Abnormal Liver Enzymes:
A Clinical Approach
David Twedd, DVM, DACVIM
Colorado State University
Fort Collins, CO

The identification of abnormal liver enzymes usually indicates liver damage but rarely provides a diagnosis or etiology. Abnormal liver enzymes are common and in a study of 1,022 blood samples taken from both healthy and sick dogs and cats, one diagnostic laboratory found 39% had ALP increases and 17% had ALT increases. When presented with a patient having abnormal liver enzymes it is important to recognize that the patient could have primary liver disease but more likely the patient has other primary non-hepatic condition resulting in secondary liver involvement. It is therefore important to perform a complete review of all other body systems.

It is also important to understand the reason for increased liver enzyme activity and the following sections will deal with liver specific tests.

Tests of heptocellular necrosis or degeneration
Increases in either alanine aminotransferase (ALT) or aspartate aminotransferase activity (AST) indicate hepatocellular membrane damage and leakage of the enzymes. This could be due to death of the hepatocyte or from hepatocyte degeneration where the membrane just has increased permeability. Conceptually ALT and AST should be thought of as hepatocellular “leakage” enzymes. Subsequent to an acute and diffuse injury, the magnitude of increase crudely reflects the number of affected hepatocytes. The plasma half-life of ALT activity is about 2.5 days (60 hours) in dogs however concentrations may take days to weeks to decrease following an acute insult based on models of acute hepatic injury. Persistent increased ALT and AST enzyme activity over weeks is characteristic of chronic hepatitis in the dog. As a general rule, ALT increases should be investigated when they are greater than twice normal or persistently abnormal over weeks to months. Hepatic AST is located predominately in hepatocyte mitochondria (80%) but is also soluble in the cytoplasm. Because of the mitochondrial location, AST elevations are more sensitive for liver disease than ALT and reflect more significant cell damage. On the other hand, AST is less specific than ALT because of the presence in other tissues (i.e., muscle so always check CK). Following an acute injury resulting in a moderate to marked increase in the serum ALT and AST concentrations, due to their difference in plasma half-life, the serum AST will return to normal more rapidly (hours to days) than the serum ALT (days).

Tests of cholestasis and drug-induction
Alkaline phosphatase (ALP) and gamma-glutamyltransferase (GGT) show minimal activity in normal hepatic tissue but can become increased in the serum subsequent to increased enzyme production stimulated by either impaired bile flow or drug-induction. These enzymes have a membrane bound location at the canicular surface; ALP associated more with the canicular membrane and GGT associated more with epithelial cells comprising the biliary ductular system. With cholestasis, surface tension in the canaliculi and bile ductules increases and production of these surface enzymes is then up-regulated. An increase in the serum ALP and GGT activity can be the result of induction by endogenous, topical or systemic glucocorticoids, anticonvulsant medications (ALP only) and possibly other drugs or herbs. The plasma half-life for hepatic ALP in the dog is 66 hours in contrast to 6 hours for the cat and the magnitude of enzyme increase (presumably a reflection of the synthetic capacity) is greater for the dog than the cat. Bone source arises from osteoblastic activity and is elevated in young growing dogs before their epiphysial plates close or in some dogs with bone tumors or lytic lesions. One study identified that increased ALP concentrations in some dogs with osteogenic bone tumors tended to indicate a poorer prognosis, probably from diffuse bone metastasis. In the adult without bone disease, an increased serum ALP activity is usually of hepatobiliary origin. Hepatic GGT is located predominately on the canicular membrane and bile ducts. Chronic elevations in GGT tend to better reflect hepatobiliary tract disease, with he most marked elevations resulting from diseases of the biliary epithelium such as bile duct obstruction, cholangiohepatitis, cholecystitis or neoplasia. In dogs, GGT has a lower sensitivity (50%) but higher specificity (87%) for hepatobiliary disease than total ALP. If ALP is elevated with a concurrent increase in serum GGT, specificity for liver disease increases to 94%. Bone does not contain GGT and the administration of anticonvulsant medications to dogs does not cause an increase in the serum GGT activity.

Evaluation of liver function
On a routine biochemical profile it is important to note the liver function tests (or tests that involve liver function) including bilirubin, albumin, glucose, BUN, and cholesterol. Bilirubin elevations can occur from hemolysis, hepatic dysfunction or extrahepatic cholestasis. Measuring the percent conjugated to unconjugated bilirubin to determine location is not useful in the dog. Albumin is exclusively made in the liver and if albumin is not lost, sequestered or diluted, a low concentration would suggest significant hepatic dysfunction. It may take greater than 60% hepatic dysfunction for albumin concentrations to decline. Cholesterol can be variable and
increased in cholestatic conditions and decreased in portosystemic shunts. When glucose and BUN activity is low from liver dysfunction suggests significant hepatic disease and a guarded prognosis.

**Bile acids**

Measurement of serum bile acids is thought to be the most sensitive function test that is readily available in small animal practice. Bile acids are synthesized from cholesterol in the liver and then conjugated and excreted into the bile. Bile acids are transported to the gallbladder and following a meal are excreted into the intestine where they emulsify fat for absorption. In the distal small intestine bile acids are actively resorbed and return to the liver where they are efficiently extracted by the hepatocytes and then re-circulated back into the bile. Only a small fraction of the total bile acid pool ever escapes into the systemic circulation. Thus, the enterohepatic circulation of bile acids occurs with a 95-98% rate of efficiency. The current suggestion for performing bile acid levels is to differentiate between congenital portal vascular anomalies and liver insufficiency, prior to the development of jaundice. The determination of total bile acids can contribute to the decision to obtain histological support for a definitive diagnosis. The fasting total serum bile acid concentration (FSBA) is a reflection of the efficiency and integrity of enterohepatic circulation. Pathology of the hepatobiliary system or the portal circulation results in an increased FSBA prior to the development of hyperbilirubinemia, therefore, bile acid measurement is not useful in the icteric patient. An increase is not specific for a particular type of pathologic process but is associated with a variety of hepatic insults or abnormalities of the portal circulation. Bile acids should be used to screen patients with persistently abnormal liver enzymes, to determine if there could be loss of hepatic function, which adds further diagnostic support during investigation of the case. It is also helpful to measure bile acids to determine level of hepatic dysfunction in animals with PSS or portal vein hypoplasia (PVH), also known as microvascular dysplasia. When the fasting value is greater than 25 µmol/L for the dog and cat, there is a high probability that the histology findings will define a lesion.

When the total fasted bile acid concentration is normal or in the “gray zone” the FSBA should be followed by a 2-hour postprandial serum total bile acid (PPSBA) looking for an increase of greater than 25 µmol/L. The diagnostic value of determining PPSBA concentration is increased sensitivity for the detection of hepatic disease and congenital portal vascular anomalies. In dogs, the specificity of fasting and postprandial bile acids for hepatobiliary disease is 95% and 100% when cutoff values greater than 15 µmol/L and 25 µmol/L are used, respectively. When using these guidelines it is prudent to recognize that a small number of apparently healthy dogs have been reported with PPSBA values above 25 umol/L or these may actually represent dogs that have PVH. Occasionally the FSBA value is greater than the PPSBA value. The reason for this non sequitur is probably multifactorial. It has been shown that (1) the peak PPSBA concentration for individual dogs is variable, (2) fasted dogs store about 40% of the newly produced bile in the gallbladder and (3) a meal stimulates the release of only between 5 to 65% gallbladder bile. Undoubtedly these physiologic variables in addition to physiological variation in intestinal transit time and concurrent underlying intestinal disease contribute to the dichotomy.

Recently, urinary bile acids have become available as a diagnostic tool. Identifying increased urinary bile acids provides similar information to what is obtained from serum bile acids and neither test appears to be better than the other. The advantage of urinary bile acid measurements would be for the screening of litters of young puppies for suspected inherited vascular anomalies where urine collection is simpler than paired serum samples.

**Coagulation panels**

Major clotting factors are synthesized in the liver (except factor 8) and therefore prolonged clotting time may suggest significant hepatic dysfunction or factor consumption. Because coagulation abilities may not be normal in patients with liver disease, it is advisable to check clotting times prior to performing liver biopsy.

**Ammonia**

High ammonia levels reflects abnormal hepatic portal shunting (acquired or congenital shunts) or significant hepatocellular dysfunction of greater than 70%. The liver detoxifies ammonia that primarily arises from the gastrointestinal tract by conversion to urea. Elevated fasting blood ammonia levels have been shown to be a sensitive (98%) and specific (89%) test for the detection of congenital or acquired portosystemic shunting in dogs. Due to problematic requirements for sample handling and submission, blood ammonia or the ammonia tolerance test is infrequently performed by some clinical practices. However, recent availability of blood ammonia for in-clinic analyzers, has helped make the test more feasible.

**Diagnostic strategies.**

In the asymptomatic patient with an increased liver biochemical test(s) the increased value should be confirmed. If no likely explanation for the laboratory abnormalities can be found there are two courses of action that one can take; either begin a diagnostic evaluation of the patient starting with bile acid determinations, or re-evaluate the patient’s liver enzymes at a later date. The diagram below depicts a general algorithm for the work-up of dogs that have abnormal liver enzymes. The identification of abnormal liver enzymes may occur when the sick patient is presented for evaluation or during a routine health screen in the healthy patient. Abnormal liver enzymes in the sick patient could either be the result of primary liver disease/damage or secondary due to a multitude of other non-hepatic disorders. The most common cause of abnormal liver enzymes is in fact, not primary liver disease at all but rather the result of reactive hepatic changes occurring secondary to other non-hepatic causes. Generally, secondary hepatic changes are
reversible once the primarily disease is treated. Successful resolution of the non-hepatic disease and continued abnormal liver enzymes would be a strong indication for further investigation of the liver for a primary disease process.

**Imaging**

Routine abdominal radiographs are helpful in determining liver size and shape and for detection of other intra-abdominal disorders. Ultrasonography is noninvasive, readily available and is the most informative initial imaging modality for liver disease. Ultrasound can determine parenchymal changes, mass lesions and disorders of the biliary system. Ultrasound however is not accurate in differentiation of the major parenchymal changes.

**Fine needle aspiration**

(FNA) for cytological evaluation is safe easily performed using ultrasound direction. One should be cautious in over interpretation of those results however. FNA is best for identification of vacuolar hepatopathies and neoplasia and is poor in detecting inflammatory hepatic changes. In one study we found FNA and cytology to only correlate in about 1/3 of the cases.

**Liver biopsy**

A biopsy is required for a definitive determination of the nature and extent of hepatic damage and to appropriately direct the course of treatment. The method for liver biopsy procurement may be surgery, ultrasound guided needle biopsy or laparoscopy. We believe if a needle biopsy is obtained that at least a 16g biopsy needle or larger be used and multiple liver lobes are biopsied. We generally take 3-4 biopsies with one split for culture and hepatic copper analysis and the remainder placed in formalin for histological evaluation.

**What you might find on a liver biopsy**

When we evaluated 150 consecutive canine liver biopsies we identified the largest category to be secondary reactive hepatopathies (25%) followed then by chronic hepatitis (23%) and then neoplasia and vascular hepatopathies making up 69% of the biopsies performed. Smaller categories included vascular anomalies, acute liver damage and other miscellaneous conditions.

**Reactive hepatopathies; a common diagnosis**

The so-called “non-specific reactive hepatopathies” (NSRH) that occur secondary to non-hepatic disease can result in increased serum biochemical hepatic tests and histomorphologic abnormalities. Most of the NSRH cause increases in laboratory tests that evaluate hepatocellular integrity (ALT, AST) and tests of hepatic cholestasis (ALP, GGT). In most cases there are little if any changes in tests that evaluate hepatic function (bilirubin, albumin, glucose, and BUN). Most of the animals with this type of secondary liver disease often retain normal hepatic function (albumin, serum bile acid concentrations), which again supports a concept that there is generally minimal loss of hepatocellular dysfunction. NSRH is often characterized by variable amount of hepatocellular degeneration or necrotic changes without evidence of significant chronic progressive inflammation. The reason the liver often undergoes these changes revolves from the fact that the liver is involved in so many metabolic and detoxification functions. Endogenous toxins, anoxia, metabolic changes, nutritional changes and endogenous stress related glucocorticoid release are all examples of conditions responsible for the majority of these changes. Gastrointestinal disease frequently results in secondary hepatic changes uptake of bacteria, toxins or nutrient abnormalities.

Histological findings associated with NSRH changes include descriptors such as vacuolar degeneration, hydropic degeneration, swollen hepatocytes, lipidosis, intracellular or intralhepatic cholestasis, mild multifocal hepatitis or periportal hepatitis or variable random hepatic necrosis. These changes are devoid of the typical progressive chronic inflammatory cell infiltrates characteristic of chronic hepatitis. Whenever I observe these changes on histology I always begin a search for an underlying etiology.

A good example that helps explain this concept is inflammatory bowel disease in which it is not unusual to observe mild inflammatory changes around portal tracts presumed to be the result of abnormal portal uptake of gastrointestinal “toxins”. Throughout the liver and closely associated with portal areas are Kupffer cells (fixed macrophages) that function to filter the blood of injurious toxins, inflammatory mediators and bacteria. When this macrophage system is abnormally insulted Kupffer cells release their own inflammatory mediators that in turn insult the hepatocytes.

In a review of consecutive liver biopsies at Colorado State University histology grouped as non-specific reactive changes made up the largest category of abnormalities (approximately 25%) In this group we were able to identify an associated disease in many that could explain the likely cause for the hepatic enzyme increases and histological changes observed. Concurrent diseases identified
included neoplasia, gastrointestinal, renal, autoimmune, dermatologic, dental, infectious and cardiac disease as a few examples. In some cases an underlying disease is not identified. The ALT values on the average are 1-2 X normal and the ALP values 1-3 X normal. It is interesting to note that in a series of 32 dogs having reactive hepatopathies, 8/8 cases in which serum bile acids were run, all were within the normal reference range again suggesting hepatic function tends to remain intact.

This category appears to be the most common histological change to occur in dogs and is by far the most common cause of elevated liver enzymes. Based on this fact, dogs presented with elevations in ALT and ALP should always have primary non-hepatic disease ruled out first. These changes are usually very reversible and no specific hepatic therapy is required short of treating the primary disease. The liver changes resolve once the primary etiology is successfully treated. Therapy providing good liver support such as antioxidants may be warranted.

Summary
Abnormal liver enzymes should not be ignored and should be investigated in a systematic manner as previously discussed. Asymptomatic animals with no evidence of significant or treatable disease or in situations where financial constraints limit further work up the patient should be fed a quality maintenance diet for the patient’s stage of life and the possibility of instituting specific liver support therapy should be explored.