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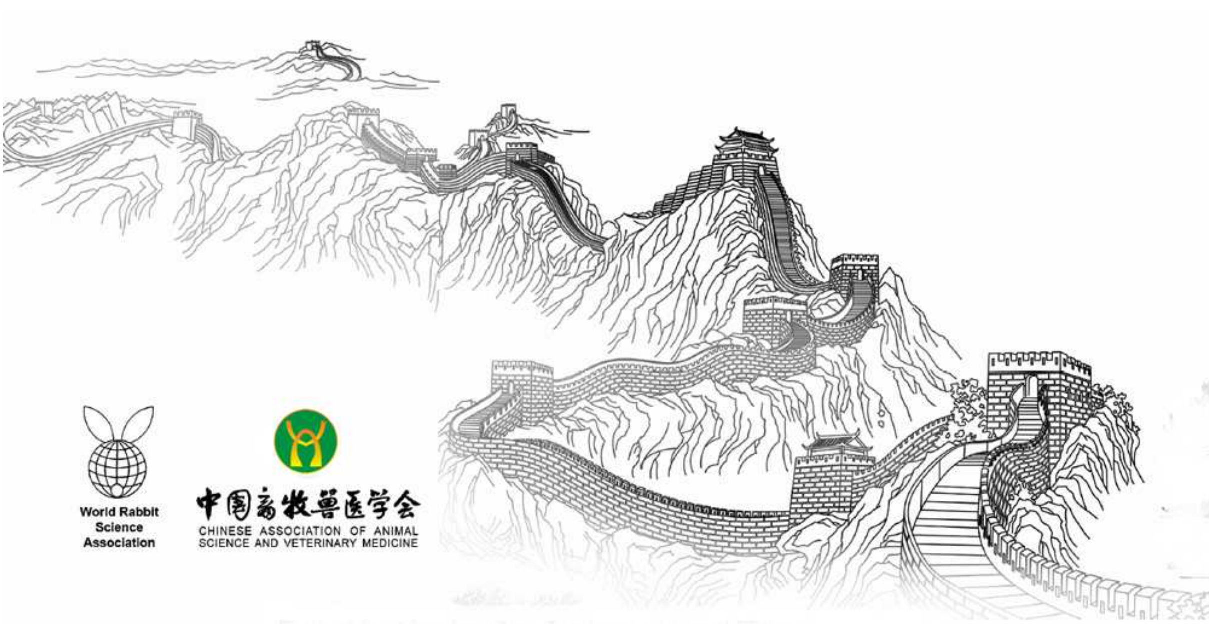

TOTAL BODY FAT CONTENT DETERMINATION BY MEANS OF COMPUTED TOMOGRAPHY (CT) IN RABBITS

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ABSTRACT

The aim of this paper was to describe a volumetric fat measurement method of rabbits *in vivo* by means of computed tomography (CT). Former studies estimated the whole body, the perirenal and scapular fat content accurately. In these studies, a few scans were used at reference sections for the estimation of fat content. Nowadays, thin slices based reconstruction is possible from the whole body using new generation CT scanners. In our study, the effect of the slice thickness was evaluated for the estimation of the dissected scapular and perirenal fat content. A morphologic method was used to segment out the non-fat voxels caused by partial volume effect. The 2 mm thin slice thickness with morphologic correction proved to be the most accurate method among the tested methods for evaluation of the fat content of the rabbits *in vivo*. The Pearson correlation coefficient and the partial correlation between the estimated fat volume of total body by CT and dissected scapular and perirenal fat weight were 0.68 and 0.49, respectively.

Key words: Rabbit, Fat content, Computed Tomography, Automated segmentation

INTRODUCTION

It is widely known that the body condition is an important property, especially for reproductive performance and lifespan of the female animals (Kasza *et al.* 2016a, b; Zomeño *et al.* 2013). For evaluation of the body condition, the most reliable methods are the experimental slaughter and chemical analysis, but these methods make the breeding of the individuals impossible or the ultrasound measurement which has moderate reliability (Pascual *et al.*, 2004).

Due to its non-invasive nature, the use of CTs enables the examination of alive animals, it is less time-consuming and reproducible than dissection based studies. For this reason several previous experiments proved the precision of *in vivo* CT methods for the determination of the fat tissue amount (Romvári *et al.*, 1996; Romvári *et al.*, 1998). In these former studies, the basis of the estimation was a pre-defined number of CT scans reconstructed on identical anatomical locations. Nowadays, the developed CT technology allows the quick data collection and reconstruction based on overlapped slices from the whole body. Szendrő *et al.* (2012) used 10 mm slice thickness in the selection program of Pannon White rabbits for the estimation of hind leg muscle volume. Due to the fat tissue distribution of the body and the small sizes of the fat depots may require to increase the resolution of the CT imaging.

Based on the results of the previous experiments and the statements above, our aim was to develop a new CT measurement protocol and 3D automated post-processing method for *in vivo* determination the body fat volume of rabbits.

MATERIALS AND METHODS

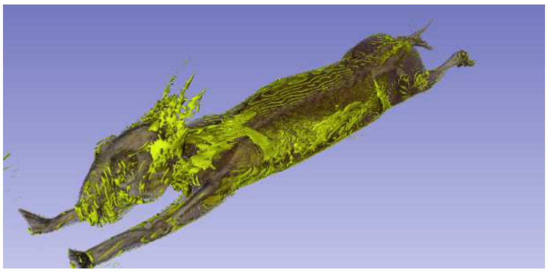
Animals and CT examinations

The investigation was carried out on 272 Pannon White growing rabbits at 11 weeks of age. The rabbits were housed in fattening cages (2-3 rabbits per cage). After weaning at 5 weeks of age all rabbits were fed *ad libitum* with a commercial pellet and water was offered *ad libitum* from nipple drinkers. Both genders were enrolled in the study. CT examinations of the animals were applied using Siemens Sensation Cardiac CT situated at the Institute of Diagnostic Imaging and Radiation Oncology (Kaposvár University) using the following settings: tube voltage: 120kV, current: 140mAs, data collection mode: spiral. During the CT scanning procedures rabbits were fixed with belts in a special plastic container, without using any anaesthetics. Three animals were scanned simultaneously. The CT measurements consisted of overlapping 10 and 2 mm thick slices covering the whole body.

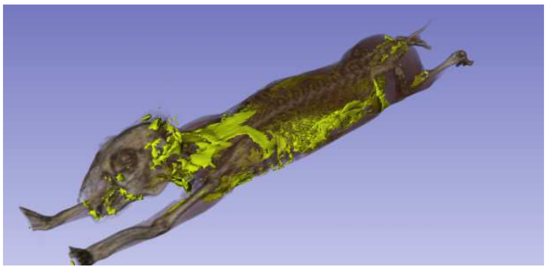
Post-processing of CT scans

After the separation of the simultaneously scanned individuals from the images and removing the troughs (plastic container image regions), the volumetric information was evaluated using a simple threshold method by OpenIP software (Kovács *et al.*, 2010), where the -200 to -20 HU range was considered as fat tissue. We analysed the data reconstructed to 2 mm and 10 mm slice thicknesses. The total body fat volume was determined from the corrected 3D data.

As it was mentioned above, scans with 10 mm slice thickness were used before in the selection work of Pannon White rabbits (Szendrő *et al.*, 2012), therefore this was the initial slice thickness. The small sizes of the fat regions seemed to need a higher resolution of the CT scans. The 2 mm thin slices seemed to have appropriate resolution, but there were much more partial volume effects predominantly on the border of the skin and air. This detrimental effect was decreased by morphologic (erosion) tools (Kovács *et al.*, 2010) as the Picture 1 shows.



(A) without correction



(B) with morphologic correction

Picture 1: The segmented fat volume visualized with yellow colour using 3D Slicer program (Fedorov *et al.*, 2012).

Test slaughter

On the day following the CT examinations, the rabbits were slaughtered at a commercial abattoir. The weights of scapular and perirenal fat were removed and measured during the dissection process according to Blasco and Ouhayoun (1996).

Statistical Analysis

Data of scapular and perirenal fat weights at slaughter and calculated total body fat volumes from CT images were analysed by Pearson correlation and partial correlation analysis (controlled by live weight) using R statistic software version 3.2.0 (R Core Team, 2013).

RESULTS AND DISCUSSION

The former studies examined the correlation of the fat content of the whole body determined by chemical analysis and the amount of fat calculated by histogram based evaluation of the CT scans (Romvári *et al.*, 1996; Romvári *et al.*, 1998). Other examination used the weight of the perirenal fat depots as reference against the segmented fat volumes of the determined region on the scans (Yonkova *et al.*, 2010). In this study, our aim was to estimate the whole body fat content. It was not possible to perform chemical analysis, therefore the total body fat volumes were segmented from the CT scans and the reference values were the weights of the dissected perirenal and scapular fat depots. In this case, we could not expect high correlation coefficients, but the result allows choosing the principally adequate method for the selection work.
Thin slice thickness resulted higher correlation with the fat weights and the morphologic correction also increased the accuracy (Table 1).

Table 1: The Pearson correlation coefficients between the estimated fat volumes of total bodies by CT and dissected scapular and perirenal fat weight ($P<0.05$).

Fat volume of total body measured by CT	Perirenal fat weight	Scapular fat weight	Perirenal + scapular fat weight
10 mm slice thickness	0.483	0.586	0.578
2 mm slice thickness	0.518	0.619	0.616
10 mm slice thickness with morphologic correction	0.524	0.623	0.622
2 mm slice thickness with morphologic correction	0.579	0.667	0.679

It is a well-known that the live weight may strongly influence the correlation; the partial correlation coefficients were calculated controlling for live weight. The partial correlation coefficients showed the same trend as the Pearson correlation coefficients (Table 2).

Table 2: The partial correlation coefficients between the estimated fat volumes of total bodies by CT and dissected scapular and perirenal fat weight ($P<0.05$) controlled by the live weights

Fat volumes of total bodies measured by CT	Perirenal fat weight	Scapular fat weight	Perirenal + scapular fat weight
10 mm slice thickness	0.276	0.349	0.350
2 mm slice thickness	0.315	0.385	0.394
10 mm slice thickness with morphologic correction	0.336	0.411	0.421
2 mm slice thickness with morphologic correction	0.400	0.455	0.488

The dissection of the perirenal fat is more complicated than that of the scapular fat, accordingly the scapular fat weights resulted higher correlation coefficients than only the perirenal fat.

CONCLUSIONS

Based on the results of the correlation analysis, the 2 mm thin slice thickness with morphologic correction proved to be the most accurate method among the *in vivo* tested methods for evaluation of the perirenal and scapular fat content of the rabbits.

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