



# CT Evaluation of Age-Related Changes in Epaxial Muscle Attenuation and Cross-Sectional Area for Sarcopenia and Myosteatosi s in Small Breed Dogs

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**Abstract** Sarcopenia and myosteatosi s can increase the risk of adverse effects in dogs and humans. However, such imaging study results for evaluating sarcopenia and myosteatosi s in small dogs have not yet been available. The objective of this study was to assess age-related changes according to the breed in epaxial muscle cross-sectional area (CSA) and fat infiltration using CT to evaluate sarcopenia and age-related myosteatosi s in small breed dogs. In 144 dogs (92 Maltese dogs, 27 Poodle dogs, and 25 Shih Tzu dogs), Hounsfield Unit (HU) values and CSA of left epaxial muscle were measured at the thirteenth thoracic vertebral level on non-contrast transverse CT images. Differences in HU values and CSA according to age and breed were analyzed. The geriatric group ( $\geq 12$  years) had significantly lower HU values of epaxial muscle than mature adult group (2 to 6 years) of all breeds. The geriatric group had significantly lower CSA of epaxial muscle than mature adult and senior groups (7 to 11 years) of Maltese dogs. HU values of epaxial muscle were not significantly different among all age groups of all breeds. Maltese dogs had significantly lower CSA of epaxial muscle than Poodle and Shih Tzu dogs in all age groups. Results of this study showed that as age increased in small breed dogs, muscle mass and density decreased.

**Key words** sarcopenia, myosteatosi s, CT, dogs.

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## Introduction

Sarcopenia is defined as age-related loss of skeletal muscle mass and strength in the absence of a disease (9,21). In this condition, decreased skeletal muscle mass is often accompanied by fat infiltration in skeletal muscle, also known as myosteatosis (20,23). Sarcopenia and myosteatosis are multifactorial conditions affecting both humans and animals. The etiology of these conditions has not been clearly identified, the multifactorial mechanisms involve physical inactivity, elevated inflammatory cytokine production, increased oxidative stress, mitochondrial dysfunction, decreased concentrations of growth hormone and sex hormone, changes in type II muscle fibers, insulin resistance, and decreased protein synthesis (9,14).

In humans, sarcopenia and myosteatosis increase the risk for numerous adverse outcomes such as physical disability, falls, fractures, poor quality of life, and even mortality. In addition, a decrease in muscle mass causes a decrease in immune function and lung capacity, increases the risk of infection such as pneumonia, and the incidence of metabolic syndrome and circulatory diseases such as cardiovascular disease, diabetes, and systemic hypertension (5,6,32).

Although sarcopenia and myosteatosis are less scientifically evaluated in dogs than in humans, several studies have revealed that dogs can also lose lean body mass, body protein, and muscle mass and increase muscle fat content during aging (12,18,22). Sarcopenia and myosteatosis could be identified in healthy geriatric dogs by clinically relevant techniques. However, mechanisms of age-related muscle changes and these clinical effects are not well understood yet. Several studies have suggested that autophagy might contribute to sarcopenia in dogs. Increased body fat may influence adipokine, insulin, and cytokine regulation related to the development of insulin resistance and heart disease (9,24). Additional research will be needed to identify both mechanisms and clinical effects of sarcopenia and myosteatosis in dogs.

Although the diagnostic criteria of sarcopenia and myosteatosis have not been completely established yet, several studies have proposed the need to assess age-related loss of skeletal muscle mass, muscle weakening, and declining physical performance (7). The most common imaging method for assessing sarcopenia and myosteatosis in humans is measuring the muscle mass and density of the thigh or paraspinal muscle via CT (4,8,21,28,31). Cross-sectional area (CSA) can be quantified by CT. Muscle density measured by CT reflects intramuscular fat infiltration (18,19). More recently, age-related muscle change has been assessed in dogs by measuring epaxial muscle size and intramuscular fat contents using CT and MRI (3,16,29). Result of these studies showed that

healthy old humans and Labrador Retriever dogs have smaller muscle CSA and lower HU values of epaxial muscle than healthy young humans and Labrador Retriever dogs (16,29).

To the authors' knowledge, effects of age on muscle mass and fat content evaluated by CT have not been reported for small dogs. As the muscle mass and fat content are related to chronic medical conditions such as chronic kidney disease, cardiac disease, cancer, hepatic disease, and orthopedic or neurologic problems, the accurate assessment of sarcopenia and myosteatosis may be helpful in small breed dogs (9,11,25). Thus, the aim of this study was to assess age-related changes in epaxial muscle CSA and fat infiltration using CT to evaluate sarcopenia and age-related myosteatosis in small-breed dogs. The hypothesis was that CSA and HU values of epaxial muscles measured by CT would decline with aging in small breed dogs, similar to reported findings for humans and Labrador Retriever dogs.

## Materials and Methods

### Animals

This multicenter retrospective study included Maltese dogs, Poodle dogs, and Shih Tzu dogs with non-contrast CT scans of the thoracic and abdomen performed for diagnostic purposes among clients of the Gyeongsang National University Veterinary Teaching Hospital from April 2015 to September 2022. To be included in this study, patients had to have a body condition score of 4 to 6 on a 9-point scale. Patients with neuromuscular disease and hormonal diseases such as hypothyroidism, hyperadrenocorticism, or diabetes mellitus were excluded. Dogs were grouped by breed (Maltese, Poodle, and Shih Tzu) and age (mature adult, 2 to 6 years old; senior, 7 to 11 years old; and geriatric,  $\geq 12$  years old). A recent study has suggested developmental stage thresholds for categorizing domestic dogs into the following age groups: puppy (0 to 5 months), juvenile (6 to 12 months), young adult (13 to 24 months), mature adult (2 to 6 years old), senior (7 to 11 years old), and geriatric ( $\geq 12$  years old) (23). All dogs included in the present study were categorized into mature adult (2 to 6 years old), senior (7 to 11 years old), and geriatric ( $\geq 12$  years old) groups based on this study (13,15).

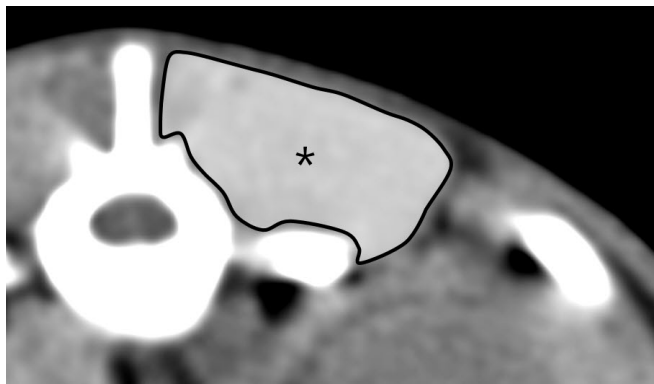
### CT examinations

Under general anesthesia, all dogs were positioned in ventral recumbency. CT data were acquired using of a two-slice helical scanner (Somatom Emotion Duo, Siemens, Munich, Germany), a 16-slice helical scanner (Activation 16, Toshiba Medical Systems., Otowara, Japan), and a 160-slice helical scanner (Aquilion Lightning 160, Canon Medical Systems,

Tokyo, Japan). Scanning parameters were 110-120 kV and 80-112 mAs. All CT data were reconstructed and evaluated with non-contrast 1 mm slice thickness transverse images on a soft tissue algorithm. All CT data were reviewed on DICOM view (ZeTTA PACS, version; TaeYoung soft co., Anyang, Korea, INFINITT Smart-Net, INFINITT Healthcare Co, Seoul, Korea) and evaluated by a single observer (LJ) without knowledge of the dog's age.

**Measurement of HU value and CSA**

HU values and CSA of the left epaxial (paravertebral) muscle over the T13 vertebral body having recognizable anatomical features at the thoracolumbar junction were measured based



**Fig. 1.** Measurements of HU value and CSA of left epaxial muscles at the T13 level. HU value and CSA were measured using non-contrast transverse CT images (window width, 400 HU; window level, 40 HU). ROI was drawn manually along the outer margin of the left epaxial muscle (asterisk). HU, hounsfield unit; CSA, cross-sectional area; CT, computed tomography; ROI, region of interest.

on previous studies (16,29). Regions of interest (ROIs) were drawn manually along the outer margin of the left epaxial muscle (Fig. 1). From these data, mean HU value, mean CSA in cm<sup>2</sup>, and their standard deviations (SD) were calculated to compare differences in epaxial muscle among breed (Maltese, Poodle, and Shih Tzu) and age (mature adult, 2 to 6 years old; senior, 7 to 11 years old; and geriatric, ≥12 years old) groups.

**Statistical analysis**

Statistical analysis was performed using the SPSS statistical computer program (SPSS for Windows, Release 27.0, standard version, SPSS Inc., Chicago, IL, USA). To compare CSA and CT attenuation for epaxial muscle among breeds and age groups, normality tests (Kolmogorov-Smirnov test and Shapiro-Wilk test) were performed to determine whether the data were normally distributed and statistical significance was analyzed using the Kruskal-Wallis test followed by post-hoc Mann Whitney U-test with Bonferroni adjustment to compare each group. In the statistical test, p-values of less than 0.05 and 0.017 were considered statistical significance for the Kruskal-Wallis test and Mann Whitney U-test, respectively.

**Results**

A total of 144 dogs met the inclusion criteria. They aged 2 years to 17 years. These dogs consisted of Maltese (n = 92), Poodle (n = 27), and Shih Tzu (n = 25) dogs. Clinical data on age, body weight, and body condition score are summarized in Table 1.

Neither body weight nor BCS differed significantly among mature adult, senior, and Geriatric groups of Maltese (p =

**Table 1.** Comparison of signalment, HU value and CSA of epaxial muscles among different dog breed groups and age groups

Variables	Group (n)	Age (years)	Weight (kg)	BCS	CT attenuation (HU)	CSA (mm <sup>2</sup> )
Maltese	Mature adult (16)	4.0 ± 1.5	3.7 ± 1.2	5.2 ± 0.4	46.4 ± 14.2	1.5 ± 0.6
	Senior (41)	9.0 ± 1.4	3.8 ± 1.1	5.1 ± 0.7	30.0 ± 18.2 <sup>d</sup>	1.5 ± 0.6
	Geriatric (35)	13.2 ± 1.4	3.4 ± 1.1	5.0 ± 0.7	20.8 ± 18.7 <sup>d</sup>	1.2 ± 0.5 <sup>ef</sup>
Poodle	Mature adult (6)	4.0 ± 2.2	5.1 ± 1.6	4.8 ± 0.7	53.8 ± 3.6	3.1 ± 1.3 <sup>g</sup>
	Senior (10)	8.9 ± 1.0	6.0 ± 1.1 <sup>b</sup>	5.2 ± 0.6	41.4 ± 11.7 <sup>d</sup>	2.6 ± 0.7 <sup>h</sup>
	Geriatric (11)	13.5 ± 1.6	6.1 ± 1.3 <sup>b</sup>	5.3 ± 0.8	28.8 ± 19.3 <sup>d</sup>	2.3 ± 0.6 <sup>h</sup>
Shih Tzu	Mature adult (3)	5.0 ± 1.0	6.4 ± 0.7 <sup>a</sup>	5.7 ± 0.6	51.5 ± 14.5	3.1 ± 0.5 <sup>g</sup>
	Senior (10)	10.5 ± 0.7	5.6 ± 1.1 <sup>b</sup>	5.2 ± 0.4	30.0 ± 19.8	2.5 ± 0.8 <sup>h</sup>
	Geriatric (12)	13.9 ± 1.7	5.3 ± 1.1 <sup>b</sup>	4.8 ± 0.6	13.0 ± 18.8 <sup>c</sup>	1.9 ± 0.8 <sup>h</sup>

Data are shown as mean ± standard deviation (SD).

BCS, body condition score; HU, hounsfield unit; CSA, cross-sectional area.

<sup>a</sup>p < 0.017, body weight compared with Maltese group; <sup>b</sup>p < 0.003, body weight compared with Maltese group; <sup>c</sup>p < 0.017, HU compared with mature adult group; <sup>d</sup>p < 0.003, HU compared with mature adult group; <sup>e</sup>p < 0.017, CSA compared with mature adult group; <sup>f</sup>p < 0.017, CSA compared with senior group; <sup>g</sup>p < 0.017, CSA compared with Maltese group; <sup>h</sup>p < 0.003, CSA compared with Maltese group.

0.336 and  $p = 0.496$ , respectively), Poodle ( $p = 0.259$  and  $p = 0.462$ , respectively), or Shih Tzu ( $p = 0.329$  and  $p = 0.053$ , respectively) dogs. BCS was not significantly different among Maltese, Poodle, and Shih Tzu dogs in mature adult ( $p = 0.111$ ), senior ( $p = 0.868$ ), or geriatric ( $p = 0.274$ ) group. However, body weights of Maltese dogs were significantly lower ( $p = 0.004$ ) than those of Shih Tzu dogs in the mature adult group. Body weights of Maltese dogs were significantly lower than those of Poodle and Shih Tzu dogs ( $p < 0.001$  and  $p = 0.001$ , respectively) in the senior group. Body weights of Maltese dogs were significantly lower than those of Poodle and Shih Tzu dogs (both  $p < 0.001$ ) in the geriatric group.

### HU value and CSA of epaxial muscle according to the age in each breed

Senior and geriatric groups had significantly lower HU values of epaxial muscles than the mature adult group of Maltese dogs ( $p = 0.003$  and  $p < 0.001$ , respectively) and Poodle dogs ( $p = 0.002$  and  $p < 0.001$ , respectively). Geriatric group had significantly lower HU values of epaxial muscles than mature adult group of Shih Tzu dogs ( $p = 0.004$ ). HU values of epaxial muscle were not significantly different between senior and geriatric groups of Maltese ( $p = 0.077$ ), Poodle ( $p = 0.061$ ), and Shih Tzu ( $p = 0.059$ ) dogs. HU values of epaxial muscles were not significantly different between mature adult and senior groups of Shih Tzu dogs ( $p = 0.217$ ).

The geriatric group had significantly lower CSAs of epaxial muscles than mature adult and senior groups of Maltese dogs ( $p = 0.012$  and  $p = 0.013$ , respectively). CSAs of epaxial muscles were not significantly different among mature adult, senior, and geriatric groups of Poodle and Shih Tzu

dogs ( $p = 0.605$  and  $p = 0.065$ , respectively) (Table 1, Fig. 2).

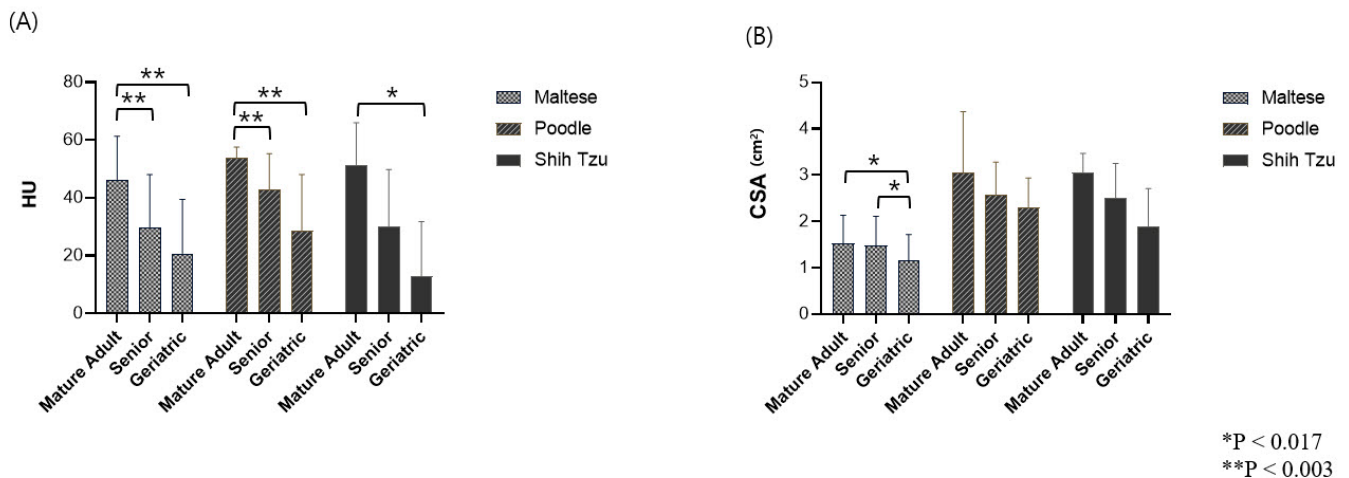
### HU value and CSA of epaxial muscles according to the breed

HU values of epaxial muscles were not significantly different among Maltese, Poodle, and Shih Tzu dogs in mature adult ( $p = 0.584$ ), senior ( $p = 0.101$ ), and geriatric ( $p = 0.129$ ) group. Maltese dogs had significantly lower CSAs of epaxial muscles than Poodle and Shih Tzu dogs in the mature adult group ( $p = 0.006$  and  $p = 0.008$ , respectively). Maltese dogs had significantly lower CSAs of epaxial muscles than Poodle and Shih Tzu dogs in the senior group (both  $p < 0.001$ ), geriatric group ( $p < 0.001$  and  $p = 0.002$ , respectively), and mature adult group ( $p = 0.013$  and  $p = 0.008$ , respectively) (Table 1, Fig. 3).

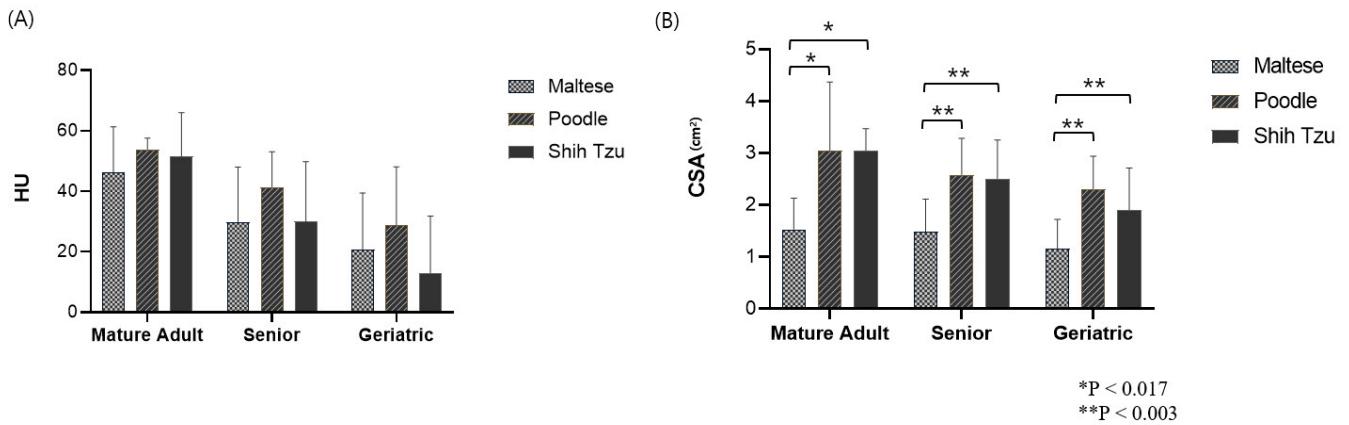
## Discussion

CT is useful for evaluating muscle quantity and quality non-invasively. In previous studies, measurements obtained from a single CSA have shown good correlations with whole-body adipose tissue and skeletal muscle distribution (33). Previous studies suggest that epaxial muscles would be less affected by muscle loss from orthopedic disease and are primarily composed of type II fibers that are more susceptible to muscle atrophy (16,26,31). In this study, the T13 level vulnerable to muscle atrophy was chosen to evaluate muscle mass and fat content.

This is the first study to analyze effects of age on muscle mass and fat content in small breed dogs using CT. In this study, geriatric and senior groups had significantly lower HU



**Fig. 2.** HU value and CSA of epaxial muscle according to the age in each breed. HU value (A), CSA (B), \* $p < 0.017$ , and \*\* $p < 0.003$ . Data in graphs are presented as mean  $\pm$  standard deviation (SD). HU, hounsfield unit; CSA, cross-sectional area.



**Fig. 3.** HU value and CSA of epaxial muscles according to the breed. HU value (A), CSA (B), \*p < 0.017, and \*\*p < 0.003. Data in graphs are presented as mean ± standard deviation (SD). HU, hounsfield unit; CSA, cross-sectional area.

values of epaxial muscles than the mature adult group of Maltese and Poodle dogs. The geriatric group had significantly lower HU values of epaxial muscles than the mature adult group of Shih Tzu dogs. Although HU values of epaxial muscles were not significantly different between senior and geriatric groups of any breeds or between mature adult and senior groups of Shih Tzu dogs, mean HU values of epaxial muscles declined with aging in Maltese, Poodle, and Shih Tzu dogs. This result is consistent with a recent study showing that healthy geriatric (>8 years) Labrador Retriever dogs have significantly lower mean HU values of epaxial muscles measured by CT than healthy young (1 to 5 years) Labrador Retriever dogs (29). Muscle density can be measured based on CT muscle HU value. It reflects the number of muscle fiber, the area of the individual muscle fiber, and the contractile ability of each muscle fiber (17). A previous study has suggested that fat tissue appears lower attenuation on CT image and muscle appears higher attenuation (30). Thus, lower attenuation muscle has a greater fat content (30). These CT-detected changes are considered to reflect infiltration of muscle with fat as previously demonstrated by comparison of HU values with results of histologic examination of muscle biopsy specimens in humans (10).

Although there was no significant difference in CSA among mature adult, senior, and geriatric groups of Poodle and Shih Tzu dogs, not Maltese dogs, the mean CSA of epaxial muscle declined with aging in all breeds. This result is consistent with previous studies showing that mean epaxial muscle area was significantly lower in healthy old (>8 years) Labrador Retriever dogs than in healthy young (1 to 5 years) Labrador Retriever dogs (16). Previous studies have suggested that loss of lean body mass is often accompanied by an

increase in fat mass (23,27). Thus, total muscle area may not change or even increase (23,27). In other words, sarcopenia might be masked by myosteatorsis. Thus, recent studies in humans have suggested cutoff values using a specific range of HU values for assessing muscle quality (1,19). The muscle area can be divided as follows according to HU: (1) normal attenuation muscle (+30 to +150 HU), reflecting healthy muscle with little intramuscular fat; (2) low attenuation muscle (-29 to +29 HU), reflecting unhealthy muscle with intramuscular lipid; and (3) intramuscular adipose tissue (-190 to -30 HU), reflecting fat tissue between muscle fibers (19). Total muscle (-190 to +150 HU) was defined as the whole area, including all skeletal muscles and fat tissues (19). These threshold ranges of muscle can be used to directly identify fatty areas inside the muscle and assess the severity of myosteatorsis (19). Additional research considering these muscle areas is needed to evaluate sarcopenia and myosteatorsis in dogs.

This is also the first study to analyze effects of breed on muscle mass and fat content in mature adult, senior, and geriatric groups based on CT. HU values of epaxial muscle were not significantly different among Maltese, Poodle, and Shih Tzu dogs in mature adult, senior, or geriatric group included in the present study.

Maltese dogs had significantly lower CSA than Poodle and Shih Tzu dogs in all age groups. These differences in CSA of epaxial muscles might be related to lighter body weight for Maltese dogs than for Poodle and Shih Tzu dogs included in this study. They might imply differences in body shapes according to breed, consistent with a recent study showing a slender body shape of Maltese dogs than Poodle and Shih Tzu dogs (24).

In previous human studies, methods of diagnosing sarcopenia by quantifying muscle mass have been suggested (2).



## References

However, it is difficult to apply these methods in dogs due to breed variation of body shape and skeletal size. In this study, intramuscular fat infiltration tendency remained the same regardless of dog breed (Maltese, Poodle, or Shih Tzu dogs). Thus, HU value is considered the most appropriate objective indicator for diagnosing sarcopenia in various breeds.

The present study has several limitations. An imbalanced number among the three age groups and the three breed groups might have affected results of this study. As there were only a small number of mature adult Maltese, Poodle, and Shih Tzu dogs and a greater number of Maltese dogs than Poodle and Shih Tzu dogs included in this study, a further study with a larger group of mature adult Poodle and Shih Tzu dogs is needed. Histopathological evaluation of the muscle for intramuscular fat infiltration was not performed. Although muscle with fat infiltration appeared as a lower HU value than healthy muscle as previously demonstrated by comparing HU values with results of histologic examination of muscle biopsy specimens (10), additional studies that perform a histopathological evaluation of the muscle will be needed to demonstrate a degree of fat infiltration with muscle.

This study demonstrates that old small breed dogs have smaller CSA and lower HU values of epaxial muscles than young small breed dogs. These changes in muscles on CT suggested that as the age increases in small breed dogs, muscle mass and density for epaxial muscle are decreased, which may suggest greater fat content of muscle and less muscle mass. This study presents descriptive data of HU values and CSA of muscle according to dog breed and age. Results of our study can be used for evaluating muscle atrophy in dogs with intervertebral disc disease and muscle-wasting diseases such as cancer, chronic kidney disease, and heart failure (11,25).

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### Conflicts of Interest

The authors have no conflicting interests.

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