Hepatic Functions
and
Laboratory Assessment-I

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Liver

- Liver is the major organ that regulates too many metabolic reactions. It is also called **Metabolic Chief**.
- **It's regeneration ability is quite high.**
  - A liver that is only 10-20% functional is sufficient for survival.
  - Complete removal of the organ results in death within 24 hours.
- **It consists of,**
  - Hepatocytes (%60),
  - Reticuloendothelial cells (Kupffer) and sellate cells (Ito cells) (%30),
  - Basolateral membrane (surface) - sinusoids, Blood circulation apical (canalicular) membrane – bile duct (%10).

Liver

- **Molecules found in liver**
  - %76 water
  - %16 protein
  - %5-10 glycogen
  - %2,2 lipids
- **Anatomic structures**
  - Hepatic cells, bile duct system, vascular siystem and reticuloendothelial system
- Blood flow is provided by portal vein, hepatic artery-veina and capillary fenestrations.

Metabolic Functions

- **Carbohydrate Metabolism**
  - Gluconeogenesis
  - Glycogenesis, glycogenolysis
- **Lipid Metabolism**
  - Fatty Acid synthesis
  - Cholesterol synthesis and excretion
  - Lipoprotein synthesis
  - Ketogenesis
  - Synthesis of Bile Acids
  - 25-hydroxylation of vitamin D

Metabolic Functions

- **Protein Metabolism**
  - Synthesis of plasma proteins
    - Coagulation factors, immunglobulins, albumin, cytokines
  - Ure Synthesis

- **Hormone Metabolism**
  - Synthesis and exceetion of steroid hormones
  - Metabolims of polypeptide hormones

- **Metabolism and disposal of drugs and foreign substances (Detoxification)**

Metabolic Functions

- **Bilirubin metabolism and its excretion**

- **Nucleic acid metabolism**
  - It is involved in the Purine and pyrimidine synthesis.
  - It's rich in *xanthine oxidase*.

- **Storage Function**
  - Glycogen
  - Vitamin A
  - Fe

Liver Diseases

- Liver pathologies include primary hepatic cells, bile duct and vascular system.
- Functional tests show mostly cellular injury and response to cellular disorders.
- The great majority of the disorders are pathologically present in the form of liver cell necrosis, intrahepatic and extrahepatic bile duct obstruction (cholestasis), liver atrophy and/or liver fibrosis.

Hepatocellular Injury

- **Hepatocyte injury is determined by measuring hepatocellular enzyme activities (leakage enzymes).**
- 3 enzymes are used routinely.
  - These are **ALT, AST, SDH**
  - In addition, **GLDH** is available.

Hepatocellular Injury

- **ALT**
  - **It is ap primary marker of hepatocellular damage. Especially in cats and dogs.**
  - Increased activity express hepatocyte death, sublethal hepatocyte damage.
  - ALT activity is sometimes the only test used to detect hepatocyte injury in dogs and cats
  - Apart from the liver, it is also found in the skeletal and heart muscles. **CK** should be evaluated for differential diagnosis.
  - Horses and ruminants have low ALT concentration in hepatocytes; consequently, serum ALT activity is not useful for detecting liver disease in these species

Hepatocellular Injury

- Moderate amounts of ALT are present in the muscle of horses and ruminants, and moderate increases in the serum ALT activity occur with muscle injury in these species; however, ALT is not included in large animal biochemical profiles. Other muscle-specific enzymes (e.g., CK) are more commonly used for detecting muscle injury in these species.
- In dogs and cats, a wide variety of liver diseases can produce increased serum ALT activity.
- **Hypoxia, metabolic alterations resulting in hepatocyte lipid accumulation, bacterial toxins, inflammation, hepatic neoplasia, and a multitude of toxic chemicals and drugs can cause hepatocyte injury, thereby resulting in ALT leakage.**

Hepatocellular Injury

- Acutely, the serum activity of ALT is proportional to the number of cells that are injured
- But the magnitude of ALT activity is not indicative of the cause of the injury.
- After acute severe injury, such as from a toxin, serum ALT activity can increase markedly within a day or two. If the injury is not ongoing, ALT activity slowly decreases over several weeks.
- There is also an increase during active hepatic regeneration. Although the half-life is 17-60 hours in dogs and 3.5 hours in cats, it is consistently high in recovery.

Hepatocellular Injury

- In chronic inflammatory conditions, ALT activities are fluctuating. Result in periodic “flares” of increased ALT activity
- Repetitive measurements should therefore be made. Thus, a deeper assessment of the disease can be made.
- It is important to recognize that in certain situations significant liver disease can occur with normal or only slightly increased serum ALT activity.
  - If hepatic mass is markedly decreased.
  - Massive acute hepatic necrosis
  - Aflatoxin

Hepatocellular Injury

- Increases in serum ALT activity can also be observed in dogs with hyperadrenocorticism or that have been administered corticosteroids.
  - These increases are generally mild (two- to fivefold), but ALT activity increases can vary widely among dogs receiving corticosteroid therapy, depending on the dose and duration of treatment.
- Anticonvulsant drugs (e.g., phenobarbital) also cause mildly increased serum ALT activity in dogs.
  - Some dogs receiving anticonvulsants will develop a toxic hepatopathy, in which case ALT activity may be markedly increased

Hepatocellular Injury

- **AST**
  - It is present at highest concentrations in hepatocytes and muscle cells (both skeletal and cardiac) of all species. Therefore, AST is not a liver-specific enzyme.
  - AST is found predominantly in the cytoplasm.
    - With about 20% located within mitochondria.
  - Increased serum AST activity can result from lethal or sublethal injury to either hepatocytes or muscle cells.
  - In dogs and cats with liver diseases, serum AST activity will generally increase parallel to ALT activity, but the magnitude of the increase may be less than that of ALT.

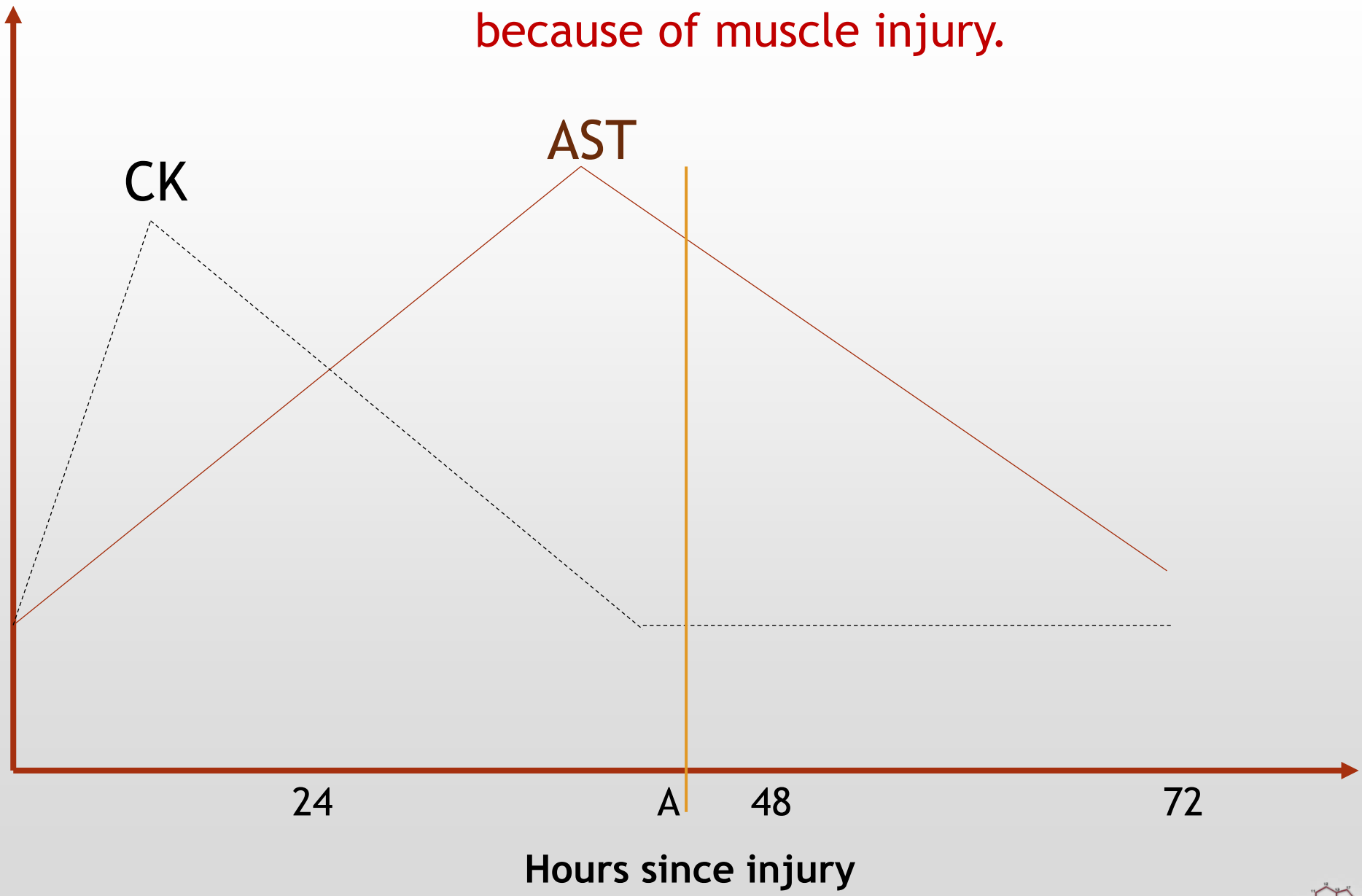
Hepatocellular Injury

- The serum AST activity may return to baseline faster than ALT following acute liver injury in some animals, making repeated measurements useful for monitoring disease resolution.
- Although AST is less liver specific than ALT, it may be more sensitive than ALT for detecting some liver diseases (e.g. Hepatic lipidosis in cats).
- Similar to ALT, mild increases in AST activity may be seen in dogs as a result of enzyme induction due to corticosteroids and possibly phenobarbital.
- CK activity measurement should be done for differential diagnosis (for muscle).

Hepatocellular Injury

- In horses and ruminants, it is routinely used (more frequently) in the assessment of hepatocellular damage and is absolutely within the biochemical profiles.
- Yinede spesifitesinin sadece karaciğer ait olmaması nedeniyle tek başına yeterli değildir. Ayırıcı tanı için CK ölçülmelidir.
- The major problem with AST in detecting hepatocyte injury is its lack of liver specificity. CK should be measured for differential diagnosis.
- Increased AST activity with normal CK activity may be seen if the source of the AST is the liver, suggesting hepatocyte injury has occurred.
  - The half-life of CK is shorter than that of AST. Serum activities of both enzymes may increase as a result of muscle injury, but the CK activity may return to normal earlier than the AST activity. These problems with use of AST in detecting hepatocyte injury in horses and ruminants have led to use of more liver-specific enzymes (such as sorbitol dehydrogenase [SDH]) in these species.

Serum activities of both AST and CK increase because of muscle injury.



Hepatocellular Injury

- **SDH**
  - It is free in the cytoplasm.
  - It is liver specific enzyme in dogs, cats, horses and ruminants.
  - Increased serum SDH activity is suggestive of either hepatocyte death or sublethal hepatocyte injury.
  - It is not used very often in cats and dogs.
  - In horses and ruminants, SDH is much more specific than AST. Thus, it is preferable to AST for detecting hepatocyte injury in horses and ruminants
  - The half-life of SDH is very short; serum activities may return to normal within 4–5 days after acute hepatocyte injury. The main disadvantage to SDH is that it is less stable in vitro than most other diagnostic enzymes

Hepatocellular Injury

- **GLDH**
  - It plays a key role in the detoxification of ammonia.
  - It is a leakage enzyme present in highest concentration within mitochondria of hepatocytes.
  - Liver specific for horse and ruminants (hepatic necrosis). Used in dogs and cats.
  - No analysis is done in each laboratory.
  - Increased serum concentration is reported to have excellent sensitivity for the detection of canine hepatic disease.
  - Serum activity of GLDH may increase in dogs with hyperadrenocorticism; increases have also been documented in dogs receiving anti-convulsants.

Cholestasis

- **Cholestasis** (impaired bile flow) can be detected by measuring the activities of serum enzymes whose increased production is induced by cholestasis or by measuring the serum concentrations of substances.
- **ALP**
  - It is attached to cell membranes and synthesized by many tissues such as liver, bone, kidney, intestine, pancreas, and placenta.
  - Most of the normal serum ALP activity originates from the liver. The half-life of other isoenzymes is short.
  - **Increased serum ALP activity commonly occur with cholestasis, increased osteoblastic activity, induction by certain drugs (primarily in dogs), and a variety of chronic diseases.**

Cholestasis

- **ALP in the liver is associated with biliary epithelial cells and canalicular membranes of hepatocytes.**
- A variety of hepatobiliary diseases can result in increased serum ALP activity due to increased enzyme production, solubilization of membranes by the action of bile salts, and release of membrane blebs after cell injury.
- Cholestatic diseases can result in marked increases in serum ALP activity in dogs (greater than 10 fold), but increases are more variable in other species.

Cholestasis

- The half-life of the cholestasis-induced ALP is approximately 3 days in **dogs** but only 6 hours in **cats**. However, ALP is still a useful enzyme for evaluation of feline cholestatic liver disease if one keeps in mind that even mild increases (2–3× URL) can be significant.
- The utility of ALP for detection of cholestasis in **horses** and **ruminants** is generally considered inferior to that of GGT.
- **When cholestasis is the cause of increased serum ALP activity, serum total bilirubin and bile acid concentrations may be increased concurrently.**

Cholestasis

- In dogs with cholestasis, serum ALP activity often increases prior to increases in serum bilirubin concentration; thus ALP is a more sensitive indicator.
- However, even if the serum bilirubin concentration is normal, bilirubinuria may accompany cholestasis-induced increases in ALP.
  - Whereas lesions primarily involving the intra- or extrahepatic biliary system are common causes of cholestasis, hepatic diseases resulting in significant hepatocyte swelling (e.g., lipidosis or inflammation of the hepatic parenchyma) can obstruct small bile canaliculi and induce increased ALP production and release.

Cholestasis

- Increased serum ALP activity associated with increased **osteoblastic activity** occurs in all species.
- **These increases are most often detected in young, growing animals.** E.g. the reference interval for total ALP activity in four-week old kittens was 97-274 U/L compared to 10-80 U/L for adult cats.
- One must remember that young animals commonly have serum ALP activities greater than adult reference intervals.
  - In puppies, kittens, and calves, ALP activity increases attributed to bone growth are generally mild (<4–5×), but foals may have increases up to 20× in the first three weeks of life.

Cholestasis

- In mature animals, oOsteosarcoma and other bone neoplasms (both primary and secondary) inconsistently result in increased serum ALP activity.
- Fracture healing usually results in localized increases in osteoblastic activity and mild increases in serum ALP that may be useful for monitoring the progression of healing.
- Canine hyperparathyroidism (primary or secondary) and feline hyperthyroidism may result in increased bone turnover and increased osteoblastic activity; mild increases in serum ALP may be detected in patients with these diseases.

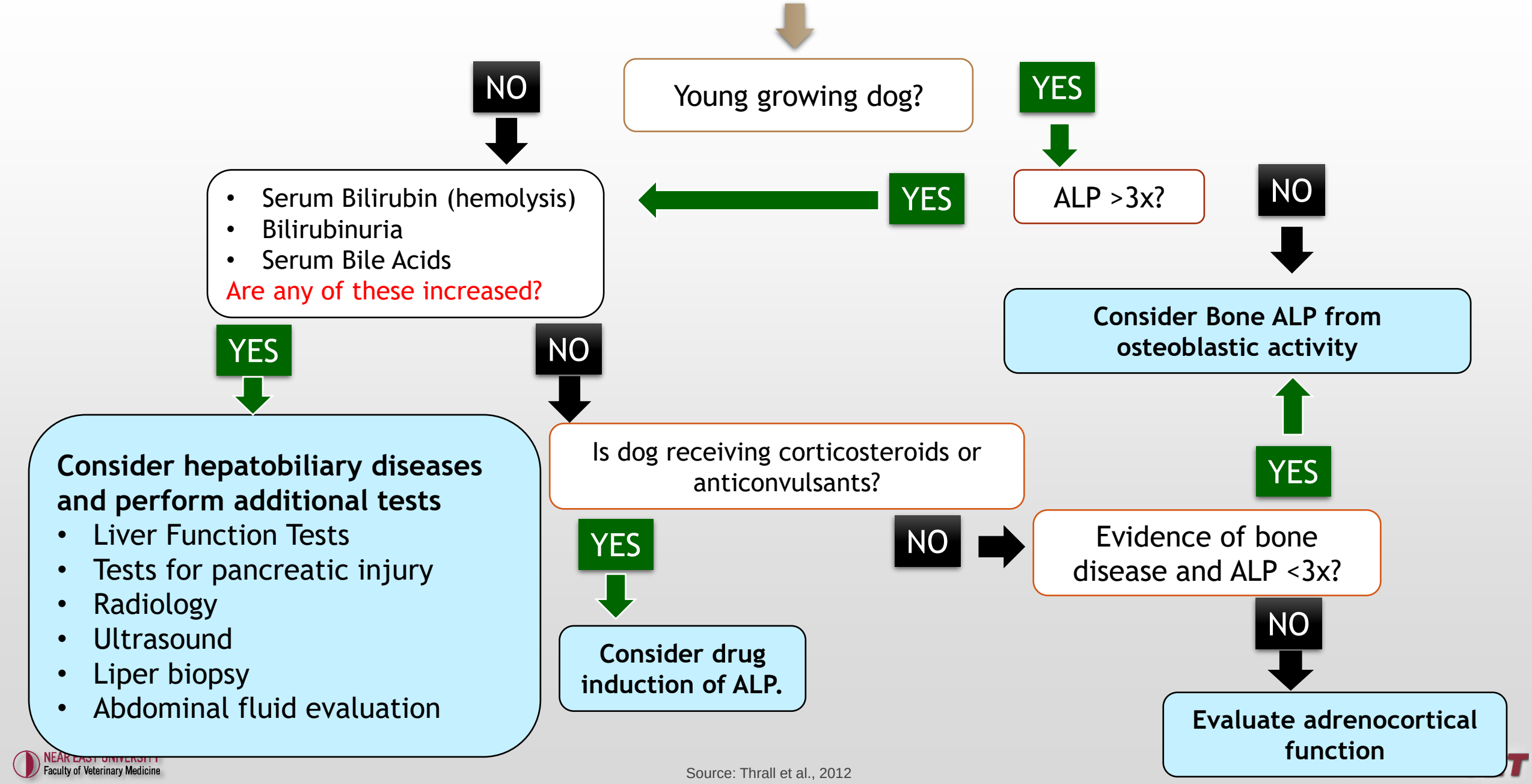
Cholestasis

- Serum ALP activity can be markedly increased when enzyme production is induced by **certain drugs**.
- Corticosteroids (exogenous or endogenous) and anti-convulsants induce increased ALP production by canine hepatocytes.
- Increased serum ALP activity induced by corticosteroids varies depending on dose and duration of exposure, but can be marked (>20×). Anticonvulsants generally cause somewhat milder increases (<10×).

Cholestasis

- Neonates of several species have high serum ALP activity following ingestion of colostrum.
  - During the first few days of life puppies, kittens, and lambs have transient marked increases in serum ALP activity (up to or >30× for adult animals).
- Hyperadrenocorticism has already been discussed as a cause of often marked corticosteroid-induced ALP activity increases in dogs.
- Diabetes mellitus, canine hypothyroidism and hyperparathyroidism, and feline hyperthyroidism result in increased ALP activity.
- Neoplasia may be associated with increased serum ALP activity (Hepatic, mammary gland, bone).

Increased Serum ALP activity in dogs



Source: Thrall et al., 2012

Cholestasis

- **GGT**
  - It is considered an induced enzyme.
  - Most body tissues synthesize GGT, with the highest concentrations occurring in the pancreas and kidney. **Most of the serum GGT activity originates in the liver.**
    - Release from renal epithelial cells results in increased urinary GGT activity, but not increased serum GGT activity. Similarly, pancreatic cells release GGT into pancreatic ducts rather than into the blood.
  - Increased GGT production, release, and subsequent increased serum GGT activity occur with cholestasis and biliary hyperplasia.
  - In dogs, increased GGT activity also occurs as a result of drug induction.
    - Dogs may show up to 50 fold GGT increase on bile duct obstruction. The cat's up to 16 fold.

Cholestasis

- It should be evaluated with ALP in cats and dogs.
  - E.g. In cats with hepatic lipidosis, ALP increase is more significant than GGT. However, if the root cause of disease is necroinflammatory, GGT activity may be higher than ALP.
- Similar to ALP, increases in serum GGT activity are seen in dogs receiving corticosteroids. Its activity increases more slowly.
- Anticonvulsants may also cause an increase in GGT (2-3x). If this increase is much higher, it can be considered to be due to cholestasis rather than drug.
  - Significant increase in drug-induced adverse may be a sign drug-associated toxic hepatopathy that it can be life-threatening.

Cholestasis

- In horses and cattle, GGT is generally considered more sensitive than ALP for detection of cholestasis. Generally higher than ALP.
- Biliary hyperplasia and liver failure develop due to alkaloid poisoning in horses and cattle. In this case, the GGT also increases considerably. However, in chronic cases ALP may increase higher than GGT.
- Cattle with moderate to severe hepatic lipidosis have only mild increases in serum GGT activity.
- Since there is high GGT activity in the colostrum of dogs, sheep and cattle, serum GGT levels in newborns may increase up to 50 fold.
  - After an average of 5 weeks, they begin to fall to normal levels.
  - **It can be used to evaluate passive immunity.**

Serum GGT Activity in Animals with Hepatobiliary Disorders

Species	Disorders
Dog	Bile duct obstruction, chronic hepatitis, lipidosis, necrosis, cirrhosis, neoplasia, corticoid therapy
Cat	Bile duct obstruction, cholangiohepatitis, cirrhosis, lymphosarcoma, necrosis
Horse	Toxic hepatic failure, subclinical hepatopathy, hyperlipidemia
Cattle	Fascioliasis, lipidosis, Metacercariae migrations, Senecio poisoning
Sheep	Bile duct obstruction, toxicity, fascioliasis,Lupinosis, Cobalt deficiency, Ketosis

Source: Kaneko et al., 2008

Your Questions?

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Next Topic;

Hepatic Functions and Laboratory Assessment-II

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