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Morphometric analysis of long bones (Femur and Tibia) of hamdani crossbred sheep fetuses (*Ovis aries*) in the second and third trimester: 3D model

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ABSTRACT

Objectives: In our study, it was aimed to obtain 3D models of femur and tibia bones using CT images in Hamdani crossbred sheep fetuses (second and third trimester) and to determine the developmental differences between sexes.

Materials-Methods: Fetuses of pregnant sheep slaughtered in Siirt province slaughterhouse were used. The gestational days of the fetuses were estimated using a previously determined formula. A total of 32 Hamdani crossbred sheep fetuses, 16 second trimester (8 females and 8 males) and 16 third trimester (8 females and 8 males) were used in the study. CT imaged the hind leg bones of the fetuses and the images were modeled in 3D Slicer software. Nine measurements were taken from the models. The measurements were evaluated statistically.

Results: In the second trimester, the FL parameter had a statistically significant difference between males and females ($p<0.01$). FDW parameter also showed a significant difference between males and females in this period ($p<0.05$). In the third trimester, the TPW measurement parameter had a highly significant difference between genders ($p<0.01$), and the ITDD parameter showed a significant difference between genders in the same period ($p<0.05$).

Conclusions: Osteometric studies are quite limited because bone development continues in the fetal period. In this sense, it is thought that the results of our research, which is the first 3D modeling study, will be useful in the fields of anatomy, zooarchaeology, taxonomy, obstetrics and gynecology, and surgery.

Keywords: *Femur, Hamdani crossbred sheep, morphometry, tibia, 3D modeling*

INTRODUCTION

Sheep breeding is an animal production activity with significant advantages in terms of cost and quality in Turkey (Aksoy and Yavuz, 2012). The short reproduction and adaptation periods of sheep, and their ability to utilize low-quality pastures effectively, reduce maintenance and feeding costs compared to cattle breeding (Dağıstan et al., 2008; Semerci and Çelik, 2016; Tamer and Sarıözkan, 2017). This situation provides sheep breeding to enterprises with small scale and low capital. In the eastern and southeastern provinces of Turkey (Hakkari, Van, Siirt, Batman, Bitlis) where different breeds are bred, the Hamdani sheep breed is a preferred breed (Örkiz et al., 1984). Considering the productivity characteristics of the breed, Hamdani crossbred sheep are preferred by the local people in Siirt province. Similar to general sheep breeds, the average gestation period in Hamdani crossbred ewes is 150 days (5 months), and growth and development are highest in the last 5 months of pregnancy (Harmeyer and Schlumbohm, 2006; Turgut et al., 2024).

Computed tomography is an imaging system in which cross-sectional images are obtained by rotating the detector and X-ray tube around the object (Adapınar, 2016). It is known that computed tomography images are converted into three-dimensional (3D) models through different programs. After the industry, medicine is also an area where 3D models are used (D'Urso et al., 1999). The accuracy and reliability of linear and angular measurements on 3D models have been clearly defined (Kim et al., 2012; Savio et al., 2016; Stull et al., 2014). There are studies in which morphometric characters of long bones were determined in sheep breeds that show great polymorphism within themselves (Alpak et al., 2009; Salami et al., 2011). In addition to these studies in adult animals, studies to be carried out in the fetal period are very important developmentally.

Most of the considered parameters such as shape, height, length, and size are easily accessible in the skeleton. This makes the skeletal system one of the body structures used in the characterization of both human and animal species (Watson, 1972; Guintard and Lallemand, 2003). Long bone structures, one of the parts of this skeletal system, can be used to determine interspecific differences and are also considered important in determining intraspecific differences (Rowley-Conwy, 1998). In previous studies, osteometric measurements were made on different bones and it was reported that the differences observed between the sexes would help determine gender (İşbilir and Güzel, 2023; Güzel and İşbilir 2024). In addition to the determination of sex characteristics, osteometric studies were carried out on different bones in different animal species, which will be useful for clinical sciences and zoo-archaeology (Dalga, 2021; Özüdoğru et al., 2023; Akçasız et al., 2024). For this reason, the osteometric properties of bones are of interest to zooarchaeologists.

In our study, we aimed to obtain 3D models of femur and tibia bones in Hamdani, crossbred sheep fetuses (second and third trimester) using CT images. The morphometric data obtained from these models will provide information about the developmental differences and will also be effective for the realization of statistical differences between sexes.

MATERIALS and METHODS

A total of 32 Hamdani crossbred sheep fetuses, 16 second trimester (8 females and 8 males) and 16 third trimester (8 females and 8 males) were used in the study. The fetuses were obtained from pregnant ewes slaughtered in the Siirt province slaughterhouse. The fetuses were removed from the uterus. The fetuses with a single pregnancy were included in the study, while the fetuses with twin pregnancies were excluded from the study. Fetal weights were determined with a scale. The gestation period was determined using the formula X=2.1(Y+17) (X=gestation period in days, Y=crown-anus length) (Noakes et al., 2001; Singh et al., 2023). Two separate groups were formed in the second trimester and third trimester. The gestation days of the fetuses used in the second trimester were estimated as 80-96 days and 105-125 days in the third trimester. No deformation was observed in the fetal bones. The bones were scanned by computed tomography. Using a 64-detector MDCT (General Electric Revolution) device, 80 kV, 200 MA, 639 mGY scan dose and protocol, 0.625 mm slice thickness was scanned. The images obtained were saved in DICOM format. Reconstructions were performed using 3D Slicer (5.0.2) software. Measurements from the obtained models were performed with reference to Bakici et al. (2021).

Second-trimester and third-trimester femur and tibia 3D model images and morphometric measurement points are presented in Figure 1 and 2.

The measurements taken were as follows:

Internal femoral diaphysis diameter (IIFD): Transverse diameter of the medullary cavity at the middle of the femur

Femoral proximal width (FPW): The maximum distance between the femoral head to the greater trochanter

Femoral head diameter (FHD): The maximum diameter at the middle of the femoral head

Femoral distal width (FDW): The maximum distance across the femoral condyles

Femoral length (FL): The maximum length of the femur

Internal tibial diaphysis diameter (ITDD): Transverse diameter of the medullary cavity at the middle of the tibia

Tibial proximal width (TPW): The maximum distance at the proximal articular surface

Tibial length (TL): The maximum length of the tibia

The results of morphometric measurements were statistically analyzed by SPSS 22.0 software. Independent samples t-test was used for gender comparison in the fetuses in the same period.

This study was approved by the Siirt University Animal Experiments Local Ethics Committee (2024/06/38).

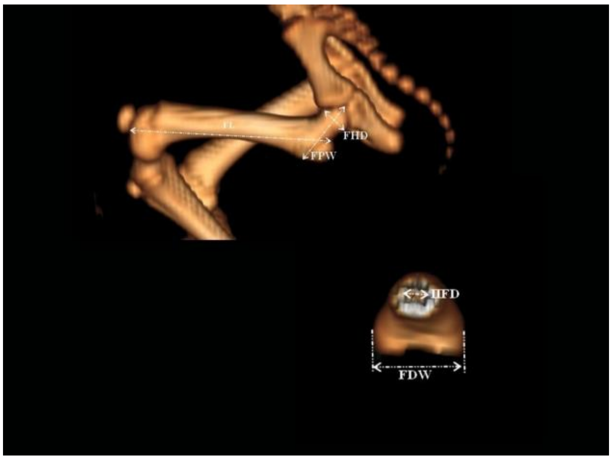

Figure 1. Osteometric measurement points of the femur in the sheep fetus.

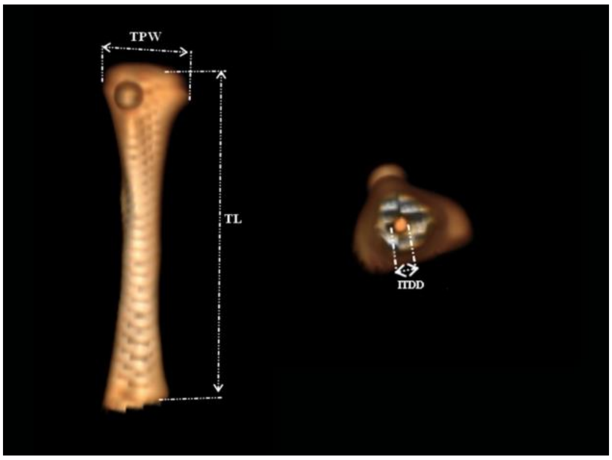

Figure 2. Osteometric measurement points of the tibia in the sheep fetus.

RESULTS

The first data obtained from the fetuses was the weight parameter. This value was determined as 284.42±276.77 g in second-trimester female fetuses and 312.30±270.52 g in males; 2255.71±549.44 g in third-trimester female fetuses and 2372.12±552.03 g in males. Osteometric measurement data were evaluated separately in two trimesters. Mean, standard deviation, and p value results of the measurement data according to gender are presented in Tables 1 and 2.

Table 1. Second trimester fetal femur and tibia osteometric measurement results (mm)

	Gender	n	Mean	Standard Deviation	P
IIFD	Male	8	0.719	0.580	NS
	Female	8	0.611	0.640	
FHD	Male	8	1.180	0.032	NS
	Female	8	1.085	0.041	
FPW	Male	8	1.896	0.052	NS
	Female	8	1.579	0.075	
FDW	Male	8	2.219	0.101	*
	Female	8	2.082	0.034	
FL	Male	8	7.942	0.061	**
	Female	8	7.744	0.177	
TL	Male	8	8.840	0.168	NS
	Female	8	8.281	0.123	
TPW	Male	8	1.921	0.071	NS
	Female	8	1.769	0.060	
ITDD	Male	8	0.357	0.034	NS
	Female	8	0.354	0.038	

NS: no significant; *: $p<0.05$; **: $p<0.01$

Table 2. Third trimester fetal femur and tibia osteometric measurement results (mm)

	Gender	n	Mean	Standard Deviation	P
IIFD	Male	8	0.863	0.331	NS
	Female	8	0.796	0.442	
FHD	Male	8	1.452	0.097	NS
	Female	8	1.267	0.049	
FPW	Male	8	2.339	0.128	NS
	Female	8	2.164	0.038	
FDW	Male	8	2.444	0.076	NS
	Female	8	2.312	0.106	
FL	Male	8	8.454	0.169	NS
	Female	8	8.292	0.074	
TL	Male	8	9.270	0.126	NS
	Female	8	9.184	0.096	
TPW	Male	8	2.419	0.056	**
	Female	8	2.222	0.101	
ITDD	Male	8	0.482	0.061	*
	Female	8	0.440	0.034	

In Table 2, it was observed that males had higher values than females in terms of femur and tibia bone lengths (TL and FL) in the third trimester, but the difference was not statistically significant. In the second trimester, the FL parameter had a statistically significant difference between males and females ($p<0.01$). FDW parameter also showed a significant difference between males and females in this period ($p<0.05$). In the third trimester, the TPW measurement parameter had a highly significant difference between genders ($p<0.01$), and the ITDD parameter showed a significant difference between genders in the same period ($p<0.05$). In the second and third trimesters, all of the statistically different values were found to be higher in male fetuses than in female fetuses.

DISCUSSION

Sheep and goats have become standard models for investigating biological responses during healing, understanding the mechanical conditions following injury, and investigating surgical treatments such as meniscal healing, ligament reconstruction, and cartilage repair (Bosch and Kasperczyk, 1992; Guisasola et al., 2002; Weiler et al., 2002; Field et al., 2003; Mora et al., 2003; Evans et al., 2004; von Rechenberg et al., 2004). For this purpose, there are studies in which long bone structures were examined in different animal species and with different techniques (Alpak et al., 2009; Kartal and Alpak, 2022; Özkadif and Halıgür, 2022; Marouf et al., 2024). However, no study was found in the literature on the fetal period.

In adult ewes, the FL parameter was analyzed by 3 different methods and reported as 204 mm (Marouf et al., 2024). The same value was reported as 188.15±4.720 mm in Morkaraman ewes (Alpak et al., 2009). In the measurements made in lambs, it was reported that the FL parameter was 134.5 mm in 1st week, 174.8 mm in 4th week, and 201.2 mm in the 8th week after birth (Eck et al., 2019). In our study, the FL parameter had higher values in male fetuses than in females in both gestational periods. While the FL parameter had a statistical difference between males and females in the second trimester, no statistical difference was determined in the third trimester.

Although distal femur width showed significant differences between genders in studies conducted in rabbits, no statistical differences were reported between laterality (dexter, sinister) and gender (Pazvant and Kahvecioğlu, 2009; Bakici et al., 2021). In addition, no statistical difference was reported between the right and left sides of New Zealand rabbits (Selçuk, 2023). In dogs, the FDW parameter was reported as 361.9±152.0 mm in males and 340.3±141.1 mm in females, while no statistical difference was observed between the sexes (Kartal and Alpak, 2022). In our study, it was determined that the FDW parameter was statistically different between the sexes in the second trimester ($p<0.05$). The distal width of the femur was higher in male fetuses than in female fetuses.

The tibial proximal width (TPW) value was found to be larger in male fetuses than in female fetuses in the third trimester. While this value was reported as 47.03±1.096 mm in the Morkaraman sheep breed (Alpak et al., 2009), 21.49±0.16 mm in Yankasa sheep and 19.10±0.29 mm in Red Sokoto goats, it was mentioned that there was a statistical difference between these two animal species (Salami et al., 2011). In our study, the ITDD parameter showed a statistically significant difference between male and female fetuses in the third trimester ($p<0.05$).

According to the results of the study, statistical differences were observed in the femur in second trimester fetuses and the tibia in third trimester fetuses. In the second trimester of pregnancy, the length and distal width of the femur were greater in male fetuses than in female fetuses. In addition, the proximal width of the tibia and the diameter of the internal tibial diaphysis were larger in male fetuses in the last period of pregnancy.

CONCLUSION

In conclusion, our study revealed the similarities and differences between the sexes in the morphometric measurements of femur and tibia in second and third trimester fetuses. The study was based on the principle of determining osteometric data by 3D modelling method from CT images. Although there are studies on this subject in adult animals, there are almost no studies in the fetal period. The data of the study will be a pioneer for developmental studies since the osteometric properties of the long bones of the lambs in the fetal period will be revealed. It is thought that the findings will contribute to studies in the field of obstetrics gynaecology and ultrasonography.

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