Update on Hepatobiliary Imaging

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Ultrasound is the most common modality used to screen animals with suspected liver disease, including vascular anomalies. Contrast-enhanced harmonic ultrasound (CEHU) is a noninvasive and highly accurate method of differentiating benign from malignant hepatic nodules in dogs. Ultrasound-guided tissue sampling has also become a mainstay of hepatic diagnostics. The use of nuclear medicine in the diagnosis of hepatic diseases in dogs and cats has become well established, and alternative imaging modalities, such as CT and MRI, are being validated for their diagnostic roles. These advanced technologies are more widely available than ever before through academic institutions and the rapid growth of specialty clinics offering these diagnostic services.

Radiography is widely available and recommended in dogs and cats suspected of having hepatic disease, but it is an insensitive method.1 Radiographic contrast studies, such as intravenous cholangiocystography and percutaneous transhepatic cholangiography, for the diagnosis of biliary obstruction are described1,2 but have not come into common use and have mostly been replaced by ultrasonography. Ultrasound is complementary to the abdominal radiograph and provides a more detailed examination of the inner structure of the liver and surrounding organs. This update on hepatobiliary imaging does not include a description of survey and contrast radiographic examinations of the liver or the basic principles of hepatic sonography. The reader is referred to the many complete and excellent sources available on these topics.1–5

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.6 An update on the sonographic, CT and MR Imaging, and nuclear medicine imaging examinations necessary to make diagnoses in each category is provided in this article.
PARENCHYMAL DISEASE

Nonneoplastic canine and feline parenchymal diseases include steroid-induced hepatopathy, hepatic lipidosis, amyloidosis, acute and chronic hepatitis, cirrhosis, necrosis (eg, toxic insult, ischemia, immune mediated), abscessation, granulomas, and metabolic storage diseases.

Sonography

Hepatic parenchymal abnormalities are characterized as being diffuse, focal, or multifocal. Ultrasound is sensitive at detecting focal and multifocal disease but can be poor at detecting diffuse changes. Therefore, a definitive diagnosis should be based on a minimum combination of ultrasound features, blood test results, and tissue sampling results.

An enlarged liver is a subjective finding and may be generalized or focal. Causes include steroid hepatopathy, lipidosis, amyloidosis, diabetes, hepatitis, congestion, neoplasia (eg, lymphoma, histiocytic sarcoma, mast cell tumor), and hepatocellular carcinoma (HCC). In cats, large amounts of falciform fat may be present and can be mistaken for an enlarged liver sonographically (Fig. 1). The ventral capsule of the liver can be observed in almost all instances and allows the liver to be visually separated from the falciform fat next to it. In obese cats, the liver may even become hyperechoic to the falciform fat.

Focal or lobar enlargement can be caused by primary or metastatic disease or by cysts, hematomas, abscesses, granulomas, lobar torsion, or thrombosis. In dogs that have portosystemic shunts (PSSs), cirrhosis, or fibrosis, the liver is typically small and the stomach appears closer to the diaphragm than usual.

Diffuse parenchymal disease

Diffuse parenchymal disease generally affects all lobes and may appear normal, isoechoic, or hyperechoic. A large group of hepatic diseases exists that can lead to infiltration of the liver without disruption of the architecture, making disease difficult to detect. These diseases include cholangiohepatitis, diffuse prenodular (early) metastatic carcinoma or sarcoma, round-cell neoplasia (eg, lymphoma, mast cell disease, histiocytic sarcoma), patchy or diffuse fatty infiltration, vacuolar hepatopathy, storage diseases (eg, amyloidosis, copper), toxic hepatopathy, and early degenerative changes associated with micronodular hyperplasia and fibrosis (Fig. 2).

Table 1

Fig. 1. Ultrasound image of the liver of a cat shows large amounts of falciform fat that can be mistaken for being part of the liver and misdiagnosed as hepatomegaly. Note the hyperechoic interface (arrows) marking the separation between the fat and the liver. The liver is of normal size.
summarizes the ultrasound findings in different causes of diffuse liver disease. Mast cell disease affecting the liver may appear sonographically normal or diffusely hyperechoic. The overall accuracy of ultrasound as the sole criterion for discriminating among the categories of diffuse liver disease is less than 40% in dogs and less than 60% in cats. Adding biochemical or hematologic information in the assessment of

<table>
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<tr>
<th>Ultrasound Findings</th>
<th>Normal</th>
<th>Enlarged</th>
<th>Hyperechoic</th>
<th>Hypoechoic</th>
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the ultrasound findings in diffuse liver disease does not seem to improve accuracy. Therefore, it is generally not possible to make a final diagnosis based on the combination of sonographic findings and biochemical and hematologic data in dogs and cats that have diffuse liver disease. Tissue sampling, preferably for histologic examination, is recommended for a definitive diagnosis in most instances, even if the liver appears sonographically normal.

Vacuolar changes in the liver associated with lipidosis and steroid hepatopathy usually cause hepatomegaly in conjunction with diffuse hyperechogenicity and rounded borders. Another feature that may occur is hyperattenuation of the ultrasound beam. This is seen as a gradual decrease in echogenicity in the far field of the image, which can be so severe that the liver in that region is not visible (Fig. 3).

Inflammatory disease can be associated with diffuse hypoechogenicity. If acute hepatitis or cholangiohepatitis is present, the liver may appear to have high contrast—a hypoechoic parenchyma with pronounced hyperechogenicity of the portal vein walls or periportal tissue (Fig. 4). Chronic inflammation of the liver usually results in hyperechoic or mixed echogenicities. When fibrosis or cirrhosis is present, the liver may be smaller and hyperechoic. If nodular hyperplasia develops, such as with vacuolar hepatopathy, the liver may appear more heterogeneous and nodular, such as in neoplastic disease. Other differential diagnoses for this pattern include amyloidosis in cats and dogs and hepatocutaneous syndrome in dogs (Fig. 5).

Quantitative determination of hepatic echogenicity has been found to be feasible through histogram analysis and may be useful for early detection of diffuse parenchymal disease and for serially evaluating disease progression. In this technique, numeric values from echogenicity data are derived that are related to the mechanical properties of the tissue being evaluated. These numeric values enhance the accuracy for differentiating between tissues with a normal and abnormal ultrasonographic appearance. The analysis is generally done with the on-board computer software of the ultrasound unit or with separate image analysis software. In people, quantitative ultrasonographic methods help to diagnose diffuse abnormalities of the liver and kidney. In cats, quantitative ultrasound video signal analysis has been used to correlate an increase in obesity-related hepatic lipid content to an increase in attenuation and backscatter of the ultrasound signal. This method of image analysis may prove useful for the evaluation of diffuse hepatic parenchymal disease.

![Fig. 3. Ultrasound image of the liver in a cat. The liver is markedly hyperechoic in the near field, with decreasing echogenicity in the far field. This is attributable to the beam attenuation commonly seen in cats that have hepatic lipidosis.](image)
Focal parenchymal disease

Focal or multifocal changes in the liver parenchyma are easier to identify sonographically than diffuse changes. Hypoechoic, hyperechoic, and anechoic lesions are easy to identify because they contrast better with the surrounding parenchyma. For this reason, cystic lesions are the easiest to detect, even when extremely small.

Anechoic cavitary structures in the liver can be attributable to necrosis, neoplasms, or cysts. Cystic structures generally have sharply defined borders, can be round or irregular in shape, and may even contain hyperechoic septa. Acoustic enhancement is typically identified in the far field, distal to the cyst. Causes include congenital cysts, posttraumatic cavitations, biliary pseudocysts, or parasitism. Unfortunately, biliary cystadenomas and cystadenocarcinomas may appear similar. Hepatic abscessation occurs rarely in small animals and may appear similar to a primary tumor, granuloma, or hematoma because of its highly variable sonographic features. It is usually the result of bacterial infections that reach the liver by means of the portal vein or umbilical vein, ascending by means of the bile system or by direct penetration of the liver. It may also occur secondarily to necrosis of hepatic neoplasms and can look similar to parasitic cystic structures. Sonographically, hepatic abscesses may be round to irregular in shape with a hypoechoic central region.

Fig. 4. (A) Ultrasound image in a dog with cholangiohepatitis diagnosed on liver tissue core biopsy samples. The liver is diffusely hypoechoic, and the portal veins seem to stand out more prominently than normal hyperechoic walls. (B) Magnification of the liver shows that the walls appear to be thickened. There are no signs of anechoic tubular structures within or adjacent to the hyperechoic walls; therefore, the intrahepatic bile ducts are likely not dilated in the periphery.

Fig. 5. Ultrasound image of the liver in a dog with chronic skin lesions. The liver has a honeycomb-like echotexture, which is commonly seen in hepatocutaneous syndrome.
or of mixed echogenicity. Reverberation artifacts may be detected because of gas accumulations within the necrotic tissue. Anechoic centers with distal acoustic enhancement also occur. Additional findings, such as regional lymphadenopathy, may be present in hepatic neoplasia and abscessation. Focal peritonitis may be seen with abscessation and may include free peritoneal fluid and focal hyperechoic mesentery.

Granulomatous causes of focal hepatic disease in dogs and cats include mycobacterial infections (Mycobacterium tuberculosis, Blastomyces dermatiditis, Cryptococcus neoformans, Histoplasma capsulatum, and Coccidioides immitis), migrating larvae, and schistosomiasis. Foreign material is another cause of granuloma formation in the liver. Sonographically, granulomas in dogs and cats may appear as multifocal hyperechoic and well-marginated parenchymal lesions.3

Liver lobe torsion occurs in the dog rarely but should be included in the differential diagnosis for acute abdomen or abdominal effusion. The torsion leads to congestion and necrosis of the affected lobe or lobes.18–22 Typically, the affected lobe appears hypoechoic, and color Doppler shows reduced or no blood flow within the lobe. Thromboembolism would have a similar appearance but is rare in the liver.

**CT**

There are few reports of the use of CT to assess canine or feline hepatic parenchymal diseases. This is likely attributable to the adequacy of ultrasonography and tissue sampling as diagnostic tests, to the fact that anesthesia is not required, and to CT’s higher cost and lack of widespread accessibility. Hepatic volumetry has recently been described in dogs using CT.23 Liver volume estimations may be helpful in dogs for assessing changes in liver size after shunt attenuation. In human beings, liver volume is used as a prognostic indicator in patients who have liver failure. More work is required in this field to determine its usefulness in veterinary medicine.

**NEOPLASTIC DISEASE**

Neoplastic disorders in dogs and cats are categorized as hepatocellular (nodular hyperplasia, adenoma, and HCC), cholangiocellular (biliary adenoma, biliary carcinoma, and mixed), hepatic carcinoids, primary vascular and mesenchymal (hemangiosarcoma and myelolipoma), hematopoietic (lymphoma and histiocytic sarcoma), and metastatic.24
Sonography

Neoplastic disease of the liver may manifest as diffuse, multifocal, or focal disease sonographically. Diffuse disease is usually attributable to round-cell neoplasia. Lymphoma, histiocytic sarcoma, and mast cell tumor are the most common neoplasms that may lead to diffuse changes and remain sonographically undetectable.\textsuperscript{9,25} Diffuse hypoechogenicity or hyperechogenicity and mixed patterns may also occur.\textsuperscript{26} Carcinomas tend to be diffusely spread throughout the liver and often lead to a mixed pattern.

Benign hyperplastic nodules are an extremely common finding in dogs, especially in older animals.\textsuperscript{5} They are generally not more than 1 cm in diameter. Malignant nodules have a highly varied appearance and size. They may appear as hypoechoic or hyperechoic nodules, target lesions, or heterogeneous ill-defined nodules. Table 2 summarizes the causes of nodules and compares their differences in echogenicity. Hypoechoic nodules can be attributable to nodular hyperplasia, metastases, lymphoma, histiocytic sarcoma, primary neoplasia, necrosis, hematomas, or abscesses. For this reason, tissue sampling is critical for a definitive diagnosis and the presence of hepatic nodules is not synonymous with malignancy. Hypoechoic lesions with a hyperechoic center are referred to as target lesions and have been associated with metastatic and benign processes (Fig. 7).\textsuperscript{27,28} Hemangiosarcoma, HCC, carcinoma, insulinomas, bile duct carcinoma, lymphoma, and histiocytic sarcoma are malignant diseases that may cause target lesions. Benign causes of target lesions include nodular hyperplasia, pyogranulomatous hepatitis, chronic active hepatitis, and cirrhosis.\textsuperscript{27} Hepatic target lesions have a positive predictive value for malignancy of 74%,\textsuperscript{27} which emphasizes the fact that histologic type cannot be predicted by the presence of target lesions.

Biliary cystadenomas and cystadenocarcinomas may appear as loculated cavitary lesions. Hepatic and biliary cysts are benign diseases that may resemble their malignant counterparts.\textsuperscript{15}

### Contrast-enhanced Harmonic Ultrasound

CEHU is a new diagnostic option that allows assessment of the perfusion patterns of organs in a noninvasive manner. It requires the use of contrast ultrasound probes and on-board software designed to receive and analyze the contrast signals. The underlying principle behind these contrast agents is based on the detection of nonlinearly

<table>
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<th>Table 2</th>
<th>Nodular hepatic infiltration: causes and sonographic appearance</th>
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<tr>
<td><strong>Disease</strong></td>
<td><strong>Nodule Echogenicity</strong></td>
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<tr>
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<td>Granuloma —</td>
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<td>Neoplasia Yes</td>
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<td>Myelolipoma Yes</td>
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scattered signals, which are harmonic frequencies generated when the ultrasound beam interacts with the contrast media. The newest agents range from 2 to 6 μm in diameter and contain air or other gases that enhance the ultrasound signal. Second-generation phospholipid shell microbubbles containing perflutren gas, such as in Definity (Lantheus Medical Imaging, Billerica, Massachusetts), elicit harmonic frequencies at much lower acoustic powers than are necessary to generate tissue harmonics. Thus, the harmonic signal of the microbubbles within the capillary bed and vessels can be separated from the tissue signals. This produces an angiogram and parenchymal perfusion effect. The contrast agents are injected into a peripheral vein in small volumes, and because of their size, they act as blood pool agents ideal for assessing organ perfusion.

Studies describing the characteristics of CEHU of the liver, spleen, and kidney in normal dogs are available. Detection and characterization of liver nodules in dogs with CEHU have been the most commonly reported uses of the technique, however. One study investigated hepatic perfusion dynamics of CEHU in normal dogs and in dogs with naturally occurring HCC and metastatic hepatic hemangiosarcoma. Another study describes the ability of CEHU for detecting hepatic metastasis not identified on gray-scale ultrasound imaging in dogs that have hemangiosarcoma.

Contrast-enhanced ultrasound in the liver is divided into two phases: early and late (or sinusoidal phase). The early phase is equivalent to the blood pool phase and is composed of an arterial phase (wash-in) and a portal venous phase (wash-out). In the early phase, the presence, number, distribution, and morphology of lesional vessels can be evaluated. The late phase corresponds to intracellular contrast media uptake in the liver. The main difference between benign and malignant lesions is that during the portal and late phases, all benign lesions, except cysts and thrombosed hemangiomas, exhibit isoenhancement or slight hyperenhancement as compared with surrounding liver tissue (Fig. 8). Malignant liver lesions exhibit hypo-enhancement or do not perfuse at all, because the perfusion of malignant tumors is provided exclusively by arterial vessels and there is no portal venous supply. The sensitivity, specificity, positive predictive value, negative predictive value, and accuracy of CEHU for diagnosing benign versus malignant liver nodules have been shown to be 100%, 94.1%, 93.8%, 100%, and 96.9%, respectively. No complications or morbidity has been reported in veterinary medicine using this agent. CEHU seems
to be accurate at discriminating between naturally occurring benign and malignant nodules in the liver of dogs, but its use is currently limited to academic institutions.

**CT and MRI**

In humans, differentiation between benign and malignant hepatic lesions is often made with CT or MRI, and the diagnosis is determined principally from vascular information obtained as a result of contrast enhancement in the arterial and portal venous phases. One study in dogs with splenic hemangiosarcoma assessed the use of CT compared with sonography for diagnosing hepatic metastases and found no significant difference between the two modalities. In a pilot study examining the specificity of MRI before and after gadolinium contrast administration for benign versus malignant hepatic lesions in dogs, MRI accurately differentiated between benign versus malignant disease in 33 of 35 lesions for a sensitivity and specificity of 100% and 90%, respectively. Both modalities show potential for diagnosing hepatic neoplasia, but more work in the field is required to validate them in dogs and cats.
BILIARY DISEASE

Biliary disease is divided into four main categories: biliary cystic disease, cholestasis, cholangitis, and diseases of the gallbladder (e.g., mucoceles, cholecystitis)\textsuperscript{15,44}

Sonography

Dogs and cats that have icterus may have cholestasis because of intrahepatic or extrahepatic disease. Extrahepatic cholestasis can be caused by intraluminal obstruction (e.g., choleliths, mucinous cystic hyperplasia, sludge) or luminal constriction (e.g., neoplasia, inflammation) of the extrahepatic biliary tree or large intrahepatic ducts\textsuperscript{1,44}. Intramural biliary obstruction may occur secondary to biliary adenocarcinomas\textsuperscript{1}. Enlarged perihilar lymph nodes may also lead to ductal obstruction. The duodenum should be examined for obstruction at the major duodenal papilla, wherein inflammatory and malignant diseases may be another source of obstruction (Fig. 9). Dilation of the ducts depends on the degree and duration of the obstruction. The common bile duct can be up to 3 mm in diameter in normal dogs and up to 4 mm in cats\textsuperscript{45,46}. Longer standing obstructions (3–7 days) of the common bile duct may lead to dilation of the extrahepatic and intrahepatic ducts\textsuperscript{1,47}. These are evident as anechoic tubular structures at the porta hepatis (extrahepatic ducts) or throughout the parenchyma (intrahepatic ducts). Color Doppler ultrasound should be used to assess any anechoic tubular structure in the liver to differentiate biliary from vascular structures. Relief of the obstruction does not lead to an immediate reduction in the diameter of the dilated biliary ducts. The gallbladder may remain a normal size or be enlarged with extrahepatic bile duct obstructions. The presence of a normal-sized gallbladder should not eliminate the possibility of an obstruction.

Neutrophilic cholangitis or cholangiohepatitis is more common in cats than in dogs\textsuperscript{44}. It is usually attributable to an ascending infection from the intestinal tract. Lymphocytic cholangiohepatitis is also common in cats. The two diseases cannot be distinguished sonographically in cats and require different treatment protocols. Therefore, it is important to perform tissue sampling to differentiate between the two diseases. Sonographic features of cholangiohepatitis in cats include a diffusely

![Fig. 9. (A) Dilated common bile duct with a small hyperechoic filing defect (arrow) close to the entrance of the duodenal papilla. Mild shadowing of this lesion could occasionally be seen during the examination (not shown). Note that the common bile duct is dilated proximal to the obstruction. A cholelith was confirmed during surgery (B) Dilated common bile duct proximal to the duodenal papilla in an icteric cat. The papilla (short wide arrow) is enlarged and moderately echogenic. The lumen of the bile duct (thin arrows) appears narrowed at the thickened and enlarged papilla. Thick arrows show the duodenal wall adjacent to the dilated bile duct. A choledochoduodenostomy was performed, and the papilla was resected. Histologic examination diagnosed a chronic inflammatory polypoid infiltration.](image)
hypoechoic liver parenchyma with prominent-appearing portal vascular structures.\textsuperscript{48} Included may be thickening of the gallbladder wall and bile duct wall and increased amounts of sludge in the gallbladder. Intra- and extrahepatic dilation of the biliary tree, in addition to pancreatitis, may also be present. Similar findings to neutrophilic and lymphocytic cholangiohepatitis may occur in liver fluke infestation (family Opisthorchiidae in endemic regions). Because these diseases appear similarly and even may appear normal sonographically, tissue sampling is critical for a diagnosis.\textsuperscript{44} Generalized gallbladder wall thickening can occur with cholecystitis, cholangiohepatitis, hepatitis, free peritoneal fluid, and hypoproteinemia.\textsuperscript{3} The wall may appear to have a “double” layer in these instances. Neoplastic disease of the gallbladder wall causing focal thickening is less common than benign cystic hyperplasia of the mucous glands, which appear as broad-based or pedunculated hyperechoic structures. Choleliths can occur, more commonly in dogs, and appear as hyperechoic structures of variable size, number, and shape that produce acoustic shadowing. They are not always associated with clinical signs and can be incidental findings, especially in older dogs. Mineralized and nonmineralized material may also be found in the bile ducts. Sludge balls are accumulations of thick or inspissated bile that can be found in the gallbladder lumen or within the bile duct, wherein they can potentially cause obstruction (Fig. 10). They appear as rounded or irregularly shaped structures of moderate echogenicity and can be found to move freely within the gallbladder.

Gallbladder mucoceles occur in dogs and are an important cause of icterus and obstructive disease. They are caused by cystic mucinous hyperplasia leading to increased mucin production that distends the gallbladder and can eventually cause wall necrosis and rupture. Sonographically, they have a varied appearance. The classic finding is that of a “kiwi fruit” pattern of hyperechoic striations radiating from a central point (Fig. 11). Variations include irregular or striated nongravitationally dependent content or content with a stellate pattern.\textsuperscript{49} They can also lead to

Fig. 10. Ultrasound image in an icteric dog. The common bile duct is dilated. There is a moderately echogenic structure filling its lumen. The structure is homogeneous and does not create acoustic shadowing. During surgery, it could be flushed through the duodenal papilla with saline. The final diagnosis was cholecystitis with a sludge ball causing obstruction of the common bile duct. Thin white arrows show the anechoic lumen of the dilated common bile duct cranial and caudal to the intraluminal structure.
extrahepatic biliary obstruction. Distention of either or both the intrahepatic or extrahepatic bile ducts may be seen. Sonographic signs of rupture include loss of the gallbladder wall continuity, hyperechoic surrounding mesentery, and free peritoneal fluid. The sensitivity of ultrasonography for diagnosing gallbladder rupture is reported as 85%. The therapeutic dilemma as to whether to perform cholecystectomy arises when a gallbladder mucocele is identified sonographically but without signs of rupture. It has been shown that they can transform into an acute clinical condition. A breed predilection has been suggested in cocker spaniels, Shetland sheepdogs and miniature schnauzers. In one study, a significant predisposition for gallbladder mucoceles in Shetland sheepdogs was shown compared with the general hospital population.

Cholecystitis is more frequent in cats than in dogs and is generally associated with bacterial infections. Because bile duct dilation and gallbladder wall changes may not occur in cats that have neutrophilic cholecystitis, bile aspirations for cytologic and bacteriologic examination in cats may be necessary to confirm a suspected diagnosis and administer appropriate antimicrobials. Emphysematous cholecystitis may result from Escherichia coli and Clostridium perfringens infections, which are gas-forming bacteria. It has also been associated with diabetes mellitus. Gas within the biliary tract, such as with mineralization, can be identified radiographically and sonographically. Ultrasonographically, it appears as irregular or pinpoint-sized hyperechoic structures that produce reverberation artifacts. The presence of gas in the gallbladder or liver

Fig. 11. (A) Ultrasound image of the gallbladder in an icteric dog. The gallbladder contents are hyperechoic and organized into a striated pattern resembling the cut surface of a kiwi fruit. The final diagnosis was gallbladder mucocele. (B) Mucocele in a dog with inspissated material filling the cystic duct and common bile duct and causing obstruction. (C) Mucocele with evidence of free gas. There are hyperechoic foci with dirty shadowing (thin arrows) adjacent to the gallbladder. Gallbladder rupture was diagnosed during surgery.
Parenchyma should alert the sonographer to the possibility of cholecystitis, cholangitis, choledochitis, or abscess formation.54

**Endoscopic Retrograde Cholangiopancreatography**

Endoscopic retrograde cholangiopancreatography (ERCP) is an established method in people for the diagnosis of biliary obstruction and chronic pancreatitis. It uses a combination of endoscopy and fluoroscopy to image the biliary and pancreatic ducts. Two studies, the first in normal dogs and the second in dogs that had gastrointestinal disease, have been performed using this technique.55,56 ERCP is technically possible in dogs, and success is influenced by the experience of the investigator. It cannot be performed in dogs with a body weight of 10 kg or less, however. In one study, 20 of 30 dogs that had gastrointestinal disease could be successfully examined using this technique. Abnormal findings compared with a group of healthy dogs included an enlarged common bile duct (n = 2), intraductal filling defects (n = 2), deviated course of the common bile duct (n = 1), and major papilla stenosis (n = 1). In 1 dog with major papilla stenosis and intraductal filling defects, endoscopic-guided sphincterotomy was performed. Endoscopic retrograde pancreatography diagnosed an abnormal course of the accessory pancreatic duct in 2 dogs. Although the use of this technique requires further investigation for validation, the preliminary findings show that changes in the biliary tree of dogs may be going undiagnosed in certain populations of dogs that have gastrointestinal disease.51,56 The need for special endoscopic equipment, fluoroscopy, and experience likely limits its use to specialty and academic centers.

**Nuclear Medical Imaging**

Hepatobiliary scintigraphy can be used to quantify liver function, evaluate hepatic morphology, assess biliary tract patency, and diagnosis cholecystitis.57,58 Cholestasis can also be diagnosed. Hepatic extraction fraction (HEF) is a quantitative measure of hepatocyte function attributable to its ability to extract a radiopharmaceutic agent from the blood by means of a peripheral intravenous injection of 99mTc-mebrofenin.59 It also assesses the ability of the hepatocyte to excrete the same radiopharmaceutic agent into the biliary tree. Indications for hepatobiliary scintigraphy include quantification of hepatic function and morphology, biliary tract patency, extrahepatic biliary obstruction, biliary kinetics, gallbladder ejection fraction, and presence of cholecystitis and intra- and extrahepatic cholestasis.50,57,60

In healthy animals, the blood pool of the radiopharmaceutic agent clears rapidly, with peak liver radioactivity at 6 to 8 minutes after injection (Fig. 12A).61 It is excreted into the biliary tree with a half-life of 19 minutes. Radioactivity should be observed in the gallbladder and small intestines by 1 hour after injection. For determining patency of the bile duct, hepatobiliary scintigraphy can show abnormalities before they are evident sonographically (Fig. 12B).61 Partial extrahepatic biliary obstruction can still have a normal HEF and prolonged radiopharmaceutic agent clearance from the liver.61 Complete obstruction is usually associated with a subnormal HEF, prolonged clearance, inability to visualize the biliary tree, and absence of radioactivity in the intestines (Fig. 12C).52,62 Because most studies have been performed in dogs at this time, the role of hepatobiliary scintigraphy in cats is not known.63

**MRI**

Magnetic resonance cholangiopancreatography (MRCP) is a newer technique in human beings for the diagnosis of bile duct obstructions. ERCP is still the “gold standard” for exploration of the biliopancreatic region in people but has a certain
complication rate associated with it.\textsuperscript{64} MRCP is a noninvasive alternative to ERCP and is currently used to diagnose many hepatobiliary and pancreatic diseases in human patients.\textsuperscript{65} Although studies in dogs and cats have not been described, this may represent a future imaging modality to diagnose hepatobiliary disease.

**VASCULAR DISEASE**

**Sonography**

Venous congestion of the liver occurs secondarily to increased resistance to flow toward the right atrium by way of the vena cava. This may be attributable to a right atrial mass causing obstruction, pericardial effusion, or invasion of the vena cava by a tumor. The hepatic vein is grossly dilated, as is the vena cava, and the liver often becomes enlarged and diffusely hypoechoic. Spectral Doppler analysis of these structures shows high-velocity retrograde flow indicating high resistance to flow toward the right heart (\textbf{Fig. 13}). Ascites is usually also present.
In veterinary medicine, operative mesenteric portography, splenoportography, and cranial mesenteric angiography are the currently well-established gold standards for depicting the anatomic details of PSSs. The reader is referred to several descriptions of the radiographic techniques. Congenital PSSs are abnormal vascular communications that allow blood from the intestine to bypass the liver and are classified as intrahepatic or extrahepatic. Diagnostic tests include serum bile acid concentrations, the ammonia tolerance test, portography, ultrasonography, and scintigraphy. Microhepatica is a common sonographic finding in dogs that have extrahepatic PSSs. In addition, bilateral renomegaly, nephrocalcinosis, nephroliths, and cystoliths attributable to urate crystals or stones may be identified. If portal hypertension is present, free fluid may be detected.

The sensitivity and specificity of sonography for the detection of extrahepatic PSSs have been reported to be 80.5% and 66.7%, respectfully. A greater sensitivity of 100% was seen for intrahepatic PSSs alone. In a second study using sonography for the diagnosis of congenital PSSs, results were improved by demonstrating a specificity of 98%, sensitivity of 95%, and accuracy of 94% in 38 dogs. Extrahepatic
shunting vessels generally originate from the portal, splenic, right or left gastric, or gastroepiploic vein in small-breed dogs. They are usually identified as tortuous-appearing vessels with hepatofugal flow. The vena cava, portal vein, and porta hepatis region should be scanned from the diaphragm to the level of the kidneys in search of an anomalous branching vessel entering the vena cava or traveling dorsally through the diaphragm toward the azygous vein adjacent to the aorta. Portocaval shunts

Fig. 12. (continued)

Fig. 13. (A) Enlarged liver with distended hepatic veins and vena cava (arrow). (B) Spectral Doppler image of a normal vena cava waveform. The waveform is triphasic, with low-velocity retrograde flow during atrial systole (arrows). (C) Spectral Doppler image with sample volume placement in the distended hepatic vein. There is high-velocity flow greater than and less than baseline, indicating high resistance to blood flow toward the vena cava, which also creates higher velocity retrograde flow (arrows). The final diagnosis was right heart failure.
terminate in the caudal vena cava, and their entrance is characterized by turbulent flow with color and spectral Doppler (Fig. 14). The size of the portal vein cranial to the shunt is generally reduced in diameter. A portal vein/aortic ratio of 0.65 or less is predictive for the presence of an extrahepatic shunt, and a value of 0.8 or greater excludes it. If the ratio is 0.80 or greater, other types of disease, such as microvascular dysplasia, intrahepatic shunt, and portal hypertension attributable to chronic liver disease with secondary shunting, could still be present.

CEHU has also been used to determine perfusion patterns in three dogs with congenital extrahepatic solitary PSSs. It was found that with coded harmonic angiographic ultrasound, the size and tortuosity of the hepatic arteries were subjectively increased. Peak perfusion times of dogs with PSSs were significantly shorter ($P = 0.01; 7.0 \pm 2.0$ seconds) than reported in normal dogs ($22.8 \pm 6.8$ seconds). Contrast-enhanced ultrasound may be a promising new method of detecting increased arterial blood flow that is an indicator of portosystemic shunting in dogs. Increased hepatic arterial blood flow alone does not confirm a diagnosis of PSS, however. Portal hypertension causes reduced portal blood flow to the liver and leads to secondary increased hepatic arterial blood flow in dogs. Prehepatic causes of chronic reduced portal flow and increased hepatic arterial blood flow include portal vein thrombosis and portal vein compression attributable to a regional primary mass or enlarged lymph node. Most of these disease processes would be easily distinguishable with a thorough sonographic examination.

Causes of portal hypertension include chronic liver disease, diffuse nodular regeneration, infiltrative neoplastic disease, congenital hypoplasia of the portal vein, arteriovenous fistula, and portal vein thrombus or extraluminal compression. Ascites is a common clinical feature and sonographic finding, and portal hypertension is suspected when flow is reduced, such as is detected with spectral Doppler. Mean velocities of 10 cm/s or less in the portal vein are highly suspicious for hypertension, but this is not always present. The midabdomen should be screened well for increased size and number of portal vessels, some of which may have a tortuous course. These may develop collateral circulation by way of the renal vein and lead to clinical signs of PSS.

Arteriovenous fistulas can be congenital or acquired and create connections between the portal vein and hepatic arteries. The ensuing high pressure overloads...
the venous side, and hypertension occurs. Acquired PSSs form much as with any other cause of portal hypertension, and clinical signs of shunting occur.

Thrombosis of the portal vein occurs with numerous diseases that are associated with the development of coagulopathies (Fig. 15). They are recognized sonographically as intraluminal structures of moderate to high echogenicity and the absence of color Doppler signals within the lumen. Thrombosis can be focal or can extend into all branches of the portal venous system and cause acquired shunting.

**CT and MRI**

Contrast-enhanced helical CT is rapidly becoming one of the more commonly used methods of diagnosing extrahepatic PSSs in dogs at academic institutions and specialty practices. It eliminates the need for invasive radiographic angiography procedures, because the contrast injections can be made by way of a peripheral vein. Other advantages include consistent and superb anatomic depiction of the origin of the anomalous vessel and its entrance into the systemic venous circulation compared with ultrasound, less operator dependency, and the potential for three-dimensional reconstructions. It also eliminates variability attributable to operator expertise, such as in sonography. Disadvantages include the need for anesthesia and possible motion artifacts requiring repeat scanning. Furthermore, access to CT scanners may be a limiting factor. Rapid scanning as afforded by helical single- or multislice scanners is critical for the procedure so that it can be done rapidly during a breath-hold procedure. It may be of great value in extra- and intrahepatic shunt detection, when multiple shunts may be present, and in unclear cases after other imaging procedures, such as ultrasonography, contrast radiographic studies, or nuclear medicine portography.

Standard protocols have been established and involve a single scan or dual-phase scanning. In both methods, a test bolus of non-ionic iodinated contrast medium (iodine, ~185 mg/kg of body weight) is made through a cephalic vein catheter to determine maximum opacification of the portal vein after injection of iodinated contrast medium. Serial axial images are made at T12 to T13 (approximate location of the porta hepatis) every second or at the shortest interval possible with a given unit’s capabilities from the onset of injection in non-helical mode. The time of maximum opacification is used to plan the helical CT study based on time-attenuation graphs. The second injection is performed

![Fig. 15. Portal vein thrombus in a dog with disseminated intravascular coagulation. The main portal vein just caudal to the liver is dilated and filled with heterogeneous material of mixed echogenicities. No flow could be detected around or within the thromboembolic material, and ascites was also present.](image-url)
with contrast medium (iodine, ~800 mg/kg), and acquisitions are made in helical mode after a breath hold and from the diaphragm to the midlumbar area. Collimation is generally set at a 3- to 5-mm slice thickness with an interval of approximately half of that.

In dual-phase computed tomographic angiography of the portal and hepatic vasculature, the test injection for timing is performed as for the single phase, but the patient is scanned twice during the second injection: first, from caudal to cranial to observe the hepatic arterial phase and then from cranial to caudal to observe the portal phase. Portal phase scanning is initiated shortly before the time of its peak enhancement based on the initial injection for determining maximal enhancement time. Median time delay for peak aortic enhancement has been shown to be 12.0 seconds after injection and 33.0 seconds after injection for the portal vein. There is an approximate 5-second delay between peak contrast attenuation in the aorta and portal vein. The portal phase of the scan is initiated with a median time of 28 seconds (range: 27.7–34.9 seconds) after injection in the cranial-to-caudal direction from the diaphragm to L5. This minimally invasive method allows complete evaluation of the hepatic arterial, venous, and portal vasculature with exquisite anatomic detail and has the potential to diagnose extrahepatic and intrahepatic shunts, arterioportal fistulas, and portal thromboembolic disease (Fig. 16).

A technique for CT splenic portography has recently been described. A 20- to 22-gauge 1.5-in needle is placed in the splenic parenchyma under CT guidance. A preloaded extension set is attached, and iodinated contrast media is administered at a concentration of iodine, 175 mg/mL. One milliliter is administered as a rapid bolus, followed by a steady manual injection of 2 mL over 5 seconds. The CT acquisition is started at the time of contrast medium injection, and images are acquired from the level of the fifth lumbar vertebra to the cranial aspect of the diaphragm. Because hand injections are used during acquisition, radiation protection procedures are important to follow and are a disadvantage of not using automatic injectors with remote activated devices. The degree of opacification of the splenic vein in all locations, and that of the main portal vein, is significantly higher in trans-splenic computed tomographic portography compared with computed tomographic angiography. Benefits include the simple technique, low dosage of contrast medium required compared with conventional computed tomographic angiography, and much better opacification of the portal system. Disadvantages include inconsistent depiction of the intrahepatic portal vasculature and parenchymal opacification attributable tostreamlining and presence of streak artifacts in addition to radiation protection. Streamlining has also been described as a cause for nonuniform distribution of radiopharmaceutic agents during per-rectal scintigraphy in dogs. These artifacts lead to preferential ventrolateral contrast medium distribution into the left divisional branch. In addition, preferential left andventral streamlining allows fewer arborizations to be detected from the right divisional branch compared with the left divisional branch. Despite these limitations, contrast medium should preferentially distribute into the shunting vessel because of hepatofugal blood flow; further studies require testing this hypothesis in dogs that have PSSs.

MRI has only rarely been reported for diagnosing PSSs in dogs. Magnetic resonance angiography (MRA) is a described method for assessment of the portal vein. MRA is a noninvasive technique that provides functional representation of blood vessels without the use of contrast. Two techniques, time-of-flight and phase contrast, can be used. At this time, MRA has not been well validated in veterinary medicine for diagnosis of PSSs and requires further investigation.
Nuclear Medical Imaging

Nuclear portal scintigraphy is a highly sensitive and minimally invasive screening method for diagnosing the presence or absence of a PSS. It does require certification for the use of radioisotopes and specialized equipment and software programs, however. Patients must also be held in isolation, generally overnight, until their radiation levels are low enough to return home or to have the shunt surgically repaired. Nuclear portal scintigraphy allows shunt fractions to be assessed before and after surgery to monitor the degree of closure of the shunt but does not allow exact anatomic descriptions to be made.

Per-rectal portal scintigraphy (PRPS) methods are performed by administering sodium $^{99m}$Tc-pertechnetate into the colon. In dogs, a dose of 5 to 20 mCi is used, whereas a dose of 5 to 10 mCi is administered in cats. The radionuclide is absorbed into the portal venous system in the distal colon, and it is then transported to the liver by way of the portal vein. After administration, dynamic acquisitions at one frame per second for 2 to 3 minutes are performed with the patient in right lateral recumbency using a $128 \times 128$ matrix and low-energy general-purpose collimator. The start of acquisition is timed with the administration of the radionuclide into the distal colon.
Radioactive markers are placed ventral to the xiphoid and apex of the heart on the gamma camera for later analysis of heart and liver location. A region of interest (ROI) is drawn over the liver and heart regions, and calculations of the time-intensity curves of the heart and liver are performed with dedicated software and provide an objective means of assessing the shunt fraction (Fig. 17). Disadvantages include lack of sufficient anatomic detail of the shunting vessel. Radiation safety concerns prevent surgical intervention after the procedure, and there is a need for sedation. Disadvantages of the technique include difficulty in identifying the liver and heart in small patients or those with poor colonic absorption or false-positive results because of rectal vein absorption of pertechnetate, which enters the systemic circulation and heart before the liver.

Positive findings for a PSS are based on the arrival of the radiopharmaceutic agent into the heart before the liver based on the time-activity curves of the ROIs drawn over the heart and liver regions. In abnormal animals, the liver is seen 10 to 12 seconds after heart activity is seen. Shunt fraction is based on the total heart counts between 8 and 16 seconds after injection divided by the total counts within the heart and liver ROIs. Dogs that have microvascular dysplasia have a normal study, with the radionuclide entering the liver before the heart. In acquired shunts, the small vessels in the middle to caudal abdomen are often difficult to visualize. Nondiagnostic or poor-quality scans have been reported at rates of 3.6% and 35.8%, respectively. These can result from poor absorption of the radionuclide, rectal administration, poor visualization of the heart and liver, fluid or diarrhea in the colon at the time of administration, or previous administration of colonic cleansing agents (oral or rectal).

Nonuniform distribution of the radionuclide attributable to portal streamlining may also cause difficulties in interpretation of the study if one is not aware of this normal phenomenon. Streamlining is a cause of nonuniform distribution of the radionuclide during portal scintigraphy within discrete channels of portal blood flow, such that they may distribute the radionuclide preferentially into one or more of the branches of the portal vein, giving a nonuniform appearance of the activity in the liver.

Transsplenic portal scintigraphy is a newly described alternative to PRPS. Compared with PRPS, it provides higher count density, consistent nuclear venograms of the splenic and portal vein, and significantly decreased radiation exposures.

Fig. 17. Normal splenic portal scintigraphy in a dog. This composite image was taken 4 seconds after injection of pertechnetate (2mCi) into the spleen. The activity in the liver ROI appears 12 seconds before the activity of the heart.
Transsplenic portal scintigraphy was found to be 100% sensitive and specific for diagnosis of congenital portosystemic shunt and significantly \((P<.05)\) more likely than PRPS to detect shunt number and termination. The technique is simple to perform and requires a lower dose of sodium \(^{99m}\)Tc-pertechnetate. A small volume (0.2 mL) of 2 mCi is injected by means of a 22-gauge 1.5-in needle into the splenic parenchyma using ultrasound guidance, and dynamic acquisitions at four frames per second for 3 minutes are acquired with the patient in right lateral recumbency. The acquisition must be started immediately before injection because of the more rapid nature of transport to the liver and heart compared with per-rectal methods. In normal animals, the radionuclide passes from the splenic vein to the left gastric vein and then into the main portal vein. One disadvantage of the splenic portal scintigraphic procedure is that shunts entering the vena cava caudal to the splenic vein could be missed. In the future, we may see the use of \(^{99m}\)Tc-mebrofenin applied, which should allow identification of the shunt and assessment of liver function.

TISSUE SAMPLING

A definitive diagnosis of most liver diseases depends on cytology and histopathology and, in some instances, bacteriology. Percutaneous ultrasound-guided aspiration and biopsy of the liver have become routine in dogs and cats. Patient preparation should include fasting for 12 hours before the ultrasound examination and tissue sampling. A coagulation profile is an important screening test before tissue core biopsy procedures, especially considering that several coagulopathies may occur with liver disease. Prothrombin time, activated thromboplastin time, and a platelet count are the minimum tests that should be performed for screening purposes. Cats and dogs should preferably be placed under general anesthesia for biopsy of the liver. Sedation with local anesthesia can also be performed on a case-by-case basis. Depending on the temperament of the dog or cat, sedation may or may not be required for fine-needle aspirations.

For diffuse lesions, the most accessible region of the liver should be sampled. Aspirations are preferred for small (<1 cm)-sized lesions, cystic structures, or lesions with high vascularity. Furthermore, fine-needle aspirations are recommended in diffuse

![Fig. 18. Splenic portal scintigraphy in a dog with an extrahepatic portocaval shunt that was confirmed and repaired surgically. The graph shows activity in the heart ROI appearing 12 seconds before that of the liver, which shows little activity over time. This is diagnostic for a PSS.](image)
lesions in which lymphoma or mast cell tumors are suspected because they generally result in diagnostic samples. Tissue core biopsies are generally recommended in most diffuse liver diseases and larger masses (>2 cm). Generally, if a tissue core biopsy is being made, fine-needle aspirations can be made at the same time, because a “preliminary” cytologic diagnosis can be made while waiting for the histopathology results, which generally take at least 24 hours or more to obtain. Touch preparations of the core biopsy sample can also be made for cytologic analysis.87

A sector, curved, or linear-array transducer may be used depending on where the lesion to be sampled is located.87 A superficial lesion can be well visualized with a high-frequency curved or linear array, whereas deeper lesions may require a low-frequency curved-array or sector format (phased-array) transducers. Tissue core needles with a 2-cm long sample notch should be used and are typically 16 or 18 gauge depending on the size of the animal. For medium- to large-sized dogs, 16-gauge needles are recommended, whereas 18-gauge needles are best for smaller dog and cats. Manual, semiautomatic, and automatic (spring-loaded gun) can be used depending on the personal preferences of the sonographer. Fine-needle aspiration is generally performed with 20- to 22-gauge 1.5-in needles for diffuse lesions, small nodules, and cystic or highly vascular structures.

After sedation or anesthesia, the skin should be clipped and cleaned in a routine sterile manner. The ultrasound probes should be covered with a sterile sleeve. For focal lesions (masses and nodules), sampling from the lesion and its periphery is recommended. Central lesional aspirations may only yield necrosis, especially with HCC. Two to three samples should be made from the affected region of the liver.

Free-handed or guided techniques can be used. In both situations, the needle should enter the plane of the ultrasound beam so that it can be visualized along its entire length. For free-handed aspirations, the needle is attached to a 3- or 6-mL syringe with the plunger pulled back before needle insertion or with a small amount of negative pressure during the aspiration. The tip of the needle is advanced under the skin and located by observing the ultrasound image and by making small “in and out” excursions under the skin. The needle is advanced until its tip is in the desired location, and in and out excursions are made within the lesion a few times, followed by removal of the needle. This is generally repeated two to three times to ensure adequate sampling for cytology. Nonaspiration techniques result in less blood dilution of the cytologic sample.

Free-handed and guided biopsy procedures may be used to obtain tissue core biopsies of the liver. Needle guidance systems are available for most ultrasound probes but are difficult to use on superficial hepatic lesions. The biopsy guide is attached to the transducer housing once the transducer is cleaned and covered with a sterile covering, such as a fitted sleeve or surgical glove. A number 11 blade should be used to make a small stab incision in the skin where the biopsy needle is to enter. This position is predetermined during the initial scanning of the liver. The needle is advanced as for fine-needle aspirations to the desired depth, taking into account the depth of the lesion and depth of penetration of the biopsy needle.87 Once the sample is taken, it should be gently removed from the sample notch and placed in formalin.

Complications of ultrasound-guided tissue sampling are rare.87 After any tissue sampling procedure, the patient should be monitored directly with ultrasound for the presence of free fluid. Small amounts of free fluid at the sampling site are not uncommon with tissue core biopsies but are less frequent with fine-needle aspirations. Small amounts of fluid are generally self-limiting when the patient’s coagulation status is normal.
Ultrasound-guided percutaneous cholecystocentesis can be performed safely and can provide valuable cytologic and bacteriologic information to make a diagnosis of cholecystitis and apply appropriate antimicrobial therapy.\textsuperscript{88} The best patient position and access to the gallbladder directly through its wall or transhepatically are determined, and this is generally a fairly simple procedure to carry out. One method is to have the patient sedated and in dorsal recumbency, with the skin of the cranioventral abdomen prepared aseptically. A 22-gauge 1.5-in needle connected to a 12-mL syringe should be guided sonographically to the gallbladder using a free-hand method. A biopsy guide can be used if preferred by the sonographer. The gallbladder must not be emptied, and depending on its size, this procedure may not be possible.

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