Ultrasonography as a complementary diagnostic method for evaluating the skin of healthy cats

Zanna, G ; Zini, E ; Scarampella, F ; Attanasi, A ; Arrighi, S ; Auriemma, E

Abstract: Ultrasonography is not often used in feline dermatology. The purpose of this study was to assess the usefulness and applicability of ultrasonography for skin evaluation in 21 clinically healthy cats. Ultrasonographic examination was conducted in 4 cutaneous regions (frontal, dorsal neck, sacral, and abdominal) using an 18-MHz linear-sequential-array transducer. Findings were assessed using histomorphometric analysis of skin samples set as reference standards. Morphologic evaluation, thickness measurements, measurement variability, and comparison between regions and genders were carried out. The ultrasonographic pattern of feline skin was characterized by 3 distinct layers of different echogenicity and echostructure. Skin was thickest at the dorsal neck region and thinnest at the abdominal region. Skin at the frontal region and dorsal neck region was thicker in males. Variability was < 10% in all regions. No apparent correspondence was found between ultrasonographic and histometric measurements of skin thickness. Collectively, these findings suggest that ultrasonography is a simple, noninvasive, and reproducible technique that allows cutaneous layers to be identified and accurately measures skin thickness in cats.

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Introduction

In recent decades the introduction of ultrasound as a diagnostic imaging tool has allowed for rapid development of it as an important instrument in dermatology as well. Owing to its versatility, repeatability, and non-invasiveness, ultrasound has become useful in the evaluation of skin thanks to technical advances that now enable very high resolution images to be obtained (1-3).

Skin ultrasonography was first proposed in human dermatology in 1979 as an addition to the dermatologic toolbox; at the time it was used for measuring skin thickness (4). Since then it has broadened its spectrum of applications, permitting examiners to qualify and quantify abnormalities within the skin layers and surrounding structures (5). In veterinary medicine, in spite of numerous studies regarding the use of ultrasound in abdominal, pleural, pericardial, and pulmonary evaluation (6), as well as in the characterization of superficial tumors (7,8), few ultrasound imaging findings in skin have been reported. Among these are a skin ultrasound image study in cattle using a 7.5 MHz transducer (9), ultrasonographic studies of skin thickness in dogs using a 13 MHz transducer (10,11), and a study in dogs, also using a 13 MHz transducer, dealing with changes in skin thickness in relation to hydration status and fluid distribution (12). Finally, a study of high-frequency ultrasound biomicroscopy of the normal canine haired skin has been documented, further proving the usefulness of this diagnostic tool in veterinary dermatology (13). Nevertheless, to the authors' knowledge, no studies of feline skin ultrasonography have been described in the literature.

The aim of this study was to assess whether ultrasonography, using a transducer frequency of 18 MHz, might improve feline skin characterization by combining echogenicity evaluation and skin thickness measurements. Furthermore, the investigation sought to correlate ultrasonographic results with gender and age of selected cats, in order to determine whether skin thickness might be affected by these variables.

Materials and methods

Study design

The investigation was performed in accordance with ethical guidelines published in no. 289 of the Italian Gazzetta Ufficiale (G.U., 10 December 1996, 289: 47-53).

Twenty-one young adult domestic shorthair cats from a feline rescue association were included in the study, and informed consent was obtained prior to any procedure. This group consisted of 10 neutered males, 9 spayed females, and 2 intact females, of known age ranging from 1 to 6 years (median 3 years) with body weight ranging from 1.9 to 6 kg (median 3.1 kg). Cats were included on the basis of the following criteria: (i) no evidence of skin lesions on physical examination; (ii) for intact female cats, not being pregnant or lactating; (iii) no clinical evidence of dehydration; (iv) normal results of complete blood count, and routine serum biochemical analysis.

Ultrasonography

A B-mode real-time ultrasound machine (GE-LogiQ S8; GE Healthcare, Italy) equipped with an 18 MHz linear-sequential-array transducer and with an axial resolution $\leq$ 0.4 mm (frequency range: 8-18 MHz) was used. A total of 4 regions, including the frontal, dorsal neck, sacral, and abdominal regions, were selected for ultrasonographic examination. In all these regions, hairs in areas of 2 x 4 cm were gently clipped at 1 mm of length and skin surface was cleaned with 70% isopropyl alcohol to remove any cutaneous debris.

A copious amount of acoustic coupling gel was applied between the transducer and skin surface and the ultrasound probe was then placed strictly perpendicular to the skin. The frontal region was examined halfway along the line connecting the rostral margins of the supraorbital processes, the dorsal neck region at the junction between the second and third cervical vertebrae, and the sacral region halfway along the line connecting the right and left *tuber coxae*. When cats were then positioned in dorsal recumbency, the abdominal region was examined along the caudal third of the *linea alba*. A series of images of the skin with a width of 26 mm and height of 10 mm were obtained and stored for subsequent off-line evaluation using a dedicated DICOM viewer (OsiriX; Pixmeo SARL, Switzerland). Three measurements of skin thickness expressed as the sum of the epidermal entry echo layer and the dermis layer were obtained at 3 different points of the same ultrasonographic image at an interval distance of approximately 5 mm.

Statistical analysis

To assess consistency of ultrasonographic measurements of the examined skin regions, the coefficient of variation was calculated for each of them. To identify possible differences in skin thickness among the 4 regions, paired comparisons were made using the Kruskal-Wallis test followed by Dunn's multiple comparison test. To verify whether skin thickness differences were present between genders (male *vs*. female) for each region, the Mann-Whitney test was used. A P-value < 0.05 was considered significant. The Spearman rank correlation coefficient (rho) was used to verify whether associations were present between age and skin thickness for each region. Analyses were performed using commercially available software (GraphPad QuickCalcs calculator; GraphPad Software Inc., La Jolla, CA).

Results

In accordance with previous studies in dogs (10-13), feline skin also showed a characteristic ultrasonographic pattern composed of 3 layers: a superficial hyperechoic linear band at the interface between the gel and the skin and corresponding to the epidermal entry echo level, beneath which there was a less echogenic band with a granular echotexture corresponding to the dermis, and, more deeply, a hypoechoic pattern separated by hyperechoic septa corresponding to the subcutaneous tissue (Figure 1). Skin thickness was measured at 3 different points of each ultrasonographic image (Figure 2). The median coefficient of variation for skin measurements in the frontal region was 8.7% (range: 0.7-15.7), in the dorsal neck 6.0% (range: 0.7-19.0), in the sacral region 6.2% (range: 0.9-22.9), and in the abdominal region 7.4% (range: 0-30.7). Median thickness of the frontal region was 1.4 mm (range: 0.8-2.0), of the dorsal neck 1.7 mm (range: 1.1-2.2), of the sacral region 1.4 mm (range: 1.0-1.9), and of the abdominal region 1.0 mm (range: 0.8-1.5). The skin of the abdominal region was significantly thinner than the frontal ($P<0.01$), dorsal neck ($P<0.001$), and sacral ($P<0.001$) regions. Thickness was not different among the frontal, dorsal neck, and sacral regions (Figure 3). Median skin thickness of the frontal region was 1.6 mm (range: 1.1-2.0) in males and 1.2 mm (range: 0.8-1.5) in females. That of the dorsal neck was 1.7 mm (range: 1.3-2.2) in males and 1.5 mm (range: 1.1-1.9) in females, that of the sacral region was 1.6 mm (range: 1.2-1.9) in males and 1.4 mm (range: 1.0-1.8) in females, and that of the abdominal region was 1.1 mm (range: 0.9-1.5) in males and 0.9 mm (range: 0.8-1.4) in females. Skin thickness of the frontal region and of the dorsal neck was significantly greater in males than females ($P<0.01$ and $P<0.05$, respectively). Thickness was not different between genders in the sacral and abdominal regions (Figure 4). Correlation coefficients (rho) between age and frontal region, dorsal neck, sacral, and abdominal regions were 0.02, -0.17, 0.18, and 0.43, respectively. The coefficients were therefore very weak to weak, and none yielded significance.

Discussion

To the best of the authors' knowledge, this is the first study to evaluate the ultrasonographic appearance of normal feline skin. In general, the superficial anatomy of skin structures is not visible using low-frequency ultrasound equipment and in humans, the mainstay is a B-mode ultrasound machine with transducers that reach frequencies of 15 MHz or higher (14-17). Therefore, findings from the present study provide additional supporting evidence that a transducer of 18 MHz may offer relevant anatomical data in feline dermatology as well. Indeed, the transducer frequency used here was higher than the 13 MHz of the linear array transducers that have normally been employed (10-12), albeit with a lower frequency than the ultrasound biomicroscopy of the 50 MHz transducer recently used in dogs (13). However and up to now, the latter is a model type less commonly available in veterinary institutions.

First, skin layers were clearly identified. A hyperechoic band corresponding to the epidermal entry echo level was first observed, followed by a second thicker and less echogenic layer band with a finely granular homogeneous echotexture compatible with the dermis. The subcutaneous tissue appeared as the deepest layer and was characterized by a hypoechoic pattern with thin linear hyperechoic bands likely corresponding to connective septa.

Secondly, the use of ultrasonography was shown to provide measurements of skin thickness that were repeatable, as indicated by the relatively low coefficient of variation; indeed, in all 4 regions the calculated coefficient was well below 10%. It cannot be ruled out that a larger degree of variation may occur if stronger pressure were used by the operator when applying the transducer on the cat skin. Nonetheless, because subcutaneous tissue, which includes more connective tissue and is therefore expected to be more compressible, was not included in the measurement because a clear demarcation of the subcutis boundary was lacking in the ultrasonographic images, as previously reported (10-13), it is very likely that irrespective of the operator the thickness would not change to any relevant degree.

In general, it is known that both cats and dogs have a skin thickness that decreases dorsally to ventrally on the trunk and proximally to distally on the limbs (18). However, in cats the reported average thickness of the general body skin ranges from 0.4 to 2 mm, with the thickest being on the back and dorsal neck and the thinnest ventrally, in the inguinal and axillary regions (19). In this study, the greatest skin thickness was demonstrated in the dorsal neck region and the least thickness in the abdominal region, in agreement with what has been described in the literature (19). Moreover, in the present series, although no correlation was detected between ultrasonographically-measured skin thickness and age in any region, probably due to the narrow age range of the cats examined, differences in skin thickness were observed between males and females. Indeed, the skin of the frontal region and dorsal neck was significantly thicker in males than in females. In general, both in humans and mice, sex steroids have been demonstrated to have significant effects on skin physiology and to modulate skin thickness, with males having a thicker dermis and females thicker subcutaneous tissues (20,21). Although from our study it may be hypothesized that gender has an influence on skin thickness and specific body regions, further studies with larger populations are needed to confirm this.

In conclusion, the results of the present study indicate that in cats, as in other species, ultrasonography represents a valid and consistent tool to investigate the skin. It allows characterization of its layers and accurately measurement of its thickness. Based on these findings, we expect that ultrasonographic examination of the skin will provide, in the future, useful information during the assessment of several dermatological disorders of cats.

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Figure legends

Figure 1. Sacral region. Ultrasonographic appearance of normal skin in cats: 3 distinct layers are recognizable, including a well-defined hyperechoic band corresponding to the epidermal entry echo, a less echogenic layer corresponding to the dermis, and a deep layer corresponding to the subcutis and containing linear hyperechoic images.

Figure 2. Dorsal neck region. Skin thickness measurements obtained from 3 points of the same ultrasonographic image and at a distance of approximately 5 mm.

Figure 3. Box and whiskers plot of skin thickness measured in the frontal, dorsal neck, sacral, and abdominal regions of 21 healthy cats. Significant *P*-values are reported.

Figure 4. Dot plots of skin thickness measured in males and females in the frontal (A), dorsal neck (B), sacral (C), and abdominal (D) regions of 21 healthy cats. Significant *P*-values are reported.