Article

Diagnostic utility of thoracic radiographs and abdominal ultrasound in canine immune-mediated hemolytic anemia

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Abstract – The utility of thoracic radiographs and abdominal ultrasound to identify abnormalities in canine immune-mediated hemolytic anemia (IMHA) is evaluated. Dogs with regenerative anemias and a clinical diagnosis of IMHA that had thoracic radiographs or abdominal ultrasound performed as part of the evaluation were included. The utility of imaging studies was assessed based on a previously utilized scheme. Patient population and clinical signs were consistent with previous reports of IMHA. In 38 out of 50 dogs, the same clinical evaluation and assessment would have been performed without thoracic radiographs. In 32 out of 64 dogs, the same clinical evaluation and assessment would have been performed without abdominal ultrasound. The results indicate that thoracic radiographs and abdominal ultrasound are of variable utility in identifying concurrent abnormalities in canine patients with IMHA. Prospective studies should be designed to further investigate whether abnormalities identified on imaging studies are related to the IMHA or affect patient prognosis.

Résumé – Utilité diagnostique des radiographies thoraciques et d'échographie abdominale lors d'anémie hémolytique à médiation immunitaire. L'utilité de radiographies thoraciques et d'échographie abdominale pour identifier les anomalies lors d'anémie hémolytique à médiation immunitaire (IMHA) est évaluée. Des chiens avec anémie régénérative et un diagnostic clinique d'IMHA qui avaient eu des radiographies thoraciques ou une échographie abdominale effectuées comme élément de leur évaluation ont été inclus. L'utilité des examens d'imagerie fut évaluée selon un système déjà utilisé. La population des patients et les signes cliniques étaient en lien avec des rapports antérieurs d'IMHA. Chez 38 des 50 chiens, la même évaluation clinique et appréciation auraient été effectuées sans les radiographies thoraciques. Chez 32 des 64 chiens, la même évaluation clinique et appréciation auraient été effectuées sans l'échographie abdominale. Les résultats indiquent que les radiographies thoraciques et l'écographie abdominale sont d'une utilité variable à identifier des anomalies concomitantes chez des patients canins avec IMHA. Des études prospectives devraient être élaborées pour étudier plus à fond si des anomalies identifiées lors d'examen par imagerie sont reliées à l'IMHA ou affectent le pronostic du patient.

(Traduit par D^r Serge Messier)

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Introduction

mmune-mediated hemolytic anemia (IMHA) is one of the most common auto-immune disorders diagnosed in canine medicine (1). This disease is characterized by production of auto-reactive antibodies against antigens on the canine erythrocyte. The stimulus for this autoimmune response can be either primary/idiopathic or secondary to an identifiable disease process in the animal (1).

Although the proportion of primary *versus* secondary IMHA cases is unknown, most cases appear to be primary or idiopathic with estimates reaching as high as 65% to 75% in some studies

(1–4). Suspected causes of secondary IMHA include infectious disease, neoplasia, drug/toxin reactions, envenomation, and vaccination (5–7). The level of evidence-based medicine in the literature supporting these as secondary causes is limited. The reported causes of secondary IMHA are mostly extrapolated from human literature and only a minority have been reported in the peer-reviewed veterinary medical literature (8–11).

Diagnosis and treatment of IMHA has been reviewed previously (1,12). When the diagnostic process is performed in a comprehensive manner it can result in a significant financial burden (1,12). Most of the financial resources during the diagnostic

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Table 1. Selecte	d descriptive data c	of complete blood ce	II count and biochemical	l profile results in th	ne 64 study dogs.
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Complete blood cell count	Mean/Median	Standard deviation/Range	Reference range
Plasma protein	71	11	56 to 73 g/L
Hematocrit	16	3.3 to 20.0	40% to 59%
MCV	74	60 to 95	62 to 77 fl
MCHC	33.9	24.1 to 42.0	330 to 361 g/L
Reticulocyte count	151.2	120.3 to 432.9	$< 105 \times 10^{9}$ /L
Platelet count	123	80 to 712	145 to 463×10^{9} /L
Total leukocyte count	31.1	4.2 to 50.7	4.8 to 13.9×10^{9} /L
Segmented neutrophil count	23.2	1.4 to 38.9	$2.6 \text{ to } 10.8 \times 10^9/\text{L}$
Lymphocyte count	1.2	0.2 to 7.8	1.0 to $4.6 \times 10^{9}/L$
Monocyte count	2.2	0.2 to 13.0	0.1 to 1.1 $ imes$ 10 ⁹ /L
Biochemistry profile	Mean	Standard deviation	Reference range
BUN	7.5	2.14 to 61.1	1.78 to 7.14 mmol/L
Creatinine	61.88	8.84 to 742.1	53.04 to 141.44 mmol/L
Phosphorus	1.25	0.75 to 4.32	1.03 to 2.62 mmol/L
Calcium (total)	2.33	1.99 to 2.89	2.33 to 2.90 mmol/L
Sodium	146	136 to 163	143 to 153 mmol/L
Potassium	3.65	2.6 to 5.7	4.2 to 5.4 mmol/L
Chloride	111	5.2	109 to 120 mmol/L
Bicarbonate	15.5	4.6	15 to 25 mmol/L
ALT	67	6.0 to 3995.0	10 to 55 IU/L
AST	59	10.0 to 2714.0	12 to 40 IU/L
ALP	298	55 to 6800	15 to 120 IU/L
CK	317	53 to 9279	50 to 400 IU/L
Cholesterol	0.15	0.06 to 0.42	0.08 to 0.21 mmol/L
Total bilirubin	22.74	1.71 to 360.1	1.71 to 6.84 µmol/L
Total protein	58	10	51 to 71 g/L
Albumin	30	5 7	29 to 42 g/L
Globulin	29	7	22 to 29 g/L
Glucose	5.88	4.72 to 23.42	4.27 to 6.99 mmol/L

BUN — blood urea nitrogen; ALT — alanine aminotransferase; AST — aspartate aminotransferase; ALP — alkaline phosphatase; CK — creatine kinase; MCV — mean corpuscular volume; MCHC — mean corpuscular hemoglobin concentration.

evaluation are aimed at identifying potential underlying causes of disease and concurrent clinically significant abnormalities. This often includes comprehensive imaging of the patient including thoracic radiographs and abdominal ultrasound. The purpose of this study was to investigate the diagnostic utility of the imaging recommended in cases of canine IMHA based on the presence and significance of abnormalities identified.

Materials and methods

The Health Information Section at The Ohio State University Veterinary Medical Center searched the medical record database between January 2005 and January 2015 for dogs with regenerative anemia (based on institutional reference ranges for hematocrit and reticulocyte count). Medical records were retrospectively reviewed (MA). To be included, the following diagnostic tests had to have been performed as part of the diagnostic work-up: a complete blood (cell) count (CBC), biochemistry profile, urinalysis, and imaging (that included at least thoracic radiographs or abdominal ultrasound). Dogs were included if they had a clinical diagnosis of IMHA that was based on conventional criteria: regenerative anemia, evidence of hemolysis (hyperbilirubinemia, hemoglobinemia, bilirubinuria, or hemoglobinuria), and 2 or more of the following clinicopathologic findings: macroscopic or microscopic agglutination, positive Coombs' test, and 2+spherocytes on slide review (1,13-15). Inclusion into the study required meeting all inclusion criteria and a confirmed diagnosis of IMHA upon review of the file by a Board-certified internist (AR). Cases were excluded if thoracic radiographs and abdominal ultrasound were not performed, if imaging studies

were not performed within the first 48 h after hospital admission, or if analyzed diagnostics were not performed at The Ohio State University Veterinary Medical Center.

Three-view thoracic radiographic and abdominal ultrasonographic studies were reviewed by a Board-certified radiologist blinded to diagnosis (EH). Radiographic reports were generated and then reviewed and used as reference for the study. Definition of normal imaging findings is outlined in Appendix A. Abnormal ultrasonographic findings were tabulated and their clinical significance regarding the diagnosis was evaluated by 4 Board-certified internists (AR, CG, CL, VP). This evaluation was performed in conjunction with case information including results of aspirates or biopsies if acquired during imaging examination. A modified version of a previously published scheme was used for evaluation (16,17). All cytologic and biopsy results used in the subjective grading scheme were reviewed by a Board-certified clinical or anatomic pathologist.

The assessment scheme consisted of 2 questions asked independently for both thoracic radiographic and abdominal ultrasonographic studies. The following example is provided in the context of thoracic radiographs.

Question 1: Did the imaging results contribute to the overall case management? [Overall Diagnostic Utility (ODU)]

Yes

No

Question 2: What was the diagnostic utility of the thoracic radiographs? [Diagnostic Utility Score (DUS)]

 Diagnosis was obtained *via* thoracic radiographs (including image guided biopsy or aspirate).

- 2. Imaging provided information that warranted additional diagnostics for further evaluation due to increased likelihood of changing case management.
- 3. Thoracic radiographs provided descriptive information that did not change case management.
- 4. Thoracic radiographs provided confounding information that did not support or contradicted the diagnosis.
- 5. The thoracic radiographs were diagnostically unremarkable.

The 2 case outcomes (ODU, DUS) were used for further reporting. The purpose of the ODU was to provide a comprehensive assessment of utility after consideration of the complete medical information. The DUS was aimed at identifying specific reasons why imaging was considered either diagnostically useful or not useful during case review. Pertinent patient information from the clinical history, physical examination, and clinicopathologic results was collected from patient files.

Statistical analyses were performed using commercially available software (IBM SPSS Statistics 2012; IBM, Armonk, New York, USA). Descriptive statistics were calculated and reported for age, gender, body weight, and clinical variables. Abnormalities identified during imaging studies as well as ODU and DUS scores are reported. Continuous variables were tested for normality using the Shapiro-Wilk test. Agreement of the 4 reviewers for the subjective utility grading scheme (ODU and DUS) was assessed using Fleiss kappa.

Results

The medical record search retrieved 187 dogs that satisfied the inclusion criteria for diagnosis of IMHA. From this population, 67 were excluded because neither thoracic radiographs nor abdominal ultrasound were completed or were not completed within time limits specified in relationship to hospital admission, 43 were excluded as the diagnostics or diagnosis were performed at another hospital, and 13 dogs had insufficient data available in the medical record. Sixty-four dogs met all inclusion and exclusion criteria and were, therefore, included in the study.

Thirty-two dogs were male with 4 of the dogs intact. Thirtytwo dogs were female with 1 female being intact. Median age was 7 y (range: 3 to 14 y). Median body weight was 19.8 kg (range: 3.5 to 53.6 kg). Breeds included 17 mixed breed dogs, 6 cocker spaniels, 5 shih tzus, 3 miniature schnauzers, 3 beagles, 2 each of following breeds (miniature pinscher, toy poodle, standard poodle, Maltese, Labrador retriever, golden retriever, miniature dachshund, and Pomeranian), and 1 each of the following breeds (Rottweiler, Australian shepherd, collie, keeshond, samoyed, Bouvier des Flanders, Siberian husky, Doberman pinscher, standard schnauzer, Boston terrier, English springer spaniel, Swiss mountain dog, German shepherd dog, and German shorthair pointer). Clinical complaints reported at hospital admission included nonspecific signs in 57 of 64 (89%) dogs (lethargy, depressed state, weakness), gastrointestinal signs in 40 of 64 (63%) dogs (inappetence, vomiting, melena, diarrhea, hematochezia), cardiorespiratory signs in 11 of 64 (17%) dogs (dyspnea, coughing, exercise intolerance, labored breathing, syncope), urinary signs in 13 of 64 (20%) dogs (hematuria, incontinence, polyuria and polydipsia), and neurologic signs in 5 of 64 (8%) dogs (seizure, seizure-like behavior). Pertinent summary data from the CBCs and biochemistry profiles are listed

Table 2. Total number of dogs with each (A) abdominal palpation finding, (B) thoracic radiographic finding, and (C) abdominal ultrasonographic finding.

	Number of dogs
A. Abdominal palpation findings	
Splenomegaly	4
Distended abdomen	3
Organomegaly (non-specific)	3
Abdominal mass	1
Hepatomegaly	1
Total number of dogs	12
B. Thoracic radiograph findings	
Sternal lymphadenopathy	5
Interstitial pattern	4
Cardiomegaly	3
Alveolar pattern	3
Cholelithiasis	2
Aerophagia	1
Megaesophagus	1
Aspiration pneumonia	1
Gastric foreign body	1
Hepatomegaly	1
Pleural effusion	1
Total number of dogs	23
C. Abdominal ultrasound findings	
Peritoneal effusion	11
Liver: Diffuse echogenecity change (hyperechoic)	10
Liver nodules	10
Splenic nodules	7
Edematous gallbladder wall	7
Nonspecific chronic renal changes	6
Hepatomegaly	5
Urinary bladder sediment	5
Renal cortical cysts	4
Gallbladder sludge	4
Lymphadenopathy	4
Splenomegaly	4
Adrenomegaly (1 – unilateral, 2 – bilateral)	3
Cystolith	3
Bilateral pyelectasia	3
Cholecystolith	3
Abdominal mass	3
Pancreatitis	2
Cholecystitis	2
Gastrointestinal wall thickening	1
Emphysematous cystitis	1
Pneumoperitoneum	1
Benign prostate hyperplasia	1
Spleen: Diffuse echogenecity change (hypoechogenecity)	1
Cystitis	1
Mottled liver	1
Splenic vein thrombus	1
Total number of dogs	104

in Table 1. Regarding pertinent clinicopathologic findings; 64 (100%) dogs had spherocytosis, 48 (75%) dogs had nucleated red blood cells noted, 50 (78%) dogs had either macro, micro, or both forms of autoagglutination, and 23 (36%) were Coombs positive.

Agreement analyses among the reviewers for the ODU and DUS are presented in Appendix B. Thoracic radiographic studies were performed in 50 of the 64 dogs (78%). In these studies, 34 (68%) dogs were deemed radiographically within normal limits or diagnostically unremarkable while 16 (32%) had radiographic abnormalities (Table 2). Majority agreement was met in 48 of 50 cases (96%) with the conclusion being that in 10 cases (20%) radiographs contributed to overall case management and

in 38 (76%) they did not contribute to overall case management. Specific findings in the 2 cases of disagreement included mediastinal fat or sternal lymphadenopathy, cholecystoliths, and hepatomegaly in 1 dog, and mild sternal lymphadenopathy and cholecystoliths in the second dog.

On question 2, pertaining to thoracic radiographs, majority agreement was reached in 47 of 50 cases (94%). Split disagreement between the final 3 cases was between an assigned score of 2 or a score of 3. These included 1 dog with mild left atrial enlargement without evidence of heart failure and multiple small metallic gastric foreign bodies, a second dog with mild sternal lymphadenopathy and cholecystoliths, and a third dog with generalized cardiomegaly and mild pleural effusion. In total, 0 dogs received a score of 1, 8 received a score of 2, 5 received a score of 3, 0 received a score of 4, and 34 received a score of 5.

Abdominal ultrasonographic studies were completed in 63 of the 64 dogs (98%). In these studies, 16 (25%) were deemed ultrasonographically within normal limits or diagnostically unremarkable, while 47 (75%) had ultrasonographic abnormalities. Ultrasonographic abnormalities are summarized in Table 2. In 23 cases (37%), there was majority agreement that the imaging study contributed to overall case management. In 32 cases (51%), there was majority agreement that the imaging study did not contribute to overall case management. Specific findings in the 8 cases of split disagreement included 3 with hepatic nodules, 2 with hepatomegaly, 2 with mild peritoneal effusion, and 1 each with gall bladder wall edema, hypoechoic liver, splenomegaly, cholecystitis, mottled liver, decreased renal corticomedullary distinction, gall bladder sludge, cystic calculi, and cholecystolith.

On question 2, pertaining to abdominal ultrasound, majority agreement was reached in 54 of 63 cases (86%). Split disagreement between the final 9 cases was between being assigned a score of 2 or a score of 3. These included the following findings: 5 with hepatic nodules, 2 each with splenomegaly, hepatomegaly, hyperechoic liver, gall bladder wall edema, splenic nodule, renal cyst, and 1 each with urine sedimentation, gall bladder sludge, decreased renal corticomedullary distinction, mild peritoneal effusion, hypoechoic spleen, mild pyelectasia, mottled liver, thrombus in splenic vein, cholecystitis, enlarged right lobe of pancreas, cystitis, and cholelithiasis. In total, 0 dogs received a score of 1, 14 received a score of 2, 23 received a score of 3, 0 received a score of 4, and 17 received a score of 5.

Discussion

The diagnostic utility of both thoracic radiographs and abdominal ultrasound to identify abnormalities was variable in canine IMHA. In total, 68% of thoracic radiographic studies and 25% of abdominal ultrasonographic studies did not reveal any abnormalities. When the findings were interpreted in conjunction with clinical case information, 76% of thoracic radiographic studies would not have changed the clinician's diagnostic and treatment plans in case management and diagnosis, while 50% of abdominal ultrasonographic studies were not useful in case management and diagnosis.

Historical clinical signs, laboratory abnormalities, and imaging findings were largely consistent with those previously reported in canine IMHA (1,12). Prior to this publication, there have been limited descriptions of thoracic and abdominal imaging in patients with canine IMHA. Many imaging findings in this study were consistent with specific disease processes and/or incidental findings, and overlapped significantly with findings reported in both normal dogs as well as dogs affected by other diseases (18–20). Subjectively, there was 1 ultrasonographic imaging finding that seemed to occur more frequently than the authors would have expected: gallbladder wall edema (17%, 11/64 dogs). The cause, relationship, and significance of this finding are unknown and cannot be addressed with this study design. Immune-mediated hemolytic anemia is an inflammatory disease and there is a potential link between gallbladder wall edema and inflammatory reactions (21,22). Further studies are required to investigate this hypothesis and determine the repeatability of this finding.

Importantly, to definitively identify whether an abnormality discovered on imaging studies is the cause of or related to IMHA will require larger, prospective clinical trials specifically designed to overcome the limitations herein. These studies may be able to provide more concrete direction in terms of the true utility of imaging studies, as well as when to proceed with imaging and when not to in the best interest of the patient and client. However, it is important to note that in individual cases, diagnostic imaging was vital to case management and until further information is published, the specifics of each individual case should be considered when recommending any diagnostic test. Furthermore, there can be inherent value in an imaging study without significant abnormalities. Therefore, while this study is able to successfully challenge the dogma that imaging studies are an imperative portion of every canine IMHA evaluation, it is unable to determine the value of imaging in individual cases.

This study is limited by its retrospective nature, which could result in either case selection bias and/or information bias. In the hospital in which the study was performed, the current standard of care for canine IMHA is that all cases receive complete diagnostic work-ups inclusive of thoracic and abdominal imaging. As such, it is standard recommendation to perform these tests unless contraindicated financially or medically. Therefore, selection bias for medical reasons should be minimized in this study as the decision to perform imaging is based to a lesser extent on clinician preference and more on financial restraints.

Furthermore, as this retrospective study describes a single clinical disease process with restrictive inclusion criteria to standardize the imaging results as much as possible, the numbers in this study are low. The reason for including only diagnostic tests that were performed and evaluated at the Ohio State University Veterinary Medical Center was to ensure validity of the imaging results. However, the restrictive inclusion criteria and resulting small number of cases in this series may have excluded some cases of IMHA, including animals which were not initially regenerative and would eventually meet IMHA criteria later in hospitalization. Additional diagnostic tests (e.g., aspirate cytology of imaging abnormalities) were performed on a case-by-case basis and were not controlled. This may have resulted in missing some pertinent findings in cases with abnormalities that were not examined further or cases with normal imaging and underlying diseases. Lastly, not all animals presented to the hospital with IMHA were included due to lack of imaging studies performed. The exact reason why imaging was not performed could not be

determined in most cases. However, the most likely reason was financial, as complete imaging is considered standard-of-care in the institution in which the study was performed.

This study relied on previously used, semi-objective outcome measures; ODU and DUS. There is inherent subjectivity to these scoring categories that can be affected by individual bias. However, the current study attempted to minimize this by using multiple reviewers (4 total) who had independent access to each medical record and evaluated each medical record. The results of the Fleiss kappa indicate that for thoracic radiographs there was consistent and reliable scoring of each case using this system amongst the 4 reviewers. Consistent scoring was not as strong for abdominal ultrasound and was potentially caused by the higher incidence of nonspecific findings on ultrasound evaluations. Therefore, confidence can be placed in the repeatability of the diagnostic utility scoring assignment for thoracic radiographs, while there was a large clinician bias towards ultrasound utility in this study.

Canine IMHA is an expensive and challenging disease to treat. Each individual case work-up will be different, being affected by a myriad of clinical variables, including attending clinician, owner financial constraints, and availability of diagnostic tests. Significant resources are dedicated to the diagnostic tests used to determine the primary or secondary nature of an IMHA case, including thoracic radiographs and abdominal ultrasound. If it is possible to identify treatable secondary causes of IMHA or significant complications of illness and/or concurrent disease, this may lead to multiple benefits to the patient including a faster recovery, enhanced quality of life, and long-term cost savings. Alternatively, if these diagnostic tests fail to provide additional information, the resources and stress to the patient may have opposite deleterious effects. This conundrum underscores the need for follow-up studies on this subject to better identify which patients will benefit from further imaging investigation.

In conclusion, this study demonstrates that the overall utility of these imaging modalities to identify abnormalities is not high. However, the findings are not consistent amongst all cases and therefore cannot be used to advise the clinician to either consistently recommend or not recommend these 2 diagnostic tests in every IMHA patient. Until further information is determined from additional studies, the choice to perform these diagnostic tests should be based on the individual patient, client, and clinician variables and expected case-by-case diagnostic utility.

Appendix A. Definitions of normal imaging findings for canine abdominal ultrasound.

Organ system	Normal description	Reference
Peritoneal space	The peritoneal space was evaluated for increased fluid or gas volume, as well as any identifiable masses or change in echogenicity.	(23)
Lymph nodes	Lymph centers examined included, but were not limited to the jejunal, hepatic, splenic, colic, mesen- teric, gastroduodenal, medial iliacs, and sublumbar lymph nodes. The lymph nodes were relatively isoechoic to surrounding normal soft tissues with regular margins. Normal lymph nodes had a short axis diameter to long axis diameter of < 0.4 cm.	(24,25)
Pancreas	Pancreatic tissue margins were indistinct, and the echogenicity was isoechoic to slightly hypoechoic to that of surrounding mesenteric fat with thickness < 1 cm.	(26)
Adrenal glands	Adrenal glands were hypoechoic to the surrounding fat. Identification of a corticomedullary rim was considered insignificant if appropriately sized. On longitudinal view, the normal glands appeared bilobed to oblong with a maximum width less than 0.81 cm for the right adrenal gland and 0.74 cm for the left adrenal gland.	(27,28)
Liver	Hepatic parenchyma was uniformly hyperechoic/isoechoic to right renal cortex and hypoechoic to spleen with more course echotexture. The caudal margin of the hepatic parenchyma was cranial to the stomach with a sharp angle.	(29)
Gallbladder and biliary tract	Gallbladder wall was a thin echogenic line between anechoic bile (in the normal patient) and the hepatic parenchyma. The gallbladder tapered into the cystic duct. The common bile duct was < 0.3 cm. Dependent echogenic luminal material in the gallbladder was interpreted as sludge and considered insignificant.	(30)
Spleen	Normal splenic architecture was homogeneous with fine echotexture that was hyperechoic the left renal cortex and liver.	(31)
Gastrointestinal tract	The GI tract was evaluated for wall thickness, appearance of wall layers, luminal contents and diameter, and motility. Wall thickness was measured from the inner luminal interface to the outer serosal surface, and considered normal if within published reference ranges (stomach: 2 to 5 mm, duodenum: 3 to 6 mm depending on body weight, jejunum: 2 to 5 mm depending on body weight, ileum: 2 to 4 mm, and colon: 2 to 3 mm). Wall layers were considered normal if all layers were clearly visible, had normal relationship with each other, and were of normal echogenicity.	(32,33)
Urinary tract	The kidneys had a distinction between the cortex and medulla with a normal shape. Size was fairly subjective and if felt abnormal then a renal length to aorta ratio was calculated; abnormal was considered < 5.5 or > 9.1 . The ureters were indistinct with acute tapering at the renal hilus. No luminal hypoechogenicity within the ureter. The renal pelves were < 2 mm on transverse image. The bladder was evaluated for content, wall layer appearance, and wall thickness according to published reference ranges depending upon bladder distention (minimally distended ~ 2.3 mm, moderately distended ~ 1.4 mm) and body weight.	(34–36)
Genitals	If present, ovaries/uterus or the testes were identified. The ovaries were hypoechoic to region peri-renal fat with homogeneous appearance with similar echogenicity to the renal cortex. Uterine horns and body did not have luminal fluid. The testes were located within the scrotum with symmetry and a distinct hyperechoic mediastinum that dissects through homogeneous parenchyma.	(37,38)

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Assessment Majority agreement			Majority agreement	It		Disagreement	Findings in cases of disagreement	Fleiss Kappa
Thoracic radiographs								
ODU			48 out of 50			2 out of 50	Mediastinal fat or sternal lymphadenopathy,	к = 0.72
		Useful		Not Useful	Ι		cholecystoliths, hepatomegaly ($n = 1$); mild sternal lymphadenoapthy, cholecystoliths ($n = 1$).	
		10 out of 48		38 out of 48	8		•	
DUS			47 out of 50			3 out of 50	Mild left atrial enlargement, metallic gastric foreign	к = 0.67
	Category 1	Category 2	Category 3	Category 4	Category 5	(*All disagreement between category 2 and	bodies $(n = 1)$; mild sternal lymphadenopathy, cholecytoliths $(n = 1)$; generalized cardiomegaly, mild	
	0 out of 48	8 out of 48	5 out of 48	0 out of 48	34 out of 48	category 3)	pleural effusion $(n = 1)$.	
Abdominal ultrasound								
ODU			55 out of 63			8 out of 63	Gall bladder wall edema $(n = 1)$, hypoechoic liver	к = 0.25
		Useful		Not Useful	1		(n = 1), splenomegaly $(n = 1)$, cholecystitis $(n = 1)$, hepatic nodules $(n = 3)$, mottled liver $(n = 1)$,	
		23 out of 63		33 out of 63	3		heptomegaly $(n = 2)$, mild peritoneal effusion $(n = 2)$, decreased corticomedullary distinction $(n = 1)$, gall bladder sludge $(n = 1)$, cystic calculi $(n = 1)$,	
							cholecystolith $(n = 1)$.	
DUS			54 out of 63			9 out of 63	Splenomegaly $(n = 2)$, hepatomegaly $(n = 2)$, urine	K = 0.31
	Category 1	Category 2	Category 3	Category 4	Category 5	(*All disagreement between category 2 and	sedmimentation ($n = 1$), gall bladder sludge ($n = 1$), hepatic nodules ($n = 5$), decreased corticomedullary	
	0 out of 63	14 out of 63	23 out of 63	0 out of 63	17 out of 63	category 3)	distinction $(n = 1)$, mild peritoneal effusion $(n = 1)$, hyperechoic liver $(n = 2)$, hypoechoic spleen $(n = 1)$, gall bladder wall edema $(n = 2)$, mild pyelectasia (n = 1), splenic nodule $(n = 2)$, mottled liver $(n = 1)$, thrombus in splenic vein $(n = 1)$, cholecystitis $(n = 1)$,	
							enlarged right lobe of the pancreas $(n = 1)$, renal cyst $(n = 2)$, cystitis $(n = 1)$, cholelithiasis $(n = 1)$.	

References

- Balch A, Mackin A. Canine immune-mediated hemolytic anemia: Pathophysiology, clinical signs, and diagnosis. Compend Contin Educ Vet 2007;29:217–225.
- Jackson ML, Kruth SA. Immune-mediated hemolytic anemia and thrombocytopenia in the dog: A retrospective study of 55 cases diagnosed from 1979 through 1983 at the Western College of Veterinary Medicine. Can Vet J 1985;26:245–250.
- Klag AR, Giger U, Shofer FS. Idiopathic immune-mediated hemolytic anemia in dogs: 42 cases (1986–1990). J Am Vet Med Assoc 1993;202:783–788.
- Reimer ME, Troy GC, Warnick LD. Immune-mediated hemolytic anemia: 70 cases (1988–1996). J Am Anim Hosp Assoc 1999;35:384–391.
- Noble SJ, Armstrong PJ. Bee sting envenomation resulting in secondary immune-mediated hemolytic anemia in two dogs. J Am Vet Med Assoc 1999;214:1026–1027, 1021.
- Lobetti RG, Schoeman T. Immune-mediated haemolytic anaemia: Possible association with *Ancylostoma caninum* infection in three dogs. J S Afr Vet Assoc 2001;72:52–54.
- McCullough S. Immune-mediated hemolytic anemia: Understanding the nemesis. Vet Clin North Am Small Anim Pract 2003;33:1295–315.
- Bloom JC, Thiem PA, Sellers TS, Deldar A, Lewis HB. Cephalosporininduced immune cytopenia in the dog: Demonstration of erythrocyte-, neutrophil-, and platelet-associated IgG following treatment with cefazedone. Am J Hematol 1988;28:71–78.
- 9. Lavergne SN, Danhof RS, Volkman EM, Trepanier LA. Association of drug-serum protein adducts and anti-drug antibodies in dogs with sulphonamide hypersensitivity: A naturally occurring model of idiosyncratic drug toxicity. Clin Exp Allergy 2006;36:907–915.
- Trepanier LA, Danhof R, Toll J, Watrous D. Clinical findings in 40 dogs with hypersensitivity associated with administration of potentiated sulfonamides. J Vet Intern Med 2003;17:647–652.
- Mellor PJ, Roulois AJ, Day MJ, Blacklaws BA, Knivett SJ, Herrtage ME. Neutrophilic dermatitis and immune-mediated haematological disorders in a dog: Suspected adverse reaction to carprofen. J Small Anim Pract 2005;46:237–242.
- Balch A, Mackin A. Canine immune-mediated hemolytic anemia: Treatment and prognosis. Compend Contin Educ Vet 2007;29:230–238; quiz 9.
- Reagan WJ, Irizarry Rovira AR, DeNicola DB, eds. Veterinary Hematology: Atlas of Common Domestic and Non-Domestic Species. 2nd ed. New York, New York: Wiley-Blackwel, 2008.
- 14. Morassi A, Bianco D, Park E, Nakamura RK, White GA. Evaluation of the safety and tolerability of rivaroxaban in dogs with presumed primary immune-mediated hemolytic anemia. J Vet Emerg Crit Care (San Antonio) 2016;26:488–494.
- 15. Wardrop KJ. The Coombs' test in veterinary medicine: Past, present, future. Vet Clin Pathol 2005;34:325–334.
- Leib MS, Larson MM, Grant DC, et al. Diagnostic utility of abdominal ultrasonography in dogs with chronic diarrhea. J Vet Intern Med 2012;26:1288–1294.
- Leib MS, Larson MM, Panciera DL, et al. Diagnostic utility of abdominal ultrasonography in dogs with chronic vomiting. J Vet Intern Med 2010;24:803–808.
- Tong NM, Zwingenberger AL, Blair WH, Taylor SL, Chen RX, Sturges BK. Effect of screening abdominal ultrasound examination on the decision to pursue advanced diagnostic tests and treatment in dogs with neurologic disease. J Vet Intern Med 2015;29:893–899.

- 19. Keyserling CL, Buriko Y, Lyons BM, Drobatz KJ, Fischetti AJ. Evaluation of thoracic radiographs as a screening test for dogs and cats admitted to a tertiary-care veterinary hospital for noncardiopulmonary disease. Vet Radiol Ultrasound 2017;58:503–511.
- Webb JA, Kirby GM, Nykamp SG, Gauthier MJ. Ultrasonographic and laboratory screening in clinically normal mature golden retriever dogs. Can Vet J 2012;53:626–630.
- Quantz JE, Miles MS, Reed AL, White GA. Elevation of alanine transaminase and gallbladder wall abnormalities as biomarkers of anaphylaxis in canine hypersensitivity patients. J Vet Emerg Crit Care (San Antonio) 2009;19:536–544.
- Walters AM, O'Brien MA, Selmic LE, McMichael MA. Comparison of clinical findings between dogs with suspected anaphylaxis and dogs with confirmed sepsis. J Am Vet Med Assoc 2017;251:681–688.
- 23. Frank P. The peritoneal space. In: Thrall D, ed. Textbook of Veterinary Radiology. 6th ed. St. Louis, Missouri: Elsevier, 2013.
- 24. Pugh CR. Ultrasonographic examination of abdominal lymph nodes in the dog. Vet Radiol Ultrasound 1994;35:110–115.
- de Swarte M, Alexander K, Rannou B, D'Anjou MA, Blond L, Beauchamp G. Comparison of sonographic features of benign and neoplastic deep lymph nodes in dogs. Vet Radiol Ultrasound 2011;52: 451–456.
- Hecht S, Henry G. Sonographic evaluation of the normal and abnormal pancreas. Clin Tech in Small Anim Pract 2007;22:115–121.
- Barthez P, Nyland T, Feldman E. Ultrasonographic evaluation of the adrenal glands in the dogs. J Am Vet Med Assoc 1995;207:1180–1183.
- Douglas J, Berry C, James S. Ultrasonographic adrenal gland measurements in dogs without evidence of adrenal disease. Vet Radiol Ultrasound 1997;38:124–130.
- Wu JX, Carlisle CH. Ultrasonographic examination of the canine liver based on recognition of hepatic and portal veins. Anatomy of the portal and hepatic veins of the dog. Vet Radiol Ultrasound 1995;36:227–233.
- Nyland TG, Mattoon JS, Herrgesell EJ, et al. Liver. In Nyland TG, Mattoon JS, eds. Small Animal Diagnostic Ultrasound. 3rd ed. St. Louis, Missouri: Saunders 2015:338.
- Nyland TG, Mattoon JS. Spleen. In: Nyland TG, Mattoon JS, eds. Small Animal Diagnostic Ultrasound. 3rd ed. St. Louis, Missouri: Saunders, 2015:400–403.
- Penninck DG, Nyland TG, Fisher PE, et al. Ultrasonography of the normal canine gastrointestinal tract. Vet Radiol 1989;30:272–276.
- Delaney F, O'Brien R, Waller K. Ultrasound evaluation of small bowel thickness compared to weight in normal dogs. Vet Radiol Ultrasound 2003;44:577–580.
- Nyland TG, Widmer WR, Mattoon JS. Urinary tract. In: Nyland TG, Mattoon JS, eds. Small Animal Diagnostic Ultrasound. 3rd ed. St. Louis, Missouri: Saunders, 2015:557–600.
- Mareschal A, d'Anjou MA, Moreau M, et al. Ultrasonographic measurement of kidney-to-aorta as a method of estimating renal size in dogs. Vet Radiol Ultrasound 2007;48:434–438.
- Geisse AL, Lowry JE, Schaefffer DJ, et al. Sonographic evaluation of urinary bladder wall thickness in normal dogs. Vet Radiol Ultrasound 1997;38:132–137.
- Davidson AP, Baker TW. Reproductive ultrasound of the bitch and queen. Top Companion Anim Med 2009;24:55–63.
- Pugh CR, Konde LJ, Park RD. Testicular ultrasound in the normal dog. Vet Radiol 1990;31:195–199.