DIAGNOSTIC TESTS FOR LIVER DISEASE IN DOGS AND CATS (PART 1/2)

The liver plays a central role in a large number of processes, for example, in the metabolism of carbohydrates, lipids and proteins. It is responsible for the detoxification of metabolites and foreign matter, and for the storage of vitamins, trace elements and glycogen. Furthermore, the liver plays an important role in lipid digestion and in immune regulation.

Introduction
The liver has a large reserve capacity for many of the functions it carries out, which results in relatively specific clinical indications of hepatobiliary diseases, such as icterus, ascites, coagulopathy or hepatic encephalopathy. However, these signs occur late in the course of the disease, once the functional reserves of the liver have already been depleted. In addition, early indications of a hepatobiliary disease, such as apathy, anorexia, polyuria/polydipsia and vomiting, are highly non-specific, and may occur as a result of diseases affecting many other organ systems. The interpretation of changes in laboratory parameters is therefore crucial to the early diagnosis of hepatopathy.

1 Liver enzymes
Liver enzymes are relatively sensitive indicators for the diagnosis of liver diseases and their concentrations are raised in many hepatopathies. Unfortunately, however, they are not very specific, and there are many other causes for raised liver enzyme levels in which the liver is not, or only secondarily, affected by the disease process.

The concentration levels to which the liver enzymes are elevated are usually proportional to the severity of damage to the liver cells and/or the biliary system, but are no measure for the capacity of the liver to fulfill its functions. Therefore, high liver enzyme values do not serve as indicators for hepatic insufficiency. Even very high liver enzyme values do not necessarily indicate a bad prognosis, due to the liver’s high capacity for regeneration.

On the other hand, liver enzymes may also be normal or only slightly raised in severe chronic liver diseases in which there is hepatic insufficiency. For example, this may be the case in liver fibrosis or cirrhosis of the liver, in which the liver cells are replaced with connective tissue, and the chronic release of liver enzymes results in depletion of the entire available pool of liver enzymes. For the reasons mentioned above, assessments of one-off tests of liver enzyme values should not be used in prognoses. In cases in which liver enzymes are measured several times during the course of the disease, the interpretation of all these values may be of relevance in prognoses, especially when the results of a liver function test or a liver biopsy are also included. The fact that only raised levels are significant should be considered when assessing liver enzymes; decreased values are of no relevance.

1.1 Alanine aminotransferase (ALT)
Alanine aminotransferase, ALT, (previously serum glutamate pyruvate transaminase, SGPT) is an enzyme specific to the liver that is found in the cytoplasm of the liver cells. The red blood cells and the cross-striated muscle cells contain only small quantities of ALT, so there is only rarely any increase in ALT in cases of severe muscle damage in dogs. An increase in ALT generally indicates the presence of hepatopathy and, in particular, damage to the actual liver cells.

For example, ALT is almost always raised in (cholangio-)hepatitis. However, it is not possible to conclude that a specific liver disease is present based on increased ALT values. The liver may also only be affected secondarily, e.g., due to hypoxia of liver cells after circulatory collapse, trauma, pancreatitis or sepsis. Hyperthyroidism should also be considered in cats.

Certain drugs, such as glucocorticoids (only in dogs) or barbiturates may also result in increases in ALT. In most dogs, the ALT increases caused by these drugs are not very high (two- to four-fold increases), although there are cases in which the values are
substantially higher. There are two different mechanisms resulting in this iatrogenic increase in ALT - an induction of the enzyme by the drugs, and the potential hepatotoxic effect of the drugs. Cushing’s syndrome may also be the cause of increases in ALT in dogs.

The serum ALT half-life is one to two days or less. A reduction in the values is to be expected within one to two weeks after the end of active liver cell damage.

1.2 Alkaline phosphatase (ALP)

Alkaline phosphatase (ALP) is found in the liver, as well as in the bones, kidneys, mucosa of the small intestine, spleen, placenta, leukocytes and erythrocytes. The specificity for hepatopathy is therefore lower than for ALT. The biliary system is usually involved when there is an increase in ALP, and this may be a case of intrahepatic or extrahepatic cholestasis. However, as with increases in ALT, it is not possible to conclude that a specific liver disease is present. Benign nodular liver cell hyperplasia, such as is often found in older dogs, can also lead to mild to moderate increases in ALP. Raised ALP levels due to increased bone metabolism in the context of pathological processes (e.g., osteomyelitis or osteosarcoma) are generally mild. Other important differential diagnoses are diabetes mellitus, sepsis, pancreatitis, hyperthyroidism in cats and Cushing’s syndrome in dogs. ALP induction may occur in dogs due to exogenous corticosteroids and phenobarbital. In experimental studies an increase in ALP was observed on the third day after administration of an immunomodulating dose of prednisolone. Increased ALP levels due to phenobarbital are usually mild (two- to six-fold increase). Any increase in feline ALP is to be regarded as significant and further examinations are indicated, as the serum ALP half-life is only 6 hours in cats. Juveniles have higher values than adult animals.

1.3 Aspartate aminotransferase (AST)

Aspartate aminotransferase, AST, (previously serum glutamate oxaloacetate transaminase, SGOT) is not specific to the liver, as it is also found in large quantities in the musculature and in the erythrocytes. Muscle disease (or even just an intramuscular injection) and haemolysis (both spontaneous and as an artifact during preliminary analysis) can therefore lead to increased AST values. As is the case for ALT, increased AST values also occur with damage to the liver cells. However, while ALT occurs in the cytoplasm of the liver cells and enters into the bloodstream after only slight damage to the cell membrane, AST occurs in the mitochondria. These are not damaged as rapidly as the liver cell membrane, which means that a significant increase in AST is an indicator for severe hepatopathy. Muscular disease should also be taken into consideration in cases in which AST is far higher than ALT. Corticosteroids and phenobarbital cause a mild increase in AST in dogs.

1.4 Gamma glutamyl transferase (GGT)

Gamma glutamyl transferase (GGT) is found in many tissues, but serum GGT originates mainly in the liver. It is predominantly found in the intrahepatic biliary epithelia. Increases in GGT are most frequently observed in cholestatic diseases (similar to ALP). In cats, it is notable that classically ALP is severely elevated in cases of hepatic lipidosis, while GGT is only mildly raised, or there is no increase at all. As is the case for the other liver enzymes, phenobarbital and corticosteroids may lead to increases in GGT (as a rule, mild, two- to three-fold increases).

1.5 Lactate dehydrogenase (LDH)

LDH occurs in many tissues and therefore has a very low specificity. Thus it is of little use in the diagnosis of hepatopathies.

2 Albumin

Albumin is produced exclusively in the liver. Hypoalbuminaemia is often only found in chronic hepatopathies, as the liver has a large reserve capacity and the half-life for albumin is eight to nine days. Hypoalbuminemia leads to a reduction in plasma oncotic pressure, which can result in ascites. Hypoalbuminaemia is not specific to liver disease and can occur due to many other diseases - the most important are protein-losing enteropathy, protein-losing nephropathy, vasculitis, large exudative skin injuries and blood loss.

3 Coagulation proteins

The liver plays a central role in the metabolism of many substances that influence blood clotting. For example, the liver synthesises all coagulation factors, except for factor VIII. Furthermore, it also synthesises both coagulation inhibitors (antithrombin III), and fibrinolysis inhibitors (antiplasmin). Fibrinolytic proteins (plasminogen) are also produced by the liver. Knowledge of the patient’s coagulation status is important as changes in haemostasis in liver patients can result in clinical symptoms or haemostatic complications when taking a liver biopsy.
Assessment of prothrombin time (PT), activated partial thromboplastin time (aPTT), fibrinogen and fibrin degradation products are most common. Determination of the coagulation parameters PT and aPTT can be conducted at the practice using CoagDx™. Alternatively, citrated plasma can be sent in to the laboratory for analysis. The interpretation of the coagulation parameters in patients with a hepatopathy is often difficult due to the complex, partially inhibitory and partially activating, effects of the liver on blood coagulation. Even though a large proportion of dogs and cats with hepatobiliary diseases exhibit changes in one or more of the coagulation tests, spontaneous haemorrhages are rare. Haemorrhages are more likely to occur after a liver biopsy.

4 Urea
For the purposes of detoxification, the ammonia (NH3) produced in protein metabolism is transformed into urea (urea cycle) in the mitochondria of the liver cells. In the case of hepatic insufficiency (e.g., due to liver atrophy caused by a portosystemic shunt, PSS), urea levels may be reduced, as the liver cells are no longer capable of producing sufficient urea. A reduced urea value, however, is not specific to liver disease, as several other factors also influence the blood urea content. A low-protein diet and polyuria/polydipsia, in particular, may result in reduced urea values. Animals with liver disease exhibit a tendency towards gastrointestinal haemorrhages and these patients may therefore also exhibit raised urea levels.

5 Glucose
The liver is also responsible for the maintenance of normoglycaemia and hepatic insufficiency or portosystemic vascular anomaly (PSVA) may therefore result in hypoglycaemia. Hypoglycaemia is observed particularly frequently in small dogs with a congenital portosystemic shunt. However, hypoglycaemia is not specific to hepatic insufficiency. Among the many other causes of hypoglycaemia are an insulinoma or some other neoplasia, sepsis, Addison’s disease, neonatal or juvenile hypoglycaemia, drugs, as well as glycogen storage diseases. When submitting samples to the laboratory for glucose estimation, please ensure that a fluoride oxalate collection tube is used.

6 Cholesterol
Liver diseases may also be associated with reduced as well as increased cholesterol values. Hypercholesterolaemia may occur in conjunction with diseases associated with a cholestasis, while more than 60 % of dogs and cats with PSVA exhibit hypocholesterolaemia. There is a whole range of differential diagnoses for increased cholesterol values, while there are only a few causes of reduced cholesterol values (protein-losing enteropathy, certain malignant tumours and severe malnutrition). The most important causes are postprandial blood sampling, hypothyroidism, diabetes mellitus, Cushing’s syndrome, pancreatitis, nephrotic syndrome and idiopathic hyperlipoproteinaemia in the miniature schnauzer.

7 Haematological changes
Anaemia may be regenerative or nonregenerative in liver patients. A regenerative anaemia is often the result of a haemorrhage, which can occur, for example, due to gastrointestinal ulceration or after a biopsy. A nonregenerative anaemia is usually triggered by the inefficient use of systemic iron reserves (anaemia due to chronic disease). Dogs and cats with congenital PSVA, dogs with an acquired shunt after cirrhosis of the liver or cats with idiopathic hepatic lipidosis, may exhibit microcytosis of the erythrocytes (small erythrocytes), which is manifested by a low mean cell volume (MCV) for the erythrocytes. The cause of microcytosis is assumed to be connected with a relative iron deficiency, due to impaired iron transport.

8 Urinanalysis
Uric acid, a product of purine metabolism, is normally transformed into allantoin by hepatic urate oxidase. There is a deficiency in this enzyme in liver diseases, resulting in a high concentration of uric acid in the blood. There is an increased concentration of both ions in the urine in cases where the plasma ammonia levels are also increased, which may then result in the precipitation of ammonium biurate crystals. Between 40 and 74 % of dogs and 15 % of cats with PSVA exhibit ammonium biurate crystalluria. Small amounts of bilirubin in the urine of dogs (mainly male animals) does not constitute a pathological finding. The reason for this is the low renal threshold for bilirubin excretion and the capacity of the renal tubuli epithelium cells to produce bilirubin. In contrast, bilirubinuria in cats is always pathological. Hepatobiliary diseases and haemolysis are the most important differential diagnoses for bilirubinuria.
**Summary**

No laboratory parameter is 100% specific to a liver disease. In cases of deviations from normal values, there are always a variety of differential diagnoses for each of the parameters discussed. However, if all haematological and biochemical laboratory parameters are considered, then a suspected diagnosis of hepatopathy is possible. It is important to distinguish whether liver function is adversely affected (hepatic insufficiency), whether PSVA might be present, or whether a disease is present in a still functional liver. Hypoalbuminaemia, reduced urea, hypoglycaemia, changes in the cholesterol values, as well as changes in the coagulation tests, may indicate hepatic insufficiency or PSVA. Urine analysis and the haemogram can also be very revealing. In contrast to the laboratory tests outlined above, raised liver enzyme values are not indicators for hepatic insufficiency.

The important contribution made by hyperbilirubinaemia and raised bile acid levels in the diagnosis of hepatopathy is discussed in part 2.

---

**Diagnostic options for liver diseases**

**IDEXX Reference Laboratory**

<table>
<thead>
<tr>
<th>Test</th>
<th>Price</th>
</tr>
</thead>
<tbody>
<tr>
<td>Liver Profile</td>
<td>£ 20.00 + VAT</td>
</tr>
<tr>
<td>Total protein, Albumin, Globulin, Albumin:Globulin Ratio, Urea, ALT, ALP, GGT, Total Bilirubin, Bile Acid, Cholesterol</td>
<td></td>
</tr>
<tr>
<td>Liver Profile plus Bile Acid Stimulation</td>
<td>£ 23.00 + VAT</td>
</tr>
<tr>
<td>as for Liver Profile + Bile Acid Stimulation Test</td>
<td></td>
</tr>
<tr>
<td>Liver &amp; Kidney Profile</td>
<td>£ 32.50 + VAT</td>
</tr>
<tr>
<td>Total Protein, Albumin, Globulin, Albumin:Globulin Ratio, Urea, Creatinine, ALT, ALP, GGT, AST, Total Bilirubin, Bile Acid, Sodium, Potassium, Sodium:Potassium ratio, Cholesterol, Calcium &amp; Inorganic Phosphate</td>
<td></td>
</tr>
<tr>
<td>Coagulation Profile</td>
<td>£ 28.50 + VAT</td>
</tr>
<tr>
<td>Platelet Count, Morphological Assessment, PT, APTT, Fibrinogen</td>
<td></td>
</tr>
</tbody>
</table>

**IDEXX VetLab® Suite**

- VetTest® Chemistry Analyser
- SNAP® Reader Bile acid and hormone analyser
- Coag Dx™ Analyser

Authors: Dr. med. vet. Cécile Rohrer Kaiser
Dipl. ACVIM (Internal Medicine) und
ECVIM-CA (Internal Medicine)

Literature on request