous studies, the UTL, ZIB, DL, and TB of the skull were the most common parameters that

showed significant differences between genders or populations. The findings concerning TB, UTL, and DL are in line with the present study (And and Reig, 1993; Aragon *et al.*, 1998; Pételis and Brazaitis, 2003; Sheremetyeva and Sheremetyev, 2008; Sabalinkiene *et al.*, 2017).

Conclusion

This investigation revealed some morphological differences between the skulls of male and female PFDs. Moreover, measured parameters in the morphometric studies, similar to other investigated members of the Cervidae family, showed a significant difference between genders. The latter result might reveal the significant role of these parameters in gender detection.

Acknowledgments

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Conflict of Interest

The authors declared that there is no conflict of interest.

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تفاوت‌های مورفولوژیک جمجمه گوزن زرد ایرانی نر و ماده (*Dama dama mesopotamica*)

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زمینه مطالعه: گوزن زرد ایرانی (*Dama dama mesopotamica*)، یکی از نادرترین اعضای خانواده Cervidae در حال حاضر توسط اتحادیه جهانی حفاظت از طبیعت (IUCN) در لیست حیوانات در معرض انقراض قرار گرفته است.

هدف: در تحقیق حاضر تفاوت‌های مورفولوژیک جمجمه گوزن زرد ایرانی نر و ماده (*Dama dama mesopotamica*) بررسی شد.

روش کار: در ابتدا با همکاری اداره حفاظت از محیط زیست، پنج جمجمه نر و چهار جمجمه ماده این حیوان تهیه شد. پس از انجام روش‌های معمول تمیز کردن استخوان، نمونه‌ها از نظر اختلافات مورفولوژیک بررسی شدند. سپس ۲۹ پارامتر در جمجمه و فک پایین به وسیله کولیس دیجیتال برای مطالعات مورفومتریک اندازه‌گیری شد. علاوه بر این، روی تصاویر جانبی فک پایین از سمت چپ و تصاویر جانبی جمجمه از سمت چپ و تصاویر پشتی جمجمه به ترتیب ده، شش و نه نقطه تعریف و به کمک نرم‌افزار TpsDig2 به صورت دو بعدی تصویرسازی شد و تفاوت‌های شکلی بین دو جنس با استفاده نرم‌افزار MorphoJ بررسی گردید.

نتایج: همانطور که مشاهدات مورفولوژیک نشان داد، سوچر بین دو استخوان پیشانی در جنس نر به طور معنی‌داری بیشتر از ماده بود. علاوه بر این، تفاوت‌های معنی‌داری در برخی از پارامترهای اندازه‌گیری شده در جمجمه و فک پایین دو جنس مشاهده شد که بیشتر تفاوت‌ها مربوط به پارامترهای اندازه‌گیری شده در فک پایین بود. نتایج بررسی‌های مورفومتریک تفاوت معنی‌داری بین جنس‌ها نشان نداد.

نتیجه‌گیری نهایی: این پژوهش نشان داد که جمجمه گوزن زرد ایرانی نر و ماده در برخی ویژگی‌های مورفولوژیک تفاوت دارند.

واژه‌های کلیدی: گوزن زرد ایرانی، دوشکلی جنسی، سر، جمجمه، فک پایین