



Morphometric evaluation of canine hepatocellular carcinoma using computed tomography: a promising tool for predicting malignancy

Rommaneeya LEELA-ARPORN^{1,2)}, Hiroshi OHTA^{2,3)}, Genya SHIMBO⁴⁾,
Noboru SASAKI²⁾ and Mitsuyoshi TAKIGUCHI^{2)*}

¹⁾Faculty of Veterinary Medicine and Applied Zoology, HRH Princess Chulabhorn College of Medical Science, Chulabhorn Royal Academy, Lak Si, Bangkok 10210, Thailand

²⁾Laboratory of Veterinary Internal Medicine, Department of Veterinary Clinical Sciences, Graduate School of Veterinary Medicine, Hokkaido University, Sapporo, Hokkaido 060-0818, Japan

³⁾Laboratory of Veterinary Internal Medicine, Department of Small Animal Clinical Sciences, School of Veterinary Medicine, Rakuno Gakuen University, Ebetsu, Hokkaido 069-8501, Japan

⁴⁾Veterinary Teaching Hospital, Graduate School of Veterinary Medicine, Hokkaido University, Sapporo, Hokkaido 060-0818, Japan

ABSTRACT. The size of canine focal liver lesions (FLLs) is known to be one of the predicting criteria for malignancy. However, there are discrepancies for the measurement of maximum lesion size, resulting in contradicting results among studies and incidences of false positive outcomes. Thus far, the morphometric changes of FLLs for distinguishing malignancy from benignancy remains undocumented. This study aimed to investigate morphometric characteristics of FLLs using computed tomography (CT). CT images of 40 dogs with histopathological confirmation of 49 liver lesions, including 39 hepatocellular carcinomas and 10 nodular hyperplasias were retrospectively reviewed. The morphometric parameters including size (long and short axis diameters measured on transverse image), shape (measured by long to short axis (L/S) ratio), volume, and surface appearance of a liver lesion were evaluated using univariate and stepwise multivariate analyses, respectively. The results of univariate analysis showed that long and short axis diameters, L/S ratio, volume, and surface appearance of a lesion were significantly different between hepatocellular carcinomas and nodular hyperplasias. Multivariate analysis revealed that short axis diameter (>3.30 cm; odds ratio (OR): 36.1, 95% confidence interval (CI): 3.36–387.05, $P=0.0031$) and L/S ratio (>1.23; OR: 18.1, 95% CI: 1.61–205.12, $P=0.0191$) were independent predictors of malignancy, with the area under the curve of 0.9154. These results suggest that the combination of short axis diameter and L/S ratio is a promising tool for predicting liver malignancy with outstanding discriminating ability.

KEY WORDS: differentiation, dog, hepatic tumor, morphometry

J. Vet. Med. Sci.

83(9): 1459–1464, 2021

doi: 10.1292/jvms.21-0218

Received: 9 April 2021

Accepted: 21 June 2021

Advanced Epub:

6 July 2021

Focal liver lesions (FLLs) are often incidentally discovered in dogs with pathological varieties, including malignant lesions such as hepatocellular carcinoma which is the most common primary liver tumor in dogs [9, 12, 15, 16], and benign liver diseases such as nodular hyperplasia, vacuolar hepatopathy, hematoma, and hepatic cyst [9]. Recently, radiological approaches, including ultrasonography and computed tomography (CT) have been reported for their ability in distinguishing between benign and malignant liver lesions [3, 5, 6, 8, 10, 11, 14, 19], which can support clinicians in making decisions for appropriate management of the patient.

The maximum size of FLLs is one of the indicators for differentiating malignant from benign liver diseases [3, 6, 10, 11, 14, 19]. However, there are discrepancies among studies in measuring the maximum diameter of the lesion due to no standardization for the measurement and subjective evaluation, leading to challenges in comparing the results among studies and incidences of false positive results.

Morphometry of the lesions such as bidirectional dimensions, shape, and volume can change due to disease progression. Previous studies reported that morphometric parameters including short axis diameter, long axis diameter and long to short

*Correspondence to: Takiguchi, M.: mtaki@vetmed.hokudai.ac.jp

©2021 The Japanese Society of Veterinary Science



This is an open-access article distributed under the terms of the Creative Commons Attribution Non-Commercial No Derivatives (by-nc-nd) License. (CC-BY-NC-ND 4.0: <https://creativecommons.org/licenses/by-nc-nd/4.0/>)

axis (L/S) ratio are able to differentiate between benign and malignant lymphadenopathy in humans and dogs [1, 7, 17, 18]. Interestingly, a recent study revealed that L/S ratio of hepatocellular carcinoma is associated with tumor protein 53 (TP53) mutation in hepatocellular carcinoma, representing that morphometric changes of the lesions might relate to growth characteristics of individual tumors and negative prognostic impact [22]. However, morphometric analysis of FLLs other than the unidirectional maximum lesion size, in relation to a broad classification of histopathological diagnosis including benignancy and malignancy remains undocumented in dogs.

CT is less operator-dependent and has greater accuracy in measuring morphometry and ability in differentiation of malignant from benign liver diseases compared to ultrasonography [5]. Therefore, due to limited information regarding morphometry of FLLs in dogs, the aims of this study were to identify morphometric parameters that could be associated with liver malignancy using hepatocellular carcinoma as a representative for malignant lesions in comparison to nodular hyperplasia which is relatively common in older dogs [12], as a representative of benign lesions via CT examination, and to evaluate diagnostic performance of independent morphometric parameters in predicting liver malignancy.

We postulated that morphometric features of FLLs could differentiate malignant and benign liver lesions by growth characteristics of the lesions, as same as in human study [22].

MATERIALS AND METHODS

Study population

A retrospective study was conducted using clinical data including demographic information (age, body weight, breed, and sex) and CT images of dogs with focal or multifocal liver lesions with histologically confirmed diagnoses for each lesion following surgical resection of the lesion between December 2016 and December 2018 at Hokkaido University Veterinary Teaching Hospital. The resected nodules or masses were histologically confirmed by a single board-certified pathologist. The dogs diagnosed histopathologically with hepatocellular carcinoma, and nodular hyperplasia were selected for image analysis in this study. Informed consent for permission to use the clinical data was obtained from the owners of all included dogs at the first time visit.

CT procedures

All CT examinations were performed with an 80-row multidetector CT scanner (Aquilion PRIME; Toshiba Medical Systems; Otawara, Japan). All dogs were examined in the position of dorsal or sternal recumbency under general anesthesia with ventilation. Helical scan of the abdomen included precontrast and postcontrast images in the arterial, portal, and delayed phases through the liver. The examinations were performed by one radiologist (GS) with 10 years of experience in diagnostic imaging using a standardized protocol. Technical setting for the scan included a slice thickness of 3 mm, reconstruction interval of 3 mm, helical pitch of 0.813, tube rotation time of 0.5 sec, X-ray tube potential of 120 kVp and X-ray tube current of 60–500 mA, which were automatically calculated by a commercial software package (Sure Exposure 3D; Toshiba Medical Systems). Following precontrast CT scanning, all dogs received iodinated non-ionic contrast medium at a dose of 600 mg iodine/kg (Omnipaque 300; Daiichi Sankyo, Inc., Tokyo, Japan or Iopamidol 150; Bayer, Osaka, Japan) intravenously with a power injector over 20–30 sec via the cephalic vein. Bolus tracking technique was used for postcontrast medium administration scan using an aortic region of interest (ROI) which placed over the abdominal aorta at the level of the diaphragm of 200 Hounsfield units (HU) as a trigger threshold for the arterial scan. The portal phase and delayed phases were obtained at 20 sec after the end of the arterial phase scan and at 180 sec after the contrast medium injection, respectively.

Image analysis

All CT images were retrieved from a computer workstation in Digital Imaging and Communications in Medicine (DICOM) for and reviewed using DICOM viewing software (OsiriX, Pixmeo SARL, Bernex, Switzerland). Morphometric features including size (long and short axis diameters), shape (measured by L/S ratio), volume, and surface appearance of a liver lesion were evaluated in transverse CT images. The definition of each parameter was determined by two authors (RL, a veterinarian with 4 years' experience in small animal internal medicine and diagnostic imaging, and MT, a veterinarian with 20 years' experience in small animal internal medicine and diagnostic imaging). For the lesion size, transverse CT images were used to evaluate and measure the long and short axis diameter of the lesion. The largest diameter of each lesion was defined as the "long axis diameter" [18, 22]. The largest diameter perpendicular to the long axis diameter of each lesion was defined as the "short axis diameter" [18, 22]. The L/S ratio of each lesion was then calculated [18, 22]. The surface appearance of each lesion was categorized as smooth or irregular margin [11]. For the volume of each lesion (cm³), delayed postcontrast images which presents maximum liver parenchymal enhancement and leads to a clear evaluation of the lesion, were used to generate volume-rendered images of the lesion. Volume-rendered images were carefully constructed by placing a closed polygon ROI on the margin of the lesion in transverse CT slices from the most cranial to the most caudal part of the lesion. Then, the volume of the lesion was automatically calculated from the sum of the volumes of all the ROIs of all the slices.

The size, shape, and surface appearance of the lesion of all CT images was measured by an experienced radiologist (GS) who performed the CT examinations and is aware of the history of the dogs, but not the final diagnosis of the lesion which was performed later. The volume of the lesion was measured by one author (RL) who is experienced in using the DICOM viewer software.

All CT data of all included dogs were collected by one author (RL).

Statistical analysis

Statistical analysis was performed by one author (RL) who is trained and experienced in biostatistics, using commercial software (JMP Pro, version 14.0.0, SAS Institute Inc., Cary, CA, USA). For comparisons of CT variables between hepatocellular carcinoma and nodular hyperplasia, morphometric features possibly associated with histopathological findings were assessed using univariate and stepwise multivariate analyses. Categorical data, including sex, breed, and surface appearance of a liver lesion were analyzed using Fisher's exact test or the χ^2 test and presented as numbers and percentages. Continuous data including age, body weight, lesion size (long and short axis diameters), L/S ratio and volume of the lesions were assessed for the normality using the Shapiro-Wilk test and were expressed as medians and ranges. The student's *t*-test and the Mann-Whitney *U* test were used to compare the normally and non-normally distributed data, respectively. The optimal cutoff values of significant continuous morphometric parameters were selected from a receiver operating characteristic (ROC) curve analysis prior to include in the stepwise multivariate analysis.

A stepwise regression analysis was performed using all significant variables from the univariate analysis with a forward selection. The selection process conducted with a *P* value threshold (*P*<0.15 for inclusion and *P*>0.2 for exclusion) to identify independent predictors with the strongest associations that can differentiate between benign and malignant liver lesions. The odds ratio (OR) and 95% confidence interval (CI) of each variable that was selected in the multivariate analysis were calculated. Then, ROC curve analysis was conducted to assess diagnostic performance of independent predictors from multivariate analysis. A value of *P*<0.05 was considered statistically significant for all statistical analyses.

RESULTS

Dogs

Forty dogs (21 males (52.5%) and 19 females (47.5%)) with 49 liver nodules or masses with histopathological confirmation were enrolled in this study. Of these, 28 dogs had a focal malignant lesion, 6 dogs had a focal benign lesion, 4 dogs had both benign and malignant lesions, and 2 dogs had multifocal malignant lesions. Histopathologic diagnosis revealed 39 hepatocellular carcinomas, and 10 nodular hyperplasias. At the time of diagnosis, the median age of all included dogs was 11 years (range, 7–16 years), and the median body weight of all included dogs was 7 kg (range, 1.7–32.5 kg). The breed distribution of included dogs was as follows: 6 Chihuahuas (15%), 4 Yorkshire Terriers (10%), 3 Miniature Dachshunds (7.5%), 3 Miniature Schnauzers (7.5%), 3 Mongrels (7.5%), 3 Shiba Inus (7.5%), 3 Shih Tzus (7.5%), 3 Toy Poodles (7.5%), 2 Welsh Corgis (5%), 2 Beagles (5%), 2 Golden Retrievers (5%), and one of each of Border Collie (2.5%), Boston Terrier (2.5%), Cairn Terrier (2.5%), Jack Russel (2.5%), Pekingese (2.5%), and Shetland Sheepdog (2.5%).

Morphometric characteristics of focal liver lesions

Morphometric characteristics of a focal liver lesion, including size, shape, volume, and surface appearance of a lesion was evaluated using univariate analysis and stepwise multivariate analysis, respectively to identify independent predictors for liver malignancy.

For univariate analysis, long and short axis diameters, L/S ratio, volume, and surface appearance of a lesion were significantly different between hepatocellular carcinomas and nodular hyperplasias as shown in Table 1. Regarding these significant parameters, the optimal cutoff value for each continuous parameter in classifying hepatocellular carcinoma based on the highest Youden's index according to ROC curve analysis was as follows: >4.40 cm for long axis diameter (OR: 26.1, 95% CI: 2.93–232.61), >3.30 cm for short axis diameter (OR: 30.0, 95% CI: 3.34–269.72), >1.23 for L/S ratio (OR: 14.4, 95% CI: 1.65–125.41), and >38 cm³ for volume of the lesion (OR: 26.1, 95% CI: 2.93–232.61).

Then, all significant parameters from the univariate results were entered in the stepwise multivariate analysis with a forward selection. Of these, only short axis diameter and L/S ratio were selected as candidate parameters for multivariate analysis and found that that short axis diameter (>3.30 cm; OR: 36.1, 95% CI: 3.36–387.05, *P*=0.0031) and L/S ratio (>1.23; OR: 18.1, 95% CI: 1.61–205.12, *P*=0.0191) were significant independent predictors for liver malignancy (Figs. 1 and 2). The diagnostic performance of the combination of both independent parameters and each independent parameter for predicting liver malignancy indicated that

Table 1. Univariate results of morphometric parameters in determining liver malignancy in 49 liver lesions

Morphometric parameters	Nodular hyperplasia (n=10)	Hepatocellular carcinoma (n=39)	<i>P</i> value
Long axis diameter (cm), median (range)	2.9 (1.2–6.1)	6.5 (1.2–13.1)	<0.0001*
Short axis diameter (cm), median (range)	2.5 (1.0–5.9)	4.9 (1.2–10.0)	0.0002*
Long to short axis ratio, median (range)	1.2 (1.0–1.3)	1.3 (1.0–2.1)	0.0300*
Volume of the lesion (cm ³), median (range)	11.2 (0.6–98.7)	102.8 (1.1–958.6)	0.0008*
Surface appearance of the lesion, n (%)			0.0435*
Smooth margin	10 (100)	27 (69.2)	
Irregular margin	0 (0)	12 (30.8)	

**P* values <0.05 were statistically significant.

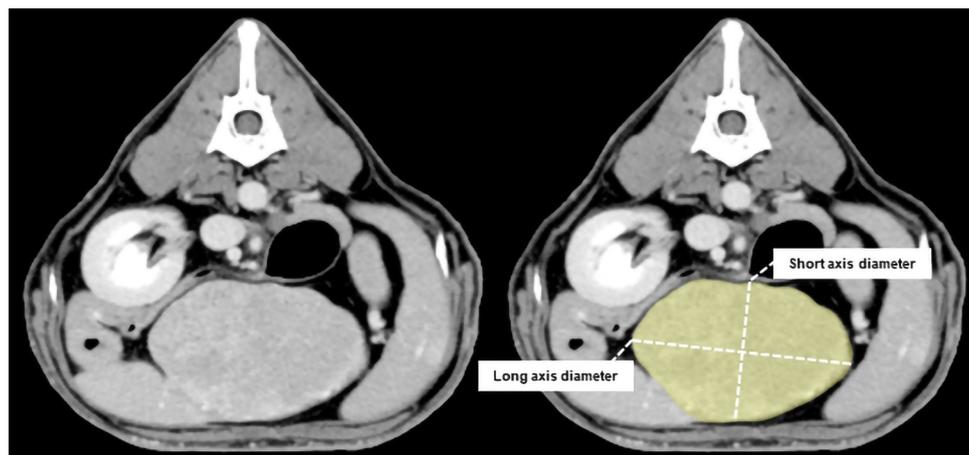


Fig. 1. Transverse CT image of a dog with hepatocellular carcinoma. The size of a massive lesion in the left lateral lobe shows a short axis diameter of 5.0 cm and a long axis diameter 7.5 cm. “Long and short axis diameters” are used in the long to short axis (L/S) ratio of the lesion, representing with the L/S ratio of 1.50.

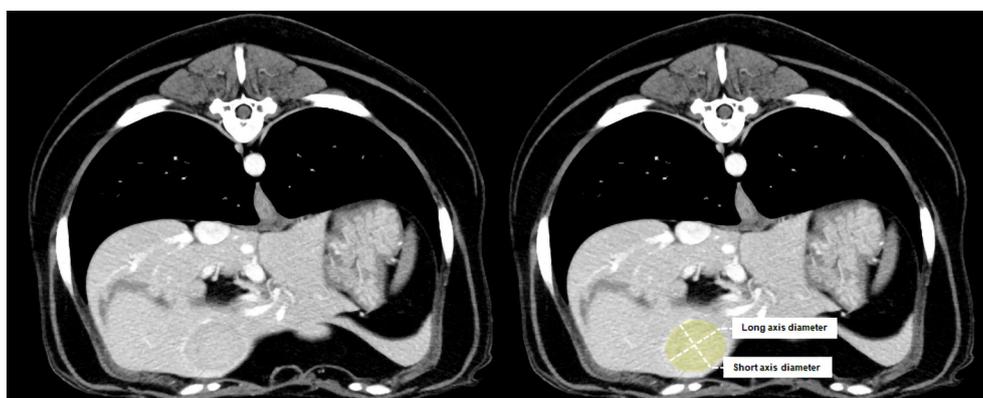


Fig. 2. Transverse CT image of a dog with nodular hyperplasia. The size of a nodule in the right lateral lobe shows a short axis diameter of 3.2 cm and a long axis diameter 3.7 cm. “Long and short axis diameters” are used in the long to short axis (L/S) ratio of the nodule, representing with the L/S ratio of 1.16.

Table 2. Diagnostic performance of independent morphometric parameters of focal liver lesions for predicting liver malignancy

Morphometric parameters	Optimal cutoff	AUC	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Accuracy (%)
Short axis diameter and L/S ratio	-	0.9154	92.3	80.0	94.7	72.3	89.8
Short axis diameter	3.30	0.8346	76.9	90.0	96.8	50.0	79.6
L/S ratio	1.23	0.7577	61.5	90.0	96.0	37.5	67.4

AUC, area under the curve; L/S, long to short axis; NPV, negative predictive value; PPV, positive predictive value.

the combination of both independent parameters had a higher diagnostic ability compared to each individual parameter, as shown in [Table 2](#) and [Fig. 3](#). The area under the ROC curve analysis of the combination of both independent parameters was 0.9154 which showed outstanding discrimination and represented the diagnostic performance as follows: 89.8% accuracy, 92.3% sensitivity, and 80.0% specificity.

DISCUSSION

In the present study, we investigated the ability of morphometric characteristics of FLLs in differentiating between benign and malignant liver lesions. Our results showed that short axis diameter >3.30 cm and L/S ratio >1.23 were significantly associated with malignancy, with the area under the ROC curve of the combination of both independent parameters of 0.9154, representing

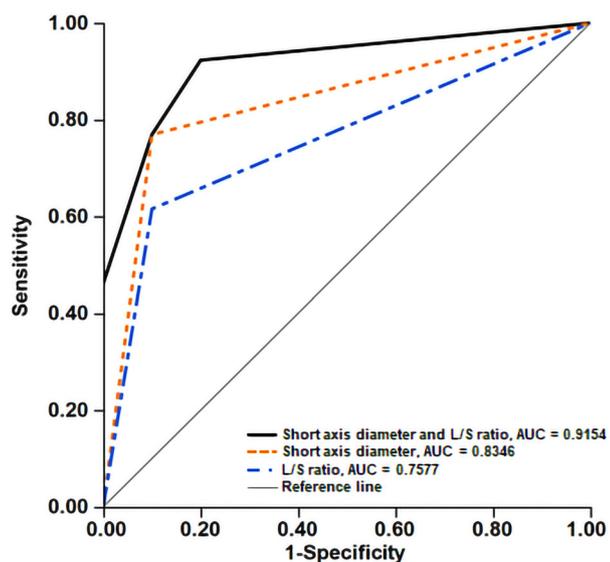


Fig. 3. The area under the receiver operating characteristic curve of independent morphometric parameters for predicting liver malignancy.

value of 1.23 had individual ability to predict hepatocellular carcinoma with a specificity of 90.0%, reflecting that malignant liver lesion becomes more elliptical in the transverse CT image, and that an increased value of the L/S ratio is associated with increasing probability of hepatocellular carcinoma. Although using the L/S ratio alone seems to be less accurate compared to the short axis diameter (67.4% for L/S ratio vs 79.6% for the short axis diameter), when using a combination of the L/S ratio and short axis diameter, the accuracy improved to 89.8%. This result supports the hypothesis that the hepatocellular carcinomas or malignant lesions showed a larger lesion compared to benign lesion, with clinical silent until more advanced disease, due to an aggressive biological behavior of liver malignancy [11, 13]. In addition, a previous human hepatocellular carcinoma study presented that the L/S ratio is associated with TP53 mutation positive hepatocellular carcinoma [22]. However, to our knowledge, there has been no published scientific report for the TP53 gene mutations in canine hepatocellular carcinoma. Although there is a report from an online dissertation, revealing the absence of TP53 mutations in canine hepatocellular carcinoma [4]. Thus, further investigation should be conducted to confirm the association between the L/S ratio and hepatocellular carcinoma in this study and to investigate the TP53 mutations in a large-scale hepatocellular carcinoma population whether there is a mutation gene similar to the presence in human hepatocellular carcinoma to determine the association between the L/S ratio and the mutation gene in the future.

Regarding the diagnostic performance, area under the ROC curve of the combination of both independent parameters is similar to the results of the previous study [11]. However, the diagnostic performance in the present study shows slightly higher with higher specificity compared to previous study, supporting that other than uniformity of the lesion after contrast uptake in the delayed phase, size of the lesion is an important factor for predicting liver malignancy, and L/S ratio is an optional novel morphometric feature which is simple and is able to predict liver malignancy as well.

Other significant morphometric parameters, including long axis diameter, volume, and surface appearance of the lesion in univariate analysis that fell out from stepwise multivariate analysis reflected that they were covariate variables which was less important compared to the independent ones. However, we did not include and analyze parameters regarding attenuation values and uniformity of the lesion after the administration of contrast medium due to the aim of study in finding morphometric characteristics for differentiating malignant from benign liver lesions. Furthermore, there is a complexity in measuring these parameters which depends on CT protocol and observer's experience in measuring the attenuation values via placing ROI on a defined region, and assessing the uniformity of the lesion, resulting in discrepancies in the results among studies. Thus, further study is needed to evaluate and compare the diagnostic performance of morphometric characteristics and the contrast uptake characteristics of FLLs in predicting liver malignancy.

Limitations of this study include its retrospective design and small number of dogs enrolled with relative lack of variety in histopathologic results, likely leading to a selection bias since some dogs with FLLs did not undergo surgery. Moreover, it is possible that other malignant pathological types of liver lesions may affect the results and accuracy of independent morphometric parameter in this study. However, we included only hepatocellular carcinomas as a representative for malignant liver lesions in the present study since we attempt to determine the morphometric characteristics of hepatocellular carcinoma in dogs whether it is similar to the result of a report on morphometry of human hepatocellular carcinoma [22]. Another limitation is that we included both focal and multifocal liver lesions of dogs in this retrospective study. It is possible that the lesion that was surgical removed for histopathology is not the same lesion presented on CT images. Thus, to minimize this limitation, we included only liver masses or

outstanding discrimination ability with an accuracy of 89.8%. Therefore, this is the first study that reports the association of short axis diameter and L/S ratio with malignant liver lesions.

The short axis diameter and L/S ratio were selected as the morphometric criteria for differentiating malignant from benign liver lesions. For the short axis diameter, a value greater than the cutoff value of 3.30 cm independently predicted hepatocellular carcinoma with an accuracy of 79.6%. Our cutoff value is consistent with the result of previous studies regarding the maximum diameter of the lesion on ultrasonography [6, 14]. In addition, this result is similar to the result from human studies on the differentiation of benign and malignant lymph nodes, suggesting that maximum short axis diameter is an accurate measurement for predicting nodal malignancy [2, 17, 20, 21]. Thus, this parameter might be applicable for the measurement of FLLs to predict liver malignancy as well. However, our result of the short axis diameter is inconsistent with previous CT studies using the maximum transverse diameter for the measurement of the longest diameter as a criterion for predicting liver malignancy [8, 11]. This discrepancy may be due to the different definition of diagnostic criteria for the size measurement. Therefore, practice guidelines for the measurement of FLL size via imaging diagnosis are needed for the standardization of the measurement method.

Interestingly, this study revealed the L/S ratio with a cutoff

nodules that was described about the size and surgical resected location for each lesion on the histopathologic report that is similar to the lesion presented on CT image for investigating the association between morphometric features and histopathologic findings. In addition, this retrospective study was conducted in one institution using a subjective evaluation of the morphometric parameters. Therefore, our results might be influenced by denominator population, and the observer's experience which could lead to different results from other studies. To decrease this impact, we used fixed criteria with simple definition for the measurement method of FLL morphometry, which can be easily repeated with minimum observer variation for image analysis of further investigations in the future.

In conclusion, morphometric parameters, including short axis diameter, and L/S ratio are independent predictors for differentiating malignant from benign liver lesions which a combination of the short axis and L/S ratio shows an outstanding discrimination ability as a promising tool for predicting liver malignancy. In addition, the L/S ratio >1.23 might be specific for increasing risk for malignancy. These results may shed insight into growth characteristics of individual lesions including benignancy and malignancy. Further studies with a large-scale population, presenting different types of lesions from multi-institutions are needed to support our findings and its clinical use.

CONFLICT OF INTEREST. The authors declare no interest or relationship, financial or otherwise that might be perceived as influencing their objectivity in the reporting of this study.

ACKNOWLEDGMENTS. The authors thank Dr. Yumiko Kagawa, an American College of Veterinary Pathologists board-certified pathologist, for her help with the interpretation of all the histopathological findings. We also thank Dr. Tithipong Plangsangmas for his assistance with English language editing.

REFERENCES

1. Belotta, A. F., Gomes, M. C., Rocha, N. S., Melchert, A., Giuffrida, R., Silva, J. P. and Mamprim, M. J. 2019. Sonography and sonoelastography in the detection of malignancy in superficial lymph nodes of dogs. *J. Vet. Intern. Med.* **33**: 1403–1413. [Medline] [CrossRef]
2. Bruneton, J. N., Roux, P., Caramella, E., Demard, F., Vallicioni, J. and Chauvel, P. 1984. Ear, nose, and throat cancer: ultrasound diagnosis of metastasis to cervical lymph nodes. *Radiology* **152**: 771–773. [Medline] [CrossRef]
3. Fukushima, K., Kanemoto, H., Ohno, K., Takahashi, M., Nakashima, K., Fujino, Y., Uchida, K., Fujiwara, R., Nishimura, R. and Tsujimoto, H. 2012. CT characteristics of primary hepatic mass lesions in dogs. *Vet. Radiol. Ultrasound* **53**: 252–257. [Medline]
4. Gomez, D. S. 2019. Mutational evaluation of TP53 gene in the canine hepatocellular carcinoma: a comparative approach. [Master's dissertation]. University of São Paulo [accessed on January 30, 2021].
5. Griebie, E. R., David, F. H., Ober, C. P., Feeney, D. A., Anderson, K. L., Wuenschmann, A. and Jessen, C. R. 2017. Evaluation of canine hepatic masses by use of triphasic computed tomography and B-mode, color flow, power, and pulsed-wave Doppler ultrasonography and correlation with histopathologic classification. *Am. J. Vet. Res.* **78**: 1273–1283. [Medline] [CrossRef]
6. Guillot, M., Danjou, M. A., Alexander, K., Bédard, C., Desnoyers, M., Beaugard, G. and Del Castillo, J. R. 2009. Can sonographic findings predict the results of liver aspirates in dogs with suspected liver disease? *Vet. Radiol. Ultrasound* **50**: 513–518. [Medline] [CrossRef]
7. Kartal, Ö., Ataş, E. and Gürsel, O. 2020. Differentiation of benign from malignant cervical lymphadenopathy by ultrasonography in children. *Arch. Argent. Pediatr.* **118**: 11–17. [Medline]
8. Kutara, K., Seki, M., Ishikawa, C., Sakai, M., Kagawa, Y., Iida, G., Ishigaki, K., Teshima, K., Edamura, K., Nakayama, T. and Asano, K. 2014. Triple-phase helical computed tomography in dogs with hepatic masses. *Vet. Radiol. Ultrasound* **55**: 7–15. [Medline] [CrossRef]
9. Larson, M. M. 2018. Liver and spleen. pp. 792–822. In: *Textbook of Veterinary Diagnostic Radiology*, 7th ed. (Thrall, D. E., ed.), Elsevier, St. Louis.
10. Leela-Arporn, R., Ohta, H., Tamura, M., Nagata, N., Sasaoka, K., Dermlim, A., Nisa, K., Morishita, K., Sasaki, N., Nakamura, K., Takagi, S., Hosoya, K. and Takiguchi, M. 2019. Predictive factors of malignancy in dogs with focal liver lesions using clinical data and ultrasonographic features. *J. Vet. Med. Sci.* **81**: 723–729. [Medline] [CrossRef]
11. Leela-Arporn, R., Ohta, H., Shimbo, G., Hanazono, K., Osuga, T., Morishita, K., Sasaki, N. and Takiguchi, M. 2019. Computed tomographic features for differentiating benign from malignant liver lesions in dogs. *J. Vet. Med. Sci.* **81**: 1697–1704. [Medline] [CrossRef]
12. Liptak, J. M. 2013. Hepatobiliary tumors. pp. 405–412. In: *Withrow and MacEwen's Small Animal Clinical Oncology*, 5th ed. (Withrow, S. J., Vail, D. M., Page, R. L., eds.), Elsevier, St. Louis.
13. Liptak, J. M., Dernell, W. S., Monnet, E., Powers, B. E., Bachand, A. M., Kenney, J. G. and Withrow, S. J. 2004. Massive hepatocellular carcinoma in dogs: 48 cases (1992–2002). *J. Am. Vet. Med. Assoc.* **225**: 1225–1230. [Medline] [CrossRef]
14. Murakami, T., Feeney, D. A. and Bahr, K. L. 2012. Analysis of clinical and ultrasonographic data by use of logistic regression models for prediction of malignant versus benign causes of ultrasonographically detected focal liver lesions in dogs. *Am. J. Vet. Res.* **73**: 821–829. [Medline] [CrossRef]
15. Patnaik, A. K., Hurvitz, A. I. and Lieberman, P. H. 1980. Canine hepatic neoplasms: a clinicopathologic study. *Vet. Pathol.* **17**: 553–564. [Medline] [CrossRef]
16. Patnaik, A. K., Hurvitz, A. I., Lieberman, P. H. and Johnson, G. F. 1981. Canine hepatocellular carcinoma. *Vet. Pathol.* **18**: 427–438. [Medline] [CrossRef]
17. Rahman, M. M., Sadeque, A., Omar, E. and Bhakta, S. K. 2009. Ultrasound differentiation of benign and malignant cervical lymph nodes. *Ibrahim. Med. Coll. J.* **3**: 40–44. [CrossRef]
18. Steinkamp, H. J., Cornehl, M., Hosten, N., Pegios, W., Vogl, T. and Felix, R. 1995. Cervical lymphadenopathy: ratio of long- to short-axis diameter as a predictor of malignancy. *Br. J. Radiol.* **68**: 266–270. [Medline] [CrossRef]
19. Taniura, T., Marukawa, K., Yamada, K., Hikasa, Y. and Ito, K. 2009. Differential diagnosis of hepatic tumor-like lesions in dog by using dynamic CT scanning. *Hiroshima J. Med. Sci.* **58**: 17–24. [Medline]
20. Toriyabe, Y., Nishimura, T., Kita, S., Saito, Y. and Miyokawa, N. 1997. Differentiation between benign and metastatic cervical lymph nodes with ultrasound. *Clin. Radiol.* **52**: 927–932. [Medline] [CrossRef]
21. van den Brekel, M. W. M., Stel, H. V., Castelijns, J. A., Croll, G. J. and Snow, G. B. 1991. Lymph node staging in patients with clinically negative neck examinations by ultrasound and ultrasound-guided aspiration cytology. *Am. J. Surg.* **162**: 362–366. [Medline] [CrossRef]
22. Wang, X., Chen, D. and Chen, B. 2020. The long-to-short-axis ratio and multifocality are associated with TP53 Mutation status in surgically resected hepatocellular carcinomas. *Acad. Radiol.* **27**: 1720–1726. [Medline] [CrossRef]