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Ultrasonography in the diagnosis of obesity in cats and its correlation with fructosamine and lipid levels

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ABSTRACT

The aim of this study was to determine whether ultrasound imaging is an efficient method to assess subcutaneous fat in cats, the anatomical sites where more significant fat deposition occurs, and if there is a correlation between subcutaneous fat thickness and serum levels of cholesterol, triglycerides, and fructosamine. A total of 30 healthy adult cats were used and divided into three groups of 10 animals each, based on the estimated body condition score (BCS). The ideal group (IG) included cats with BCS 3; the overweight group (OWG), with BCS 4; and the obese group (OG), with BCS 5. Ultrasonographic measurement of subcutaneous fat was conducted in five anatomical regions: lumbar, abdominal, thoracic, femoral, and pectoral. We observed that obese cats had greater fat deposition in the abdominal and thoracic regions when compared to those with ideal weight, and that cholesterol and triglyceride levels were higher with the increase in subcutaneous fat thickness in the thoracic region. Nonetheless, there were no differences in fat deposition in the OWG compared to cats from the IG and OG. Ultrasonography made it possible to associate cholesterol and triglyceride levels with local fat deposits and to differentiate obese cats from those with ideal weight by analyzing the thickness of subcutaneous fat in the abdominal and thoracic regions, making this method efficient and less subjective.

1. Introduction

Obesity is defined as a positive energy balance disorder, where an excessive accumulation of body fat occurs, and is established when the animal exceeds 20% of its ideal body weight (Little, 2012; Jericó et al., 2015). The disease is commonly diagnosed in routine clinical practice. In Brazil, one study carried out with 50 cats evidenced a prevalence of 14% of obesity/overweightness (Mendes-Junior et al., 2013). In New Zealand, 23,794 cats had their body condition score (BCS) evaluated, and the authors found that 21.9% of the animals were overweight and 2.6% were obese (Gates et al., 2019).

This disorder can reduce the quality of life and survival of animals by compromising normal organic functions (Little, 2012; Jericó et al., 2015) since it produces a chronic inflammatory condition through the release of inflammatory cytokines by adipocytes, which predisposes the development of some diseases (Cline et al, 2021). Thus, the maintenance of lean muscle and ideal weight is essential for the cat's health and they are determining factors for endocrine homeostasis, especially for normal lipid and glucose metabolism (Christopher, 2018).

Obesity can cause an increase in serum lipid concentrations compared to cats within the ideal weight, rendering this parameter a suitable indicator for its assessment (Chala et al., 2021). Also, the disorder can induce a state of insulin resistance, triggering an increment in glucose levels (Clark; Melissa, 2021), which, if sustained for more than 3 to 4 days (Link and Rand, 2008), can lead to increases in fructosamine levels, which is not influenced by hyperglycemic states resulting from acute stress (Clark; Melissa, 2021).

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Methods for assessing obesity in cats are varied and seek to estimate the amount of body fat. Some of these techniques, such as the evaluation of body weight, determination of the BCS, and morphometric measurements, are low-cost and easy to perform. However, due to their subjective character, they do not allow such accurate assessments. Imaging tests are also used to diagnose obesity and provide more accurate results, including dual-energy x-ray absorptiometry, computerized tomography, and magnetic resonance imaging. Nevertheless, these tests are more costly and are not easily available in routine clinical practice (Santarossa et al., 2017).

In dogs, Payan-Carreira et al. (2016) showed that ultrasonography is a sensitive method to detect small variations in subcutaneous fat when compared to the BCS, providing more reliable and accurate values with minimal changes. Thus, ultrasound imaging can enable the assessment of the degree of obesity in a less subjective and more accessible than other diagnostic tests in cats.

Therefore, the aim of the present study was to determine whether ultrasonography is an efficient method to assess subcutaneous fat in cats, as well as the anatomical sites where greater fat deposition occurs and if there is a correlation between subcutaneous fat thickness and serum levels of cholesterol, triglycerides, and fructosamine.

2. Material e Methods

This study was approved by the Ethics Committee on the Use of Animals of the Institution where it was performed under protocol number 002/2018, and included a total of thirty domiciled adult cats. Their owners were informed regarding the purpose of the study and signed a free and informed consent form.

Animal inclusion was carried out with previous anamnesis and a complete physical examination. Blood sample was also collected for the hematological and biochemical analyses of the following parameters: complete blood count, alanine aminotransferase, alkaline phosphatase, albumin, total protein, urea, creatinine and glucose. Animals that showed alterations during selection, suggestive of clinical disease, were excluded from the study. The selected cats were also weighed on a calibrated electronic scale (Blue One 300®, São Paulo, Brasil).

The groups were divided based on the BCS scale of 1 to 5 (LaFlamme, 1997), where BCS 1 represents cachectic animals and BCS 5 obese animals. The evaluation was conducted by inspecting the cats' abdominal silhouette and palpating the ribs, lumbar vertebrae, and iliac wing. In the end, the cats were divided into three groups, each containing 10 animals.

Group I (IG) consisted of cats within the ideal weight (BCS 3), with seven females and three males, and nine of which were neutered. The animals had a mean weight of 3.85 ± 0.49 Kg and aged 12 to 72 months (34.6 ± 17.93 months). Meanwhile, OWG comprised overweight cats (BCS 4), with six females and four males, all of which were neutered, a mean weight of 4.85 ± 0.75 Kg, and age between 18 and 84 months (52.10 ± 22.36 months). Lastly, Group O (OG) included obese cats (BCS 5), with four females and six males (all of which were neutered) with an average weight of 6.2 ± 0.98 Kg and age ranging from 24 to 84 months (58.8 ± 18.29 months). Each cat was submitted to a biochemical evaluation and ultrasound imaging of subcutaneous fat thickness.

Prior to blood collection, the cats fasted for 12 hours. During sampling, the animals were physically restrained, underwent local trichotomy and antisepsis and sustained puncturing of the jugular or cephalic vein with the aid of a sterile disposable syringe and needle. From 3 to 5 mL of blood were collected, which were placed in tubes without anticoagulant for later centrifugation and obtention of serum for determining the dosages of total cholesterol, triglycerides, and fructosamine. The laboratory tests were performed using conventional techniques and commercial kits.

The ultrasound images were always taken by the same experienced operator, using ultrasound equipment Phillips HD5 (Phillips Healthcare, United States) coupled with a linear transducer at a frequency of 7.5 to 12 MHz. The images were acquired with the cat in the right lateral position and obtained from five anatomical regions (Figure 1), previously determined in dogs, as described by Payan-Carreira et al. (2016). The anatomical regions were:

- 1. Pectoral Region: at the entrance of the thorax, from the midline towards the left side, with the transducer transversally positioned in relation to the manubrium of the sternum, over the cleidocephalic muscle;
- 2. Thoracic Region: on the cat's left side, on the ninth intercostal space, just above the costochondral junction, with the transducer transversally positioned in relation to the ribs on the latissimus dorsi muscle;
- 3. Abdominal region: on the left lateral wall of the abdomen, with the transducer in a vertical position on the external abdominal oblique muscle;
- 4. Femoral Region: in the inner portion of the right thigh, with the transducer placed transversally in relation to the femur, between the gracilis and semimembranosus muscles;
- 5. Lumbar Region: between the third and fifth lumbar vertebrae, on the longissimus dorsi muscle, 2 to 3 cm to the left of the midline, with the transducer parallel to the spinous process of the lumbar vertebrae.

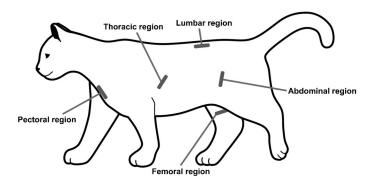


Figure 1 – anatomical representation of the regions used to evaluate the ultrasound images of subcutaneous fat in cats. Fonte: Author illustration

Image measurements were performed using the RadiAnt DICOM Viewer (64-bit) program. In each analyzed region, three measurements were made, with one at the central point and the others laterally equidistant by 1 cm. The mean of these measurements was calculated to obtain the subcutaneous fat thickness value of each anatomical region.

A descriptive statistical analysis was conducted with the calculation of the mean, standard deviation and coefficient of variation (CV) for the analyzed parameters. The subcutaneous fat thickness values were submitted to the Shapiro-Wilk normality test. The means of subcutaneous fat thickness were compared between groups by Analysis of Variance and Tukey's post-hoc; the means of the biochemical measurements were compared using the Friedman test. Pearson's correlation test was used to compare the value obtained from the thickness of te subcutaneous fat layers and the BCS with serum levels of cholesterol, triglycerides, and fructosamine.

3. Results

The mean values, standard deviation and CV of the subcutaneous fat thickness of the cats with different BCS are shown in Table 1. The lowest CV among the evaluated anatomical sites was found in the abdominal region and the highest in the pectoral and thoracic regions.

The subcutaneous fat thickness values in the thoracic and abdominal regions were significantly higher in cats in the OG than those in the IG, however, the fat layer thickness in these regions in the overweight cats (OWG) did not differ statistically compared to the other groups (Figure 2). The other anatomical sites did not show significant differences between groups.

Although there was no difference in subcutaneous fat thickness between groups considering the five anatomical areas evaluated, the lumbar region was where the greatest deposition of fat was observed in the three groups, with the IG differing significantly regarding the pectoral, thoracic, and femoral regions. In the OG, the values differed significantly considering the pectoral and femoral regions and the lumbar region differed from all the others in the OWG (Table 1).

The means, standard deviations and CV of the serum concentrations of total cholesterol, triglycerides, and fructosamine are shown in Table 2. No significant difference was found between the groups.

A positive correlation was observed between subcutaneous fat thickness and levels of cholesterol (r=0.39; p=0.02) and triglycerides (r=0.41; p=0.02), that is, the higher the levels of cholesterol and triglycerides, the greater the thickness of the fat in this region. However, this correlation was not observed in the other anatomical regions, nor was there an association between fructosamine levels and thickness in the different regions. Yet, there was a positive correlation of the thoracic (r=0.48; p=0.007), lumbar (r=0.39; p=0.02), and abdominal (r=0.57; p=0.001) regions with the different BCSs.

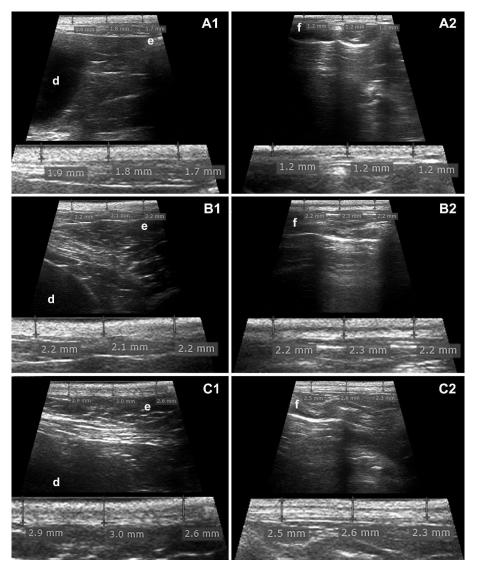


Figure 2 – Ultrasonographic evaluation of subcutaneous fat thickness in cats in the abdominal and thoracic regions considering the different groups of BCS, represented by 1 (abdominal regions); 2 (thoracic regions); A (ideal weight); B (overweight); C (obese); d (bladder); e (external abdominal oblique muscle); f (latissimus dorsi muscle).

Anatomical region	IG		owg		OG		l
	mean ± SD	CV (%)	mean ± SD	CV (%)	mean ± SD	CV (%)	_ p-value
Pectoral	1.44 ± 0.4^{Aa}	28.31	1.41 ± 0.48 ^{Aa}	34.25	1.31 ± 0.60^{Aa}	46.12	0.83
Thoracic	1.27 ± 0.45^{Aa}	35.23	$1.87 \pm 0.53^{\text{ABa}}$	28.56	$2.11 \pm 0.90^{\text{Ba}}$	42.46	0.02
Lumbar	2.21 ± 0.52^{Ab}	23.73	2.60 ± 0.55^{Ab}	21.22	2.89 ± 0.88^{Ab}	30.62	0.09
Abdominal	$1.79\pm0.35^{\mathrm{Ab}}$	19.39	2.15 ± 0.36^{ABb}	16.84	$2.43 \pm 0.46^{\text{Bb}}$	18.98	0.005
Femoral	1.16 ± 0.4^{Aa}	35.03	1.33 ± 0.52^{Aa}	39.30	1.46 ± 0.55^{Aa}	37.87	0.59

Table 1 – Means, standard deviations (SD) and coefficient of variation (CV) of the subcutaneous fat thickness in different anatomical sites measured by ultrasonography in cats within the ideal BCS (IG), overweight (OWG) and obese cats (OG). The measurements are shown in millimeters.

Legend: IG – ideal group; OW; G – overweight group; OG – obese group. The superscript letters represent the statistical difference between regions (lowercase letters – columns) and between BCS groups (uppercase letters – lines).

Biochemicals	IG		OWG		OG		р-
	mean ± SD	CV (%)	mean ± SD	CV	mean ± SD	CV value	value
				(%)	mean ± SD		
Cholesterol	116.7 ± 26.67^{a}	22.85	129.10 ± 33.77^{a}	26.15	158.4 ± 75.17^{a}	47.46	0.14
Triglycerides	95 ± 55.72^{a}	58.65	138.60 ± 174.24^{a}	124.27	331.11 ± 586.02^{a}	176.99	0.12
Fructosamine	299.5 ± 52.98^{a}	17.69	293.90 ± 29.88^{a}	10.17	312.11 ± 36.80^a	11.12	0.20

Table 2 – Means, standard deviations (SD), and coefficient of variation (CV) of the serum concentrations of cholesterol, triglycerides, and fructosamine in cats within the ideal weight (IG), overweight (OWG) and obese (OG).

Legend: IG – ideal group; OWG – overweight group; OG – obese group. The superscript letters represent the statistical difference between BCS groups.

4. Discussion

Ultrasound imaging is considered an accessible, easy to apply, and low-cost method when compared to other imaging techniques. In addition, there was good acceptance by the cats, which favored its use in this study. Ultrasonography has the advantage of not emitting radiation like other possible diagnostic methods, rendering it a preferred technique for the long-term monitoring of animals undergoing treatment for weight reduction.

In this study, it was possible to determine that the thoracic region is the most prone to fat accumulation in obese animals, complementing the BCS classification of the corporal score proposed by LaFlamme (1997), since the ultrasonography allowed to precisely measure the thickness of the fat in this region. Ultrasound imaging of the thoracic region was also performed by Iwazaki et al. (2018) as a way of analyzing subcutaneous fat deposit in cats. Interestingly, the authors did not evaluate animals with distinct BCS, as done in our study, but used young and elderly animals, as well as two thoracic regions for the assessment. Although there is no standardized technique for the species, the results of our study indicate that the thoracic region is an important fat deposition site in cats, corroborating with Iwazaki et al. (2018).

In obese dogs, which underwent the same evaluation method as that applied in the current study, a more significant deposition of fat was also observed in the abdominal region, corroborating our results, as well as in the lumbar region (Payan-Carreira et al., 2016). Thus, it can be inferred that the abdominal region is possibly a location of greater fat accumulation in both species. The differences found in fat deposition in the thoracic (in cats) or lumbar (in dogs) regions may be inherent characteristics of the species, indicating that cats have a greater tendency to accumulate fat in the thorax, and dogs, in the lumbar region.

The abdominal and thoracic anatomical sites were found to be the areas of more pronounced fat deposition in obese cats. However, the CV revealed to be lower in the abdominal region than the thoracic region, suggesting that the subcutaneous fat thickness in the abdomen has a more homogeneous deposition character and, therefore, stands out as the most reliable anatomical site for fat layer assessments.

Our results also showed that the subcutaneous fat layer thickness in overweight cats (BCS 4) did not differ from those with ideal weight or obese. Thus, the ultrasonography was not sensitive enough to identify cats with smaller accumulations of fat, possibly because a considerable fat deposit had not yet occurred, which would enable the distinction of subcutaneous fat thickness by ultrasonography in these animals.

When comparing the five anatomical sites evaluated, the lumbar region showed the highest deposition of subcutaneous fat in the three analyzed groups, suggesting that the evaluation of this region is unable to differentiate cats with ideal weight or different degrees of overweightness. This indicates that, regardless of the cat's BCS, the accumulation of subcutaneous fat will be the same in the lumbar region, a fact that hampers the diagnosis of obesity through ultrasonographic evaluation in that region.

In addition, the thoracic, lumbar, and abdominal anatomical regions showed a significant correlation between the different body condition scores, i.e., with the increase in subcutaneous fat thickness in cats in these regions, there is a concomitantly proportional increase in BCS. Also, the abdominal region presented the lowest coefficient of variation among the three anatomical sites, possibly indicating that it is the region of greatest reliability for assessing obesity in cats by ultrasound since different sizes of subcutaneous fat thickness were observed among ideal weight, overweight, and obese animals.

Regarding the analyzed biochemical levels, no significant difference was observed in the cholesterol, triglyceride, and fructosamine values in the different groups. The correlation observed between cholesterol and triglyceride levels and the thickness of subcutaneous fat deposited in the thoracic region indicated that the larger the fat layer in this region, the higher the lipid levels. Hyperlipidemia may or may not occur in obese cats, as evidenced in previous studies (Freitas et

al., 2018; Chala, 2021), but its investigation is important in cats with excess subcutaneous fat. Our results show that, among the five regions analyzed, only the thoracic region can be related to hyperlipidemia in cats, and its investigation is relevant when used as a method of diagnosing obesity.

However, the subcutaneous fat thickness of the cats in the different groups did not show an increase in serum fructosamine concentrations. In a previous study, Hoenig et al. (2013) also noted that obese cats did not present increases in serum fructosamine concentrations. The absence of hyperlipidemia or increase in serum fructosamine concentrations in our results may be due to subcutaneous fat being less metabolically active than visceral fat in releasing various pro-inflammatory cytokines and fatty acids (Okada, 2019).

As a limitation of this study, it was not possible to assess visceral fat, which could show better correlations with the biochemical tests evaluated.

5. Conclusion

It can be concluded that ultrasonography is an effective method for evaluating subcutaneous fat and allows to differentiate obese cats from those with ideal weight in the abdominal and thoracic regions. Furthermore, it is concluded that cholesterol and triglyceride levels are associated with the thickness of fat in the thoracic region, while there is no correlation with fructosamine levels.

Conflict of interest: There were not conflict of interest or sources of funding for the development of this study.

Declarations and Ethics: Our study was done based on the ethical board approval of "Universidade Federal do Espírito Santo – Campus Alegre". The scientific committee in the department of veterinary, was approved the study of "Ultrasonography in the diagnosis of obesity in cats and its correlation with fructosamine and lipid levels" under (reference number 002/2018).

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