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Three-dimensional modelling of the femur and humerus in adult male guinea pigs (guinea pig) with computed tomography and some biometric measurement values

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Background: Guinea pig is a species belonging to the Caviidae family of the Rodentia order and is frequently used in experimental studies. Biomedical imaging methods are used in the diagnosis and treatment of many diseases in medicine. Among these methods, computed tomography (CT) is one of the most important imaging methods. In this study, it was aimed to perform the three-dimensional (3D) modelling of the CT images, obtained from the humerus and femur in the guinea pigs, via the MIMICS programme, and to make some biometric measurements regarding the bones over these models.

Materials and methods: In the present study, 12 male adult guinea pigs were used. The soft tissue on the humerus and femur bones of the guinea pigs was removed. After this procedure, CT images at a 0.5 mm-thickness were obtained from the animals. The images were recorded in DICOM format. Then, the reconstruction process was performed from the images by using the 3D modelling programme MIMICS® 13.1. On the 3D model of the humerus and femur (right-left), volumes, surface areas and lengths as well as other biometric parameters were measured separately, and the values were recorded. In addition, measurements of the bones were made with the help of a digital calliper.

Results: Among the parameters obtained from 3D models, a statistical difference was observed between the right and left cortical thicknesses of the femur from the measurements of calliper and the right and left humerus volumes (p < 0.05); whereas, no statistical difference was found in other parameters of both measurements (p > 0.05).

Conclusions: It can be stated that CT and 3D modelling can be used for the measurement of some parameters in the long bones of the guinea pigs. (Folia Morphol 2019; 78, 3: 588–594)

Key words: computed tomography, femur, humerus, guinea pig, three-dimensional reconstruction

INTRODUCTION

Guinea pig is a species belonging to the Caviidae family of the Rodentia order [22]. Guinea pigs that

are frequently used in the experimental studies such as immunology, toxicology, pharmacology, and physiology are native to South America and their wild ones are rodents that inhabit in Peru [41]. Their heads are quite large and their ears and tails are quite small [22]. They are herbivores. Adult guinea pigs have a body length of 27–33 cm and weigh of 700–1200 g [34]. They live for 2–8 years [41].

Bones, which are the passive elements of the locomotor system, are composed of organic and inorganic substances. In a guinea pig that has completed its growth, 1/3 of the bone is organic substances (ossein); whereas, 2/3 is inorganic substances (85% — calcium phosphate, 10% — calcium carbonate, magnesium phosphate, calcium fluoride). While the organic substances provide the elasticity of the bone, inorganic substances provide hardness and durability to the bone [12, 34]. Appendiculare skeleton is divided into two parts as bones of thoracic limb and pelvic limb. One of the bones of thoracic limb is the humerus. The longest bone of the body, which is included in the bones of the pelvic limb, is the femur [12, 26].

Morphometry is a research method which examines the shape differences of the objects and organisms and the correlations of the shape differences with other variations. This method is sometimes used to analyse the correlation between gender and growth factors of the species and sometimes the effects of the treatment applied [6, 36]. Conventional morphometry is the multiple variance statistical analysis that is performed with quantitative (length, width, height) variables [16]. Depending on the environmental components and the genetic structure, the differences in the growth of the living being can be easily revealed by morphology and morphometric studies [39].

Biomedical imaging methods are used in the diagnosis and treatment of many diseases in medicine [13, 40]. The working principle of computed tomography (CT), which is one of the medical imaging methods, is based on the detection of the non-absorbable part of the collimated X-rays passing through any part of the body in the tissues by the detectors as well as the conversion of the electrical signals into a cross-sectional image by means of computer operations [21]. With this method, the structures are displayed in a cross-sectional view and the images are much more detailed than the X-ray [1]. In recent years, CT has played an important role in the anatomical studies and in the correct diagnosis of the diseases in veterinary medicine. In addition to the diagnosis of diseases, CT is also used effectively in many biometric researches [13, 27, 28].

Three-dimensional (3D) formation of the transversal and longitudinal sections obtained from CT and other medical imaging methods is referred as reconstruction [2, 15, 32]. Thanks to this method, organisation and processing of the 3D information obtained by the reconstructions of the first cross-sectional images and enabling them to be presented in various shapes have become an acceptable method in medical sciences, especially in anatomy, surgical and clinical treatment [14, 20, 31, 38]. Anatomical details that cannot be observed in 2D images can be observed in more details with 3D reconstructed models. Today, MIMICS is one of the computer software used to create 3D models of two-dimensional (2D) images that are obtained from CT and magnetic resonance imaging methods. This software is a medical imaging and control system developed by Materialise (Mimics, Materialise Inc., Leuven, Belgium) for the 3D modelling of anatomical structures. One of the most important features that distinguishes MIMICS from other 3D modelling software is that it is a segmentation programme using Hounsfield values [3, 15, 18, 24, 31, 33].

In recent years, various studies have been conducted on the 3D modelling of anatomical structures in the field of veterinary anatomy. In a study conducted on the pelvic cavity of New Zealand rabbits, pelvis diameters and pelvic inclination were measured on a 3D model and evaluated in terms of gender. It has been suggested that the models obtained by the present study will contribute to the anatomical training and also in the diagnosis and treatment of diseases and to the knowledge of today's anatomy [30]. In addition, 3D modelling of penile urethra, stomach and intestine, respiratory system, paranasal sinus and antebrachium was performed in the rabbits [9, 10, 19, 29, 31]. In addition, a 3D model of the nasal airway was also performed in Nycticebus pygmaeus (Slow Ioris) [11]. 3D modelling of the thick filaments of the cardiac muscle in zebrafish, paranasal sinuses and diverticulum tubae auditivae in the horses and the nephron in the rat was done [4, 5, 8, 17]. As a result, it is seen that the 3D modelling method has started to be frequently used in the field of veterinary anatomy.

In the literature reviews, it was observed that CT and 3D modelling of the femur and humerus, which are the long bones of the body, in the guinea pigs, and the information regarding the measurements to be obtained over these models were limited [35, 40].

The purpose of the present study is to perform the 3D modelling of the CT images obtained from the humerus and femur in guinea pigs via the MIMICS programme and to calculate some of the biometric measurements of the bones such as volume, surface area and length over these models.

MATERIALS AND METHODS

In the study, 12 adult male guinea pigs were used. The study procedure was approved by the Ethics Committee of Selçuk University Faculty of Veterinary Medicine (09/03/2017, 2017/26). After the soft tissue on the humerus and femoral bones of the guinea pigs was removed, morphological differences were recorded. After this procedure, CT images were obtained from the animals at 0.5-mm thickness. The images were recorded in DICOM format. Then, the reconstruction process was performed from the images transferred to a portable external disk by using the MIMICS® 13.1 (The Materialise Group, Leuven, Belgium), a 3D modelling programme (Fig. 1).

In the reconstruction processes, the bone edges to be selected by using the "Thresholding" command from the "Segmentation" menu of MIMICS programme were automatically masked. Bones that could not be masked were corrected manually by the computer mouse by selecting the "Edit mask" command. After the manual correction process was completed, independent masking was reapplied to the selected bone by the "Region Growing" command. Finally, 3D modelling was performed by the "Calculate 3D" command. On the 3D model of the humerus and femur (right-left), their volumes, surface areas and lengths as well as other biometric parameters were automatically measured separately by using the "Info" command and the values were recorded. In addition, measurements of the bones were made by the help of a digital calliper. The diameter and cortical thickness of the humerus and femur were measured from the diaphysis section of the bones (Fig. 2).

Measurements performed on the bone (Figs. 3, 4):

- 1: maximum length of the humerus;
- 2: maximum length of the femur;
- 3: proximal width of the humerus;
- 4: proximal width of the femur;
- 5: distal width of the humerus;
- 6: distal width of the femur;
- 7: cortical thickness of the humerus;
- 8: cortical thickness of the femur;
- 9: medullary cavity diameter of the humerus;

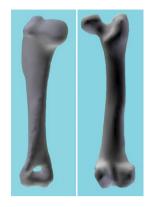


Figure 1. Three-dimensional models of the humerus and femur.



Figure 2. Humerus (left) and femur (right).

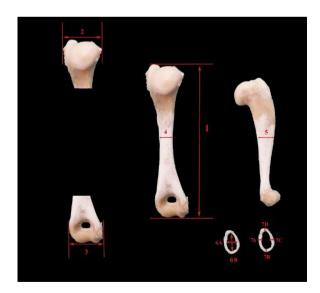


Figure 3. Measurement points taken from the humerus; 1 — maximum length of the humerus; 2 — proximal width of the humerus; 3 — distal width of the humerus; 4 — diaphysis diameter of the humerus (medio-lateral); 5 — diaphysis diameter of the humerus (cranio-caudal); 6 — medullary cavity diameter of the humerus [6A (mediolateral) + 6B (craniocaudal)/2]; 7 — cortex thickness of the humerus [7A (medial) + 7B (lateral) + 7C (caudal) + 7D (cranial)/4].



Figure 4. Measurement points taken from the femur; 1 — maximum length of the femur; 2 — proximal width of the femur; 3 — distal width of the femur; 4 — diaphysis diameter of the femur (medio-lateral); 5 — diaphysis diameter of the femur (cranio-caudal); 6 — medullary cavity diameter of the femur [6A (mediolateral) + 6B (craniocaudal)/2]; 7 — cortex thickness of the femur [7A (medial) + 7B (lateral) + 7C (caudal) + 7D (cranial)/4].

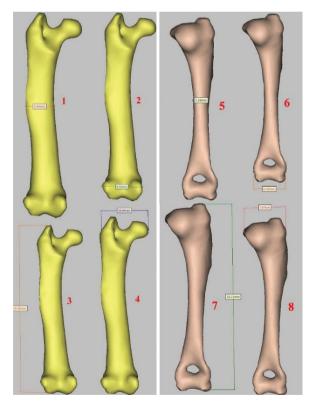


Figure 5. Measurements taken from the three-dimensional models of the femur and the humerus; 1 — diaphysis diameter of the femur (medio-lateral); 2 — distal width of the femur; 3 — maximum length of the femur; 4 — proximal width of the femur; 5 — diaphysis diameter of the humerus; 6 — distal width of the humerus; 7 — maximum length of the humerus; 8 — proximal width of the humerus.

- 10: medullary cavity diameter of the femur;
- 11: diaphysis diameter of the humerus;
- 12: diaphysis diameter of the femur.

On the 3D model, above mentioned parameters 7, 8, 9, and 10 could not be measured.

Anatomic terms used in this study were written by taking Nomina Anatomica Veterinaria as a reference [25].

Normality test was applied on the measurements taken on the 3D model and the morphometric measurements obtained from the bones via the MIMICS programme. While the normally distributed values were evaluated by the paired t test, the values that did not show normal distribution were evaluated by the Wilcoxon test (SPSS 22.0). The value of p < 0.05 was accepted as the statistically significant limit.

RESULTS

It was observed that the head of the humerus in the proximal extremity section of the shoulder joint with the scapula was quite large in guinea pig. Deltoid tuberosity was not significant. Radial fossa and olecranon fossa were associated with supratrochlear foramen.

It was observed that greater trochanter in the proximal extremity section of the femur was higher than the head of the femur and lesser trochanter was a circular crest. The presence of fovea of the head was determined on the articular surface of the femoral head. On the other hand, in the distal extremity section, each condyle had articular surfaces specific to articulation of the sesamoid bones.

Table 1 shows the measurement values performed on the 3D models of the humerus and femur in the study. While there was a statistical difference in the right and left humeral volume (p = 0.046) from the parameters obtained from 3D models, no difference was determined in other parameters (p > 0.05).

Table 2 shows measurement values of the humerus and femur measured with digital calliper. While a statistically significant difference was observed between the cortical thickness of the femur (p < 0.05), no difference was found in other parameters (p > 0.05).

DISCUSSION

While the guinea pig is being fed as an exotic animal, it is an animal model that is used in many experimental studies in the field of health [41]. The humerus and the femur are the two long bones that are frequently preferred in morphometric studies [23]. CT and 3D modelling are currently used in the

Table 1. Measurement results performed on the three-dimensional models of the humerus and femur in adult male guinea pigs

Parameters	Right	Left	Р
Maximum length of the humerus [mm]	32.22 ± 1.16	32.15 ± 0.85	> 0.05
Maximum length of the femur [mm]	38.75 ± 1.65	38.61 ± 1.83	> 0.05
Proximal width of humerus [mm]	7.72 ± 0.74	7.79 ± 0.41	> 0.05
Proximal width of the femur [mm]	10.69 ± 0.89	9.74 ± 0.61	> 0.05
Distal width of the humerus [mm]	6.28 ± 0.59	6.17 ± 0.30	> 0.05
Distal width of the femur [mm]	8.16 ± 0.38	7.80 ± 0.41	> 0.05
Diaphysis diameter of humerus (lateral-lateral) [mm]	2.89 ± 0.4	3.10 ± 0.30	> 0.05
Diaphysis diameter of the femur (cranio-caudal) [mm]	4.07 ± 0.33	4.16 ± 0.40	> 0.05
Diaphysis diameter of the femur (lateral-lateral) [mm]	5.03 ± 0.49	5.09 ± 0.55	> 0.05
Diaphysis diameter of the humerus (cranio-caudal) [mm]	3.78 ± 0.40	4.11 ± 0.30	> 0.05
Surface area of the humerus [mm²]	515.74 ± 59.63	597.49 ± 140.24	> 0.05
Surface area of the femur [mm²]	823.88 ± 84.70	802.54 ± 158.88	> 0.05
Volume of the humerus [mm³]	447.70 ± 52.70	601.85 ± 183.50	< 0.046
Volume of the femur [mm³]	770.12 ± 148.18	853.84 ± 85.88	> 0.05

Data are shown as mean \pm standard deviation.

Table 2. Morphometric measurement results of the humerus and femur in adult male guinea pigs

Right	Left	Р
32.92 ± 0.83 (32.78)	32.88 ± 0.63 (32.70)	> 0.05
39.66 ± 1.47 (39.52)	39.48 ± 1.70 (39.36)	> 0.05
8.23 ± 0.52	8.36 ± 0.34	> 0.05
10.00 ± 0.30	9.92 ± 0.17	> 0.05
$7.01 \pm 0.52 (6.92)$	7.05 ± 0.66 (6.77)	> 0.05
$8.40 \pm 0.20 (8.34)$	$8.40 \pm 0.40 (8.37)$	> 0.05
2.91 ± 0.16	2.95 ± 0.20	> 0.05
4.11 ± 0.28	4.13 ± 3.23	> 0.05
5.11 ± 0.43	5.22 ± 0.35	> 0.05
3.95 ± 0.30	3.91 ± 0.17	> 0.05
0.67 ± 0.09	0.74 ± 0.15	> 0.05
0.84 ± 0.11	0.76 ± 0.10	< 0.039
2.49 ± 0.46	2.30 ± 0.36	> 0.05
3.36 ± 0.25	3.23 ± 0.35	> 0.05
	$32.92 \pm 0.83 (32.78)$ $39.66 \pm 1.47 (39.52)$ 8.23 ± 0.52 10.00 ± 0.30 $7.01 \pm 0.52 (6.92)$ $8.40 \pm 0.20 (8.34)$ 2.91 ± 0.16 4.11 ± 0.28 5.11 ± 0.43 3.95 ± 0.30 0.67 ± 0.09 0.84 ± 0.11 2.49 ± 0.46	$32.92 \pm 0.83 (32.78) \qquad 32.88 \pm 0.63 (32.70)$ $39.66 \pm 1.47 (39.52) \qquad 39.48 \pm 1.70 (39.36)$ $8.23 \pm 0.52 \qquad 8.36 \pm 0.34$ $10.00 \pm 0.30 \qquad 9.92 \pm 0.17$ $7.01 \pm 0.52 (6.92) \qquad 7.05 \pm 0.66 (6.77)$ $8.40 \pm 0.20 (8.34) \qquad 8.40 \pm 0.40 (8.37)$ $2.91 \pm 0.16 \qquad 2.95 \pm 0.20$ $4.11 \pm 0.28 \qquad 4.13 \pm 3.23$ $5.11 \pm 0.43 \qquad 5.22 \pm 0.35$ $3.95 \pm 0.30 \qquad 3.91 \pm 0.17$ $0.67 \pm 0.09 \qquad 0.74 \pm 0.15$ $0.84 \pm 0.11 \qquad 0.76 \pm 0.10$ $2.49 \pm 0.46 \qquad 2.30 \pm 0.36$

Data are shown as mean \pm standard deviation and (median).

diagnosis, and treatment of many diseases [13, 15, 20]. In addition, bone morphometric measurements are made with the help of the digital calliper [23]. In the present study, the humerus and femur bones of 12 adult male guinea pigs were modelled as 3D and various parameters were measured. Some morphometric parameters of the same bones that were measured by using a digital calliper were also evaluated.

In the present study, it was determined that while a statistical difference was observed in the left and right

humeral volume parameters obtained from the data of 3D models (right: 447.70; left: 601.85, p = 0.046), no difference was observed in other parameters (maximum length of the humerus [mm] right: 32.22; left: 32.15, maximum length of the femur [mm] right: 38.75; left: 38.61, proximal width of the humerus [mm] right: 7.72; left: 7.79, proximal width of the femur [mm] right: 10.69; left: 9.74, distal width of the humerus [mm] right: 6.28; left: 6.17, distal width of the femur [mm] right: 8.16; left: 7.80, diaphysis diameter

of the humerus (lateral-lateral) [mm] right: 2.89; left: 3.10, diaphysis diameter of the humerus (cranio-caudal) [mm] right: 4.07; left: 4.16, diaphysis diameter of the femur (lateral-lateral) [mm] right: 5.03; left: 5.09, diaphysis diameter of the femur (cranio-caudal) [mm] right: 3.78; left: 4.11, surface area of the humerus [mm²] right: 515.74; left: 597.49, surface area of the femur [mm2] right: 823.88; left: 802.54, volume of the femur [mm³] right: 770.12; left: 853.84) (Table 1, p > 0.05). In the literature reviews, several studies in which CT was used in guinea pigs were accessed [7, 37, 42], a limited number of studies investigating the humerus and femur were accessed. The humerus and femur lengths calculated by Witkowska et al. [40] in 1-year-old guinea pigs via this model were similar to the present study. Pazvant [35] stated that the difference between the maximum lengths of the right and left humerus and the anterio-posterior diameter of the diaphysis was significant in the measurement results of the CT images of the guinea pig. Pazvant [35] reported that the difference observed in results of the measurement performed between medio-lateral and anterio-posterior diameters of the diaphysis of the femur on CT images of guinea pig was statistically significant; whereas, no such difference was determined in measurements performed on bone and model as a result of the study. The differences between the results of the study may be caused by the difference in methodology.

Among the measurements performed by means of the digital calliper in the present study, a statistically significant difference was found between right (0.84 mm) and left (0.76 mm) cortical thickness of the femur (p < 0.05); whereas, no difference was found in other parameters (maximum length of the humerus [mm] right: 32.92; left: 32.88, maximum length of the femur [mm] right: 39.66; left: 39.48, proximal width of the humerus [mm] right: 8.23; left: 8.36, proximal width of the femur [mm] right: 10.00; left: 9.92, distal width of the humerus [mm] right: 7.01; left: 7.05, distal width of the femur [mm] right: 8.40; left: 8.40, diaphysis diameter of the humerus (lateral-lateral) [mm] right: 2.91; left: 2.95, diaphysis diameter of the humerus (cranio-caudal) [mm] right: 4.11; left: 4.13, diaphysis diameter of the femur (lateral-lateral) [mm] right: 5.11; left: 5.22, diaphysis diameter of the femur (cranio-caudal) [mm] right: 3.95; left: 3.91, cortex thickness of the humerus [mm] right: 0.67; left: 0.74, medullary cavity diameter of the humerus [mm] right: 2.49; left: 2.30, medullary cavity diameter of the femur [mm] right: 3.36; left: 3.23) (Table 2, p > 0.05). In the studies conducted between the sexes in guinea pigs, it has been specified that there is no difference in the maximum length of the right and left humerus on the direct osteometric and radiography images and the right and left values in all measurements of the femur [34, 35] and the femur and humerus lengths in both genders of nandrolone-administered rats [23].

CONCLUSIONS

In conclusion, it was determined that among the data obtained from 3D modelling, there was a statistical difference in the right and left humeral volume parameter and also in the right and left cortical thickness from the measurements performed by the digital calliper; whereas, no statistical difference was observed in other parameters. It can be asserted that CT can be used in the modelling of both bones; however, 3D modelling in some parameters in small bones and micro CT in measurements may be more beneficial.

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