



Ultrasonographic examination in liver and gallbladder pathology in dogs. Clinico-ecographic correlations

Daniela M. Neagu, Robert Purdoiu, Cristian Popovici, Razvan Codea, Alexandra Biris

Department of Clinical Sciences, Faculty of Veterinary Medicine, University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, Cluj-Napoca, Romania. Corresponding author: D. M. Neagu, daniela.neagu@usamvcluj.ro

Abstract. Hepatobiliary disorders in dogs evolve with a variety of clinical signs, many of them non-specific, such as lethargy, nervous signs, weight loss, jaundice, ascites, gastrointestinal disturbances (anorexia, vomiting, diarrhea). The diagnosis of certainty needs complementary examinations, and among these, at least in human medical practice, ultrasonographic examination stands out. The observational study involved 50 owner-owned dogs of various breeds, ages (over 1 year old), sexes, and body weights, divided into two groups: group 1 - 20 patients with severe clinical symptoms; and group 2 - 30 patients with moderate clinical symptoms. The patients of both groups were clinically and paraclinically investigated by laboratory explorations and abdominal ultrasonographic examination. The diagnosis of hepatobiliary disorders is established by correlating the anamnesis and the results of paraclinical examinations with ultrasonographic changes. Ultrasonography is often the first method of investigation of the hepatobiliary system and intra-abdominal organs. The non-invasive method is the most commonly used screening method in animals suspected of hepatobiliary disorders, allowing the differentiation of focal from diffuse abnormalities, and cystic from solid and vascular abnormalities.

Key Words: dog, hepatobiliary disorders, ultrasonography.

Introduction. One of the biggest challenges in veterinary medicine is to detect diseases in an organ as metabolically active as the liver. The second largest organ in the body, it performs approximately 1500 essential biochemical functions. From metabolizing drugs, neutralizing exogenous and endogenous toxicants, to synthesizing substances - albumin, blood clotting factors - the liver is the key factor in digesting food and metabolizing nutrients. In other words, this organ influences the nutritional status through its role in the metabolism of proteins, carbohydrates, lipids, vitamins, as well as in the elaboration of bile salts (Barone 1997; Meyer 2000; Mannion 2006).

Hepatobiliary disorders in dogs evolve with a variety of clinical signs, many of them non-specific, such as lethargy, nervous signs, weight loss, jaundice, ascites, gastrointestinal disturbances (anorexia, vomiting, diarrhea). Anorexia together with gastrointestinal disorders and metabolic disturbances associated with liver disease contribute to chronic weight loss. Jaundice may be seen especially in severe cholestasis and discolored feces are the result of bile duct obstruction (Codreanu 2000c; Codreanu 2010; Rothuizen 2006). Nervous signs of hepatic encephalopathy are commonly seen in animals with portosystemic vascular abnormalities, and in all situations that progress to reduced liver function. These include: ataxia, lethargy, fatigue, ptyalism, altered consciousness (disorientation, stupor, rarely coma), head compressions of surrounding objects, gait in maneuver and tremors (Mannion 2006; Larson et al 2021).

The normal liver is quite difficult to examine (palpation) in dogs due to its anatomical position (Barone 1997; Codreanu 2000a; Constantinescu 2005). Hepatomegaly of various causes (passive venous congestion, inflammation, neoplasia, nodular hyperplasia, fatty infiltrates, amyloid or glycogen) is relatively easy to palpate.

On the other hand, the reduced liver size associated with abdominal distension due to ascites makes palpation difficult (Haroutunian 1995; Codreanu 2010; Kumar et al 2012).

A large number of conditions lend themselves to confusion with liver disease because of clinical manifestations and paraclinical changes. Thus, increased transaminases: alanine aminotransferase (ALT) and aspartate aminotransferase (AST) may be found in quadriplegic patients with pancreatitis, diabetes mellitus, hyperthyroidism, cardiac pathology (Rothuizen & Meyer 2000). High values of alkaline phosphatase (ALP) associated with moderate values of alanine aminotransferase (ALT) may be found in dogs with hyperadrenocorticism, while bilirubinaemia is also present in prolonged anorexia or infectious diseases (Mwanza 1998; Syakalina 1998).

Another important aspect is that the diagnosis of the type of disease, parenchymal, vascular or biliary liver disease on the basis of the anamnesis, clinical and laboratory examination alone can only be presumptive. The diagnosis of certainty needs complementary examinations, and among these, at least in human medical practice, ultrasonographic examination stands out.

Radiography is widely available and recommended in dogs and cats suspected of liver disease, but is an insensitive method (Codreanu 2000b; Larson 2007). Contrast radiographic studies, such as intravenous cholangiography and percutaneous transhepatic cholangiography for the diagnosis of biliary obstruction are described (Partington & Biller 1995; Rothuizen 2006), but have not entered common use and have largely been replaced by ultrasonography. Over time, this has become a complementary technique to abdominal radiography providing a more detailed examination of the internal structure of the liver and adjacent organs (Partington & Biller 1995; Nyland et al 2002; Rothuizen 2006; D'Anjou 2008).

Liver ultrasonography, as a diagnostic technique, may be used routinely or in the context of abnormalities detectable on inspection, palpation and percussion, the presence of liver failure syndromes, and positive functional tests. It can be concluded that ultrasonography is the most commonly used modality for the detection of animals suspected of liver disease, including vascular abnormalities (Rothuizen & Meyer 2000; Larson 2016).

The present scientific endeavor aims to highlight the role of ultrasonographic examination in the diagnosis of hepatobiliary disorders in dogs, and what it involves: establishment of correlations between clinical manifestations and the level of hepatic biochemical parameters in canine patients under investigation; establishment of correlations between clinical aspects and ultrasonographic changes identified; identification and specification of the performance of the ultrasonographic method in canine hepatobiliary diseases; prevalence of the type of liver and biliary diseases in canine patients under investigation.

Material and Method. The observational study was conducted at the Internal Medicine Clinics and the Radiology and Imaging Department of the Faculty of Veterinary Medicine of the University of Agricultural Sciences and Veterinary Medicine, Cluj-Napoca, Romania between 2021-2023. It involved 57 owner-owned dogs of various breeds (German shepherd, cocker spaniel, Rottweiler, Saint-Bernard, fox terrier, Dog de Bordeaux, poodle, bichon, Doberman, as well as their mixes), ages (over 1 year old), sexes, and body weights.

Inclusion criteria: patients with general clinical and digestive signs that are equally attributable to hepatobiliary disorders. Exclusion criteria: patients with other (renal, cardiac and endocrinological) disorders other than digestive disorders. From the initial group of 57 dogs, only 50 met the criteria for inclusion in the study.

Based on the provided criteria, the patients (n=50) were divided into two groups: group 1 consisted of 20 patients with severe clinical symptoms: fever, anorexia, vomiting with a glabrous/bilious appearance, dehydration, abdominal pain, jaundice, diarrhea/discholored feces (acholi), septic shock; group 2 consisted of 30 patients with moderate clinical symptoms: inconstant fever, vomiting, right hypochondrial tenderness, anorexia, weight loss, jaundice, lethargy, polyuria/polidipsia, dehydration, ascites.

The patients of both groups were clinically and paraclinically investigated by laboratory explorations and abdominal ultrasonographic examination. The research was supported by the Clinical Laboratory of the Faculty of Veterinary Medicine Cluj-Napoca. Various parameters, including haematological (CBC), coagulation time (PT; APTT) and biochemical parameters, albumin (ALB), glucose (GLU), triglycerides (TRG), cholesterol (COL), aspartate aminotransferase (AST)/glutamic oxaloacetic transaminase (GOT), alanine aminotransferase (ALT)/glutamic-pyruvic transaminase (GPT), gamma-glutamyl transferase (GGT), urea (U), creatinine (Cr), and bilirubin (Bil), were quantified in the serum samples using specific assay kits from Elabscience Biotechnology Inc. (Texas, HT, USA). All biochemical analyses were rigorously validated and conducted via spectrophotometry with the SPECTROstar® Nano microplate spectrophotometer from BMG Labtech in Germany. The kits utilized in the current study were procured from Elabscience Biotechnology Inc., located in Texas, HT, USA. The additional chemicals were acquired from Sigma Aldrich and Merck, both located in Darmstadt, Germany.

Statistical analysis was performed using GraphPad Prism 9 software (San Diego, CA, USA). Data were evaluated with an unpaired t-test with Welch correction. Significance levels were set at $p < 0.05$, $p < 0.01$, $p < 0.001$, and $p < 0.0001$, to assess differences between the two groups of patients. The two-stage step-up method (Benjamini, Krieger, and Yekutieli) was used for all determinations, and results were presented as mean values \pm standard deviations.

Preparation of patients for the ultrasonographic examination included: diet 6-12 hours before the examination, defecation (but not urination) before the examination, toileting of the examination area (trimming or shaving, degreasing with hydroalcoholic solution), restraint (right, left or dorsal lateral decubitus, sedation for non-cooperative patients) and application of ultrasound gel (with respect to the waiting time for penetration into the cutis).

Abdominal ultrasonographic exploration was performed in the Medical Pathology Clinic and in the Radiology and Medical Imaging Department with MindRay DC-6 and Esaote MyLab™ 40 VET (Esaote SpA, Genoa, Italy) ultrasound scanners, both equipped with 3C5A convex probe 2-5 MHz, CA123 microconvex probe 9-3 MHz and 7L4A linear probe 5-10 MHz in B-mode and color Doppler.

Results and Discussion

Clinical-anamnestic results. Of the 50 patients enrolled in the two groups, 22 were male and 28 were female. The mean age of the investigated patients was 8.3 ± 3.68 years with extremes of 2 years and 16 years. By sex, the mean age did not differ significantly, being 8.92 ± 3.32 years in females and 8.77 ± 3.77 years in males. The mean age between the two groups did not show significant differences, being 8.63 ± 3.44 years for group 1 and 8.5 ± 4.01 years for group 2.

Clinical signs had a different frequency in the two groups of patients investigated: Lot 1 patients presented with high frequency jaundice (90%), abdominal pain (95%), diarrhea (95%), fever (90%), vomiting (80%), anorexia (75%), while Lot 2 patients presented with high frequency lethargy (90%), anorexia (86.67%) weight loss (93.33%), polyuria/polydipsia (50%), abdominal discomfort (60%) and ascites (40%) (Table 1).

Blood metabolic results. Paraclinical evaluations were performed haematologically, biochemically and ultrasonographically. Haematologically, moderate non-regenerative anaemia (haemoglobin $< 14 \text{ g dL}^{-1}$) was found in 74% of group 2 patients and 27% of group 1 patients. In group 2, the anaemia was accompanied by erythrocyte morphological changes (poikilocytes, acanthocytes, target erythrocytes). Marked leukocytosis ($> 19000 \text{ mm}^{-3}$), accompanied by neutrophilia with leftward deviation of the Arneth curve, was seen in 83% of patients in group 1, while only 29% of patients in group 2 developed moderate leukocytosis ($> 16000 \text{ mm}^{-3}$) with predominantly rightward deviation. The haematological changes observed in the patients included in the study are consistent with data reported in the literature.

Table 1

Prevalence of clinical symptoms in the studied groups

Group 1	Percentage	Group 2	Percentage
Fever	90%	Fever	16.67%
Vomiting	80%	Vomiting	33.3%
Anorexia	75%	Abdominal tenderness	60%
Dehydrate	50%	Disapetența	86.67%
Abdominal Pain	95%	Weight loss	93.33%
Jaundice	90%	Jaundice	33.33%
Diarrhea	95%	Lethargy	90%
Septic shock.	25%	Polyuria/polidipsia	50%
Weight loss	50%	Ascites	40%

The biochemical parameters quantified in relation to clinical manifestations showed some differences in the two groups. The mean slightly elevated glycemc mean values did not differ significantly between the two groups, being 132.04 ± 22.91 mg dL⁻¹ in group 1 patients and 132.46 ± 35.42 mg dL⁻¹ in group 2 patients ($p < 0.05$). Hyperglycemia was observed in dogs with cirrhosis and in those with portosystemic shunts, in contrast to human patients with cirrhosis in whom stored glycogen is rapidly mobilized and consumed (within 10-12 hours), leading prematurely to protein catabolism with gluconeogenesis from amino acids. Much more common than hypoglycemia in cirrhotic patients is glucose intolerance, with about 80% of them having this disorder. Glucose intolerance in dogs with liver disease is poorly documented in terms of importance and causes.

Transaminases, enzymes that quantify the hepatocyte status (especially in dogs ALT), showed increased mean values in both groups: group 1 with 149.95 ± 74.04 u L⁻¹ and group 2 with 204.21 ± 158.60 u L⁻¹ for ALT, and for AST of group 1 with 108.97 ± 28.30 u L⁻¹ and group 2 with 138.87 ± 105.56 u L⁻¹. Compared with reference values of ALT < 40 u L⁻¹, AST < 30 u L⁻¹ (Ghergariu et al 2000) the increase of transaminase values is statistically ensured ($p < 0.05$).

Unlike in human medicine, where both transaminases quantify the liver, ALT is more specific for liver disease in dogs than AST. This is due to the different intrahepatocyte localization of the two enzymes: ALT is present in the cytoplasm, being released at the smallest hepatocyte membrane injury, whereas AST has a mitochondrial localization and is released under conditions of cell death. AST is present in large amounts in other organs, like heart and skeletal muscle. Increased serum activity of both enzymes seems to indicate severe liver damage, in contrast only to increased ALT.

Coagulation parameters, PT and APTT had mean values above baseline, but without statistically significant differences between the two groups. In dogs with chronic hepatitis, with or without cirrhosis, there was a reduction in coagulation factors as a result of impaired hepatic synthesis and reduced protein synthesis.

Biliary enzymes showed increased mean values with statistically assured differences between the two groups and correlated with the intensity of clinical manifestations: PAL in group 1 was 658.75 ± 628.20 U L⁻¹ and in group 2 it was 308.03 ± 269.55 U L⁻¹ ($p < 0.05$). GGT had values of 80.03 ± 41.56 U L⁻¹ in group 1 and 48.45 ± 35.87 U L⁻¹ in group 2 ($p < 0.05$). In dogs, the two enzymes increase in parallel, one of the factors being cholestasis. In humans, increased bile enzymes are associated with liver cirrhosis, metastatic carcinoma and liver infiltrates.

The liver is the major site of cholesterol synthesis. Therefore, cholesterol is an important marker of lipid metabolic processes in the liver. Its mean values showed statistically significant differences between the two groups: group 1 with 376.82 ± 48.33 mg dL⁻¹ and group 2 with 588.47 ± 354.34 mg dL⁻¹ ($p < 0.05$). Hypercholesterolaemia correlates positively with jaundice, which is a major indicator of bile duct obstructions. In humans, hypercholesterolaemia is associated with atherosclerosis or ischaemic heart disease, associations that have not been found in animals.

Albumin, a marker of hepatic synthetic function, showed statistically significant low mean values in group 2 patients ($1.34 \pm 0.24 \text{ g dL}^{-1}$) ($p < 0.001$), in contrast to group 1 ($1.77 \pm 0.34 \text{ g dL}^{-1}$) and to the reference level: $1.81\text{--}3.13 \text{ g dL}^{-1}$ (Ghergariu et al 2000). Hypoalbuminaemia implies more than 70% loss of liver functional capacity as a result of cirrhosis, systemic porto-systemic encephalopathy and severe necrosis. An element indicating hepatobiliary impairment is bilirubin, whose mean values significantly increased in both groups (group 1: $15.02 \pm 3.12 \text{ mg dL}^{-1}$; group 2: $10.19 \pm 3.46 \text{ mg dL}^{-1}$), with statistically significant differences ($p < 0.001$).

There is an important correlation between the mean values of this parameter and the mean values of albumin: as bilirubin values increase, albumin values decrease. High values correlate positively with haemolysis, hepatocellular dysfunction and extrahepatic bile duct cholestasis or obstruction.

Ultrasound results. Canine patients from the two groups were evaluated by abdominal ultrasonography. The ultrasonographic changes observed involved the hepatic parenchyma, cholecyst and bile ducts. The World Small Animal Veterinary Association's Liver Standardization Group recently categorized canine and feline hepatic disease into four main groups: parenchymal disease, neoplastic disease, biliary disorders, and vascular disorders (Larson et al 2021).

Patients in group 1 ($n=20$) were predominantly categorized in the third category, biliary disorders. Thus, inflammatory gallbladder changes, cholecystitis, were identified sonographically in 50% of the canine patients ($n=10$), lithiasic cholecystitis in 30% ($n=6$), mucoceles in 10% ($n=2$), and in two patients (10%) no hepatobiliary changes were identified, their suffering being pancreatic or gastrointestinal.

Of the inflammatory changes, 41% ($n=8$) were acute cholecystitis and 19% ($n=4$) were chronic cholecystitis. Ultrasonographically dominant for acute cholecystitis was parietal thickening with inflammatory infiltrative or oedematous appearance with parietal halo with the presence of sludge and varying degrees of tone and contractility impairment (Figure 1a). In chronic cholecystitis the parietal thickening is dominated by the presence of connective fibers (muscular and subserosal) resulting in a scleroatrophic appearance.

Cholelithiasis present in 30% of the patients was characterized ultrasonographically by the presence of an intravesical hyperechoic hyperechoic focus, with regular and invariable contour and a posterior acoustic shadow dependent on gravity (Figure 1b). The ultrasonographic findings of the gallbladder wall and its contents are consistent with those described in the literature (Codreanu 2000a; Brömel et al 1998).

In a rather small percentage of 10%, mucocele, an important cause of jaundice and obstructive disease in dogs, was identified. It takes on varied sonographic appearances. In the patients studied, we did not identify the classic "kiwi fruit" appearance with hyperechogenic striations radiating from a central point with depletion of the gallbladder walls, but the appearance of hyperechogenic intravesical sediment defying gravity as opposed to sludge (Gaschen 2009). Biliary sludge had a prevalence of 60% ($n=12$) in patients in this group. Ultrasonographic examination provided information about the changes in the cholecyst, namely parietal thickening, sometimes with a double contour appearance, the nature of the intravesical contents being sludge or calculi (Figure 2).

Statistical significance analysis for the identification of cholecystitis showed a sensitivity of 89.6%, specificity of 87.7%, positive predictive value (PPV) of 87.7%, negative predictive value (NPV) of 86.7% and accuracy of 82.5%. For the identification of biliary lithiasis, a sensitivity of 98.9%, specificity of 100%, PPV of 98.5%, NPV of 97.9% and accuracy of 95.2% were assessed.

It can be appreciated that the sonographic aspects identified in patients in group 1 correlate with the clinical manifestations presented and with the level reached by some biochemical parameters.

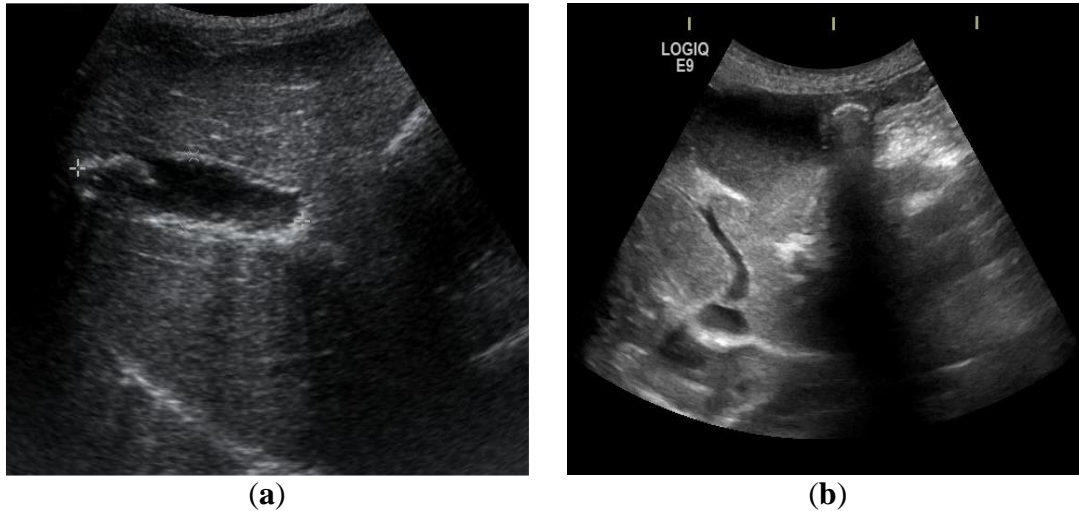


Figure 1. (a) Acute cholecystitis - Schnauzer dog, 10 years old, thickened cholecyst with hyper-echogenic wall; (b) gallstone - German shepherd, 9 years old, hyper-echogenic formation with posterior acoustic umbra (Internal Medicine Clinic and Radiology and Medical Imaging Department).

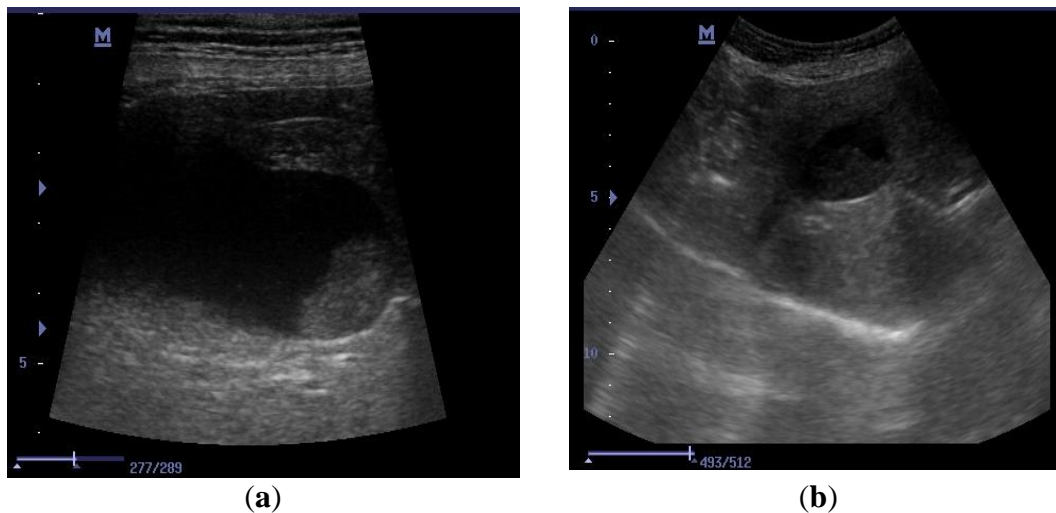


Figure 2. (a) Mucocell - Cocker dog, 9 years old, intravesical formation, hyper-echogenic, non-gravitational; (b) sludge - Cocker dog, 10 years old (Internal Medicine Clinic and Radiology and Medical Imaging Department).

In group 2 patients (n=30), parenchymal liver diseases with diffuse echogenicity changes and, in a reduced proportion, vascular liver diseases predominated. Of the diffuse parenchymal liver diseases, liver cirrhosis (macronodular/micronodular) was identified in 40% (n=12) and cholangiohepatitis in 10% (n=3) of the patients. Cardiac liver is the vascular hepatopathy present in 13.4% (n=4) of group 2 patients, a higher prevalence holds lymphoma 36.6% (n=11) as secondary tumoural hepatopathy. Biliary sludge had a prevalence of 56.6% (n=17) without cholecystitis involvement. Hepatic cirrhosis, the end-stage of chronic hepatitis, was characterized by diffuse hyperechogenic, relatively homogeneous liver parenchyma, the homogeneity of the parenchyma being affected by the presence of regenerating nodules and foci of steatosis (Figure 3a). Ascites was identified clinically and sonographically in 33.3% (n=10) and only sonographically in 6.66% (n=2).

In cholangiohepatitis, the sonographic changes consisted of hypoechoic liver parenchyma in contrast to the pronounced hyperechogenicity of the portal vein walls, abnormalities of the cholecystis and intrahepatic bile ducts.

Cardiac liver is an established vascular hepatopathy following right heart failure. Hepatomegaly, the dilatation of the hepatic venous system (the veins lose respiratory kinetics, which indicates the cardiac origin of the hepatomegaly) and abundant ascites were found. Malignant lymphoma was characterized by decreased hepatic echogenicity with the presence of a solitary mass with a hypererectogenic centre (target lesion) (Figure 3b).

The ultrasonographic examination of patients in group 2 allowed the identification of diffuse changes of the liver parenchymal parenchyma-hepatic cirrhosis. Therefore, the sensitivity of the method was assessed as 75.6%, specificity of 100%, PPV of 100%, NPV of 88.9% and accuracy of 85.7%. In the identification of mixed liver parenchymal lymphoma changes, the sensitivity was 90.1%, specificity 100%, PPV 100%, NPV 78.9% and accuracy 93.8%.

Regarding the prevalence of hepatobiliary disorders in canine patients in the study, liver cirrhosis was diagnosed in 24% (n=12), followed by malignant lymphoma in 22% (n=11), cholecystitis in 20% (n=10), cholelithiasis in 12% (n=6), cardiac liver in 8% (n=4) and mucocell liver in 4% (n=2) (Table 2).

Table 2

Prevalence of hepatobiliary disorders in study dogs

<i>The condition</i>	<i>Percentage</i>
Liver cirrhosis	24%
Malignant lymphoma	22%
Cholecystitis	20%
Cholelithiasis	12%
Cardiac liver	8%
Cholangiohepatitis	6%
Mucocell	4%

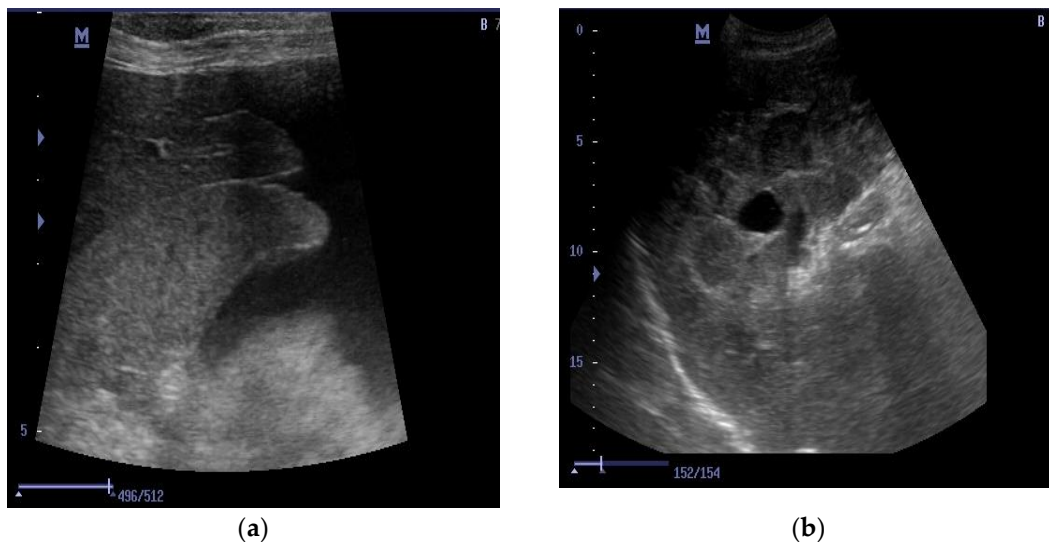


Figure 3. (a) Hypotrophic macronodular cirrhosis - Amstaff dog, 10 years old- hyperechogenic, relatively homogeneous liver with lobar evidence and ascites; (b) Lymphoma - Rottweiler dog, 9 years old, enlarged liver, relatively homogeneous echostructure, slightly hyperechogenic and presence of hypoechoic masses (Internal Medicine Clinic and Radiology and Medical Imaging Department).

Hepatobiliary disorders in dogs, unlike in humans, are characterized by a polymorphic and non-specific symptomatology. Recognizing hepatobiliary disorders on the basis of

history and clinical manifestations is difficult (Brömel et al 1998; Codreanu 2000c). However, there are some clinical features that point towards liver disease, namely jaundice with or without anemia, hepatomegaly without the presence of cardiac disease and ascites (resulting from the combination of hypoalbuminemia and portal hypertension) (Barrera 1994; Mihai & Andronie 2000; Gaschen 2009). These clinical features were also observed in the studied dogs. The statistical analysis of the paraclinical explorations carried out on the two groups of canine patients indicates that the parameters analyzed are useful in detecting hepatobiliary disorders.

Non-regenerative anaemia, recorded in a high proportion in dogs in group 2, accompanies the course of chronic hepatobiliary disorders, chronic haemorrhage and malnutrition. Leukocytosis, with higher prevalence in dogs from batch 1, characterizes acute forms of hepatobiliary diseases (Goggin et al 1997).

Hyperglycaemia has been observed in dogs with cirrhosis and in those with portosystemic shunts, in contrast to human patients with cirrhosis, where stored glycogen is rapidly mobilized and consumed (within 10-12 hours), leading prematurely to protein catabolism with gluconeogenesis from amino acids. Much more common than hyperglycaemia in cirrhotic patients is glucose intolerance. About 80% have this disorder. Glucose intolerance in dogs with liver disease is poorly documented in terms of importance and causes (Pop 1998; Meyer et al 2010).

With regard to coagulation parameters, no statistically significant differences were found in dogs with chronic hepatitis, with or without cirrhosis. Reduced coagulation factors were found as a result of impaired hepatic synthesis and reduced protein synthesis. Disorders in coagulation indicate severe liver dysfunction with reduced protein synthesis (Codreanu & Mihai 2000; Lessa et al 2010).

Significantly low albumin values ($p < 0.001$) in group 2 dogs are an indicator of 70% loss of liver function (Nyland et al 2002). There is an inverse correlation between mean albumin and mean bilirubin value. Bilirubin, a parameter that quantifies biliary function, showed statistically significant increases ($p < 0.001$) in patients in both groups compared to the reference mean (Ghergariu et al 2000), but non-significant between groups. High values correlate positively with haemolysis, hepatocellular dysfunction and extrahepatic bile duct cholestasis or obstruction as documented by Smith et al (1998).

Hepatic cytolysing enzymes showed increases in mean values more markedly in group 2, indicative of hepatocyte distress. Unlike in human medicine, where both transaminases quantify the liver, ALT is more specific for liver disease in dogs than AST (Pop 1998; Codreanu 2000c; Codreanu 2010). This is due to the different intrahepatocyte localization of the two enzymes: ALT is present in the cytoplasm, being released at the smallest hepatocyte membrane injury, whereas AST is mitochondrially localized and is released under conditions of cell death. Analyses of serum activity of both enzymes seem to be more indicative of severe liver damage, in contrast to only that of ALT (Larson 2007; Kumar et al 2012). Biliary enzymes ALP and GGT by higher values recorded in group 1 patients plead for a predominantly biliary distress. In pets, GGT increases in parallel with ALT.

We can state with certainty that serum levels of bilirubin, albumin, transaminases, biliary enzymes along with the coagulation profile provide information about the functional capacity of the liver. These tests taken separately have no diagnostic relevance, each being rather taken out of context (Mwanza 1998; Meyer 2000).

The ultrasonographic examination of the two groups of dog patients allowed the identification of the type of disease: hepatic, biliary or hepatobiliary, responsible for their clinical condition. In the patients of group 1, ultrasonographic examination revealed with high specificity, inflammatory gallbladder changes, cholecystitis and cholelithiasis. The dominant sonographic feature of the reactive process was the thickening of the parietal components and the change in the bladder content, which became inhomogeneous (Weill 1991; Smith et al 1998). Statistical analysis showed a sensitivity of 89.6% and a specificity of 87.7%.

The changes in the bile, in the sense of loss of an anechoic appearance with the identification of elements in suspension or sediment or calculi, were easily highlighted due to the high degree of specificity of these changes (Gaschen 2009; Codreanu 2010).

Biliary sludge is defined as a solid or semi-solid suspension of particles in more or less viscous fluid. Ultrasonographically, it is defined as a low-amplitude echo pattern without acoustic shadow (Weill 1991; Partington & Biller 1995). In human patients, biliary sludge positively correlates with acute cholangitis, acute cholecystitis being the precursor of cholelithiasis. Studies have shown that its prevalence in patients without gallbladder disease is 1.7%, and in patients with other gastrointestinal disorders is 5.1% (Brömel et al 1998; Pop 1998). In dogs, its presence is not associated with hepatobiliary disease. Brömel et al (1998) found that 58% of dogs without hepatobiliary disease had sludge. In the present study, we found 17 dogs (56.6%) with sludge without biliary involvement in dogs in group 2. It can be concluded that biliary sludge in dogs rarely causes gallstone formation.

Cholelithiasis was evidenced ultrasonographically by the presence of hyperechogenic formations in the gallbladder lumen, with regular contour that respects gravity, with posterior acoustic shadow and evident parietal involvement by thickening (as a side effect) (Penninck & Finn-Bodner 1998). In the identification of lithiasis, the ultrasonographic examination had a sensitivity of 98.9% with a specificity of 100%.

In the second group of patients, the ultrasonographic changes were mostly diffuse parenchymal - liver cirrhosis. The ultrasonographic criteria for liver cirrhosis were increased echogenicity of fibrous tissue, lack of visualization of the portal vein walls, attenuation phenomenon and macrogranular appearance. These criteria had a sensitivity of 75.6% and a specificity of 100%. According to Lessa et al (2010), ultrasound criteria for the diagnosis of liver cirrhosis had a sensitivity of 70.6%, with a specificity of 100%.

Liver cirrhosis was accompanied by the onset of ascites identified clinically and ultrasonographically in 58.4% (n=7) of patients with cirrhosis, and only ultrasonographically in 41.6% (n=5). In dogs, the regenerative nodules in liver cirrhosis have a distinct outline, compared to the regenerative nodules in humans, which are iso- or hypoechoic in relation to the rest of the liver parenchyma (Nyland & Matoon 1995).

In dogs, metastatic liver tumours are more prevalent than primary tumours. These include lymphoma characterized sonographically by decreased liver echogenicity with a solitary mass (Codreanu & Mihai 2000; Nyman et al 2004). Ultrasonography had 90.1% sensitivity and 100% specificity in detecting focal lesions. Lessa et al (2010) assessed ultrasonographic examination as having 90% sensitivity and 100% specificity in identifying focal lesions. Focal liver lesions have a positive predictive value for malignancy of 74%. Hepatic and biliary cysts are benign diseases that may resemble malignancy (Meyer 2000).

The prevalence of clinically and paraclinically suspected and ultrasonographically confirmed hepatobiliary disorders in the dogs in the study is in line with the literature. Thus, liver cirrhosis had a prevalence of 24%, lymphoma 22%, cholecystitis 20%. Meyer (2000) and Larson (2016) identified hepatic cirrhosis at 17%, malignant lymphoma at 14% and cholecystitis 12%.

Finally, we can say that the diagnosis of hepatobiliary disorders is established by correlating the anamnesis and the results of paraclinical examinations with ultrasonographic changes. On the basis of these correlations, it is possible to say whether a disease is acute or chronic (active). Ultrasonography is often the first method of investigation of the hepatobiliary system and intra-abdominal organs. Non-invasive is the most commonly used screening method in animals suspected of hepatobiliary disorders. It allows the differentiation of focal from diffuse, cystic from solid and vascular abnormalities.

Conclusions. Clinical and ultrasonographic investigations in different hepatobiliary disorders in dogs, performed on a number of 50 cases, belonging to different breeds and ages, have captured a wide range of pathological processes, mainly functional, inflammatory, lithiasic and neoplastic. The clinical expression of the patients was rather polymorphic and inconsistently expressed clinically, in relation to the type and degree of morphological involvement, the changes being appreciated sonographically. Abdominal ultrasonographic examination confirmed the presumptive diagnosis on the basis of anamnestic data, clinical and paraclinical examinations or refuted the suspicion on the

basis of hepatobiliary functional morphological features. Statistical analysis of the ultrasonographic method demonstrated that it is an easy and necessary method in the detection of hepatobiliary disorders with high sensitivity and specificity. The high prevalence of sludge without biliary involvement on the one hand and low prevalence of cholelithiasis on the other hand in dogs compared to humans suggests that sludge in dogs causes stone formation; its presence is not associated with cholecystic parietal changes and is considered incidental. Ultrasonographic exploration is the first method of imaging investigation of the abdomen in general and of the hepatobiliary system in particular, with high efficiency, safety, repeatability and non-invasiveness.

Conflict of Interest. The authors declare that there is no conflict of interest.

References

- Barone R., 1997 [Comparative anatomy of domestic mammals - Volume 3 - Splachnology 1: Digestive apparatus and respiratory apparatus]. 3rd Edition. Vigot-Maloine, 853 p. [In French].
- Barrera R., 1994 Ultrasound parameters of the hepatic vascular pattern in the dog. *Veterinaria Technica* 4(6):87-89.
- Brömel C., Barthel P. Y., Léveillé R., Scrivani P. V., 1998 Prevalence of gallbladder sludge in dogs as assessed by ultrasonography veterinary. *Radiology & Ultrasound* 39(3):206-210.
- Codreanu M. D., 2000a [Ultrasound examination-complementary method of diagnosis in hepato-biliary diseases in dogs]. A X-a Sesiune Științifică, Facultatea de Medicină Veterinară Spiru Haret, Bucharest. [In Romanian].
- Codreanu M. D., 2000b [Ultrasound diagnosis in some hepatobiliary diseases in dogs]. Al VII-lea Congres Național de Medicină Veterinară, Băile Felix, Romania. [In Romanian].
- Codreanu M. D., 2000c [Ultrasound diagnosis of internal diseases in animals]. Coral Sanivet, Bucharest, pp. 37-101. [In Romanian].
- Codreanu M. D., 2010 [Clinical and sonographic aspects in cholecystopathies in dogs]. *Practica Veterinară Romana* 1(1):20-24. [In Romanian].
- Codreanu M. D., Mihai D., 2000 [Ultrasound aspects recorded in the case of abdominal tumors in dogs]. Al VIII-lea Congres Național de Medicină Veterinară, Băile Felix, Romania. [In Romanian].
- Constantinescu G. M., 2005 [Practical guide to dog and cat anatomy]. Med`Com Editions. [In French].
- D'Anjou M. A., 2008 Liver. In: Atlas of small animal ultrasonography. Penninck D., d'Anjou M. A. (eds), Blackwell, pp. 217-262.
- Gaschen L., 2009 Update on hepatobiliary imaging. *The Veterinary Clinics of North America. Small Animal Practice* 39(3):439-467.
- Ghergariu S., Kadar L., Pop A., Spînu M., 2000 [Veterinary clinical laboratory manual]. Editura ALL, Bucharest. [In Romanian].
- Goggin J. M., Biller D. S., Rost C. M., De Bey B. M., Ludlow C. M., 1997 Ultrasonographic identification of *Drofilaria immitis* in the aorta and the liver of a dog. *Journal of the American Veterinary Medical Association* 210(11):1635-1637.
- Haroutunian G., 1995 [Echography of dog and cat]. Vigot, Paris. [In French].
- Kumar V., Kumar A., Varshney A. C., Tyagi S. P., Kanwar M. S., Sharma S. K., 2012 Diagnostic imaging of canine hepatobiliary affections: A review. *Veterinary Medicine International* 2012:672107.
- Larson M. M., 2007 The liver and spleen. In: Textbook of veterinary diagnostic radiology. 5th Edition. Thrall D. E. (ed), Saunders Elsevier, pp. 667-693.
- Larson M., 2016 Ultrasound imaging of the hepatobiliary system and pancreas. *Veterinary Clinics of North America. Small Animal Practice* 46(3):453-480.
- Larson M., Mattoon J., Lawrence Y., Sellon R., 2021 Chapter 9: Liver. In: Small animal diagnostic ultrasound. Elsevier, pp. 355-421.

- Lessa A., Paredes B. A., Dias J., Carvalho A. B., Quintanilha L. F., Takiya C. M., Tura B. R., Rezende G. F. M., Campos de Carvalho A. C., Resende C. M. C., Goldenberg R. C. S., 2010 Ultrasound imaging in an experimental model of fatty liver disease and cirrhosis in rats. *BMC Veterinary Research* 6:6.
- Mannion P., 2006 *Diagnostic ultrasound in small animal practice*. Blackwell Science, pp. 50-69.
- Meyer H. P., 2000 Hepatic encephalopathy: An overview. *Proceeding of the Hill's European Symposium on Canine and Feline Liver Disease*, Amsterdam, pp. 24-28.
- Meyer H. P., Twedt D. C., Roudebush P., Dill-Mackey E., 2010 Hepatobiliary disease. In: *Small animal clinical nutrition*. Mark Morris Institute, pp. 1156-1167.
- Mihai D., Andronie V., 2000 [Animal internal medicine]. Geea, Bucharest, pp. 45-90. [In Romanian].
- Mwanza T., 1998 Evaluation of experimentally induced canine liver disease and portal hemodynamics using ultrasonography as a noninvasive diagnostic method. *Japanese Journal of Veterinary Research* 46(2-3):111-112.
- Nyland T. G., Mattoon J. S., 1995 Ultrasonography of the general abdomen. In: *Veterinary diagnostic ultrasound*. Nyland T. G., Mattoon J. S. (eds), W.B. Saunders Company.
- Nyland T. G., Mattoon J. S., Wisner E. R., Herrgesell E. J., 2002 Ultrasonography of the liver. In: *Small animal diagnostic ultrasound*. 2nd Edition. Nyland T. G., Mattoon J. S. (eds), WB Saunders, pp. 93-127.
- Nyman H. T., Kristensen A. T., Flagstad A., McEvoy F. J., 2004 A review of the sonographic assessment of tumor metastases in liver and superficial lymph nodes. *Veterinary Radiology & Ultrasound* 45:438-449.
- Partington B. P., Biller D. S., 1995 Hepatic imaging with radiology and ultrasound. *Veterinary Clinics of North America. Small Animal Practice* 25(2):305-335.
- Penninck D. G., Finn-Bodner S. T., 1998 Updates in interventional ultrasonography. *Veterinary Clinics of North America. Small Animal Practice* 28(4):1017-1040.
- Pop T., 1998 [Diagnostic and interventional clinical ultrasound]. Ed. Medicală, Bucharest, pp. 120-147. [In Romanian].
- Rothuizen J., 2006 Introduction—background, aims and methods. In: *Standards for clinical and histological diagnosis of canine and feline liver diseases—WSAVA Liver Standardization Group*. Rothuizen J., Bunch S. E., Charles J. A., et al. (eds), Saunders Elsevier, pp. 5-14.
- Rothuizen J., Meyer H. P., 2000 History, physical examination and signs of liver disease. In: *Text book of veterinary internal medicine: Diseases of the dog and cat*. 5th Edition. Ettinger S. J., Feldman E. C. (eds), WB Saunders, pp. 1272-1277.
- Smith S. A., Biller D. S., Kraft S. L., Goggin J. M., Hoskinson J. J., 1998 Diagnostic imaging of biliary obstruction. *Compendium on Continuing Education for the Practicing Veterinarian* 20(11):1225-1234.
- Syakalina M., 1998 Comparison of attenuation and liver-kidney contrast of liver ultrasonographs with histology and biochemistry in dogs with experimentally induced steroid hepatopathy. *Veterinary Quarterly* 20(1):18-22.
- Weill F. S., 1991 [Precise digestive and renal ultrasound. First part: Digestive ultrasound]. Vigot, Paris. [In French].

Received: 10 July 2024. Accepted: 25 July 2024. Published online: 05 August 2024.

Authors:

Daniela Mihaela Neagu, Department of Clinical Sciences, Faculty of Veterinary Medicine, University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, 400372 Cluj-Napoca, Romania, e-mail: daniela.neagu@usamvcluj.ro

Robert Purdoiu, Department of Clinical Sciences, Faculty of Veterinary Medicine, University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, 400372 Cluj-Napoca, Romania, e-mail: robert.purdoiu@usamvcluj.ro

Cristian Popovici, Department of Clinical Sciences, Faculty of Veterinary Medicine, University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, 400372 Cluj-Napoca, Romania, e-mail: cristian.popovici@usamvcluj.ro

Razvan Codea, Department of Clinical Sciences, Faculty of Veterinary Medicine, University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, 400372 Cluj-Napoca, Romania, e-mail: razvan.codea@usamvcluj.ro

Alexandra Biris, Department of Clinical Sciences, Faculty of Veterinary Medicine, University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, 400372 Cluj-Napoca, Romania, e-mail: Alexandra.biris@usamvcluj.ro

This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution and reproduction in any medium, provided the original author and source are credited.

How to cite this article:

Neagu D. M., Purdoiu R., Popovici C., Codea R., Biris A., 2024 Ultrasonographic examination in liver and gallbladder pathology in dogs. Clinico-ecographic correlations. ABAH Bioflux 16(1):14-25.