ULTRASONOGRAPHIC EVALUATION OF CANINE HEPATIC DISORDERS

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DECLARATION

I hereby declare that the thesis entitled **"ULTRASONOGRAPHIC EVALUATION OF CANINE HEPATIC DISORDERS"** is a bonafide record of research work done by me during the course of research and that this thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

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CERTIFICATE

Certified that this thesis, entitled **"ULTRASONOGRAPHIC EVALUATION OF CANINE HEPATIC DISORDERS"** is a record of research work done independently by **Dr. Renjith. R** under my guidance and supervision and that it has not previously formed the basis for the award of any degree, diploma, associateship or fellowship to him.

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CONTENTS

LIST OF ILLUSTRATIONS

List of Plates

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Renjith, R.

Introduction

1. INTRODUCTION

Liver is the largest parenchymatous organ in the body comprising almost 3.4 per cent of body weight in adult animals. Liver performs a wide range of functions including filtration and storage of blood, metabolism of carbohydrates, proteins, fats, hormones and foreign chemicals, storage of iron and vitamins and formation of bile, plasma proteins and coagulation factors.

Liver is positioned immediately caudal to the diaphragm and lies entirely within the intrathoracic portion of the abdominal cavity covered by bony thorax. Liver in dogs is composed of six lobes, being right lateral, right medial, quadrate lobe, left medial, left lateral and caudate lobe. Another anatomical peculiarity of liver is that it is supplied by both arterial and venous system.

Diagnosis of liver disease has always been a challenging task for veterinarians. Hepatic diseases occur quite frequently in dogs. Important causes reported for hepatic injury were infectious agents like Canine adenovirus, Leptospires, Mycobacterium, drugs, heavy metals, toxins and inherited defect in copper excretion leading to excess copper accumulation in liver. Other causes included diseases of other organs and cholestasis (Center, 1999).

A spectrum of clinical signs is associated with liver diseases. Liver is the central organ of metabolic and detoxifying pathways. Therefore, a failing liver would cause dysfunction of other organs. Diseases of other organs would secondarily affect liver. Non-specific clinical signs recorded were depression, anorexia, weight loss, vomiting, diarrhoea, polyuria and polydypsia. Clinical signs suggestive of liver disease were jaundice, ascites, alteration in liver size, drug intolerance, coagulopathies and hepatic encephalopathy (Rutgers, 1996).

Most of the hepatic disorders were not diagnosed or were presented with clinical symptoms at a terminal stage. Early diagnosis and treatment of liver diseases was important for a favourable prognosis (Voros *et al.,* 1991).

In addition to routine clinical examination, other diagnostic procedures like diagnostic ultrasound and ultrasound-guided biopsy could be used for the early identification of hepatic diseases. Ultrasound offers a safe, non-invasive method to detect many liver diseases. Ultrasonographic changes were classified into parenchymal and vascular changes. Ultrasound provided information on the gross changes noticeable in liver. Ultrasound might suggest the disease process, but cannot provide a histological diagnosis (Nyland, 1984). Liver biopsy was required for a confirmatory diagnosis of the changes that were detected on ultrasound examination. Ultrasound guided biopsy can safely and effectively furnish diagnostic cytological material from focal changes (Nyland, 1984; Barr, 1995).

Abnormal liver function tests would also point towards an underlying liver disease, which could be used in conjunction with ultrasonography.

Considering these views, the present study on "Ultrasonographic evaluation of canine hepatic disorders" was taken up for:

1. Detection of changes in diseased liver with ultrasound scans and classification of liver diseases.
2. Confirmation of tentative diagnosis with the help of ultrasound guided biopsy (in selected cases).
3. Correlation between ultrasound image, gross and histopathological changes in liver.

Review of Literature

2. REVIEW OF LITERATURE

2.1 GROSS ANATOMY OF LIVER

The falciform ligament is a delicate membrane, which leaves the liver between the left medial and quadrate lobe to the diaphragm. The free border of falciform ligament has the round ligament, which was the vestige of the umbilical vein (Schummer and Nickel, 1979).

Cartee and Robert (1981) opined that knowledge of hepatic structures was essential for proper interpretation of ultrasonographic patterns.

Carlisle *et al.* (1995) studied the anatomy of portal and hepatic veins for systematic ultrasonographic evaluation of canine liver and showed that cranial end of liver began at the sixth intercostal space and extended caudally to the 11^{th} intercostal space. Gall bladder could be seen in the right ventrolateral approach at seventh intercostal space.

Liver is positioned immediately caudal to the diaphragm and laid entirely within the intrathoracic portion of the abdominal cavity covered by bony thorax. Liver is composed of six lobes that are deeply separated from each other, being right lateral, right medial, quadrate lobe, left medial, left lateral and caudate lobe which bear the renal impression of right kidney along with right lateral lobe (Smith, 1999).

2.2 ETIOLOGY

2.2.1 Infectious Causes

Infectious causes of liver disease included Canine Adenovirus-1 and Leptospires (Rutgers and Haywood, 1988; Johnson, 1995; Rutgers, 1996; Boisclair *et al.*, 2001).

Canine Adeno virus 1 (CAV 1) produced cytotoxic effects in liver, which included widespread centrilobular to panlobular hepatic necrosis in acute cases. But chronic latent hepatic infection could develop in dogs with moderate level of immunity resulting in chronic active hepatitis, hepatic fibrosis and persistent inflammation (Greene, 1990).

Hepatotrophic virus like Canine Adenovirus 1(CAV-1) and Corona virus, bacteria like Leptospira sp, Mycobacterium sp, enteric organisms, Nocardia sp, fungal, parasitic and protozoal infection could cause hepatic infection (Center, 1999).

2.2.2 Drugs and Toxins

Potential causes for liver insult included long-term use of anticonvulsant drugs like Primidone alone or in combination with Phenytoin (Rutgers and Haywood, 1988; Andersson and Sevelius, 1991; Rutgers, 1996).

Toxins that could cause acute hepatic insult were dimethylnitrosamine (Boothe *et al.*, 1992) heavy metals like copper, iron, lead, mercury, chlorinated biphenyls, naphthalenes (Rutgers, 1996) carbon tetrachloride (Voros *et al.*, 1997), tetrachloroethylene (Kim Young Bum *et al.*, 1999) and pyrrolidizine alkaloids (Rutgers, 2000).

Griseofulvin, Tetracycline and heavy metals like copper, lead (Johnson, 1995) iron and mercury could cause acute liver disease (Rutgers, 1996).

Johnson (1995) and Center (1995 and 1999) found that aflatoxin and other mycotoxins would cause hepatic injury in humans, dogs and cats while Rutgers (1996) reported that in addition to this algatoxin (blue-green algae) and bacterial endotoxin were associated with liver damage.

Liver injury could be associated with the use of drugs like Acetaminophen, Cimetidine, Diazepam, Halothane, Mebendazole, Oxibendazole, Phenytoin, Phenobarbital, Primidone, Sulphonamides and Thiacetarmide (Center, 1999) due to hepatocellular injury following metabolic activation by cytochrome p450 enzyme system either through direct damage to plasma membrane or through immune reactivity.

Ingestion of toxic plants like Amanita mushrooms, cyad palm tree, fruits of chinaberry tree and sting from Hymenopteran insects could cause acute hepatic failure (Bunch, 2000).

2.2.3 Hereditary Causes

Rutgers and Haywood (1988); Rutgers (1996); Center (1999); Johnson (2000) and Boisclair *et al.* (2001) described chronic hepatitis in certain breeds like Bedlington terriers, West highland terrier, Dobermann pinschers and Skye terriers with accumulation of excess copper due to inherited defect in copper transport or excretion or excessive copper intake.

Excessive accumulation of iron in hepatocytes due to an inherited defect causing increased alimentary uptake of iron could lead to chronic liver disease in humans. Hepatocyte damage was related to free radical generated oxidant injury to cell membrane (Center, 1999).

2.2.4 Secondary Liver Diseases

Rutgers (1996) reported that central role of liver in metabolism made it vulnerable to diseases of other systems like acute pancreatitis, chronic or acute small intestinal diseases, extra hepatic bacterial infection, shock, anaemia and congestive heart failure.

Center (1999) observed that secondary hepatic injury occurred subsequent to systemic infections, disseminated neoplasm or pathological changes in other organs like cardiovascular, gastro-intestinal, uro-genital and endocrine systems.

2.2.5 Other Causes

Liver failure subsequent to autoimmune haemolytic anaemia was reported by Okin (1984) while Center (1999) discussed immune mediated hepatocyte damage due to cytokines and complement activation especially during endotoxaemia and oxidative damage, while immunological reaction against liver specific antigens were attributed to liver injury in chronic hepatitis.

Injection of serum or liver homogenates from dogs with naturally occurring chronic liver disease produced canine acidophil hepatitis manifested as acute hepatitis or chronic hepatitis or cirrhosis (Jarrett and O'Neill, 1985; Rutgers and Haywood, 1988) while Rutgers (1996) and Boisclair *et al.* (2001) postulated a viral cause for canine acidophil cell hepatitis.

Rutgers (1996) and Center (1999) opined that alpha$_1$-antitrypsin deficiency in liver led to hepatic cirrhosis particularly in Cocker spaniels.

Chronic liver injury could occur secondary to intra-hepatic or extra-hepatic cholestasis (Center, 1999). Intrahepatic cholestasis could be due to drugs, toxins or steroid hormones and endotoxins which caused accumulation of poorly water soluble lithocholic acid. This resulted in metabolic and structural damage to hepatocytes. Extra-hepatic cholestasis was due to mechanical obstruction to bile flow.

2.3 SIGNALMENT

2.3.1 Incidence

Andersson and Sevelius (1991) noted a higher incidence of chronic liver disease and cirrhosis in Cocker spaniels and Labrador retrievers, while West highland white terriers and Dobermann pinschers had copper associated hepatitis.

Farrar *et al.* (1996) reported 14 cases of hepatic abscesses over a study period of 12 years and found that overall prevalence of hepatic abscesses for the period was 0.56/10000 cases while Schwarz *et al.* (1998) observed 13 cases of hepatic abscesses over a period of 8 years.

2.3.2 Age

Rutgers and Haywood (1988) reported that the average age of presentation of idiopathic Chronic Active Hepatitis (CAH) was four to seven years.

Andersson and Sevelius (1991) reported the average age at which liver disease was diagnosed in Cocker spaniels was five years while that in Labrador retrievers and West highland terriers was 6.9 and 4.8 years respectively.

Mean age of presentation of dogs with portosystemic shunts ranged from 2 to 84 months (Mean, 17 months) (Holt *et al.*, 1995).

The average age of dogs presented with hepatic abscesses ranged from 8 to 15 years with a mean of 10.6 years (Farrar *et al.*, 1996) and 4 to 16 years with a mean of 11.5 years (Schwarz *et al.*, 1998).

Lamb *et al.* (1996) observed mean age of incidence of congenital portosystemic shunts (PSS) in dogs was 1.7 (0.2 to 10) years.

Lucena *et al.* (2001) reported hepatic cirrhosis in a five month old dog.

2.3.3 Sex

Rutgers and Haywood (1988) reported higher incidence of chronic active hepatitis in females.

Incidence of liver disease in Cocker spaniels was higher in males, while in Labrador retrievers it was higher in females (Andersson and Sevelius, 1991).

2.3.4 Breed

Rutgers and Haywood (1988); Rutgers (1996); Center (1999); Johnson (2000); Boisclair *et al.* (2001) described chronic hepatitis in certain breeds like Bedlington terriers, West highland terrier, Dobermann pinschers and Skye terriers with accumulation of excess of copper due to inherited defect in copper transport or excretion.

Cocker spaniels and Labrador retrievers had a higher incidence of chronic liver disease and cirrhosis, while copper associated hepatitis was reported in West highland white terriers and Dobermann pinschers (Andersson and Sevelius, 1991).

2.4 CLINICAL SIGNS

Clinical signs associated with liver disease in canines included depression, weakness, anorexia, polydypsia, polyuria, jaundice, vomiting, ascites, weight loss, hepatic coma and hepatomegaly (Strombeck and Gribble, 1978). One two-year-old bitch was anoestrus for 18 months after contracting liver disease.

Canine superficial necrolytic dermatitis or hepatocutaneous syndrome manifested as ulcerative, erythematous lesions in footpads, mucocutaneous junction, genital area and at pressure points (Nyland, 1996).

Non-specific clinical signs of liver disease in dogs were depression, anorexia, weight loss, vomiting/diarrhoea, polyuria/polydypsia and pigmented urine. Specific clinical signs were jaundice, ascites, hepatic encephalopathy, hepatomegaly or microhepatica, drug intolerance and coagulopathy (Rutgers, 1996). Rothuizen and Meyer (2000) reported that splenomegaly and pale mucous membrane might also point towards an underlying liver disease.

Hoque and Varshney (2001) observed vomiting, jaundice, hepatomegaly, distended abdomen, bilateral hind limb edema, anorexia, melaena, constipation and signs of encephalopathy in their study on canine hepatopathies.

History and clinical signs suggesting hepatopathies in 24 cases included nausea / vomiting, jaundice, mild anaemia, evidence of peritoneal fluid accumulation, abdominal distension, constipation, diarrhoea, head pressing, convulsion, hypersalivation, muscular tremor, apparent blindness, melaena (Varshney and Hoque, 2002). Nonspecific signs included chronic anorexia, weakness, emaciation and depression.

2.4.1 Alteration in Liver Size

Differential diagnosis of hepatomegaly included inflammatory disease, venous congestion, diffuse infiltrative diseases like lymphosarcoma and fatty infiltration (Nyland, 1984), hepatic abscess (Farrar *et al.*, 1996) glycogen accumulation in Cushings disease and caudal venacaval obstruction proximal to liver (Rothuizen and Meyer, 2000).

Portal venous anomalies caused decreased hepatic blood flow, thus reducing hepatotrophic factors such as insulin, glucagon and nutrients resulting in hepatic atrophy (Johnson, 1995).

Size of liver might be normal to small in cirrhosis (Nyland *et al.*, 1995).

Sen *et al.* (2001) reported that polyphagia and hepatomegaly were the most prominent clinical findings in steroid induced hepatopathy while hepatomegaly was the non- specific clinical manifestation of hepatic lymphosarcoma in canines (Lamb *et al.*, 1991).

2.4.2 Hepatic Lymphosarcoma

Fever, severe depression, vomiting, anorexia, weight loss and hepatomegaly were the clinical signs associated with canine hepatic lymphosarcoma (Nyland, 1984).

Lamb *et al.* (1991) reported that dogs with histologically confirmed lymphosarcoma had a variety of non-specific clinical signs. Most common signs were weight loss, anorexia, lethargy or depression, hepatomegaly and lymphadenopathy.

2.4.3 Chronic Hepatitis

Rutgers and Haywood (1988) observed that clinical signs in idiopathic chronic active hepatitis were vague gastro-intestinal signs along with ascites and portal hypertension. In early stages of chronic hepatitis, clinical signs were vague and asymptomatic like depression, anorexia, weight loss, vomiting and diarrhoea while icterus, ascites, polyuria, polydypsia and neurological signs of hepatic encephalopathy were the major clinical manifestations in advanced liver disease (Rutgers and Haywood, 1988; Johnson, 2000).

Center (1995) and Varshney and Hoque (2002) observed jaundice and melaena in chronic active hepatitis.

Sevelius (1995) reported cirrhosis as the end stage of chronic hepatitis, leading to liver failure with jaundice, ascites and hepatic encephalopathy.

2.4.4 Acute Liver Disease

Abnormal physical findings in early phase of Infectious Canine Hepatitis (ICH) were increased rectal temperature, transient or biphasic fever, tonsillar enlargement, subcutaneous edema of head, neck and dependant parts, hepatomegaly and petechial and ecchymotic haemorrhages. Icterus was uncommon in acute ICH (Greene, 1990).

Acute hepatic injury referred to illness of generally less than or equal to two weeks duration with no previous evidence of hepatobiliary disease (Bunch, 2000).

2.4.5 Hepatotoxicosis

Boothe *et al.* (1992) reported clinical signs indicative of liver disease in experimentally induced hepatotoxicosis in relative order of appearance were fever, weight loss, ascites, icterus, vomiting, gastro-intestinal hemorrhage and hepatic encephalopathy. The author also observed icterus, fever, weight loss, ascites, anaemia and pale mucous membrane and gastro-intestinal bleeding (tarry faeces) on physical examination of dogs with experimentally induced hepatotoxicosis.

2.4.6 Portosystemic Shunts

Hunt *et al.* (1993) opined that liver disease could clinically manifest as ascites and development of portosystemic shunts due to sustained portal hypertension.

Clinical signs in dogs with portosystemic shunts included stunted growth, weight loss, polyuria and polydypsia, vomiting, inappetance and neurologic signs like bizarre behaviour, ataxia, seizures and signs of depression (Lamb *et al.*, 1996).

2.4.7 Hepatic Abscess

Physical examination of 14 dogs with hepatic abscesses showed fever, dehydration, signs of abdominal pain, hepatomegaly and signs of bleeding-epistaxis, ecchymoses or haematochezia (Farrar *et al.*, 1996).

Clinical signs associated with hepatic abscess included lethargy, vomiting, anorexia, diarrhoea, trembling, polyuria and polydypsia(Schwarz *et al.*, 1998).

2.4.8 Cirrhosis

Ascites developed due to portal hypertension, hypoalbuminemia or hormonal changes associated with hepatic dysfunction causing excessive sodium retention (Hunt *et al.*, 1993).

Clinical signs of hepatic cirrhosis were initially vague and nonspecific. Anorexia, lethargy, depression, vomiting, diarrhoea, polyuria and polydypsia were noticed. Signs of overt liver disease were ascites, jaundice and hepatic encephalopathy. Ascites was the most consistent finding in dogs with cirrhosis, while icterus was a less common clinical sign (Johnson, 2000).

Lucena *et al.* (2001) reported a case of hepatic cirrhosis in a five month old male pure-bred spanish mastiff which had a week old history of anorexia, constipation and melaena. Physical examination of the pup showed ascites, peripheral edema and pale icteric mucous membrane.

Clinical examination of dogs with ascites revealed generalized edema with pendulous abdomen, hollow flank with prominent spines and enlarged abdomen filled with fluid. Fluid thrill was noticed on percussion and body temperature ranged from 38.6 to 40.6° C (Shukla and Sisodia, 2001).

Melaena and, or, haematemesis consequent to gastric ulceration were observed in cirrhosis, and ascites was common clinical finding. Hepatic encepahalopathy was noticed only in two dogs with cirrhosis (Sevelius, 1995).

2.4.9 Hepatic Encephalopathy

Carmichael *et al.* (1996) reported cases of hepato-cerebellar degeneration in Bernese mountain dogs where clinical signs varied from mild ataxia to truncal ataxia, stumbling, incoordination, head tremors, nystagmus and falling sideways or backward. Neurological examination revealed slight proprioceptive deficits in forelimbs and hindlimbs; cranial and spinal reflexes were normal but patellar reflex was exaggerated, hypermetria and hypotonia of hindlimbs.

Clinical signs of hepatic encephalopathy included mild signs like lethargy, inappetence, vomiting, behavioural changes, ataxia, weakness to severe signs like amaurosis, head pressing, pacing, seizures and coma (Rutgers, 1996).

2.5 DIAGNOSIS

2.5.1 Haematology

Normal leucocyte count in dogs was 6000 to 17,000 per micro litre. Normal differential count in dog was neutrophils 3000 to 11,500, band cells 0 to 300, lymphocytes 1000-4,800, monocytes 150 to 1350 and eosinophils 100 to 1250 per micro litre (Meinkoth and Clinkenbeard, 2000).

Leucocytosis with neutrophilia was the most common non-specific finding in liver disease (Voros *et al.*, 1991).

Sevelius (1995) reported that total leucocyte counts would be higher in dogs with chronic progressive hepatitis and cirrhosis.

Hepato-cerebellar degeneration in dogs were characterised by leucocytosis with normocytic, normochromic anaemia (Carmichael *et al.*, 1996).

Farrar *et al.* (1996) found that in case of hepatic abscess, the important hematological abnormities were leucocytosis, neutrophilia with shift to left, mild monocytosis, mild lymphopenia, mild to marked thrombocytopenia and slight anaemia.

The most common complete blood count (CBC) abnormalities observed by Schwarz *et al.* (1998) in dogs with hepatic abscesses were increased white blood cell count with neutrophilia, left shift and thrombocytopenia.

In case of cirrhosis in a five month old pup Lucena *et al.* (2001) observed neutrophilic leucocytosis (36.2 X 10^3 leucocytes per ml.).

2.5.2 Ascitic Fluid

Ascitic fluid in liver disease was characterized as transudate and contained less than 1.0 g protein /dl. (Boothe *et al.*, 1992).

Presence of pure abdominal transudate (low protein ascites) in the absence of severe hypoalbuminemia suggested pre-sinusoidal portal hypertension. (Hunt *et al.*, 1993).

Spaulding (1993b) reported the biochemical and pathological differences between transudate, modified transudate and exudate and different conditions leading to the formation of these. Transduates were fluids with protein concentration of less than 2.5g/dl and low nucleated cell count (less than 1000cells/µl) while modified transudates contained protein up to 3.5 g/dl and cell count up to 5000cells/µl. Exudate contained a high protein concentration of greater than 3g/dl and nucleated cell count greater than 5000 cells/µl.

Ascitic fluid with a high protein content of 3.9g/litre identified as modified transudate was reported in a five month old Rottweiler dog, caused by post-sinusoidal hypertension due to obstruction of blood flow in the caudal venacava. (Smith, 1994)

Lucena *et al.* (2001) reported a case of cirrhosis in a five month old pup where abdominal paracentesis retrieved fluid consistent with a transudate.

2.5.3 Serum Biochemistry

2.5.3.1 Total Protein and Albumin

Kaneko *et al.* (1997) reported that normal level of total protein in canine serum was 61 ± 5.2 g/L, albumin 29.1 ± 1.9 g/L, globulin 34.0 ± 5.1 g/L and A:G ratio was 0.83 ± 0.16.

Strombeck and Gribble (1978) reported slight hypoalbuminemia (mean 2.5g/dl) and hypergammaglobulinemia (mean 3.9 g/dl.) in dogs with chronic active hepatitis.

Boothe *et al.* (1992) noticed low serum protein and low albumin concentration by 14.7 ± 11.5 weeks and 16.8 ± 10.8 weeks respectively in canines, following a hepatic insult.

Sevelius (1995) observed that mean serum albumin concentration in dogs with cirrhosis was significantly decreased (18.08 ± 6.40 g/litre) when compared to other liver diseases. Sensitivity of albumin to detect cirrhosis was 96 per cent while for chronic nonspecific hepatitis it was 16.7 per cent. Hypoalbuminemia was an important marker of chronic inflammatory liver disease.

In a retrospective study on hepatic abscesses in dogs, hypo-albuminemia and hypoproteinemia was detected (Farrar *et al.*, 1996).

2.5.3.2 Bilirubin

Kaneko *et al.* (1997) reported that normal level of total serum bilirubin in canines was 0.2 ± 0.1 mg/dl (0.1 to 0.5 mg/dl) while that of conjugated and unconjugated bilirubin were 0.06-0.12 and 0.01-0.49, mg/dl respectively.

Increased serum levels of unconjugated bilirubin, ALT and ALP were observed in intra and extra-hepatic cholestasis, whereas with hepatocellular damage, serum levels of conjugated, unconjugated bilirubin and ALT were increased, while ALP was not significantly elevated (Milne, 1985).

Serum bilirubin peaked six to eight days following liver damage in leptospirosis (Greene *et al.*, 1990)

High bilirubin concentration was observed by 25.1 ± 11.4 weeks, following liver damage (Boothe *et al.*, 1992).

Bilirubin levels were always normal in the subclinical stage of liver disease (Speeti *et al.*, 1996) while hyperbilirubinemia was seen in hepatic abscess (Schwarz *et al.*, 1998; Farrar *et al.*, 1996).

Speeti *et al.* 1996 studied subclinical and clinical hepatitis in Dobermann and found that mean bilirubin levels were significantly higher ranged from 9.4 to 163.9 mmol/litre in dogs with clinical hepatitis.

2.5.3.3 Liver Enzymes

Milne (1985) observed that a combination of serum ALP and ALT measurements could accurately diagnose 80-100 per cent of cases of fatty change, hepatic neoplasia, hepatoma and cirrhosis in dogs.

Jones (1988) reviewed various enzymes used for diagnosis in veterinary medicine and their clinical significance and normal values in dog, cat, horse, cattle and sheep.

Increased serum concentration of Alanine amino transferase (ALT), Aspartate amino transferase (AST), Alkaline phosphatase (ALP) and bilirubin were seen in liver damage following leptospirosis (Greene *et al.*, 1990).

Chronic persistant hepatitis and chronic active hepatitis (CAH) were characterised by elevation in levels of serum aminotransferase and alkaline phosphatase. In cirrhotic patients the values of liver enzymes might be normal despite severe liver injury (Andersson and Sevelius, 1991).

Twelve out of fifteen cases of hepatic lymphosarcoma in dogs were characterised by elevation in serum enzymes and icterus (Lamb *et al.*, 1991).

An increased serum ALT and ALP activity with few or no clinical signs suggested some underlying liver disorder (Cornelius, 1997).

Bromel *et al.* (1998) observed no change in serum levels of ALT, AST and ALP in association with gall bladder sludge.

2.5.3.3.1 Alkaline Phosphatase (ALP)

Normal level of Alkaline phosphatase (ALP) in canines was 66 ± 36 U/L (Kaneko *et al.*, 1997).

Strombeck and Gribble (1978) found that 100 per cent of dogs with chronic active hepatitis had elevated serum Alkaline phosphatase levels almost five times the normal (Mean 695 U/L, range 170-2000 U/L).

Elevated levels of ALP was seen in dogs with liver disease subsequent to haemolytic anaemia (Okin, 1984) and hepatic abscess (Farrar *et al.*, 1996) and in hepatic abscess, level of ALP ranged from 275 – 5485 U/L (Schwarz *et al.*, 1998).

Alkaline phosphatase activity was found in small intestinal mucosa, kidney, pancreas, colon, testicle, muscle, brain, spleen, lung and liver and higher levels were seen in young dogs. Half-life of liver AP was found to be approximately three days. Acute hepatocellular damage would not produce much change in serum ALP, while in congenital porto-systemic vascular shunts, serum ALP was occasionally elevated. (Milne, 1985).

Increased ALP activity was observed in endogenous and exogenous glucocorticoid administration (Milne, 1985; Center *et al.*, 1992) and steroid induced hepatopathy (Sen *et al.*, 2001). Enzyme activity declined slowly over two or three months after discontinuation of glucocorticoid treatment.

Concurrent increase in serum ALP and bilirubin levels were seen in dogs with intrahepatic cholestasis, due to inflammatory infiltrates and fibrosis (Rutgers and Haywood, 1988).

Increase in serum ALP activity was often proportionally greater than that of ALT activity in liver damage following leptospirosis (Greene *et al.*, 1990).

High ALP activity was noticed by Boothe *et al.* (1992) 15.2 ± 10.6 weeks following hepatic injury.

For healthy dogs, maximal activity of GGT considered to be normal was 10 IU/L and that for ALP was 107 IU/L. Specificity of GGT (87per cent) exceeded that of ALP (51per cent) while ALP was more sensitive than GGT in detecting hepatic diseases (Center *et al.*, 1992).

Solter *et al.* (1994) found that hepatic activities of liver ALP (LALP) and GGT was increased in prednisone treated dogs and in dogs with bile duct obstruction.

Mean ALP concentration was significantly higher (19.74 ± 18.62 µkat/litre) in chronic cholangiohepatitis when compared to other chronic liver diseases. A high level of ALP in cirrhosis (3.42 ± 2.70 µkat/litre) was due to cholestasis secondary to intrahepatic biliary obstruction (Sevelius, 1995).

ALP levels increased only after the development of clinical symptoms when inflammation had spread to bile duct. Dobermann with clinical hepatitis had significantly higher levels of serum ALP (1751 ± 1061 U/L) when compared to subclinical hepatitis (797 ± 626 U/L) (Speeti *et al.*, 1996).

Cornelius (1997) reported that glucocorticoid administration caused increase in both LALP (liver ALP) and CALP (corticosteroid induced ALP) while pathologic conditions that caused increase in serum ALP levels were intrahepatic or extra hepatic cholestasis, liver disease and increased osteoblastic activity.

Solter and Hoffmann (1999) showed that increased solubilisation of liver ALP (LALP) during cholestasis was due to increased cleavage of its membrane anchor by endogenous glycosylphosphatidylinositol phospholipase C (GPI-PLD) activity, which was enhanced by increased concentration of hepatic bile acids during cholestasis.

Serum ALP and bilirubin levels were increased following cholestasis secondary to intrahepatic biliary obstruction (Lucena *et al.*, 2001).

2.5.3.3.2 Alanine Amino Transferase (ALT)

Normal level of ALT in serum of carnivores was 47 ± 26 U/L (Kaneko *et al.*, 1997).

Strombeck and Gribble (1978) observed that in chronic active hepatitis, mean elevation of ALT was 819 U /L with a range 50-2,238U/L.

Elevated levels of ALT was seen in a dogs associated with liver disease subsequent to haemolytic anaemia (Okin, 1984).

Milne (1985) reported that acute hepatocellular damage would result in high serum ALT activity.

Persistently elevated level of serum Alanine aminotransferase (ALT) was observed in chronic hepatitis, (Rutgers and Haywood, 1988) hepatocellular degeneration or necrosis (Center *et al.*, 1992) and hepatic abscess (Farrar *et al.*, 1996).

Boothe *et al.* (1992) described the temporal change in clinical laboratory test results following hepatic injury. High serum AST and ALT values were observed by 6.2 ± 5.6 weeks and 6.4 ± 6.2 weeks respectively.

Hepatic ALT activity did not increase following prednisone treatment (Solter *et al.*, 1994) and steroid induced hepatopathy (Sen *et al.*, 2001).

In terminal stages of liver disease, ALT level was decreased as very few hepatocytes were left to secrete intracellular enzymes (Centre, 1995; Speeti *et al.*, 1996).

Sevelius (1995) in a retrospective study on chronic hepatitis reported that sensitivity of ALT for detection of liver disease was fairly high and ranged from 84.6 to 68.2 per cent. In dogs with chronic cholangiohepatitis mean ALT concentration was significantly higher (19.74 ± 18.62 µkat/litre) than in dogs with other chronic liver diseases like chronic nonspecific hepatitis (7.24 ± 4.09 µkat/litre), cirrhosis (3.42 ± 2.70 µkat/litre) and chronic progressive hepatitis (2.99 ± 4.25 µkat/litre).

Specti *et al.* (1996) observed that ALT could be used for screening liver diseases prior to liver biopsy, but was of little use in determining the prognosis of liver diseases. They also reported that there was no significant difference in ALT levels between dogs with subclinical versus clinical Dobermann hepatitis.

2.5.4 Ultrasonography

2.5.4.1 Principle

Cartee and Robert (1981); Lamb (1990) and Cartee *et al.* (1993) explained the principles of ultrasound production and transmission through tissues. Barr (1988); Park *et al.* (1981) and Rantanen and Ewing (1981) explained various modes of display, equipment and principles of image interpretation.

When viewing ultrasound images, the scanning surface or near field should be uppermost. Longitudinal, sagittal or parasagittal scan should be oriented with the cranial aspect of the animal to the viewer's left and caudal aspect to the viewer's right. When viewing transverse scans, the left and right sides should be oriented as though the viewer is observing the animal from the caudal aspect (Park *et al.*, 1981). He also suggested that while evaluating a possible lesion ultrasonographically, the internal echo pattern, borders and adjacent echo patterns should be observed. Internal echo patterns are anechoic, hypoechoic, echoic and complex. Margins of lesion may be well defined or irregular and ill defined.

2.5.4.2 Indications

Indications of hepatic ultrasonography were hepatomegaly, abdominal mass or mass in area of liver, identification of possible hepatic metastasis, icterus, unexplained weight loss, fever of unknown origin, pain or gastrointestinal signs, assessing integrity of diaphragm, biopsy guidance, ascites and suspected diaphragmatic hernia (Nyland and Park, 1983; Lamb, 1990).

Center *et al.* (1992) reported that hepatobiliary disease was suspected based on history, hepatomegaly, bilirubinuria, jaundice, abnormal activity of ALT, AST, ALP and hyperbilirubinemia.

Indications of ultrasonography as a primary diagnostic technique include discrimination of cystic and solid mass, exploration of fluid filled body cavities, discrimination of texture of solid mass and biopsy guidance (Cartee *et al.*, 1993).

Center (1995) and Nyland *et al.* (1995) opined that hepatic ultrasonography could be used for identification of mineral densities, detection and confirmation of portosystemic vascular anomaly, assessment of liver in conditions of elevated serum enzymes and evaluation of response to treatment.

2.5.4.3 Limitations

Park *et al.* (1981) found that bone and air had highly reflective interfaces and effective examination with ultrasound could not be done through either bone or air.

Nyland (1984) opined that ultrasound might suggest the disease process, but cannot provide a histopathologic diagnosis.

Godshalk *et al.* (1988) and Lamb (1990) opined that ultrasonography did not appear to be a viable method for estimating hepatic volume in dogs.

Lamb (1990) reported that confirmatory diagnosis of focal and diffuse hepatic lesions in ultrasonography could not be established without histopathological examination.

Lamb *et al.* (1996) opined that diagnosis of PSS with ultrasonography depended on operator's experience and use of high resolution equipment .

Canine liver was much more difficult to scan than the human liver as it was located further cranially under the rib cage and is oriented in an upright position. Poor image quality would be the result in fatty animals due to excess amount of subcutaneous fat giving rise to hyperechoic image between skin and hepatic parenchyma (Bhadwal *et al.*, 1999).

2.5.4.4 Procedure

A 7.5 MHz transducer was best for detailed examination of liver. Transverse oblique positioning of the transducer along the costal arch was the best diagnostic position while lateral intercostal approach was also possible. Transducer was positioned longitudinally in the right hypochondriac and right lateral region for longitudinal imaging of the caudal venacava and portal veins (Cartee and Robert, 1981).

Nyland and Park (1983) reported that transverse scan was done at 1 cm interval by angling the sound beam 25-35° cranially under the sternum and rib cage and longitudinal sector scans were done with the transducer in a horizontal position directed cranially. Optimal image was produced when the transducer face remained perpendicular to the diaphragm during the entire sector sweep. Scanning liver in longitudinal and transverse planes by rotating the transducer face in an arc from the tip of the ventral liver lobes to the caudal extent of the dorsal lobes and also from the right and left to include lateral liver lobes.

Lamb (1990) opined that examination of liver could be done by placing ultrasound probe immediately caudal to the xiphoid and directing cranially. A right intercostal approach was also explained.

Carlisle *et al.* (1995) reported that right lateral and caudate lobes were better imaged in ultrasound through the right intercostal space approach. Gall bladder was

identified slightly to the right and in a cranio-ventral position and right medial lobe was to the right and quadrate on the left.

Jian-Xin WU and Carlisle (1995) described the procedure for ultrasonographic examination of liver in dogs in different planes. Keeping dog in dorsal recumbancy, the transducer was placed immediately caudal to xiphisternum in a longitudinal plane and angled dorsocranially between 30-40° to locate liver and diaphragm. Transducer head was angled to the right to locate gallbladder, and then rotated into a transverse plane. Transverse and sagittal scans through right and left intercostal space commencing from 5th intercostal space and progressing caudally were also explained.

Liver was scanned from a point just caudal to xiphisternum with the sound beam directed cranio-dorsally. Transverse and sagittal scan were taken by rotating the transducer scanhead in an arc fashion on successive scans and areas not accessible from xiphisternum was scanned through 12th intercostal space close to sternal margin. There was difficulty with 7.5 MHz transducer in scanning the farthest part of the liver close to diaphragm in dogs while with 3.5 MHz transducer both liver and diaphragm were easily scanned, but architectural details were better visible with 7.5 MHz than with 3.5 MHz transducer (Bhadwal *et al.*, 1999).

Varshney and Hoque (2002) in their study scanned liver in dorsal, right and left intercostal approach, in sagittal, transverse and dorsal planes.

2.5.4.5 Normal Ultrasonographic Pattern of Liver

2.5.4.5.1 Parenchyma

Normal ultrasonographic appearance of liver in dog was transonic (dark) with multiple echoes from fibrous portions of hepatic architecture (Cartee and Robert, 1981).

Barr (1988) and Nyland *et al.* (1995) opined that normal liver was slightly more echogenic than the renal cortex and slightly less echogenic than the spleen

Lamb (1990) reported that normal hepatic parenchyma had uniform slightly coarse echotexture and was less echogenic than spleen while hyperechoic or isoechoic compared to right kidney, with larger blood vessels and gall bladder appearing anechoic.

Normal liver was having a homogenous echotexture (Cartee *et al.*, 1993) resulting from nonspecular reflectors. This uniform echo pattern was interrupted by portal and hepatic vessels (Nyland *et al.*, 1995).

England (1996) reported that neonatal liver had a similar ultrasonographic appearance to that of adult, although in the first eight weeks, parenchymal echogenic stippling was less coarse.

Liver parenchyma was uniform, homogenous with specked echoes (Bhadwal *et al.*, 1999) and was isoechoic to right renal cortex but hypoechoic than spleen.

2.5.4.5.2 Hepatic Vasculature

Barr (1988) and Cartee *et al.* (1993) observed that the portal vessels had hyperechoic walls while hepatic veins did not have a distinct wall.

Hepatic veins could be distinguished from portal veins because portal veins had a well defined echogenic wall while hepatic veins lack a well defined wall (Lamb, 1990; Jian-Xin WU and Carlisle, 1995).

Carlisle *et al.* (1995) detailed the anatomy of the portal and hepatic veins in the dogs in order to establish a procedure for the systematic evaluation of liver by ultrasonography.

Jian-Xin WU and Carlisle (1995) showed that right medial, quadrate, left medial and lateral hepatic and portal veins could be identified with dog in dorsal recumbancy while right lateral and caudate hepatic veins were seen from the right side with the transducer positioned between the ninth and the eleventh intercostal space. He also reported that left lateral, left medial, quadrate and right medial hepatic veins were observed between seventh to the ninth intercostal space on both sides. Visibility was best at seventh and eighth intecostal space with transducer at one third of the way along the rib dorsal to the sternum. The right lateral and caudal hepatic veins were best seen between the ninth to eleventh intercostal spaces half way along the ribs, cranial to right kidney on the right side using sagittal scan. Main portal vein could be seen from right eighth to eleventh intercostal space half way up the ribs on sagittal scan. Portal hepatis was best seen with the transducer located half way up from the sternum at the eighth intercostal space.

England (1996) reported that in pups, during the first eight weeks, portal veins were not well delineated in ultrasound scanning.

Bhadwal *et al.* (1999) observed caudal venacava as a anechoic circle right of the midline close to diaphragm while portal veins were seen as round structures with anechoic core and echogenic periphery while small hepatic veins in liver parenchyma appeared as anechoic structures.

2.5.4.5.3 Gall Bladder

Gall bladder appeared as pear shaped or oval to round anechoic structure to the right of the mid line and appeared full in all the animals as they were fasted for more than 12 hours (Cartee *et al.*, 1993; Bhadwal *et al.*,1999).

Normal position of gall bladder was in a fossa between the quadrate lobe and the right lateral lobe and normal gallbladder wall sonographically appeared as thin echogenic line and measured about 2-3 mm in thickness (Spaulding, 1993a).

Sometimes normal gall bladder wall was not visible sonographically (Nyland, *et al.*, 1995).

Jian-Xin WU and Carlisle (1995) found that gall bladder was located from the seventh to ninth intercostal space and visibility was not related to thoracic conformation, the amount of fat or fullness of stomach.

Bhadwal *et al.* (1999) observed distal acoustic enhancement of gall bladder was consistent in all the animals.

Hoque and Varshney (2001) examined gall bladder of 35 dogs and reported that normal gall bladder was anechoic with smooth well-defined margin and produced distal acoustic enhancement.

2.5.4.5.4 Ascitic Fluid

Park *et al.* (1981) reported that fluid filled structures have an anechoic ultrasonographic appearance and fluid with this appearance was usually nonviscous.

Spaulding (1993b) reported that anechoic effusions might be either a transudate, modified transudate, while echogenic or septated and homogenously echogenic appearances were typically associated with exudates.

Hoque and Varshney (2001) reported that detection of peritoneal fluid accumulation even in traces was best seen between liver and diaphragm suggesting ascites.

2.5.4.6 Abnormal Liver Sonogram

Voros *et al.* (1991) in his study observed focal alterations in 11 dogs (50 per cent) while 50 per cent (11 cases) showed diffuse ultrasonographic alterations.

Liver parenchymal abnormalities were broadly divided into focal, multifocal or diffuse lesions (Nyland *et al.*, 1995) and intensity as anechoic, hyperechoic and hypoechoic (Nyland *et al.*, 1995; Hoque and Varshney, 2001).

2.5.4.6.1 Focal Lesions

Studies in man indicated that hepatic ultrasonography had a sensitivity of approximately 80 per cent for focal hepatic lesions larger than two centimetre (Lamb, 1990).

Lamb (1990) reported that focal lesions were described according to their number, size and echotexture as anechoic, hypoechoic, hyperechoic or complex.

Hoque and Varshney (2001) studied ultrasonographic appearance of liver in 35 dogs and found focal alteration in two dogs (5.71per cent) and diffuse alterations in 32 dogs (91.43per cent).

2.5.4.6.1.1 Hepatic Abscess

Farrar *et al.* (1996) found ultrasonographic appearance of hepatic abscess was hypoechoic or anechoic hepatic masses in eight cases while one had heteroechoic mass. In four cases, the hepatic lesions became more echogenic from hypoechoic or anechoic to heteroechoic or hyperechoic in follow up abdominal ultrasonography.

Schwarz *et al.* (1998) reported the appearance of hepatic abscess with size ranging from 1 to 11 cm shape varying from round, oval or irregular with irregular internal margins or echogenic rim. Echogenicity of the contents varied from poorly echogenic to anechoic, with far enhancement artifact that was helpful in the diagnosis of hepatic abscess.

2.5.4.6.1.2 Hepatic Neoplasia

Nyland and Park (1983) observed multiple well defined irregularly shaped masses in liver, some of which were hypoechoic while others had an echogenic centre with a hypoechoic rim.

Nyland (1984) reviewed three sonographic patterns of canine hepatic lymphosarcoma. The first pattern reflected a normal hepatic parenchymal appearance or a slight reduction in echogenicity, while second pattern consisted of anechoic or hypoechoic poorly marginated lesion. A third pattern was characterized by multiple round echodense centers and hypoechoic margins or target lesions.

Nyland (1984) and Lamb (1990) reported that lymphosarcoma produced hypoechoic foci in the liver.

Multiple small hypoechoic or hyperechoic nodules within the hepatic parenchyma was characteristic of hepatic metastasis. (Nyland and Hager, 1985; Lamb, 1990)

Barr (1988) observed that hepatic neoplasia whether primary or secondary might cause multiple focal areas of both increased or decreased echogenicity. Diffuse hepatic lymphosarcoma had been associated with a general decrease in echogenicity of liver parenchyma.

Out of the 14 patients with hepatic lymphosarcoma only three had abnormal echotexture with two having diffuse hypoechogenicity and one had focal hypoechoic lesions (Lamb *et al.*, 1991).

2.5.4.6.1.3 Haematoma

Haematomas presented a spectrum of sonographic appearance; initially it appeared cystic, but internal echoes appeared later with clotting of blood or necrosis and size decreased over a period of weeks or months (Nyland and Park, 1983).

van Sonnenberg *et al.* (1983) reported that rounded echogenic foci was seen sonographically after the injection of 0.5 to 2.0 ml of blood into liver and concluded that ultrasound was useful in the diagnosis of haematoma following blunt penetrating trauma.

Nyland *et al.* (1995) observed that haematomas generally had highly irregular and poorly defined margins. The appearance of the internal contents was variable over time. Acute parenchymal haemorrhage was echogenic. Later appeared anechoic or hypoechoic until clot organization occurred when the internal contents became echogenic.

2.5.4.6.1.4 Calcification

Nyland *et al.* (1995) reported that dystrophic calcification of liver parenchyma produced diffuse increase in the echogenicity of liver parenchyma; sometimes shadowing was seen distally if there was enough sound attenuation.

Pai and Bude (2002) opined that extensive hepatic arterial calcification should be considered when intrahepatic linear echogenic regions producing distal acoustic shadowing were found sonographically. Pneumobilia would also produce a similar sonographic appearance.

Hepatic artery calcification in human patients subsequent to chronic renal failure produced a large acoustic shadow, which corresponded to calcified hepatic

artery. It was concluded that calcification of hepatic artery could be identified by its acoustic shadow (Okuda *et al.*, 2003).

2.5.4.6.2 Diffuse Lesion

In a retrospective study of ultrasound images of liver of patients with hepatitis Kurtz *et al.* (1980) found that in acute hepatitis there was accentuated brightness and more extensive demonstration of the portal vein walls and overall decreased echogenicity of the liver parenchyma. In chronic hepatitis, there was decreased brightness and number of portal veins and overall increase in echogenicity of hepatic parenchyma.

Diffuse echoic pattern of an organ might decrease due to infiltrative processes such as edema (Park *et al.*, 1981) while fat infiltration, fibrosis or lymphosarcoma could produce a diffuse increase in echogenicity (Lamb, 1990).

Diffuse hepatic lymphosarcoma had been associated with a general decrease in echogenicity of liver parenchyma. Diffuse fibrous or fatty infiltration (Barr, 1988) cirrhosis, hepatitis, cystic or portal fibrosis might be associated with an overall increase in echogenicity (Nyland and Park, 1983).

Diffuse parenchymal abnormalities were much more difficult to detect as there was no gross disturbance in hepatic architecture, but a rather subtle change in echogenicity (Barr, 1988).

Fat accumulation in the liver could produce a hyperechoic appearance of hepatic parenchyma in ultrasound (Lamb, 1990).

Nyland *et al.* (1996) reported "honey comb" pattern on ultrasonography of liver of five dogs with canine superficial necrolytic dermatitis.

Sen *et al.* (2001) opined that hyperechoic liver with hepatomegaly was the ultrasonographic finding in steroid hepatopathy.

2.5.4.6.2.1 Cirrhosis

Diffuse 'bright' or hyperechoic or small liver could be diagnosed as cirrhosis (Cartee and Robert, 1981; Lamb, 1990; Voros *et al.*, 1991; Hoque and Varshney, 2001).

Barr (1988) observed a mixed pattern of increased and decreased echogenicity in cirrhosis. A similar pattern of solitary echogenic patches was also seen in fatty infiltration of liver in dogs.

Lamb (1990) observed that hepatic cirrhosis produced a characteristic diffuse patchy echogenicity and lobulated surface to the liver, which was easily appreciated when ascites was present.

Lamb *et al.* (1996) observed that the visibility of intrahepatic portal vessels was reduced in 23/38 (68 per cent) dogs with hepatic cirrhosis.

2.5.4.6.3 Hepatic and Portal Vein Abnormality

Distended hepatic vein and caudal venacava was observed in hepatic venous congestion secondary to right sided cardiac failure (Barr, 1988; Lamb, 1990) or obstruction of the venacava between heart and liver. The enlarged hepatic veins were best visualised near diaphragm where they enter the caudal venacava and enlargement of hepatic veins was judged subjectively (Nyland *et al.*, 1995).

Lamb *et al.* (1996) observed that the visibility of intrahepatic portal vessels was reduced in 23/38 (68 per cent) dogs with congenital portosystemic shunts, cholangiohepatitis and hepatic cirrhosis.

2.5.4.6.3.1 Portosystemic Shunts

Portal veins near the porta hepatis became tortuous and enlarged secondary to acquired liver disease or portal hypertension. Patent ductus venosus or extrahepatic shunt might be visible near caudal venacava. Sometimes the actual communication between the hepatic vein and portal system might be difficult to demonstrate (Nyland and Park, 1983) but, a dilated vascular segment might be suggestive of a shunt (Nyland *et al.*, 1995).

In intra-hepatic portosystemic shunts, the liver echotexture was diffuse or patchy hyperechoic with an overall reduction in liver area and shunting vessel was more distended and tortuous (Voros *et al.*, 1991; Hoque and Varshney, 2001).

Holt *et al.* (1995) and Varshney and Hoque (2002) reported that intrahepatic PSS was more common between intrahepatic portion of venacava and portal vein than between portal and hepatic vasculature. The shunting vessels were distended and tortuous.

Extrahepatic portosystemic shunts were identified with an accuracy of 92 per cent in dogs. Ultrasonographic features in dogs with congenital portosystemic shunts included small liver, reduced visibility of intrahepatic portal vessels and anomalous vessel draining into the caudal venacava between the right renal and right hepatic vein using a right intercostal window (Lamb *et al.*, 1996).

2.5.4.6.4 Liver Size

Ultrasonographic assessment of canine liver size was of little value in predicting actual liver weight (Godshalk *et al.*, 1988).

Cartee *et al.* (1993) reported that normal liver size was not established on ultrasonographic examination but usually it should not extend caudally beyond xyphoid and hypochondriac regions and cranially from seventh rib to diaphragm.

Hepatomegaly was assessed subjectively by comparing the caudal position of the organ to the costal arch (Nyland *et al.*, 1995; Bhadwal *et al.*, 1999).

Bhadwal *et al.* (1999) estimated the mean liver size as 7.63 ± 1.48 cm and a regression equation to calculate liver size based on body weight and found that there was marked variation in the normal liver size for any given weight.

2.5.4.6.5 Gall Bladder Sludge

Bromel *et al.* (1998) observed gallbladder sludge as echogenic material in the gall bladder without acoustic shadowing. Gallbaldder sludge in dogs was not associated with hepatobiliary disease and should be considered as an incidental finding.

Gall bladder sludge was observed as mobile hyperechoic sediment within the lumen without shadowing (Spaulding, 1993a; Bhadwal *et al.*, 1999).

Gall bladder sludge was documented as well defined hyperechoic content, which gravitated to the dependent portion of the gall bladder (Hoque and Varshney, 2001).

2.5.4.6.6 Gall Bladder Wall Edema

Acute cholangiohepatitis in dogs produced edema of gall bladder wall that was appreciated sonographically as thickened gall bladder wall with a double rim produced by the inner and outer walls (Nyland and Park, 1983).

Lamb (1990) and Spaulding (1993a) observed gall bladder wall thickening in ultrasound appeared a hypoechoic region between two echogenic lines and this 'halo' was reported in 26 per cent of cases diagnosed as acute cholecystitis. Gall bladder wall thickening was diagnosed when wall thickness was more than 3–3.5 mm (Spaulding, 1993a).

Spaulding (1993a) described causes for gall bladder thickness as hypoproteinemia, right-sided cardiac failure, hepatic dysfunction associated with hypoalbuminemia, hepatitis, ascites, renal disease, venous hypertension, gall bladder tumors and infectious canine hepatitis.

2.5.5 Liver Biopsy

Percutaneous liver biopsy could safely and effectively furnish cytological material for confirmatory diagnosis (Nyland, 1984), treatment and prognosis in liver disease (Hitt *et al.*, 1992; Sevelius, 1995; Rutgers, 1996).

Rutgers and Haywood (1988) stated that liver biopsy was required for definitive diagnosis and biopsies could be obtained percutaneously, by laparoscopy or laparotomy.

Else (1989) reviewed advantages and disadvantages of various needle biopsy techniques and different methods for obtaining liver biopsy like percutaneous blind technique, 'keyhole' technique, laparotomy and laparoscopy.

Indications of liver biopsy were generalized hepatic hypoechogenicity during ultrasound examination, unexplained hepatomegaly, presence of solitary or multiple nodular mass, abnormal serum enzyme activities and hyperbilirubinemia (Day, 2000.)

The site for liver biopsy was an area between the xiphoid cartilage and the angle formed by the left costal arch. Biopsy was taken using a Tru-cut biopsy needle (Nambi *et al.*, 1994).

2.5.5.1 Ultrasound Guided Liver Biopsy

Ultrasound could be used to guide the needle during percutaneous liver biopsy especially when biopsying focal lesions in liver (Nyland and Park, 1983; Rutgers, 1996).

Hager *et al.* (1985) reported the procedure for ultrasound guided liver biopsy and out of sixty-nine hepatic biopsies performed under ultrasound guidance, multiple attempts were required and adequate samples obtained in 94 per cent of hepatic biopsies. Problems encountered during the ultrasound guided liver biopsy procedure were overlying bowel gas obscuring the target organ and poor visualization of the biopsy needle.

Hoppe *et al.* (1986) compared manual, ultrasound guided biopsy technique with an automated method and reported that the automatic method yielded better quality samples.

Lamb (1990) opined that ultrasonography provided an accurate, relatively non-invasive means of guiding liver biopsy and described the procedure for ultrasound guided liver biopsy with Tru-Cut biopsy needle, fine needle aspiration and automated biopsy devices.

Allen and Kramer (1993) reported that improved visualization of needle tip in ultrasound could be obtained with roughened or scored needle surface, side hole or presence of an intraluminal guide wire or stylet.

Free hand approach allowed greater flexibility to compensate for the patient's movement and slight deflections of the biopsy needle, but required more skill (Leveille *et al.*, 1993).

Percutaneous ultrasound guided tissue core biopsy was minimally invasive and cost effective method of obtaining specimens for histological evaluation and bacteriologic culture (Leveille *et al.*, 1993), required no general anesthesia (de Rycke *et al.*, 1999) and complication rate is low (Barr, 1995).

Menard and Papageorges (1995) described the procedure for ultrasound guided fine needle aspiration biopsy. Surgical gloves, surgical drapes and sterile gel were unnecessary as long as contact with puncture site was avoided.

de Rycke *et al.* (1999) described the procedure for tissue core biopsy using needle guide fastened to transducer as well as free hand method.

Out of the total 30 liver biopsies performed 23 (77 per cent) contained liver sample, four (13 per cent) were devoid of tissue, while three remaining samples contained either skeletal muscle, blood or small intestine (de Rycke *et al.*, 1999).

2.5.5.2 Complications

Complications were not encountered with the liver biopsy procedure (Hager *et al.*, 1985).

Hitt *et al.* (1992) reported that possible complications of percutaneous liver biopsy were haemorrhage, laceration of hepatic parenchyma, bile peritonitis, septic contamination and perforation of other abdominal or thoracic organs.

Leveille *et al.* (1993) observed that out of 246 cases three animals (1.2 per cent) had major post biopsy complications. Mild haemorrhage and formation of haematoma at biopsy site were minor complications.

Puncturing of aorta and gall bladder were reported as complication of liver biopsy. (Nambi *et al.*, 1994).

de Rycke *et al.* (1999) observed that complications following ultrasound guided biopsy in dogs included haemorrhage or haematoma at biopsy site, local or generalized peritonitis, tumor seeding, pancreatitis, haematuria and hydronephrosis, but the frequency of complication was less.

2.5.6 Gross Appearance

Strombeck and Gribble (1978) observed that in chronic active hepatitis, at necropsy the liver was small and subdivided into small variable sized nodules.

Liver with diffusely infiltrated white-yellow rounded nodules was diagnosed as pancreatic islet cell tumor with liver metastasis (Nyland and Park, 1983)

Greene (1990) reported that the post-mortem lesions in dogs that died in acute phase of Infectious canine hepatitis were enlarged liver with dark and mottled appearance. Thickened gall bladder had a bluish white opaque appearance. Abdominal cavity contained fluid that varied from clear to bright red in colour, but icterus was not usually apparent. Dogs that survived the acute phase had lesions of chronic hepatic fibrosis where liver was small, firm and nodular.

Small nodular proliferations throughout the liver and a yellowish discolouration were suggestive of fully developed stage of cirrhosis (Andersson and Sevelius, 1991).

Gross changes observed at necropsy in steroid hepatopathy were grossly pale liver with no lobular changes (Sen *et al.*, 2001).

2.5.7 Histopathology

Boothe *et al.* (1992) opined that histologic evaluation was an important tool for a definitive diagnosis of hepatic disease; yet it could not predict the effect of disease on hepatic function as determined by laboratory test results.

de Rycke *et al.* (1999) reported that all tissue samples including those taken with reused or resterilized needle had sharp cut edges. Histopathologic sections that were cut transversely to the long axis were better than that of the sections cut transversely to the short axis.

2.5.7.1 Chronic Active Hepatitis

Strombeck and Gribble (1978) reported that chronic active hepatitis (CAH) was histologically marked by foci of hepatocellular necrosis and a cellular response consisting of lymphocytes and diffuse fibrosis. Another important feature of CAH was 'limiting plate necrosis' characterized histologically as necrosis of the hepatocytes that formed a plate adjacent to the portal triad. Intrahepatic cholestasis and bile duct proliferation was seen in CAH. Leucocytic reaction of varied intensity was seen with lymphocytes and monocytes predominating with plasma cells and neutrophils to a lesser number.

Rutgers and Haywood (1988) observed that idiopathic CAH was characterized by dissection of lobular parenchyma by reticulin and fine collagen fibres and by a modest mixed inflammatory infiltrate.

Andersson and Sevelius (1991) reported that CAH was characterised by portal inflammation with mononuclear or mixed inflammatory cells along with, piecemeal necrosis, bridging necrosis and periportal fibrosis extending from the portal triads into the hepatic parenchyma. Chronic progressive hepatitis had a similar pattern, but

piecemeal and bridging necrosis were absent while, chronic hepatitis referred to later stages of chronic active or chronic progressive hepatitis.

2.5.7.2 Cirrhosis

Abnormal deposition of collagen without the loss of normal architecture was fibrosis while cirrhosis was more diffuse and caused distortion of the normal hepatic architecture. Fibrosis changed the microscopic architecture of the liver, creating a pattern of pseudolobulation with bands of fibrous connective tissue extending from one central vein to another or increased in amount along portal tracts and hepatic veins or encircle individual or small group of hepatocytes (Strombeck and Gribble, 1978).

In hereditary copper induced hepatitis histopathological changes ranged from normal to centrilobular hepatitis to bridging necrosis and cirrhosis (Rutgers and Haywood, 1988).

Cirrhosis refers to end stage liver with necrosis, fibrosis, hepatocyte degeneration and marked architectural distortion (Andersson and Sevelius, 1991).

Boothe *et al.* (1992) in studies on experimentally induced hepatotoxicosis with Di Methyl Nitrosamine (DMNA) found that hepatospecific nature of DMNA suggested it as a preferred model to study liver disease and centrilobular necrosis was the primary lesion of DMNA induced hepatotoxicosis. Biliary hyperplasia, fibrosis and lobulation and inflammation were the important histological findings. Hepatotoxicosis has been described as progressive, with cirrhosis representing the end point of disease.

Sevelius (1995) suggested that hepatic cirrhosis was characterized by loss of lobular architecture, nodular regeneration and bile duct proliferation and fibrosis.

Hepatic fibrosis represented net increase in extracellular matrix in the liver and the most common progression of chronic hepatitis in the dog was fibrosis bridging between portal triads (Center, 1999).

2.5.7.3 Vacuolar Degeneration

Lucena *et al.* (2001) reported that histopathological changes seen in cirrhosis were loss of hepatic lobular architecture along with multiple regenerative nodules composed of large hepatocytes with severe fatty change. Abundant lymphoplasmacytic infiltrate in portal spaces, bile duct proliferation and fibrosis were also reported.

Non-specific vacuolar hepatopathy was the histopathological finding in liver of dogs with secondary liver disease subsequent to diseases of other systems like acute pancreatitis, chronic or acute small intestinal disease, extrahepatic bacterial infection, shock, anaemia and congestive heart failure (Rutgers, 1996).

Sen *et al.* (2001) stated that histological changes in steroid hepatopathy was ballooning of hepatocytes with eccentric nucleus, disorganized hepatic cords and obliterated sinusoids.

2.6 TREATMENT

2.6.1 Drug Therapy

2.6.1.1 Antibiotics

Metronidazole and Neomycin reduced the growth of anaerobic bacteria in the small bowel (Johnson, 2000; Rutgers and Haywood, 1988) while Ampicillin, Amoxycillin, Cephalexin and Enrofloxacin were preferred antibiotics for the treatment of hepatitis, cholangiohepatitis, cholycystitis and hepatic abscess (Rutgers, 1996).

2.6.1.2 Corticosteroids

Rutgers and Haywood, (1988); Rutgers (1996) and Varshney (2001) opined that corticosteroids could be used for modulation of inflammation and fibrosis in chronic hepatitis. Prednisolone might be given either at the dose rate of 1-2 mg / kg bodyweight once daily for two to four weeks, followed by tapering dose or in combination with Azothioprine at the dose rate of 1-2 mg/kg body weight. Progression of fibrosis could be prevented by administration of Colchicine at the dose rate of 0.03mg/kg/day orally which inhibits microtubular assembly necessary for extracellular secretion of procollagen and increase collagenase activity (Rutgers, 2000).

2.6.1.3 Copper Chelation Therapy

D-penicillamine was commonly used as copper chelating agent at the dose rate of 10-15 mg/kg while trientine or 2,3,2 tetramine could also be used as decoppering agents at the dose rate of 10-15 mg/kg given orally atleast one hour before meal (Rutgers, 1996; Johnson, 2000; Varshney, 2001)

2.6.1.4 Ursodeoxycholic Acid

Ursodeoxycholic acid prevented reduction in levels of hepatic cytochrome p450 isoenzyme following hepatocyte damage due to hydrophobic deoxycholic acid especially during cholestasis (Tanaka *et al.*, 1999) at the dose rate of 10-15 mg/kg orally (Rutgers, 1996; Johnson, 2000; Leveille, 2000).

2.6.1.5 Fluid Therapy

Hartmann's solution 40-60ml/kg/day, dextrose 2.5-5 per cent or 0.9 per cent NaCl could be used depending on degree of dehydration (Rutgers, 1996).

Hypokalemia associated with hepatic failure was treated with 20 to 30 mEq of potassium chloride per litre of fluid given (Bunch, 2000).

2.6.1.6 Control of Coagulopathies

Treatment of coagulopathies in dogs with hepatic failure included administration of plasma to replenish coagulation factors along with heparin to prevent disseminated intravascular coagulation. Subcutaneous or intramuscular administration of Vitamin K at the dose rate of 2 mg/kg was recommended (Rutgers, 1996).

Bunch (2000) recommended administration of histamine receptors antagonists like Cimetidine at the dose rate of 5-10 mg/kg, Ranitidine at the dose rate of 2-4 mg/kg or Famotidine at the dose rate of 0.5 mg/kg and Sucralfate 0.5-1.0 g TID orally to prevent gastrointestinal bleeding.

2.6.1.7 Control of Ascites

Ascites in chronic hepatic failure could be managed with restriction of salt intake (Rutgers and Haywood, 1988). Furosemide given at the dose rate of 1-2 mg/kg as diuretic, but has to be supplemented with potassium chloride (Varshney, 2001) or a combination of Spiranolactone and Furosemide was used instead (Rutgers, 1996; Johnson, 2000).

2.6.1.8 Nutritional Management

Nutritional management of hepatic encephalopathy consisted of diets high in carbohydrate, low in protein with high biological value and moderately low in fat like home made diets based on cottage cheese and rice (Strombeck and Gribble, 1978; Rutgers and Haywood, 1988) while Rutgers (1996) opined that dairy and vegetable proteins were better tolerated. Diets low in aromatic amino acids, high in branched

chain amino acids and arginine supplemented with adequate amount of vitamin A, B, C, D, E and K was preferred in patients with liver disease (Bunch, 2000).

2.6.1.9 Dietary Fibre

Inclusion of moderate amount of insoluble dietary fibre increased bulk of the stool and prevented constipation which was a risk factor for development of hepatic encephalopathy (Rutgers, 1996).

Soluble fibres like lactulose at the dose rate of 0.25-0.5 ml/kg, p.o. (Johnson, 2000) in diet increased nitrogen incorporation by intestinal bacteria and acidified the luminal colonic pH and increased conversion of ammonia (NH_3) to poorly absorbed ammonium ion (NH_4) (Rutgers, 1996; Varshney, 2001).

2.6.1.10 Zinc

Oral administration of zinc increased the activity of an enzyme ornithine transcarbamylase needed for urea synthesis thereby reducing the plasma concentration of ammonia (Riggio, 1992) and reduced intestinal absorption of copper (Rutgers, 1996).

Zinc was supplemented as zinc sulphate at the dose rate of 2 mg/kg/day or zinc gluconate at the dose rate of 3 mg/kg/day for 30 days followed by 50 mg orally at 12 hour interval (Johnson, 2000)

2.7 PROGNOSIS

Rutgers and Haywood (1988) reported that prognosis of idiopathic CAH was guarded.

Mean survival time of dogs with liver cirrhosis was one month, while in chronic progressive hepatitis the mean survival time was 21.1 months and that of dogs with chronic cholangiohepatitis was 25.8 months (Sevelius, 1995).

Rutgers (1996) reported that the prognosis for a full recovery depended upon the severity of the original insult and was poor in the event of massive nepat:. necrosis.

Center (1999) opined that chronic injury induced a self-perpetuating cycle of cell injury, cytokine production and inflammatory cell accumulation culminating in hepatobiliary fibrosis.

Lucena *et al.* (2001) opined that infection in vaccinated dogs without adequate immunity led to subclinical progressive hepatitis that might result in chronic active hepatitis or cirrhosis.

Materials and Methods

3. MATERIALS AND METHODS

The study was conducted in the Department of Clinical Medicine, College of Veterinary and Animal Sciences, Mannuthy during the period from September, 2002 to September, 2003.

Dogs brought to the University Veterinary Hospital, Kokkala and Veterinary College Hospital, Mannuthy with clinical signs suggestive of liver diseases were selected and utilized for the present study.

3.1 SELECTION OF CASES

Criteria for selection of clinical cases for the study were

1. Dogs showing specific signs of liver involvement, such as jaundice, ascites, hepatomegaly, hepatic encephalopathy, (Rutgers, 1996; Rothuizen and Meyer, 2001).

2. Non-specific clinical signs including vomiting, diarrhoea, polyuria, polydypsia, anorexia, depression, weight loss, which do not respond to routine treatment (Rutgers, 1996).

Animals with these clinical signs were subjected to ultrasound scanning of abdomen, in addition to detailed clinical examination. Of these, cases with findings suggestive of liver diseases formed material for this study.

3.2 OUTLINE OF STUDY

3.2.1 Clinical Examination

Detailed clinical examination of patients was conducted as per Houston (2000) and significant changes, if any were recorded.

3.2.2 Ultrasound Scanning

3.2.2.1 Equipment

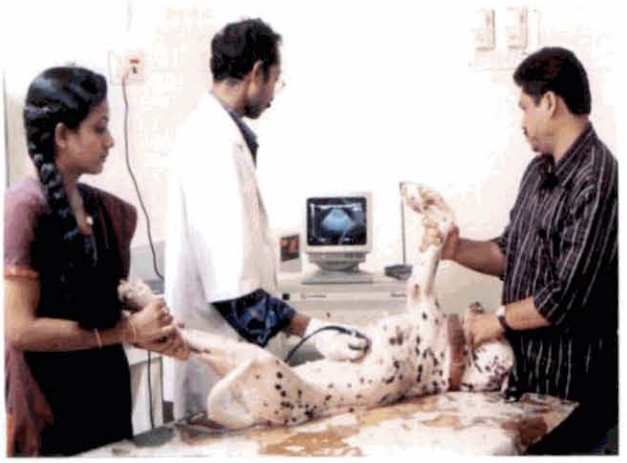
Selected animals were subjected to ultrasound scanning. Hepatic ultrasonography was done using L&T SYMPHONY 4.0 ultrasound scanner using 3.0, 5.0 and 7.5 MHz transducer.

3.2.2.2 Ultrasound Scanning Procedure

Animal was placed in dorsal recumbency. Hair in the abdominal area was removed and acoustic coupling gel was liberally applied to the skin. Animal was placed such that the cranial portion of the image was oriented to the viewers left on sagittal scan and right side of animal to viewers left on transverse scan. Imaging of liver was done by placing the transducer head directly under the sternum and directing the beam cranially. Liver was also scanned in right dorsal and lateral intercostal approach using transverse and sagittal planes (Nyland and Park, 1983; Nyland *et al.* 1995; Varshney and Hoque, 2002) (PLATE 1).

The ultrasonograms were reviewed for alterations in the echogenicity of liver parenchyma, contour, hepatic vasculature and liver size. The parenchymal lesions were classified into focal and diffuse. The echogenicity of liver parenchyma was decribed as normal, hypoechoic, hyperechoic or mixed echogenicity. Gall bladder wall thickness and echogenicity of internal contents were also evaluated. Liver size was assessed subjectively by comparing the caudal position of liver to the costal arch (Nyland *et al,* 1995).

The image was recorded in electromagnetic tape and later photographed.



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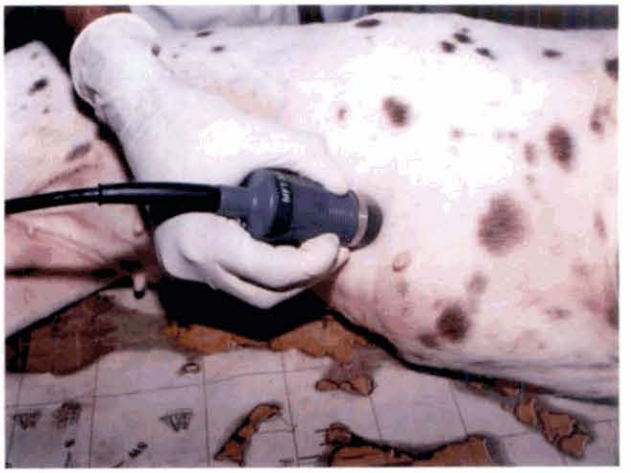

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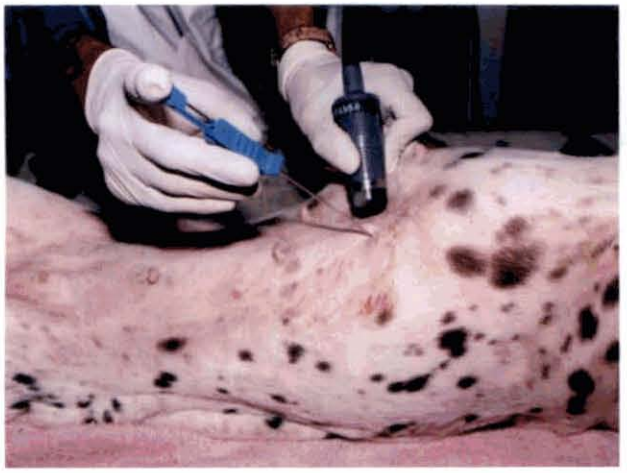

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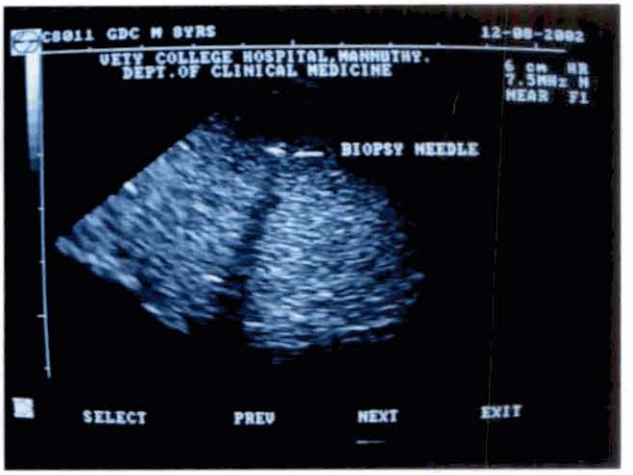

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Plate 1. Ultrasound Guided L iver Biopsy Technique

A- Positioning for ultrasound examination of the liver- Dorsal recumbency.
B- Positioning for ultrasound examination of the liver- right lateral approach at 11th or 12th intercostal space.
C- Position for ultrasound guided biopsy-free hand approach.
D- Ultrasound guided biopsy-position of tip of needle seen in scan (arrow).

Classification of Liver Disorders

Liver diseases were classified into primary and secondary based on etiology (Rutgers, 1996). Primary liver disorders were grouped on the basis of ultrasonographic changes into parenchymal and vascular changes. Parenchymal changes were subdivided into focal and diffuse lesions.

Secondary liver disorders were also grouped into parenchymal and vascular changes. Parenchymal changes were divided into focal and diffuse (Nyland *et al.*, 1995).

3.3 ULTRASOUND GUIDED BIOPSY

Ultrasound guided liver biopsy was performed in cases where it was found necessary with the consent from the owner. Liver biopsy was done with a 14 G Tru-Cut biopsy needle[1]. Haemostatic profiles including whole blood clotting time by capillary tube method and platelet count by direct method using Rees–Ecker solution were evaluated (Benjamin, 1998) before liver biopsy to rule out coagulopathies due to liver disease.

For liver biopsy, the scan head was placed on anterior abdomen, just caudal to the xiphoid and abdomen was scanned sagitally. The area to be biopsied was identified avoiding major hepatic vessels. The biopsy site was prepared in a sterile manner. Local anaesthesia with two per cent Lignocaine solution was attained through ring block. Patient was placed on dorsal recumbancy. A small skin incision was made with No 11 BP blade. The biopsy needle was advanced through this skin incision directing the needle to the left to avoid accidental puncturing of gall bladder, in a freehand approach. The needle was introduced at an angle of 15° or 30° to the transducer. Biopsy was made when the needle tip and the target organ/lesion could

[1]Angiomed,Gmbh & Co. Medizintechnik KG

be seen clearly (Barr, 1995; Hager *et al.*, 1995; de Rycke *et al.*, 1999) (PLATE 1). Tissue was immediately transferred to 10 per cent formalin solution and labelled.

3.4 CLINICAL PATHOLOGY

3.4.1 Collection of Clinical Material

Relevant clinical materials were collected at the time of admission. Five ml of whole blood was collected from saphenous or cephalic vein of the affected dog in dry glass vials with EDTA at the rate of 1-2 mg per milliliter as anticoagulant (Benjamin, 1998).

Ten ml of blood was collected in another test tube for separating serum for biochemical analysis. Sera thus collected were stored at -20°C till further analysis.

In case of ascites, five ml of ascitic fluid was tapped with a sterile 20 G needle into a screw-capped vial.

From cases suspected for Leptospirosis, two ml of serum was collected and sent to Department of Microbiology, College of Veterinary and Animal Sciences, Mannuthy for confirmatory diagnosis.

3.4.2 Haematology

Haematocrit, haemoglobin, total leucocyte count (TLC) and differential leucocyte count were estimatied as per the method described by Schalm *et al.* (1975). Only cases with marked changes were reported.

3.4.3 Serum Biochemistry

Total serum protein, albumin, globulin, A:G ratio, total and conjugated bilirubin and liver enzymes including Alanine aminotransferase (ALT) and Alkaline phosphatase (ALP) were estimated.

All biochemical estimations were done by spectrophotometry in Merck 200 spectrophotometer using commerically available kits.

Serum total protein[1] was estimated by modified Biuret method described by Weichselbaum (1946) while albumin[2] was estimated by bromocresol green dye binding method as described by Doumas *et al.* (1971). Total bilirubin[3] was estimated using the method of Jendrassik and Grof (1938) and direct bilirubin[3] was estimated as described by Schellong and Wende (1960).

Alanine amino transferase (ALT)[4] was measured based on the reference method of International Federation of Clinical Chemistry (IFCC). Alkaline Phosphatase (ALP)[5] was measured in accordance with the recommendations of Deutsche Gesellschaft fur Klinische Chemie (1970).

3.4.4 Ascitic fluid

The ascitic fluid was visually assessed for colour and turbidity. Total protein was estimated by Biuret method as described by Weichselbaum (1946).

3.4.5 Diagnosis of Leptospirosis

Confirmation of Leptospirosis was done with Microscopic Agglutination test (MAT) or Polymerase Chain Reaction (PCR) at Department of Microbiology, College of Veterinary and Animal Sciences, Mannuthy.

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[1] Merck Ecoline Total Protein

[2] Merck Ecoline, Albumin

[3] Merckotest Bilirubin

[4] Merck Ecoline ALAT Tris (GPT)

[5] Merck Ecoline Alkaline Phosphatase

3.5 HISTOPATHOLOGY

Tissue collected was then processed for histologic examination; embedded in paraffin, four micrometer sections were made and stained with haematoxylin and eosin (Bancroft and Cook, 1984).

In the event of mortality during the course of clinical investigation and treatment, liver tissue was collected at necropsy. Liver was examined both grossly and microscopically. The correlation between ultrasonographic and histopathological appearance was studied.

Line of Treatment and Assessment of Response to Therapy

On the day of admission, after making tentative diagnosis, all the cases were administered routine treatment. After detailed investigations, necessary changes were adopted in the therapeutic regimen in consultation with the owners. The routine treatment included administration of five per cent Dextrose Normal saline, Vitamin B complex, liver extracts and symptomatic treatment.

Response to treatment was assessed by follow up examination and noting clinical improvement of the animal

Results

4. RESULT

Dogs brought to the Veterinary College Hospital, Mannuthy and University Veterinary Hospital, Kokkala that exhibited clinical signs suggestive of hepatopathy were subjected to detailed clinical examination and forty eight cases were selected. These animals were subjected to ultrasonography, biopsy (in selected cases), laboratory investigation and serological tests for leptospirosis and tentative diagnosis was confirmed.

Primary liver diseases were diagnosed in thirty five cases, nine were secondary, while in four cases, although there was hepatomegaly no other definite indications of hepatic involvement were available and hence a specific diagnosis could not be made.

Based on ultrasonographic findings, primary and secondary liver diseases were classified into parenchymal and vascular changes. Parenchymal changes were grouped into focal and diffuse.

Primary liver diseases that produced focal parenchymal changes were hematoma and hepatic neoplasia whereas, acute hepatitis in leptospirosis, cirrhosis, fibrosis, fatty infiltration, chronic active hepatitis produced diffuse parenchymal changes. Portal hypertension and portosystemic shunts were the vascular changes observed.

Diffuse parenchymal changes (hypoechogenicity) were observed secondary to pyometra, proctitis and cholecystitis. The focal parenchymal change noticed was hepatic calcification. Passive venous congestion secondary to right sided heart failure was the vascular change observed.

Among 48 selected cases, twenty two (45 per cent) were females while, twenty six (54 per cent) were males.

Highest incidence was found in German shepherd dog (40 per cent), followed by Spitz (21 per cent), Dobermann (13 per cent) and Dachshund (eight per cent). Rottweiler, and mixed breed had 4 per cent incidence while, Great dane, Labrador, Cocker spaniel, Dalmatian and non descript had two per cent incidence.

Age of affected animals ranged from two months to 12 years with a mean age of 4.4 ± 3.3 years.

Non-specific signs reported were anorexia (81 per cent), polydypsia (47 per cent), vomiting (29 per cent), polyuria (25 per cent), diarrhoea (20 per cent), dysuria (six per cent), abdominal distension (25 per cent) and pale mucous membrane (18 per cent). Specific signs of liver involvement noticed were icterus (18 per cent), ascites (23 per cent), hepatomegaly (10 per cent) and hepatic encephalopathy (six per cent).

4.1 PRIMARY LIVER DISEASES

Out of 35 cases of primary liver diseases, parenchymal changes were noticed in 26 and vascular changes in nine. Parenchymal changes were focal in five cases and remaining 21 were of diffuse nature.

4.1.1 Focal Parenchymal Changes

Among the five cases with focal parenchymal changes, acute change was noticed in one case, while the rest of the cases had chronic progression.

4.1.1.1 Haematoma

The case was reported in a six year old male dachshund presented with history that the animal had been hit by a stone on its lateral abdomen one week back and was anorectic for the past three days. Physical examination of the animal revealed mild abdominal pain in its anterior abdomen.

Ultrasonography

Ultrasonography revealed a unifocal lesion having highly irregular margins. The internal contents were hyperechoic surrounded by an anechoic irregular margin and the case was diagnosed as haematoma. (PLATE 5) The ultrasonographic finding was backed by the history and clinical examination.

Clinical pathology

Total leucocyte count of 7,350 cells per micro litre was observed in this case. Differential count was in normal range with 4,410 neutrophils per micro litre.

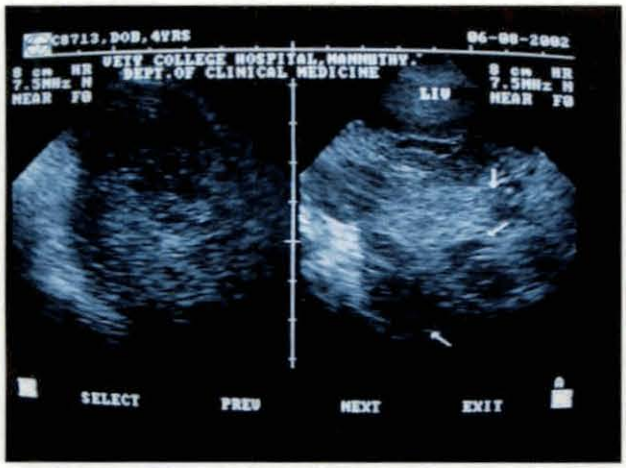
Level of serum total protein was 4.9 g/dl. Level of albumin and globulin was 2.1 and 2.9 g/dl respectively with an A:G ratio of 0.7. Serum activity of ALP and ALT were 102 and 215, IU/L respectively.

Treatment and Response

Animal was treated for three days with parenteral fluids (five per cent Dextrose normal saline) and antibiotics (Amoxycillin and Cloxacillin at the dose rate of 10 mg/kg body weight) given intravenously and tab Amoxycillin 250 milligram orally in the afternoon) for three days. Animal showed improvement. Advised to continue oral medication for two more days. The animal did not turn up for review.

4.1.1.2 Hepatic Neoplasia

Hepatic neoplasia was diagnosed in four cases by ultrasonography and confirmed only in two cases, one with biopsy and the other at postmortem examination.



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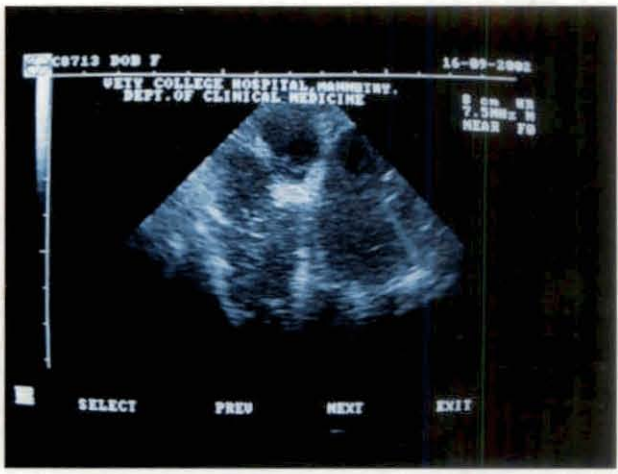

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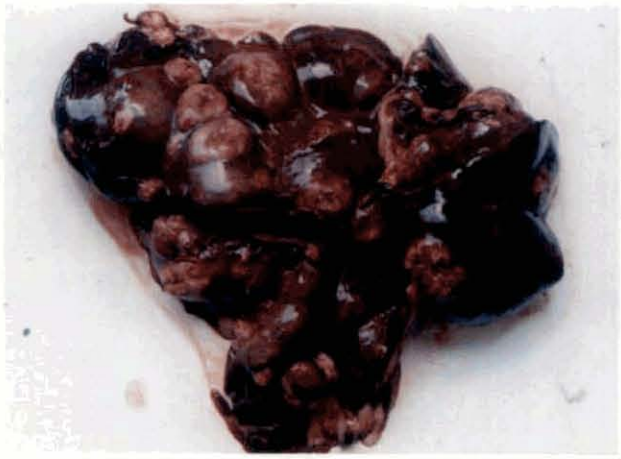

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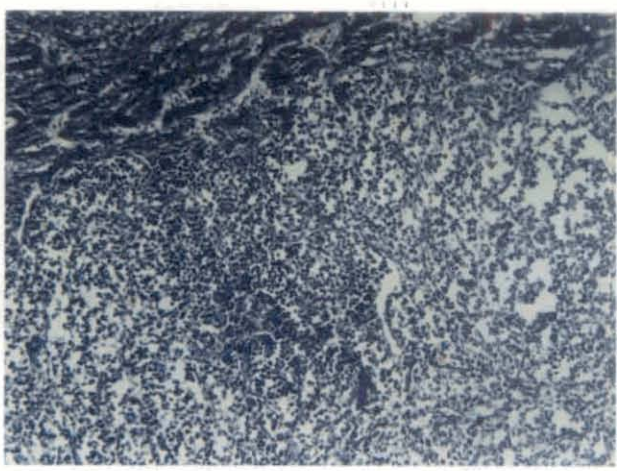

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Plate 2. Hepatic lymphosarcoma

A- Transverse scan of liver showing multifocal anechoic areas with indistinct margins.

B- Transverse scan of same area after one month.

C- Gross appearance of liver with multiple nodules and necrotic centre.

D- Biopsy of liver showing infiltration of liver parenchyma with cells having scanty cytoplasm and hyperchromatic nuclei (H&E, X 100).

The average age of animals with chronic focal changes in parenchyma was 5.5 $\pm$ 2.9 years (3.0-9.0 years). Two of the cases were reported in Dobermann while one each in Dachshund and mixed breed. One was a female and rests were males.

Patients had the history of occasional anorexia. Presenting clinical signs were anorexia (n=4), lethargy (n=4), vomiting (n=2), diarrhoea (n=2) and pale mucous membrane (n=2) while specific signs like abdominal distension and icterus were noticed in one case each. Physical examination showed a hard palpable mass in the anterior abdomen just caudal to costal arch.

Ultrasonography

Four cases with focal parenchymal changes had a chronic course and were diagnosed as hepatic lymphosarcoma. Ultrasonography revealed multifocal circular area of about two to four centimeter diameter in liver parenchyma, with identifiable but indistinct margins. Internal echoes ranged from anechoic to hypoechoic. Ultrasonographic examination was repeated after one month. Transverse and sagittal ultrasound scans revealed that multiple heteroechoic circumscribed areas previously noticed had increased in size. Some of the focal areas had a "target" appearance, which suggested lymphosarcoma. (PLATE 2).

The other three cases with focal parenchymal changes in ultrasonography revealed circular hypoechoic lesion of 4.5 to 5 centimetre diameter having a distinct margin in one of the cases, while the other two had circular unifocal areas in liver having mixed echogenicity. These were diagnosed as hepatic lymphosarcoma.

Ultrasound guided biopsy was attempted in one of the cases.

Clinical Pathology

Mean Total Leucocyte Count (TLC) was 30,460 ± 5,600 cells per micro litre Differential Count was 16,755 ± 2,560 neutrophils and 13,250 lymphocytes per micro litre. Two animals had haemoglobin level of 4.5 and 4.9 gram per cent and Packed Cell Volume (PCV) of 20 and 24 per cent respectively.

Total protein level of 6.1 ± 0.9 g/dl was observed in case of lymphosarcoma. Mean albumin and globulin level in serum was 2.6 ± 0.3 and 5.5 ± 0.2 g/dl. respectively. Albumin: Globulin ratio was 0.6 ± 0.2. ALP and ALT activity were 250 ± 170 and 380 ± 120 IU/L respectively.

Biopsy

Biospsy of liver showed diffuse infiltration of liver parenchyma with hyperchromatic cells having scanty cytoplasm and hyperchromatic nuclei suggestive of lymphosarcoma.

Treatment and Response

In all the confirmed cases, the owners were not willing to administer specific treatment for lymphosarcoma. Therefore, symptomatic therapy was adopted with parenteral fluids, Vitamin B injections and antibiotics (Amoxycillin and Cloxacillin at the dose rate of 10 mg/kg body weight) given intravenously. The animals showed slight improvement. Biopsy was attempted in one case on second day of treatment. Feeding had improved by day three and the animals were discharged.

One of these animals was again presented one month later with similar clinical symptoms. Administered symptomatic therapy. Animal died after two days. Other three cases did not turn up for further evaluation.

Gross and histopathology

Autopsy revealed multiple nodules of varying sizes ranged from three to five cm in diameter. The nodules appeared pale and some had a white necrotic centre. Similar nodules were observed in lungs, heart and kidneys. Histopathological examination of liver showed diffuse infiltration of liver parenchyma with hyper chromatic cells having scanty cytoplasm and hyperchromatic nuclei suggestive of lymphoblasts. Normal hepatic architecture was lost with only remnants of hepatocytes seen in parenchyma. Tumor cells were detected within blood vessels suggestive of possible metastasis. Haemorrhage and sinusoidal dilation were noticed in areas where normal hepatic architecture could be appreciated.

4.1.2 Diffuse Parenchymal Changes

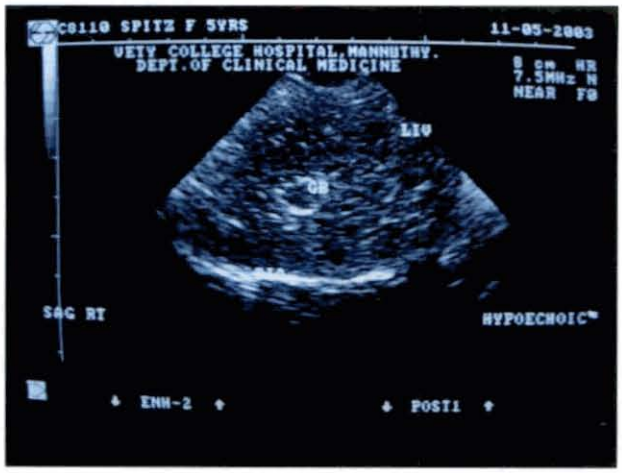
Primary liver disease with diffuse parenchymal changes were sonographically detected in 21 cases, seven of which were due to acute liver insult and 14 cases were diagnosed as chronic liver disease.

4.1.2.1 Leptospirosis

Seven cases were diagnosed as acute liver damage due to leptospiral infection.

The average age of affected animals were 3.02 $\pm$ 2.0 years ranging from 1.5 to 8 years. Among affected animals four were males and three females. Four were German shepherd dogs and one each of Cocker spaniel, Rottweiler and Dachshund.

Animals were presented with varying clinical symptoms and history revealed that none was vaccinated against leptospirosis and had the habit of catching bandicoots (*Bandicoota bengalensis*). Non-specific clinical signs were inappetence (n=6), polydypsia (n=4), vomiting (n=4), polyuria (n=1) and dysuria(n=2). Clinical signs specific to liver disease observed were icterus (n=4). Clinical examination



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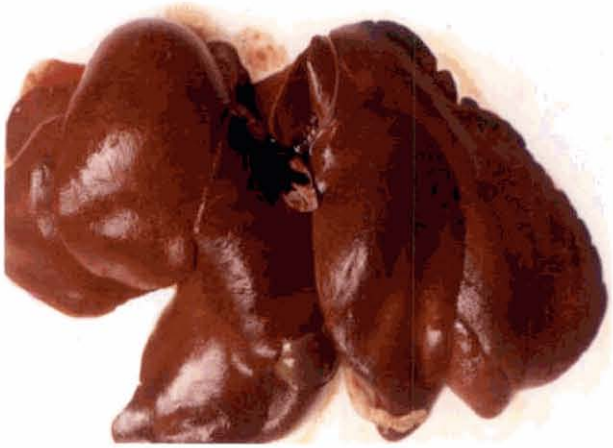

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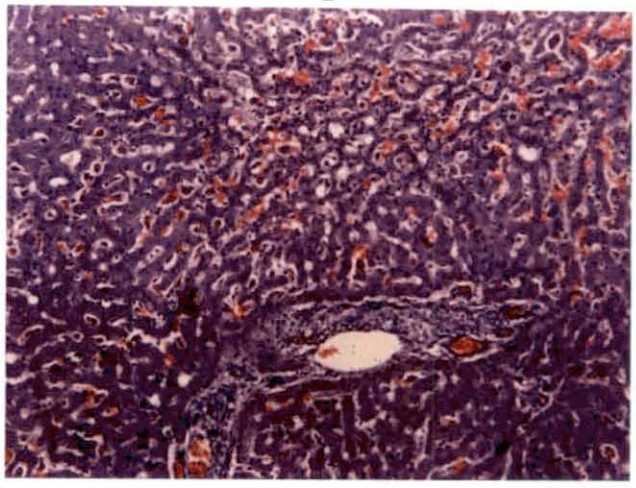

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Plate 3. Leptospirosis

A -Sagittal scan of liver showing hypoechoic parenchyma and echogenic portal vessel walls ("starry-night appearance").

B- Gross appearance of same liver on post-mortem examination - bright red in colour and congested with rounded margins.

C- Histopathology of same liver - venous congestion, sinusoidal dilatation and congestion, degeneration of hepatocytes and perivascular fibrosis (H&E, X100).

showed elevated body temperature (104°F) in two animals while remaining had subnormal temperature. Enlarged kidneys were palpable in three cases while one passed dark brown coloured urine that was positive for blood pigments in benzidine test. Skin and sclera were icteric in four cases. Two animals showed signs of encephalopathy that is, occasional convulsions, tremors and salivation just before death.

Ultrasonography

Hepatic ultrasonography of four cases showed that the parenchyma was uniformly hypoechoic (4/7) in both transverse and sagittal scans. The echogenic portal vessel wall appeared as white specks in hypoechoic liver parenchyma giving a "starry night" appearance (PLATE 3). Liver was hyperechoic than normal in two cases, while in one of the cases right lobes were hypoechoic and left lobes were hyperechoic.

Four out of seven cases had rounded liver borders and hepatic vasculature was not visible in three cases. Changes in kidney were a consistent ultrasonographic finding in all the seven cases. Kidneys were enlarged with irregular margins, had hyperechoic cortex and loss of corticomedullary distinction.

Clinical Pathology

In leptospirosis, TLC was 24, 500 $\pm$ 3,000 with a neutrophil count of 19,600 $\pm$ 2,500 cells per microlitre. Two cases had a PCV of 22 and 26 per cent. Haemoglobin level was 6.8 and 7.2 gram per cent respectively. Rest had a PCV ranging from 36-40 per cent.

Total protein, albumin and globulin level were 5.2 $\pm$ 0.8, 2.5 $\pm$ 0.8 and 3.6 $\pm$ 0.5, g/dl respectively. A:G ratio in case of leptospirosis was 0.7 $\pm$ 0.1. ALP and ALT level in serum were 853 $\pm$ 189 and 283 $\pm$ 90, IU/L respectively. Blood urea

nitrogen, was elevated with a mean of 206 ± 156.7 mg/dl and creatinine level of 7.8 ± 3.4 mg/dl.

Treatment and Response

The cases were tentatively diagnosed as leptospirosis on the first day and were treated with parenteral fluids (five per cent dextrose) and Inj. Benzyl penicillin at the dose rate of 40,000 IU/kg body weight b.i.d intravenously. Supportive therapy was given with Vitamin B injections and Ranitidine at the dose rate of 0.5-1 mg/kg body weight b.i.d intramuscularly.

Five of the animals died during the course of treatment and two cases showed improvement after five days of treatment. They were advised to continue tab Doxycycline at the dose rate of 10 mg/kg body weight daily orally for one week. Dead animals were subjected to postmortem examination.

All these seven cases were positive for various serovars of Leptospira sp to Microscopic Agglutination test (MAT) or Polymerase Chain Reaction (PCR).

Gross and Histopathology

Five out of seven animals died within one to three days during the course of treatment. Postmortem examination of the dogs revealed icterus (3/7) of skin, subcutaneous fat and omentum. Grossly, liver appeared bright red in colour and congested with rounded margins. Kidneys were enlarged (3/5) or shrunken (2/5). Cortex was uniformly pitted and showed longitudinal white necrotic streaks. Urinary bladder showed haemorrhagic streaks.

Histopathology of liver showed extensive congestion, sinusoidal dilatation and congestion, bile duct hyperplasia, atrophy and degeneration of hepatocytes. Diffuse interlobular, perivascular and periductular fibrosis was evident in one case

where ultrasonography revealed hyperechoic specks in the hypoechoic liver parenchyma.

Kidneys showed extensive haemorrhage and coagulation of parenchyma. Glomerular congestion, tubular hyalinisation, interstitial and intratubular infiltration with inflammatory cells were noticed.

4.1.2.2 Cirrhosis

Six cases were diagnosed as cirrhosis based on ultrasonographic changes (PLATE 4).

The average age of animals with cirrhosis was 3.5 $\pm$ 3.1 years (2months to 10 years). Highest incidence was found in Spitz (n=3) followed by German shepherd (n=2) and Dobermann (n=1). Among the reported cases four were males while two were females.

Presenting clinical sign was abdominal distension due to ascites. Other non-specific clinical signs were anorexia (n=4), polydypsia (n=4), polyuria (n=2), pale mucous membrane (n=2), melaena (n=1) and vomiting (n=1). Physical examination revealed pendulous abdomen and fluid thrill was perceivable on abdominal percussion. Temperature of most of the animals was normal (102.2-102.6°F) but three cases showed subnormal temperature (98-99°F). One animal showed ataxia, amourosis, staggering gait and inability to hold head upright during terminal stages.

Ultrasonography

Ultrasonography showed diffuse hyperechogenicity of liver parenchyma along with free peritoneal fluid in abdominal cavity, which appeared anechoic. These were diagnosed ultrasonographically as cirrhosis with ascites. Liver size was reduced

in three cases. Irregular serrated margins with surface nodularity were observed in four cases. Intrahepatic vasculature could be poorly visualised in all the cases.

Clinical pathology

Total leucocytic count in cases of dogs with cirrhosis was 15,440 ± 6,650 cells per micro litre. Differential count was 11,735 ± 2,305 neutrophils per micro litre. Haemoglobin content in three cases ranged from 5.6-5.8 gram per cent.

Mean total protein was 4.3 ± 0.2 g/dl. Albumin and Globulin levels were 1.3 ± 0.1 and 3.1 ± 0.1, g/dl respectively. A:G ratio was 0.4 ± 0.1. Serum ALP activity was 285 ± 80 IU/L while ALT levels were 250 ± 152 IU/L.

Ascitic fluid

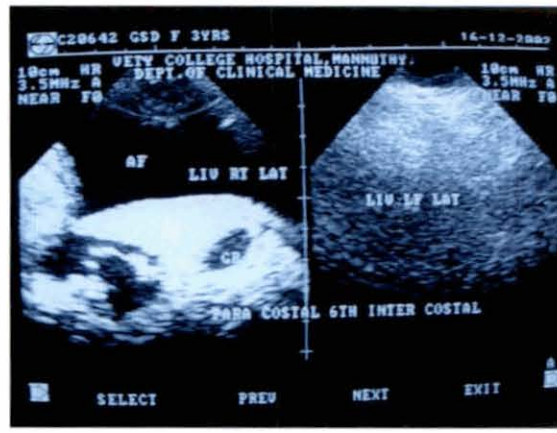
Ascitic fluid was examined in six cases of cirrhosis. In five cases the fluid aspirated were clear transudate with protein content of 0.23g/dl (0.2-0.3g/dl). The ascitic fluid protein to total serum protein ratio in these cases was six per cent indicating low protein ascites.

Treatment and Response

Paracentesis of ascitic fluid was done in animals with dypsnoea. All the cases were treated with tab Lasilactone[1] one tab b.i.d orally, tab Silymarin[2] (70mg), one tab daily and Liv-52 vet syrup, one teaspoon b.i.d orally. Three cases died during the course of treatment. All other cases showed clinical improvement and reduction in fluid accumulation. Ultrasonography of abdomen did not reveal much change during the subsequent examination.

[1]contains Furosemide 20 mg and Spiranolactone 100 mg.

[2] Active principle from *Silybum marianum* used as liver protectant.



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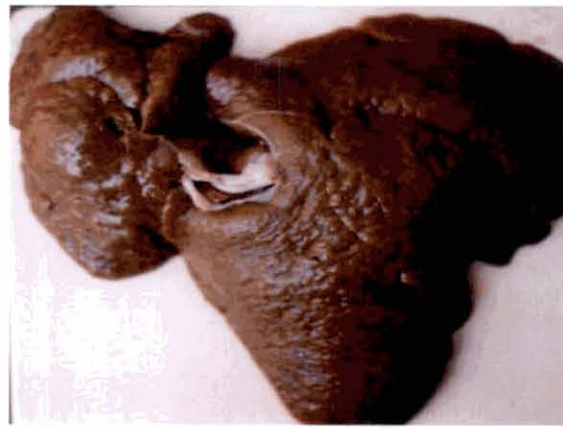

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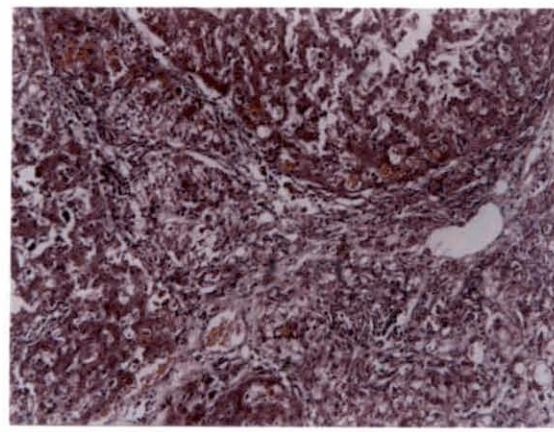

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Plate 4. Cirrhosis

A. Cirrhosis- transverse sonogram showing diffuse hyperechoic liver and anechoic ascitic fluid (AF). Irregular margins could be appreciated.

B. Gross appearance of same liver- showing surface nodularity and reduced size.

C. Histopathology- proliferation of connective tissue fibres, pseudolobulation, atrophy of hepatocytes and central venous congestion (H&E, X 100).

Gross and histopathology

Autopsy showed gelatinisation of subcutaneous fat. Peritoneal cavity contained about one to four litres of clear ascitic fluid.

Grossly, liver was red to yellow in colour, shrunken and surface had nodular appearance.

Histopathology revealed proliferation of connective tissue fibres encircling group of hepatocytes to form pseudolobulation. Fatty changes, atrophy of hepatocytes, central venous congestion, haemorrhage, haemosiderosis, bile duct dilatation and scanty mononuclear infiltration in interstitium were also evident in one of the cases.

4.1.2.3 Hepatic Fibrosis

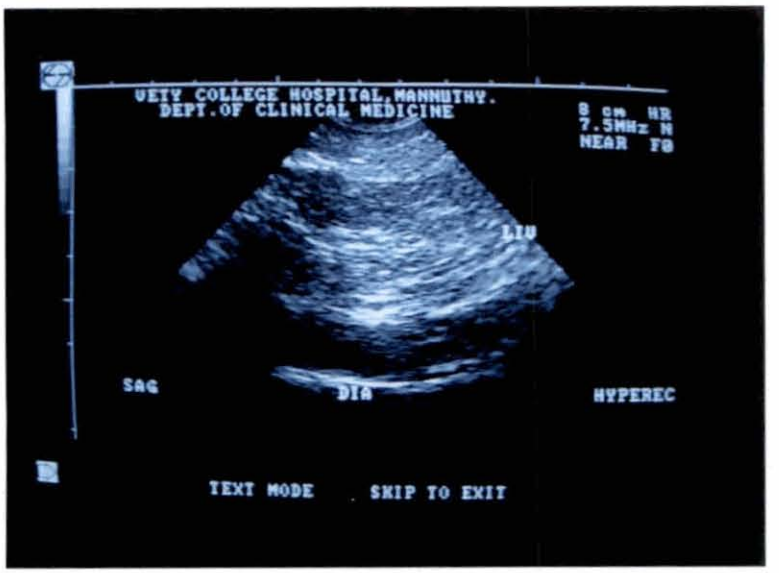
Two cases were diagnosed as hepatic fibrosis.

Hepatic fibrosis was detected in a four year old female Dobermann and five year old male mixed breed.

All the animals were presented with non-specific clinical signs like anorexia, vomiting and polydypsia.

Ultrasonography

Hepatic parenchyma was coarsely hyperechoic and interspersed with hyperechoic patches. Liver margins were normal but hepatic artery and vein could not be distinguished (PLATE 5). These were diagnosed as cases of hepatic fibrosis.



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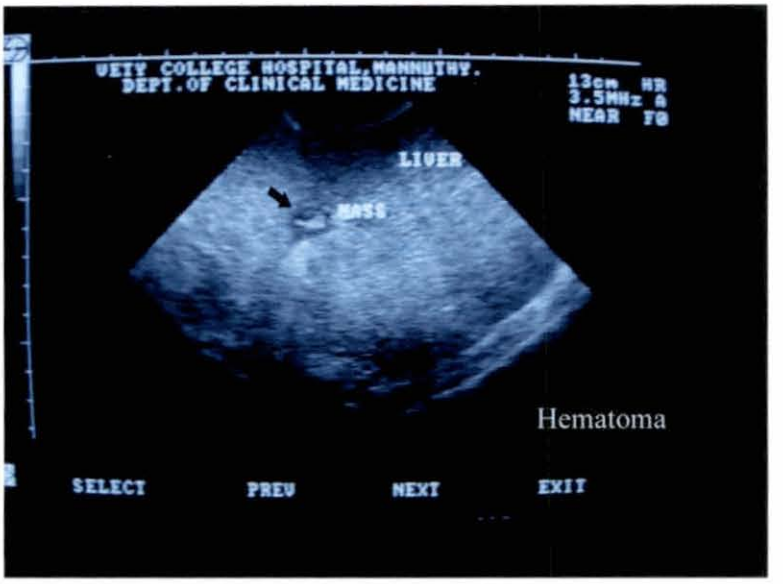

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Plate 5. Hepatic Fibrosis and Haematoma

A- Hepatic fibrosis-linear bands of increased echogenicity and reduced visualisation of hepatic vasculature (sagittal view).

B- Haematoma- transverse view of liver showing irregular margins with echogenic and anechoic contents (arrow).

Clinical Pathology

Haematology

Total leucocyte count of 14,250 ± 2,350 cells per micro litre was observed in cases of hepatic fibrosis, while neutrophil count was 7,250 ± 1,325 cells per micro litre.

Total protein level was 5.4 ± 0.1g/dl. Albumin and globulin level in serum was 2.1 ± 0.3 and 3.1 ± 0.8, g/dl respectively. A:G ratio was 0.7 ± 0.1. ALP and ALT levels were 285 ± 80 and 230 ± 153, IU/L respectively.

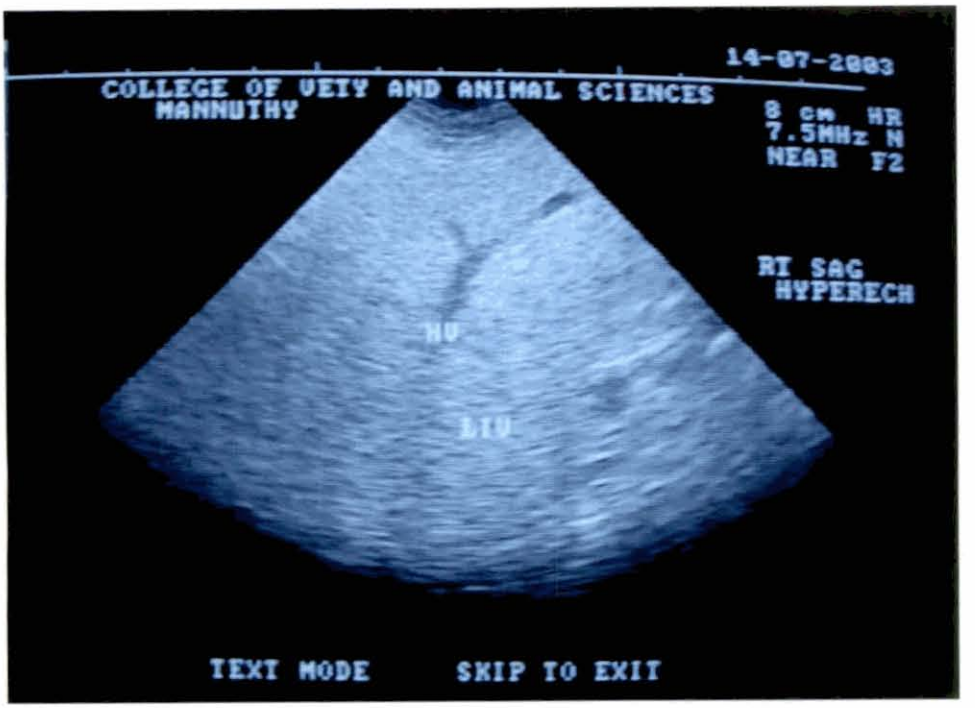
Treatment and Response

Both the animals were given symptomatic treatment with parenteral fluids to correct dehydration along with Vitamin B complex injection. Patients showed improvement from the second day onwards. Discontinued treatment after three days and advised to continue oral medication with Liv-52 vet syrup at the dose rate of one teaspoon full twice daily.

4.1.2.4 Fatty infiltration

Two cases were diagnosed as fatty infiltration. The cases were reported in a three year old male German shepherd dog and 11 month old female Dalmatian.

Both the animals in this group had the history of anorexia for almost a week. The animals showed non-specific clinical signs like anorexia, listlessness and had an emaciated appearance. Physical examination showed marked hepatomegaly, which was the only clinical sign, indicative of liver disease.



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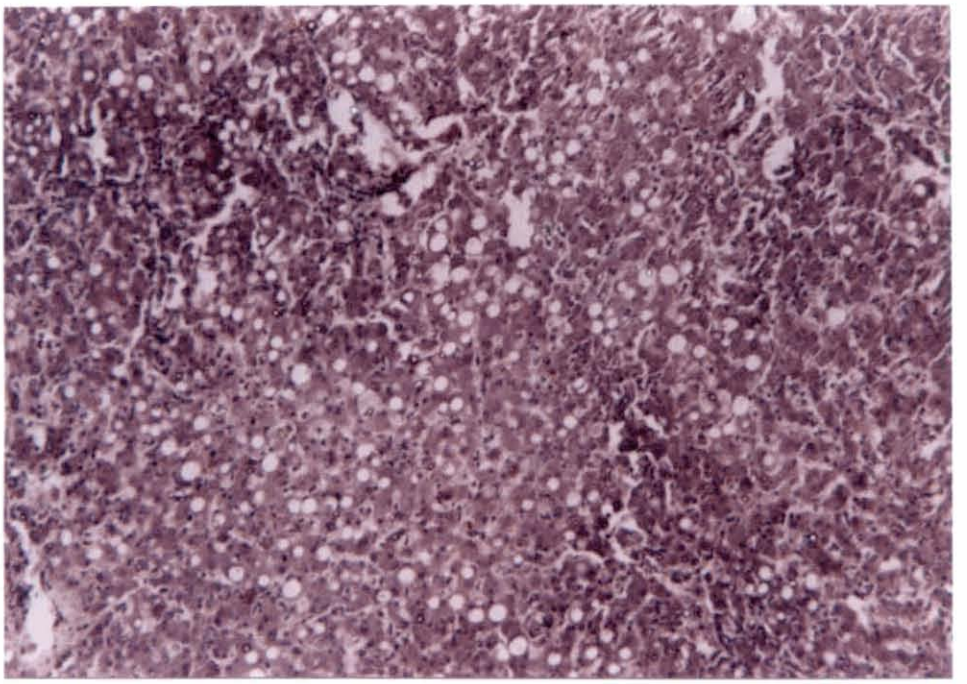

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Plate 6. Fatty infiltration

A. Sagittal scan of liver showing hyperechoic parenchyma.

B. Biopsy of liver showing extensive vacuolation of hepatocytes and kupffer cell reaction (H&E, X 100).

Ultrasonography

Liver parenchyma showed diffuse hyperechogenicity and hepatomegaly, but no change detected in vasculture and margins. The cases were tentatively diagnosed as fatty infiltration of liver (PLATÉ 6) .

Clinical Pathology

Total leucocyte count was 12,340 ± 3,600 cells per micro litre, while neutrophil count was 10,350 ± 2,600 cells per micro litre.

Total protein, albumin and globulin level were 5.1 ± 1.1, 2.8 ± 0.3 and 2.2 ± 0.3 g/dl respectively. A:G ratio was 0.8 ± 0.2. ALP activity was 289 ± 30 IU/L while ALT activity was 153 ± 30 IU/L.

Biopsy

Liver biopsy in one of these cases revealed extensive vacuolation of hepatocytes and kupffer cell reaction.

Treatment and Response

Symptomatic treatment was given with parenteral fluids supplemented with Vitamin B injection. Both the animals showed improvement by next day. Discharged after three days of treatment. Advised to give high quality protein diet.

4.1.2.5 Chronic Hepatitis

Four cases were diagnosed as different stages of chronic hepatitis.

Chronic hepatitis was diagnosed at an average age of 5.9 ± 3.1 years ranging from 1.5 to 8 years. Two were Dobermann while other two were Labrodor. Two were females.

Presented with a history of anorexia, listlessness and reduced activity for more than one week. Hepatomegaly was a consistent clinical finding in chronic hepatitis. Mild ascites (n=2) was detected in ultrasound scan which could not be appreciated during physical examination. Icterus (n=1) was also detected in chronic active hepatitis. Other non-specific findings were listlessness, occasional inappetence and pale mucous membrane. One case was presented after a month with relapse of clinical symptoms. The animal was markedly anaemic and slight icterus of mucous membranes was noticed. Hepatomegaly and enlarged popliteal lymph nodes were detected on physical examination.

Ultrasonography

All four showed hypoechoic to normoechoic hepatic parenchyma and was tentatively diagnosed as hepatitis. Out of this, two showed hepatomegaly and mild degree of ascites. Ultrasound guided biopsy was done in these two cases (PLATE 7).

Clinical Pathology

Total leucocyte count of 15,160 $\pm$ 2,140 cells per micro litre was observed in chronic hepatitis, while neutrophil count was 11,475 $\pm$ 750 cells per micro litre. In two cases the haemoglobin level was 4.5 and 4.9 gram per cent and total red blood cell count was 1.3 and 1.5 million cells per micro litre.

Total protein was 5.2 $\pm$ 0.1g/dl. Albumin and globulin level was 2.1 $\pm$ 0.1 and 3.1 $\pm$ 0.1, g/dl respectively. A:G ratio was 0.7 $\pm$ 0.1. ALP and ALT activity was 452 $\pm$ 70 IU/L and 332 $\pm$ 170 IU/L. One dog showed an elevated ALP level of 1120 IU/L during the course of treatment.

In one case that showed icterus, serum total bilirubin was 1.9 mg/dl and conjugated bilirubin was 0.8 mg/dl. After one month the values had come down to 1.3mg/dl (total) and 0.8 mg/dl (conjugated).

Biopsy

Histopathology of one of these cases showed total disruption of hepatic architecture with a few number of heptocytes having vacuolations. There were areas of necrosis with dense infltrate of lymphocytes, plasma cells and polymorphs. Bile duct proliferation and hemosiderosis were also seen. These changes were suggestive of chronic active hepatitis.

In the second case, hepatic architecture was normal but hepatocytes showed vacuolation in cytoplasm and diffuse collection of inflammatory cells in the parenchyma. This suggested fatty degeneration and chronic hepatitis.

Treatment and Response

One case was treated with 10 per cent dextrose, Inj. Calcium Sandoz 10 per cent, 10ml intravenously and Inj. Belamyl, two ml intramuscularly on the first day. In this case Electrocardiograph (ECG) showed prolongation of QRS duration suggestive of left ventricular hypertrophy. Advised Digoxin at the dose rate of 0.01-0.02 mg/kg body weight orally, Furosemide at the dose rate of 2-4 mg/kg body weight b.i.d orally, Potklor[1] syrup, one teaspoon b.i.d and Tab Silymarin[2] 70 mg, one tab b.i.d. Animal was presented again after one month with similar signs but showed slight icterus. Ultrasound guided liver biopsy was taken. Animal was treated with parenteral fluids (5 per cent dextrose normal saline), Vitamin B injection, Inj. Imferon[3] and Sulfa-trimethoprim. Advised to give tab Ursofac (ursodecholic acid) one tab daily for 30 days.

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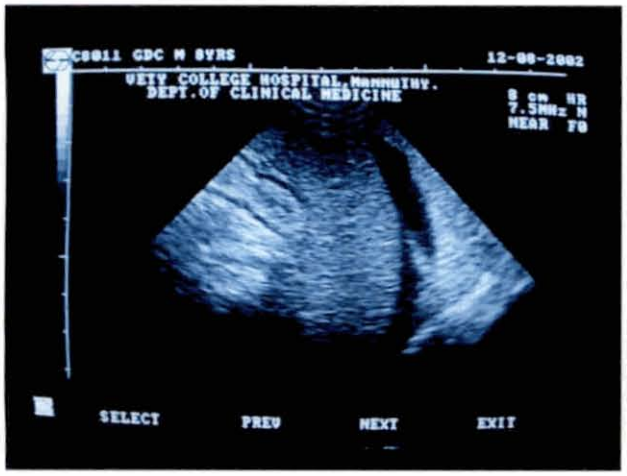
[1]Potassium chloride 3g equivalent to 40mEq. elemental potassium.

[2] Active principle from *Silybum marianum* used as liver protectant.

[3] Iron dextran equivalent to 50mg elemental iron, per ml.



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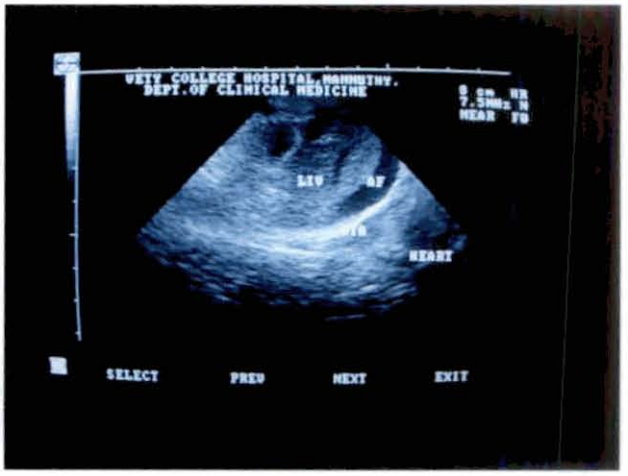

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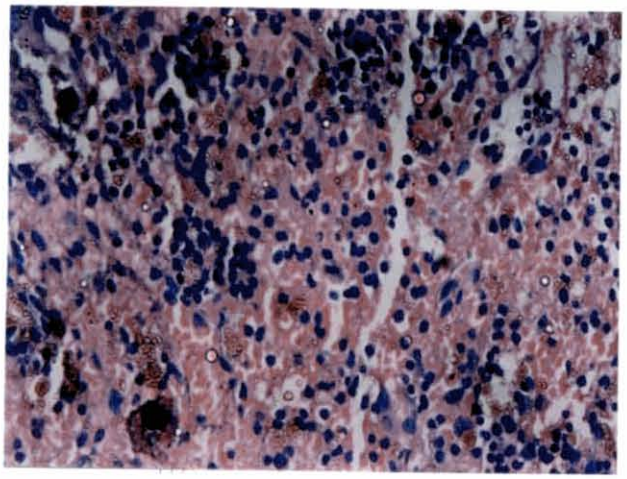

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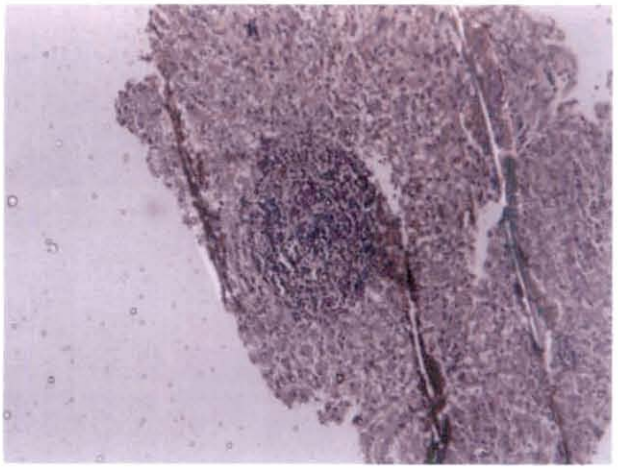

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Plate 7 Chronic hepatitis

A. Chronic active hepatitis- hypoechoic to normoehcoic liver parenchyma. Mild degree of ascites fluid could also be seen.

B. Biopsy of liver in A – disruption of hepatic architecture, scanty hepatocytes showing vacuolation, diffuse infiltration of lymphocytes, plasma cells and polymorphs, hemosiderosis and bile duct proliferation suggestive of chronic active hepatitis (H&E, X 250).

C. Chronic hepatitis- Transverse sonogram of liver showing hypoechoic to normoechoic parenchyma with mild ascites.

D. Biopsy of liver in C- hepatocytes showed vacuolation and diffuse collection of inflammatory cells in the parenchyma suggestive of fatty degeneration and chronic hepatitis (H&E, X100).

Remaining three cases were treated with parenteral fluids (five per cent dextrose), antibiotics (Amoxycillin and Cloxacillin at the dose rate of 10 mg/ kg intravenoulsy, later changed over to Cephalexin at the dose rate of 15 mg/kg body weight intravenously). Biopsy was attempted in one case. One of them showed clinical improvement but other two did not show any improvement even after one week of treatment. Both were later reported dead.

4.1.3 Vascular Changes

Nine cases were diagnosed to have vascular changes, primarily due to hepatic involvement

4.1.3.1 Portal Hypertension

Seven cases were diagnosed as portal hypertension.

Average age at which portal hypertension was reported was 3.2 $\pm$ 3.1 years (10years to 2 months). Four were German shepherd dog, two were Spitz while one was non-descript. Three were females and remaining males.

Animals were presented with the history of ascites, listlessness, failure to gain weight and diarrhoea. Non-specific signs like anorexia (n=6), polydypsia (n=2), diarrhoea (n=2), vomiting, polyuria and pale mucous membrane were observed. Abdominal distension due to ascites (n=3) was the only clinical symptom suggestive of liver disease.

Ultrasonography

Vascular changes noticed in ultrasound included dilated hepatic veins. Mild to moderate degree of ascites was present in three cases of portal hypertension (PLATE 8). Hepatic parenchyma was either normal (3/7), hypoechoic (3/7) or

hyperechoic (1/7). Electro cardio graphy was done in three cases with ascites and no change suggestive of right-sided heart failure was detected.

Clinical Pathology

A TLC of 13,735 ± 1,200 cells per microlitre was observed in portal hypertension while neutrophil count was 11,475 ± 759 cells per microlitre.

Total protein, albumin and globulin levels were 5.7 ± 0.8, 2.0 ± 0.8 and 3.7 ± 0.4 g/dl respectively. A:G ratio was 0.6 ± 0.2. ALP level was 250 ± 25 IU/L while ALT level was183 ± 25 IU/L.

Ascitic Fluid

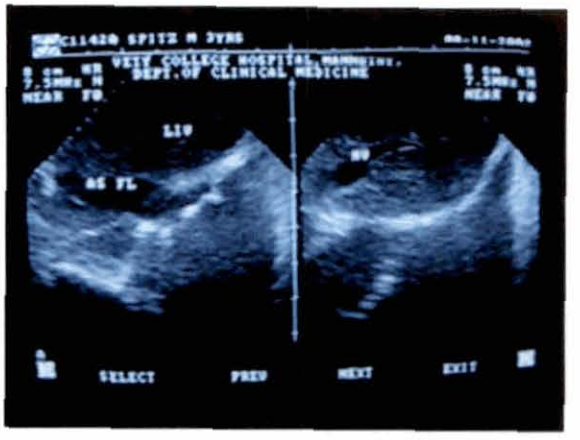
Ascitic fluid from three cases was examined for protein content. In two cases, ascitic fluid was clear transudate with a protein content of 0.2 and 0.3 g/dl. The ascitic fluid protein to total serum protein ratio in these cases were six per cent. In one case ascitic fluid was serosanguineous exudate with a protein content of 4.2 g/dl. Here the ascitic fluid protein to total serum protein ratio was 67 per cent suggestive of high protein ascites.

Treatment and Response

All the cases were given only symptomatic treatment. Resolution of clinical symptoms was noticed within three to five days of treatment.

4.1.3.2 Portosystemic Shunts (PSS)

Two cases with changes similar to PSS were observed in two German shepherd dogs of age 1.5 and 3 years.



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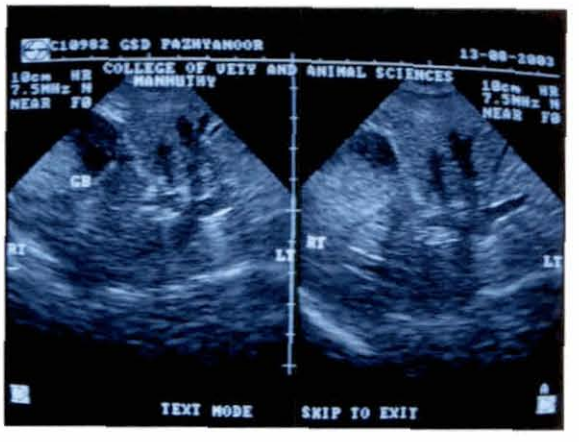

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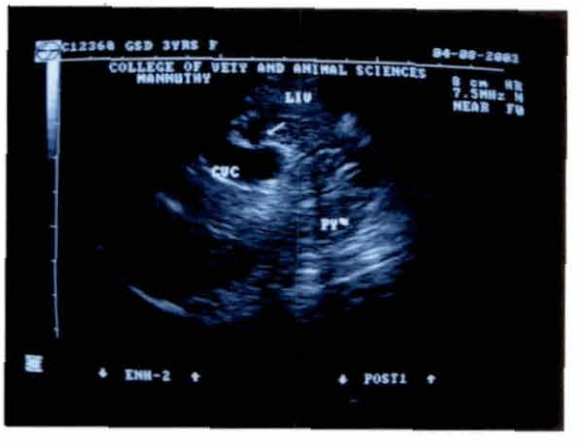

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Plate 8. Vascular changes

A. Portal hypertension- dilated hepatic vein (HV) with presence of anechoic ascitic fluid (AS FL). Liver parenchyma (LIV) was hypoechoic.
B. Portosystemic shunt- A sagittal view of liver showing tortuous portal vessels.
C. Portosystemic shunt- Communication between portal vein (PV) and caudal venacava (CVC) shown by arrow. PY-pyloric part of stomach.

One case was presented with the history of chronic diarrhoea and voluminous semisolid faeces. Clinical signs were non-specific comprising of anorexia, vomiting, diarrhoea, polyuria and polydypsia.

Physical examination showed engorged peripheral vessels on skin and all other clinical parameters were normal.

Ultrasonography

Ultrasonographic changes detected were dilated caudal venacava and a dilated portal vein draining into venacava, suspected to be an extra hepatic portosystemic shunt (PSS). One of these cases had tortuous vessels in hepatic parenchyma suggestive of intrahepatic shunt (PLATE 8).

Clinical pathology

Total leucocyte count was 13, 240 $\pm$ 3,450 cells per micro litre while, neutrophil count was 10, 250 $\pm$ 1,356 cells per micro litre. Haemoglobin level was 5.2 and 5.4 gram per cent.

Total protein, albumin and globulin levels were 5.8 $\pm$ 0.1, 2.1 $\pm$ 0.2 and 3.7 $\pm$ 0.8 g/dl while A:G ratio was 0.6 $\pm$ 0.2. ALP and ALT activity was 210 $\pm$ 30 and 193 $\pm$ 50 IU/L respectively.

Treatment and Response

Confirmation of PSS with the help of portography could not be done. Owner was not willing for exploratory laparotomy or further diagnostic procedures. The animal was treated symptomatically with Metronidazole, Ranitidine and Sulphosalazine. Animal showed improvement and owner preferred to repeat medication at local veterinary hospital.

4.2 SECONDARY LIVER DISEASES

Cases in which hepatic parenchyma was secondarily involved subsequent to failure of some other systems were studied. Amongst nine cases of secondary liver diseases, six cases showed parenchymal changes, one had vascular changes while other two had both parenchymal and vascular changes. Focal parenchymal change observed was one case of hepatic calcification. Diffuse parenchymal changes were noticed in five cases.

4.2.1 Focal Parenchymal Changes

4.2.1.1 Hepatic Calcification

Calcification of hepatic vessels was observed in a 12 year old Spitz.

The animal was presented with non-specific signs like anorexia and difficulty in walking.

Ultrasonography

Transverse abdominal scans showed a linear hyperechoic streak casting a distal acoustic shadow suggestive of calcification (PLATE 9). Sonogram of right and left kidneys revealed hyperechoic cortex and loss of corticomedullary distinction, suggestive of end stage kidney. Biopsy could not be obtained in this case because of poor condition of the animal.

Clinical Pathology

Total leucocyte count was 11,780 cells per micro litre while neutrophil count was 8,500 cells per micro litre.

Total protein level in serum was 4.6 g/dl. Albumin, Globulin and A:G ratio was 2.5 g/dl 2.1 g/dl and 1.2 respectively. ALP and ALT level was 210 and 183IU/L respectively.

Treatment and Response

Animal was treated symptomatically with parenteral fluids. Improvement in condition was noticed from third day onwards.

4.2.2 Diffuse Parenchymal Changes

The average age of animals in this group was 7.8 $\pm$ 3.1 years. Pyometra was noticed at an average age 8.3 years, while age of animal with haemorrhagic proctitis and cholecystitis was three years and five months respectively.

Clinical signs of secondary liver diseases were more related to the primary system affected. Polydypsia, polyuria and vomiting were noticed in pyometra. The case of proctitis showed extensive pain over the body and melaena.

Ultrasonography

Hepatic parenchyma uniformly hypoechoic to anechoic with portal vessel wall appearing as white speck, was the prominent ultrasonographic change detected in acute liver disease secondary to pyometra (3/6), haemorrhagic proctitis (1/6) and cholecystitis which were detected by ultrasound scan. (PLATE 9)

Abdominal scan in three cases of pyometra revealed enlarged uterus, hyperechoic uterine walls with homogenous hypoechoic internal contents having a slow, swirling movement. Transverse abdominal scan of the fourth case showed a tubular organ with hyperechoic wall and anechoic contents in the pelvic region which was suspected to be enlarged uterus, but on postmortem it was found to be enlarged colon which was haemorrhagic, diagnosed as haemorrhagic proctitis. Sagittal scan of

a case of cholectystits revealed distended gall bladder with hyperechoic wall and neck.

Clinical pathology

The mean TLC in cases of pyometra, hemorrhagic proctitis and cholecystitis were 24,650 ± 3,400, 60,035, and 13,450 cells per micro litre respectively. Neutrophil count in pyometra, proctitis and cholecystitis were 22,765 ± 1,200, 58,835 and 10,450 cells per micro litre respectively.

Total protein level in case of pyometra, proctitis and cholecystitis were 4.8 ± 0.2, 5.1 and 4.9 g/dl respectively.

The mean albumin levels in pyometra, proctitis and cholecystitis were 2.3 ± 0.2, 2.5 and 2.1 g/dl respectively

The mean globulin levels in pyometra, proctitis and cholecystitis were 2.5 ± 0.1, 2.6 and 2.8 g/dl respectively

A:G ratio observed in pyometra, proctitis and cholecystitis was 0.9 ± 0.1, 0.9 and 0.8 respectively.

Serum level of ALP in pyometra, proctitis and cholecystitis was 289 ± 15, 263 and 1106 IU/L respectively.

In pyometra, proctitis and cholecystitis, mean elevation in level of ALT was 235 ± 120, 246 and 750 IU/L respectively.

Treatment and Response

The three cases of pyometra were referred to Department of Surgery for panhysterectomy, one of which died post-operatively. The case of haemorrhagic proctitis was treated with parenteral fluids (five per cent Dextrose, dextrose normal

saline) and Amoxycillin and Cloxacillin at the dose rate of 10 mg/kg body weight intravenously. But the condition of the animal did not improve and it died three days later. Cholecystitis was treated with antibiotics (Cephalexin at the dose rate of 15 mg/kg body weight intravenously, b.i.d) and improvement was reported after a week but did not turn up for review.

Gross and histopathology

Autopsy was done in one case of pyometra and in case of haemorrhagic proctitis. Grossly, liver had turgid appearance, yellowish discolouration and was soft to touch, suggestive of hepatosis. Histopathological examination showed vacuolation of hepatocytes indicating fatty degeneration (PLATE 9).

4.2.3 Vascular Changes

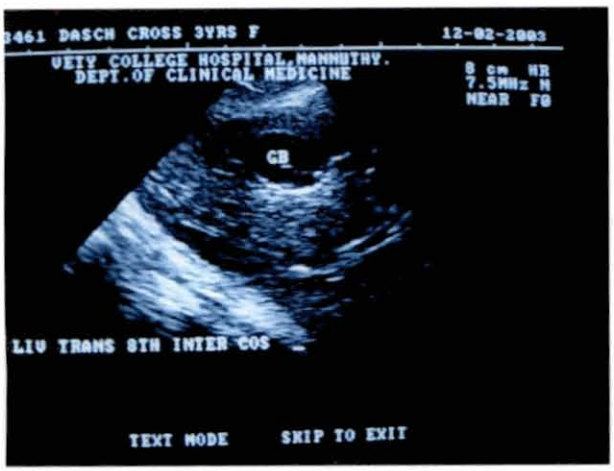
4.2.3.1 Passive Venous Congestion

Two cases were reported in Spitz and one was a Great dane. The change was detected at an average age 10.3 $\pm$ 1.2 years (9 to 12 years). Two were females while one was male.

Animals were presented with clinical history of occasional night cough, difficulty in rising and walking. ECG revealed prolongation of P-wave amplitude and duration, in second lead.

Ultrasonography

Dilated hepatic veins were detected in both transverse and sagittal scan in three cases with right-sided congestive heart failure (PLATE 9). Cardiac enlargement was confirmed by lateral chest radiograph. Electrocardiogram (ECG) that showed increased P-wave amplitude and duration and 'W' pattern of QRS complex suggested



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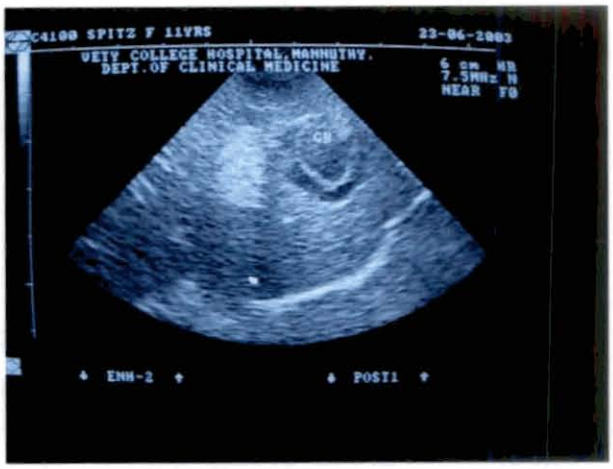

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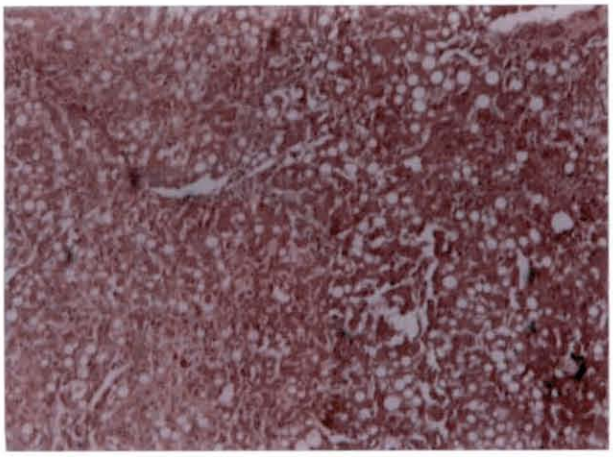

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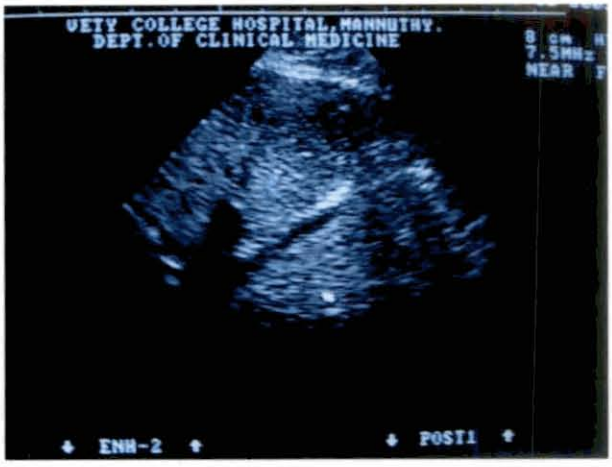

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Plate 9. Secondary liver diseases

A. Diffuse parenchymal changes-Hepatosis-Transverse view of liver showing diffusely hypoechoic to anechoic parenchyma secondary to pyometra.

B. Histopathology of liver in A- showing vacuolation of hepatocytes indicating fatty degeneration (H&E, X 100).

C. Focal parenchymal changes- Hepatic calcification- focal increase in hepatic echogenicity due to dystrophic calcification with distal acoustic shadowing (between arrows).

D. Vascular changes- dilated hepatic vein and caudal venacava secondary to right heart failure.

right-sided heart failure. Liver showed gross enlargement with normal parenchymal echotexture, but in other two cases parenchyma were diffusely hypoechoic.

Clinical Pathology

Total leucocyte count was 9,250 ± 1,250 cells per micro litre while; neutrophil count was 5,985 ± 1,200 cells per micro litre.

Total protein, albumin and globulin in serum was 4.8 ± 0.5, 2.5± 0.4 and 2.3± 0.1, g/dl respectively. Mean A:G ratio was 1.1± 0.2.

The mean serum activity of ALP was 243 ± 159 IU/L while that of ALT was 180 ± 25 IU/L.

Treatment and Response

The animals were treated for congestive heart failure with Digoxin at the dose rate of 0.01-0.02 mg/kg and Furosamide at the dose rate of 1-2 mg/kg body weight bid orally. The animal showed clinical improvement within a few days but was not brought for review.

Hepatomegaly (in four dogs)

Four dogs with clinical history of anorexia and other variable clinical symptoms had normal echotexture of hepatic parenchyma, but hepatomegaly was a consistent finding in these animals that could not be attributed to any cause. These animals were treated symptomatically for a few days and then discharged.

4.3 ULTRASONOGRAPHIC CHANGES IN GALL BLADDER

Twelve cases revealed ultrasonographic changes in gall bladder also along with changes in liver. (These cases were discussed elsewhere along with other hepatic disorders.)

Average age of animals with affections of gall bladder was 3.5 $\pm$ 3.0 years. Among the animals five were German Shepherd, three Spitz and one each of Dachshund, Dobermann and mixed breed. six each from both sexes.

Presented with nonspecific signs like anorexia (n=10), vomiting(n=4), polydypsia (n=4), polyuria (n=4) and diarrhoea (n=3).

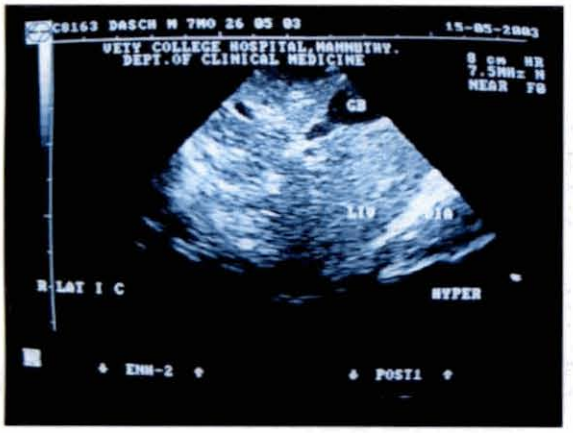
Ultrasonography

In one of the cases, a hyperechoic area was observed in the dependant part of gall bladder casting a distal acoustic shadowing along with dilated neck of gall bladder suggestive of biliary obstruction due to cholelithiasis (PLATE 10).

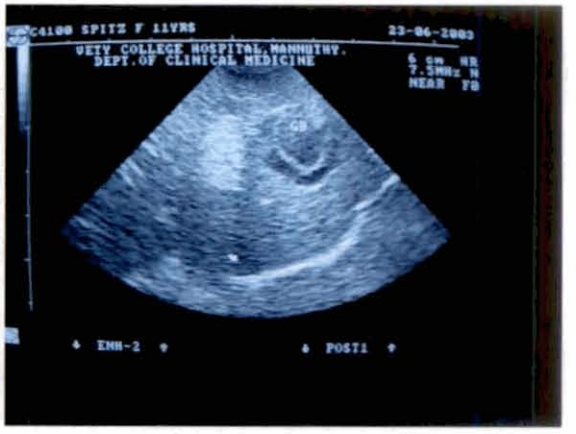
Thickening of the gall bladder wall with hyperechoic strands floating freely inside the lumen (noted in five cases) suggested cholecystitis (PLATE 10).

On transverse scan, in two cases, gall baldder showed hyperechoic wall surrounded by hypoechoic rim designated as 'halo' suggestive of gall bladder wall edema. (PLATE 10)

Gall bladder sludge was observed in one cases and ultrasonographically appeared as hyperechoic mass inside the lumen of gall bladder, which gravitated when position of animal was changed.



A



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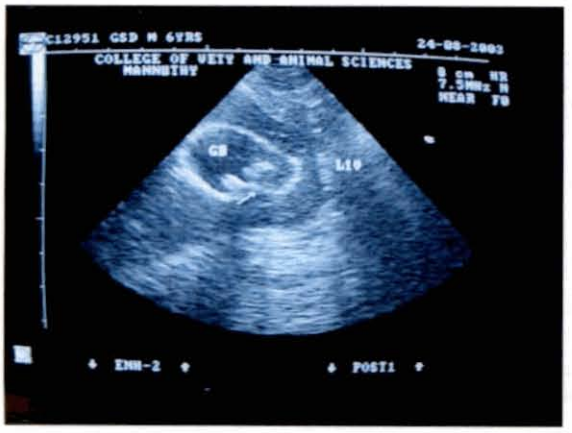
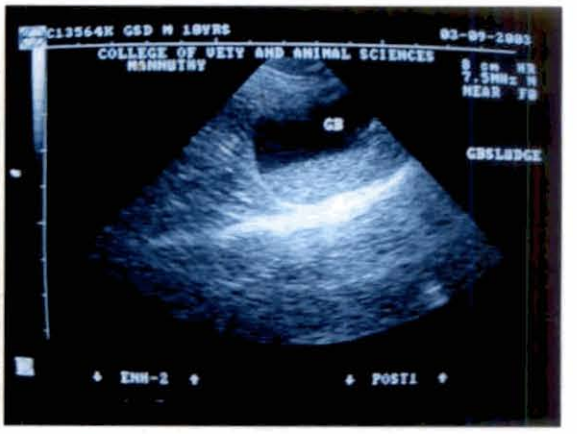


C



D

Plate 10. Ultrasonographic changes in gall bladder.

A. Cholelithiasis- calculi in the dependent portion of gall bladder (GB) casting distal acoustic shadowing (between arrows). Dilated gall bladder neck could also be seen.

B. Cholecystitis- thickened gall bladder wall with a hyperechoic strands floating freely inside the lumen (arrows).

C. Gall bladder wall edema- gall baldder showing hyperechoic wall surrounded by hypoechoic rim designated as 'halo' suggestive of gall bladder edema.

D. Gall bladder sludge- sediment seen within gall bladder lumen. No shadow could be seen.

Discussion

5. DISCUSSION

Hepatopathies observed in this study were grossly classified into primary liver diseases and secondary liver diseases based on whether liver was the primary organ affected or affected secondarily as a consequence of disease of some other organs. Primary liver diseases were again divided into acute and chronic based on progression of disease (Rutgers, 1996).

In the present study, forty-eight cases were selected, out of which thirty-five cases were diagnosed as primary liver disease, nine as secondary while in remaining five cases no specific reasons could be attributed to the changes detected in hepatic sonogram.

Changes in liver detected by ultrasound were further divided into parenchymal lesions and vascular lesions. Parenchymal changes can be divided into focal and diffuse disorders (Nyland *et al.*, 1995). Focal changes were further divided into unifocal and multifocal (Nyland and Park, 1983; Lamb, 1990). Voros *et al.* (1991) classified the changes detected in ultrasound into focal and diffuse and reported that 50 percent of cases were focal while remaining were diffuse.

In the present study parenchymal changes were observed in 26 cases of the primary liver disease, of which five were focal while 21 were diffuse. Hoque and Varshney (2001) reported that focal alterations were detected in 5.71 percent (2/35) dogs whereas diffuse changes were observed in 91.43 percent (32/35) dogs.

5.1 PRIMARY LIVER DISEASES

5.1.1 Focal Parenchymal Changes

5.1.1.1 *Haematoma*

Anorexia was the presenting clinical sign in the case of haematoma. The animal had the history of being hit by a stone on its lateral chest a few days back. Physical examination of the animal showed mild pain in its anterior abdomen. Drazner (1985) reported that even though haematomas were rare in animals, automobile accidents, trauma, fights, fall from height, inhumane acts could produce haematoma in liver, which could be transcapsular, subcapsular or parenchymal.

Sonographic picture revealed a unifocal lesion, having highly irregular margins. The internal contents were hyperechoic surrounded by an anechoic irregular margin and the case was diagnosed as hematoma. Hematomas present a spectrum of sonographic appearance. van Sonnenberg (1983) reported that acute parenchymal bleeding would be echogenic. Later internal contents appeared anechoic or hypoechoic until clot organisation occurred. Later on, a mixed pattern of echogenic and echo-free components were seen when clot retraction occurred (Nyland *et al.*, 1995). Tumor, cyst and abscess also present a similar picture, but abscess and cyst produced posterior acoustic enhancement (Nyland and Park, 1983), which was absent in this case. Clinical signs and history also supported tentative diagnosis with ultrasound examination as suggested by Nyland and Park (1983). Since the physical insult had occurred a few days back, the hematoma formed might have organised to form clot producing the mixed pattern of echogenicity as described by Nyland and Park (1983) and Nyland *et al.* (1995).

Kaneko *et al.* (1997) reported that normal level of total protein in canine serum was 61 $\pm$ 5.2 g/L, albumin 29.1 $\pm$ 1.9 g/L, globulin 34.0 $\pm$ 5.1 g/L and A:G ratio was 0.83 $\pm$ 0.16. Level of total serum protein observed in the present study was

4.9 g/dl. Level of albumin and globulin was 2.1 and 2.9 g/dl respectively with an A: G ratio of 0.7. The level of total protein was low compared to reference value. Acute liver insult rarely produced change in total protein and albumin levels (Tennant, 1997).

Serum activity of ALP and ALT were 102 and 215 IU/L respectively. Normal level of Alkaline phosphatase (ALP) and Alanine amino transferase (ALT) in canines were 66 $\pm$ 36 and 47 $\pm$ 26, U/L respectively (Kaneko *et al.*, 1997). ALP showed only a marginal increase. Elevation in ALT might be due to the damage to hepatocytes and subsequent leakage of cytosolic enzymes (Tennant, 1997).

5.1.1.2 Hepatic Neoplasia

Clinical signs observed in two cases of lymphosarcoma were occasional anorexia, depression, vomiting and diarrhoea. Similar non-specific symptoms were reported by Nyland (1984). Nyland (1984) and Lamb *et al.* (1991) reported hepatomegaly as a consistent finding in lymphosarcoma, but in the present study it was observed only in one case. Lymphoadenopathy was not observed in the present study as reported by Lamb *et al.* (1991). Icterus noticed in one case suggested impaired hepatic function.

Other two cases also showed similar clinical symptoms. The clinical symptoms were anorexia, listlessness, mild anaemia and icterus towards later stages. In addition to this Nyland (1984) reported other non-specific symptoms like fever, depression, tonsillar enlargement, vomiting and severe depression. There was slight distension of abdomen, which on palpation revealed a hard mass on the anterior abdomen immediately behind the xiphoid suggestive of hepatomegaly or a mass in liver. Distended abdomen and a palpable mass in the midabdomen was reported by Nyland (1984) in canine hepatic lymphosarcoma. The differential diagnosis of hepatomegaly in canines were inflammatory disease, venous congestion, diffuse

infiltrative diseases like lymphosarcoma and fatty infiltration (Nyland, 1984), hepatic abscess (Farrar *et al.*, 1996) glycogen accumulation in Cushings disease and caudal venacaval obstruction proximal to liver (Rothuizen and Meyer, 2000). Although Nyland (1984) reported that hepatomegaly was consistent with hepatic lymphosarcoma in canines, such a conclusion could not be drawn from the present study due to the small population studied.

Autopsy of one case that died during treatment had multifocal nodules with necrotic center in liver. But enlargement of lymphnodes could not be appreciated as reported by Nyland (1984).

Histopathology of liver revealed massive infiltration of neoplastic lymphocytes that had compressed and destroyed surrounding hepatocytes. A similar picture was seen in both the cases of lymphosarcoma where histopathology was studied. Histopathological findings were similar to those reported by Nyland (1984)

Ultrasound image of lymphosarcoma showed multifocal circular area of about two to four centimeters diameter in liver parenchyma, with indistinct margins. Internal echoes ranged from anechoic to hypoechoic as described by Nyland and Hager (1985). Transverse and sagittal ultrasound scans were repeated after one month. Multiple heteroechoic circumscribed areas previously noticed had increased in size, suggestive of expansion of the tumor mass similar to the one observed by Nyland and Park (1983). Some of the focal areas had echodense center with hypoechoic peripheral margins, a "target" appearance, which suggested lymphosarcoma. Nyland and Park (1983) also made similar observations. Inaddition to this, Nyland and Park (1983) reported a case of lymphosarcoma characterised by hepatomegaly and reduced echogenicity of parenchyma while Voros *et al.* (1991) reported both enlarged and normal sized liver with hyperechoic parenchyma in case of lymphosarcoma.

Canine lymphosarcoma was difficult to diagnose since clinical signs, laboratory analysis and radiographic findings did not present a true picture of the severity and location of lesion. Ultrasonography to a certain extend could find changes in liver produced by lymphosarcoma (Nyland and Park, 1983). Hepatic metastasis, nodular hyperplasia and lymphosarcoma could produce multiple small hypoechoic or hyperechoic nodules in hepatic parenchyma. Ultrasonography has got 80 percent sensitivity in detecting focal lesions in liver larger than two centimetre (Lamb, 1990). So, many of the tumor conditions could be overlooked.

Total protein level of 6.1 $\pm$ 0.9 g/dl was observed in cases of lymphosarcoma. Mean albumin and globulin level in serum were 2.6 $\pm$ 0.3 and 5.5 $\pm$ 0.2 g/dl respectively. Total protein and albumin level were in the normal range as reported by Kaneko *et al.* (1997). A slight increase in globulin levels was observed. This could be due to increase in amount of gammaglobulins seen with primary hepatocellular carcinomas (Strombeck and Guilford, 1991). Albumin: Globulin ratio was 0.6 $\pm$ 0.2 while Kaneko *et al.* (1997) reported a value of 0.83 $\pm$ 0.16. The low A: G was due to increased globulin level.

ALP and ALT activity were 250 $\pm$ 170 and 380 $\pm$ 120 IU/L respectively. Normal levels of ALT and ALP in serum of carnivores were 47 $\pm$ 26 and 66 $\pm$ 36 U/L respectively (Kaneko *et al.*, 1997). ALT was elevated almost five times the normal while ALP had increased almost six times. These findings agreed with that of Evans (1988) and Leveille (2000). Strombeck and Guilford (1991) reported up to 50 fold increase in level of ALP in hepatic carcinomas.

5.1.2 Diffuse Parenchymal Changes

5.1.2.1 Leptospirosis

Leptospirosis is an important infectious cause of acute hepatitis (Leib, 1997 and Bunch, 2000).

Clinical signs noticed in leptospirosis were inappetence (n=6), polydypsia (n=4), vomiting (n=4), polyuria (n=1) and dysuria(n=2). Rutgers (1996) reported similar symptoms. Polyuria, polydypsia, vomiting and dysuria were clinical symptoms associated with renal failure due to leptospirosis. Birnbaum *et al.* (1998) also expressed a similar view that most common clinical presentation of leptospiral infection was acute renal failure. Clinical examination showed icterus and high temperature (104°F) which was also reported by Navarro *et al.* (1981). Salivation, convulsions and tremors shown by two animals might be clinical manifestations of hepatic encephalopathy (Rutgers, 1996) or due to benign meningitis produced by leptospires invading nervous system (Greene *et al.,* 1990).

The sonographic appearance of liver showed diffusely hypoechoic liver parenchyma (4/7) in both transverse and sagittal scans. Observations were consistent with that of Kurtz *et al.* (1980) that acute hepatitis produced an overall reduction in echogenicity of liver parenchyma and attenuated brightness and more extensive demonstration of portal vein walls. Changes in histopathology correlated well with the changes observed in ultrasonography. Histopathology of liver showed extensive congestion, chronic venous congestion, sinusoidal dilation and congestion, bile duct hyperplasia, atrophy and degeneration of hepatocytes. Biller *et al.* (1992) reported that congestion, suppurative hepatitis and lymphoma caused diffusely hypoechoic liver in ultrasonography. Diffuse interlobular, perivascular and peritubular fibrosis was evident in one case where ultrasonography revealed hyperechoic specks in the hypoechoic liver parenchyma. Lamb (1990) opined that echogenic appearance of portal vein was due to adjacent fat and fibrous tissue.

In two cases liver was uniformly hyperechoic than normal and this might be due to chronic hepatitis produced by leptospiral infection (Greene, 1990). Increased echogenicity of liver parenchyma was reported in chronic hepatitis of dogs (Nyland *et al.*, 1995) and in humans (Kurtz *et al.*, 1980).

On postmortem examination, liver was bright red in colour and congested with rounded margins. Adamus *et al.* (1997) reported firm, tan coloured liver in dogs infected with *Leptospira interrogans* var *canicola* and *icterohaemorrhagiae.*

Profound hepatic dysfunction might occur in leptospirosis due to subcellular damage produced by leptospiral toxins. Leucocytosis with left shift was noticed as suggested by Greene (1990). Serum total protein, albumin and globulin were within normal range as reported by Kaneko *et al.* (1997). An elevated ALP and ALT levels of 853 $\pm$ 189 and 283 $\pm$ 90, IU/L respectively were observed. Similar findings were also reported by Navarro *et al.* (1981), Birnbaum *et al.* (1998) and Saravanan *et al.* (1998).

Blood urea nitrogen and creatinine were also elevated, suggestive of concurrent renal damage (Benjamin, 1998). Elevated bilirubin level was noticed as quoted by Greene (1990).

5.1.2.2 Cirrhosis

Cirrhosis was recorded at an average age of 3.5 $\pm$ 3.1 years. Varshney and Hoque (2002) reported incidence of cirrhosis at an average age of 78 months ranging from 72 to 84 months. Lucena *et al.* (2001) reported hepatic cirrhosis in a five-month-old dog. Ascites was noticed in a two month old pup with no signs of hepatic cirrhosis. Hunt *et al.* (1993) reported ascites due to non-fibrosing liver disease in three young dogs in the age group of three to nine months.

Clinical signs observed in cirrhosis included non-specific signs like anorexia, polydypsia, pale mucous membrane, melaena and vomiting. Similar symptoms were reported by Baba and Matsuda (1987), Sevelius (1995) and Lucena *et al.* (2001). Physical examination revealed pendulous abdomen and fluid thrill on abdominal percussion as observed by Varshney *et al.* (2002). One of the cases showed melaena along with ascites, which was strongly suggestive of cirrhosis (Sevelius, 1995).

Haematemesis and melena might have developed as a result of coagulopathies and gastrointestinal ulceration resulting from liver disease (Center, 1995). Animals with extensive hepatocyte damage had deranged nitrogen metabolism, causing hyperammonemia and signs of hepatic encephalopathy (Strombeck and Guilford, 1991). Deposition of collagen and extracellular connective tissues might cause impaired perfusion of hepatocytes and resultant hepatic insufficiency (Strombeck and Guilford, 1991). This might be the reason for the signs of hepatic encephalopathy shown by one of the dogs in terminal stages.

Ultrasonography showed diffuse hyperechogenicity of liver parenchyma. Cartee and Robert (1981) and Lamb (1990) previously reported similar changes in cirrhosis. Liver size was reduced in three cases as reported by Voros *et al.* (1991). Liver parenchyma was coarse and inhomogenous in three cases. Coarsening of liver echotexture and inhomogenity of the liver parenchyma were less specific abnormalities in cirrhosis (Kurtz and Middleton, 1996). Hepatic cirrhosis produced a characteristic diffuse patchy echogenicity and a lobulated surface of liver that was easily appreciated in presence of ascites (Lamb, 1990). Surface nodularity was observed in four cases, which was more clearly visible in cases with ascites. Kurtz and Middleton (1996) opined that surface nodularity could be easily detected sonographically in the presence of ascites and considered as a fairly reliable sign of cirrhosis.

Ascitic fluid appeared anechoic in ultrasound scans as described by Park *et al.* (1981) and Spaulding (1993b).

In all the six cases intrahepatic vasculature was poorly visualised in ultrasound Lamb *et al.* (1996) reported similar results in 23/38 (68 percent) dogs with hepatic cirrhosis. Pressure from the increased accumulation of collagen and other extracellular matrix would increase pressure on adjacent hepatocytes and compromise

circulation (Stombeck and Guilford, 1991). This might be the reason for decreased visibility of hepatic veins.

Leucogram showed moderate leucocytosis with left shift as reported by Sevelius (1995). Anaemia observed in two cases might be due to myelosupression by circulating endotoxins or gastrointestinal bleeding (Baba and Matsuda, 1987; Center, 1995).

Hypoproteinemia and hypoalbuminemia were observed as reported by Sevelius (1995) and Varshney *et al.* (2002). ALP showed five fold increase while ALT showed a six fold increase. This was similar to the findings of Sevelius (1995) but disagreed with that of Varshney *et al.* (2002). Increased ALP level in cirrhosis might be due to cholestasis secondary to intrahepatic biliary obstruction (Sevelius, 1995).

In five cases, ascitic fluid aspirated was clear transudate with protein content of 0.23g/dl (0.2-0.3g/dl). The ascitic fluid protein to total protein ratio in these cases were six percent indicating low protein ascites. Ascites might be due to the low albumin content resulting from impaired protein assimilation. Decreased albumin contributes to formation of ascites and could cause ascites if serum albumin level falls below 1.0 to 1.5 g/dl (Leib, 1997).

5.1.2.3 Fibrosis

Important clinical signs observed were similar to those reported by Sevelius (1995). But other symptoms like ascites and hepatic encephalopathy reported by Rutgers *et al.* (1993) was not observed in the present study. Abnormal deposition of collagen without the loss of normal architecture was explained as fibrosis while cirrhosis was more diffuse and caused distortion of the normal hepatic architecture (Strombeck and Gribble, 1978). Hepatic fibrosis might progress to cirrhosis, which represents the end stage of liver disease (Boothe *et al.*, 1992) if the inciting factor was

present for a prolonged time. Fibrosis would lead to a self-perpetuating cycle of hepatocyte injury, repair and fibrogenesis that was initiated and sustained by a variety of mechanism (Center, 1999).

Hepatic parenchyma was coarsely hyperechoic and interspersed with hyperechoic patches. Fibrosis produced diffuse increase in echogenicity of liver parenchyma (Lamb, 1990). Portal and hepatic vessels interrupted normal homogenous echotexture of liver (Nyland *et al.*, 1995). Echogenicity of the portal vessels was due to surrounding fat and fibrous tissue as suggested by Barr (1988). Hyperechoic patches seen might be due to increased amount of collagen and elastin around the walls of hepatic and portal veins as suggested by Center (1999).

Liver margins were normal but hepatic and portal vein could not be distinguished. This might be due to the inability of sinusoids, hepatic and portal veins to distend owing to increased intrahepatic pressure from collagen fibres (Center, 1999).

Total leucocyte count of 14,250 $\pm$ 2,350 cells per micro litre was observed in case of hepatic fibrosis. Normal leucocyte count in dogs was 6000 to 17,000 per micro litre (Meinkoth and Clinkenbeard, 2000). Leucogram was in normal range. But, mild neutrophilia was reported by Baba and Matsuda (1987) and Lucena *et al.* (2001).

Total protein, albumin and globulin were within the normal range. But Baba and Matsuda (1987) and Rutgers *et al.* (1993) reported hypoproteinemia due to hypoalbuminemia.

Almost five fold increase in level of ALP and ALT was observed. Rutgers *et al.* (1993) reported high serum activites of ALP and moderate elevation of ALT.

5.1.2.4 Fatty Infiltration

A three year old male German shepherd dog and 11 month old female Dalmatian were diagnosed to have fatty infiltration of liver.

Hepatic lipidosis in small animals was caused by undernutrition (starvation), overnutrition (obesity), protein-calorie malnutrition, diabetes mellitus, drugs and toxins. Important clinical symptoms were depression, anorexia, vomiting, diarrhoea and icterus. It was difficult to determine whether observed clinical signs were due to hepatic lipidosis or lipidosis developed secondarily (Strombeck and Guilford, 1991). Both the studied cases had a history of prolonged anorexia and clinical signs were anorexia and listlessness. Physical examination revealed hepatomegaly. Fatty infiltration was one of the causes attributed to hepatomegaly (Nyland, 1984; Center, 1995).

In both cases, liver parenchyma showed diffuse hyperechogenicity and hepatomegaly. Fatty infiltration produced diffuse increase in echogenicity of hepatic parenchyma (Lamb, 1990). This might be due to higher percent of sound waves being reflected at the liver- fat interface (Nyland *et al.,* 1995). Diffuse hyperechoic liver and hepatomegaly was diagnosed with ultrasound in two cases of hepatic lipidosis secondary to diabetes mellitus (Voros *et al.,* 1991). Severe cases of fatty infiltration would cause significant sound beam attenuation, making the visualization of hepatic vessels difficult (Kurtz and Middleton, 1996). Since such a change was not detected, this could be inferred as a less severe case of fatty infiltration.

Other conditions that would cause increase in echogenicity of hepatic parenchyma were chronic hepatitis, cystic and portal fibrosis and cirrhosis (Nyland and Park, 1993). Therefore, biopsy was indicated to confirm the exact cause for hyperechogenicity. Histopathology revealed extensive vacuolation of hepatocytes

and kupffer cell reaction suggesting fatty infiltration or broadly, vacuolar hepatopathy.

Accumulations of fat in the hepatocytes were lost during tissue processing and histologically appear as a void in stained preparation. Since histopathological finding did not appreciate the vesicles containing lipid (in hepatic lipidosis) it could be diagnosed as vacuolar hepatopathy (Strombeck and Guilford, 1991).

Clinical history of prolonged anorexia (Strombeck and Guilford, 1991), sonographic changes (Lamb, 1990) supported with histological appearance suggested fatty infiltration of liver.

Total leucocyte count was within the normal range. Total protein, albumin and globulin levels were also within the normal range. ALP showed a five fold increase and ALT had three fold increment. Strombeck and Guilford (1991) and Leib (1997) also reported similar findings.

5.1.2.5 Chronic hepatitis

Chronic hepatitis was diagnosed at an average age of 5.9 ± 3.1 years while Rutgers and Haywood (1988) reported that the average age of animal presented with idiopathic Chronic Active Hepatitis (CHA) was four to seven years. Speeti *et al.* (1998) reported subclinical Dobermann hepatitis in the age group of 2.5 to 7 years.

Presenting clinical signs were vague and non-specific like intermittent anorexia and lethargy. Rutgers and Haywood (1988) reported vague gastrointestinal signs in chronic active hepatitis. Specific clinical signs noticed on physical examination were hepatomegaly and icterus. Strombeck and Gribble (1978), Sevelius (1995) and Crawford *et al.* (1985) reported similar symptoms in addition to polydypsia, polyuria, vomiting and weight loss in chronic active hepatitis in dogs.

Rothuizen and van den Ingh (1998) and Johnson (2000) reported hepatic encephalopathy in terminal stages.

In two cases, mild degree of ascites was detected sonographically in peritoneum, wherein physical examination failed to establish ascites. Varshney and Hoque (2002) had similar findings. Sevelius (1995) reported ascites as an important clinical finding in dogs with chronic hepatitis.

All four animals showed hypoechoic to normoechoic hepatic parenchyma and was tentatively diagnosed as hepatitis. Voros *et al.* (1991) reported hepatitis or hepatosis resulted in diffuse fine hypoechoic liver that was indistinguishable from normal liver. In the present study two animals showed hepatomegaly and mild degree of ascites. Kurtz and Middleton (1996) stated that hepatitis resulted in normal echogenicity of liver parenchyma with no detectable sonographic abnormality.

The results obtained were in contradiction to the reports of Kurtz *et al.* (1980) that chronic hepatitis produced increased echogenicity of liver parenchyma, but agreed with the findings that it produced decreased brightness and number of portal vein walls.

TLC of 15,160 $\pm$ 2,140 cells per micro litre and neutrophil count of 11,475 $\pm$ 759 cells per microlitre was observed in chronic hepatitis. Mild leucocytosis with neutrophilia was also reported by Sevelius (1995).

Total protein and albumin values were slightly reduced when compared with normal reference value, while globulin was almost normal. Reduction in total protein might be due to reduced levels of albumin as a result of reduced hepatic synthesis (Strombeck and Guilford, 1991).

Increased serum ALP (seven fold) and ALT (eight fold) activity was observed. Speeti *et al.* (1998) reported increased serum ALT activity at onset of

chronic active hepatitis, while ALP activity was increased both in clinical and subclinical Dobermann hepatitis. Increased levels of ALP and ALT were reported by Strombeck and Gribble (1978), Sevelius (1995) and Evans (1988). One dog showed an elevated ALP level of 1120 IU/L during the course of treatment. This could be due to prednisolone given as part of treatment (Solter *et al.,* 1994; Milne, 1985).

Histological changes were disruption of hepatic architecture with few hepatocytes having vacuolations. There were areas of necrosis with dense infiltrate of lymphocytes, plasma cells and polymorphs suggesting a chronic course. Second case showed vacuolation of hepatocytes and focal collection of inflammatory cells in the parenchyma which suggested fatty degeneration and chronic hepatitis. Similar changes were reported by Strombeck and Gribble (1978), Sevelius (1995) and Speeti *et al.* (1998). Bile duct proliferation and hemosiderosis were also seen which was reported to be rare (Speeti *et al.,* 1998).

5.1.3 Vascular Changes

5.1.3.1 Portal Hypertension

Average age at which portal hypertension reported was 3.2 $\pm$ 3.1 years (10years to 2 months). Hunt *et al.* (1993) reported ascites and portal hypertension in three dogs of three, six and nine months of age. Smith (1994) reported a case in a five year old Rottweiler.

Animals were presented with a history of ascites, listlessness, failure to gain weight and diarrhoea. Non-specific clinical signs observed were anorexia, polydypsia, diarrhoea, vomiting, polyuria and pale mucous membrane. Abdominal distension due to ascites was noticed in three cases. Hunt *et al.* (1993) and Lamb *et al.* (1996) also noticed similar symptoms.

Vascular changes noticed in ultrasound included dilated hepatic veins in seven cases. Portal hypertension causes vasodilatation of venous capacitance vessels in liver and reduction in arterial flow. There would be passive congestion and hepatomegaly with dilated sinusoids (Strombeck and Guilford, 1991). Dilated hepatic vein and prominent portal vein with a hypoechoic liver parenchyma was reported in post-hepatic portal hypertension. Caudal venacaval occlusion also presented a similar ultrasonographic picture (Center, 1995). Schabel *et al.* (1980) reported that umbilical vein might dilate as collateral in portal venous hypertension and become visible within echodense fat of falciform ligament giving a bulls-eye appearance, but such changes could not be appreciated in the present study.

Ascites is defined as transudation of large amount of fluid and protein into the abdominal cavity, caused by the combined effect of decreased plasma protein concentration and high portal capillary pressure (Guyton and Hall, 2000). Three cases showed ascites. Two of them had clear transudate with a protein content of 0.2-0.3 g/dl. while, one was serosanguineous exudate with a protein content of 4.2 g/dl.

Protein content of the ascitic fluid is divided by the plasma protein concentration and ratio was expressed in percentage. Ascitic fluid protein to plasma protein ratio would help to differentiate low protein ascites from high protein ascites in the presence of hypoproteinemia (Hunt *et al.*, 1993). The ascitic fluid protein to total protein ratio in these two cases were six and sixty-seven percent indicating a low protein and high protein ascites respectively. Ascites due to portal hypertension could be due to pre-sinusoidal, sinusoidal or post-sinusoidal obstruction. Post-sinusoidal hypertension resulted in exudation of hepatic lymph having high protein concentration (Center, 1995). Other two cases where protein content of ascitic fluid was 0.2 and 0.3 gram/ dl, might be due to hepatic or pre-hepatic portal hypertension (Center, 1995).

Ultrasonography revealed normal hepatic echotexture only in three cases as described by Nyland and Park (1983). Transverse scan showed that hepatic veins were distended which suggested portal hypertension (Center, 1995). Therefore, the case could be high protein ascites caused by portal hypertension due to post-sinusoidal obstruction. Findings were similar to those reported by Holt *et al.* (1995) and Smith (1994) due to occlusion of caudal venacava. Portal hypertension resulted from collagenization of sinusoids and fibrosis in liver (Center, 1995). Diffuse fibrosis resulted in the hyperechoic liver parenchyma noticed in one of the cases as described by Nyland and Park (1983) and Lamb (1990). Literature review did not give any possible explanation for the hypoechoic liver noticed in remaining three cases.

Total leucocyte count was normal. Total protein, albumin and globulin levels in serum were within the reference range. This was contradictory to the findings of Hunt *et al.* (1993) who reported hypoproteinemia (4.4g/dl) and slightly reduced albumin concentraion. Smith (1994) reported hypoalbuminemia (1.2g/dl) in caudal venacaval occlusion. Moderate increase was noticed in serum levels of ALP and ALT. Similar results were observed by Hunt *et al.* (1993) and Center (1995).

5.1.3.2 Portosystemic Shunts

Two cases of PSS were tentatively diagnosed in two German shepherd dogs of age 1.5 and 3 years. Holt *et al.* (1995) observed that age of dogs presented with portosystemic shunts ranged from 2 to 84 months (median, 17 months), while Lamb *et al.* (1996) reported mean age of 1.7 (0.2 to 10) years. Varshney and Hoque (2002) noticed PSS at a mean age of 26.8 months (3 to 96 months).

Congential intrahepatic PSS was a communication between the portal vein and caudal venacava while extrahepatic PSS connected portal vein or one of it's

tributaries with the caudal venacava . Acquired multiple PSS resulted from portal hypertension (Center, 1995).

Presenting clinical signs were anorexia, vomiting, diarrhoea, polyuria and polydypsia. Most important clinical signs associated with PSS included neurological signs like head pressing, apparent blindnesss or seizures, gastrointestinal signs like diarrhoea, vomiting and poor appetite (Holt *et al.*, 1995; Lamb *et al.*, 1996). Neurological symptoms were not noticed in the present study. Bostwick and Twedt (1995) reported absence of clinical signs related to a small shunting vessel.

Ultrasonographic change detected were dilated caudal venacava and a dilated portal vein draining into venacava, suspected to be an extra hepatic portosystemic shunt. Lamb (1996) reported similar findings and that right intercostal space could be used as a window. One of these cases had tortuous vessels in hepatic parenchyma suggestive of intrahepatic shunt. Hoque and Varshney (2001) and Varshney and Hoque (2002) expressed similar views that shunting vessels were more distended and tortuous.

Size of liver was very much reduced because of decreased hepatic blood flow and lack of hepatotropic factors such as insulin, glucogon and nutrients resulted in hepatic atrophy as opined by Johnson (2000). In dogs with PSS, liver would be grossly small and often mottled. Sonography of hepatic parenchyma showed diffuse or patchy hyperechoic small bright area with an over all reduction in scanning area (Varshney and Hoque, 2002).

Haemoglobin level was 5.2 and 5.4 gram per cent, indicating anaemia. But Bostwick and Twedt (1995) reported normal PCV. Total protein, albumin and globulin levels were in the normal range. ALP and ALT activity were 210 ± 30 and 193 ± 50, IU/L respectively which were above the reference range. Bostwick and

Twedt (1995) too reported a high serum ALP (3.2 ± 3.7 times normal) and ALT (2.4 ± 3.5 times normal) concentration in dogs with PSS.

5.2 SECONDARY LIVER DISEASES

Center (1999) observed that secondary hepatic injury occurred subsequent to systemic infections, disseminated neoplasm or pathological changes in other organs like cardiovascular, gastro-intestinal, urogenital and endocrine systems. Rutgers (1996) reported that central role of liver in metabolism made it vulnerable to diseases of other systems.

5.2.1 Focal Parenchymal Changes

5.2.1.1 Hepatic Calcification

A case of hepatic calcification secondary to chronic renal damage was studied. 1, 25 dihydroxycholecalciferol or calcitrol is produced in kidney by hydroxylation of 25 hydroxycholecalciferol. Calcitrol limits Parathyroid hormone (PTH) production. In chronic renal failure, calcitrol production is inhibited. Thus there was no inhibition on PTH. This would lead to secondary hyperparathyroidism and dystrophic calcification of soft tissues including kidney (Polzin *et al.*, 1995).

Transverse abdominal scans showed a linear hyperechoic streak casting a distal acoustic shadow suggestive of calcification. Nyland *et al.* (1995) reported that dystrophic calcification of liver parenchyma produced diffuse increase in the echogenicity of liver parenchyma; sometimes shadowing was seen distally if there was enough sound attenuation. Similar findings were also made by Pai and Bude (2002) and Okuda *et al.* (2003).

Hematological parameters studied were within the normal range. Reduction in total protein, albumin and globulin were noticed. This could be due to reduced

hepatic synthesis of protein fraction due to old age and malnutrition. Slight elevation in ALP and ALT activity were observed.

5.2.2 Diffuse Parenchymal Changes

Primary organs affected were uterus, rectum and gall bladder. Affections of these organs secondarily affected liver as opined by Rutgers (1996) and Center (1999). Secondary hepatitis might have additional clinical signs. In the present study, bacterial infection in uterus and rectum had resulted in sepsis or endotoxemia (Leib, 1997).

The presenting clinical symptoms were polydypsia, polyuria and vomiting in case of pyometra. Physical findings included vaginal discharge, abdominal distension and dehydration as described by Johnson (1995). Clinical findings in proctitis included pain and tenesmus (Willard and Fossum, 1995), while in cholecystitis it was non-specific.

Ultrasonography revealed uniformly hypoechoic to anechoic hepatic parenchyma with portal vessel wall appearing as white speck. Observations were consistent with that of Kurtz *et al.* (1980) that acute hepatitis produced an overall reduction in echogenicity of liver parenchyma and attenuated brightness and more extensive demonstration of portal vein walls.

Leucogram showed severe leucocytosis with left shift consistent with an acute septicemia (Benjamin, 1988)

Histopathology revealed fatty degeneration and congestion suggestive of acute hepatitis (Boothe *et al.*, 1992).

Animals with pyometra were referred to Department of Surgery for pan hysterectomy. One among this died during treatment. The animal with proctitis died

after three days while case of cholecystitis showed improvement after a week's treatment.

5.2.3 Vascular Changes

5.2.3.1 Passive Venous Congestion

Three cases were presented, two were Spitz and one Great dane. The mean age of animals at presentation was 10.3 $\pm$ 1.2 years (9 to 12 years). Two were females while one was male.

Secondary hepatic disorders could result from acute right sided heart failure (Leib, 1997). Electrocardiogram (ECG) that showed increased P-wave amplitude and duration and deep Q and S wave with a 'W' pattern suggested right-sided heart failure (Bolton, 1975).

Heart failure causes pooling of blood in all the visceral organs leading to congestion and alterations in the size of the organs. Nyland (1984) reported that dilated hepatic veins and hepatomegaly were the ultrasonographic changes noticed due to passive venous congestion. Lamb (1990) also reported that dilated hepatic vein could be seen draining into the caudal venacava close to the diaphragm.

Ultrasound examination showed dilated hepatic veins in transverse and sagittal scan in three cases with right-sided congestive heart failure. Other ultrasound changes noticed were hepatomegaly with hypoechoic liver parenchyma. Sinusoids and vascular spaces in liver expand subsequent to heart failure and this might be responsible for hepatomegaly (Strombeck and Guilford, 1991). Hypoechoic liver parenchyma might be due to passive congestion as suggested by Nyland *et al.* (1995). Venous congestion diagnosed ultrasonographically by dilated hepatic veins along with hypoechoic parenchyma indicated a cardiac rather than hepatic

abnormality. These were in accordance with the observations made by Nyland (1984) and Nyland *et al.* (1995).

The dogs showed a normal leucogram. Slight reduction in total protein, albumin and globulin levels were noticed. This could be due to reduced hepatic synthesis of proteins due to malnutrition and old age (Benjamin, 1998). Two fold increase in level of ALP and ALT were observed in the present study.

Hepatomegaly (in four dogs)

Hepatomegaly was defined as an enlargement of the liver. The most common casues of hepatomegaly were vascular congestion subsequent to right sided heart failure, pericardial tamponade and cardiomyopathies. Post-sinusoidal blocks like kinking of venacava, hepatic vein thrombus and Budd-chiari syndrome could result in hepatomegaly. Reticuloendothelial hyperplasia casued by hypertrophy or hyperplasia of Kupffer cells subsequent to septicemias, hepatitis, malignancies and hypovitaminosis A could cause enlargement in liver size. Hepatomegaly was reported in infiltrative diseases including lipid or glycogen storage disease, inflammatory diseases and neoplastic diseases (Nyland, 1984; Strombeck and Guilford; 1991, Center, 1995; Leib, 1997). Hepatomegaly is a consistent finding in lymphosarcoma (Nyland, 1984).

Hepatomegaly was a subjective term because no reliable method exists for quantifiation of liver size in dogs (Voros *et al.* 1991).

Ultrasonographic examination revealed no abnormality in echotexture, except for increased liver size. Serum biochemistry and liver function test were also normal. Biopsy that was necessary for confirmatory diagnosis could not be done.

5.4 ULTRASONOGRAPHIC CHANGES IN GALL BLADDER

Involvement of gall bladder was not a separate entity and it was associated with other hepatic disorders.

Clinical signs noticed in animals with gall bladder involvement were anorexia, vomiting, polydypsia, polyuria and diarrhoea. Forrester *et al.* (1992) reported similar symptoms in addition to fever and icterus.

Varshney and Hoque (2002) observed distended gall bladder with patent bile duct in anorectic dogs.

A hyperechoic area was observed in the dependant part of gall bladder casting a distal acoustic shadowing along with dialated neck of gall bladder suggestive of biliary obstruction due to cholelithiasis. Nyland and Park (1983) and Lamb (1990) reported similar findings. Lamb (1990) also reported that pancreatitis could cause biliary obstruction but, choleliths were uncommon in dogs.

Thickened gall bladder wall was noticed in five cases with hyperechoic strands floating freely inside the lumen in one case suggestive of cholecystitis. Gall bladder wall normally appeared as thin echogenic line and measured about 2-3 mm in thickness (Spaulding, 1993a). Thickening of gall bladder wall was nonspecific, associated with acute or chronic hepatitis, cholecystitis or cholangiohepatitis (Nyland *et al.*, 1995).

In two cases, gall bladder wall was surrounded by hypoechoic rim designated as 'halo' suggestive of gall bladder wall edema. Nyland and Park (1983) reported similar change associated with acute cholangiohepatitis in dogs. Spaulding (1993a) described causes of gall bladder thickness as hypoproteinemia, right sided cardiac failure, hepatic dysfunction associated with hypoalbuminemia, hepatitis, ascites, renal diseases, venous hypertension, gall bladder tumor and infectious canine hepatitis.

Gall bladder sludge was observed in one case and ultrasonographically appeared as hyperechoic mass inside the lumen of gall bladder that did not cast any acoustic shadowing. Spaulding (1993a), Bhadwal *et al.* (1999) and Hoque and Varshney (2001) observed similar changes in gall bladder. Bromel *et al.* (1998) opined presence of gall bladder sludge as an occasional finding.

Summary

6. SUMMARY

The study on 'Ultrasonographic evaluation of canine hepatic disorders' was conducted to evaluate the usefulness of ultrasonography as a tool for early diagnosis of liver diseases and classify liver diseases based on ultrasonographic findings. Ultrasound guided biopsy was performed in selected cases for confirmation of tentative diagnosis and to correlate between ultrasound image and histopathologic changes in liver.

Following parameters were studied

History and clinical signs,

Physical examination,

Ultrasonography of liver,

Ultrasound guided biopsy (in selected cases),

Haematology and serum biochemistry,

Postmortem examination and histopathology.

Subjects of the present study were selected based on clinical signs, history, serum biochemistry, serology and ultrasonographic changes. Liver diseases identified were broadly classified into primary and secondary liver diseases. Based on ultrasonography these were divided into parenchymal and vascular changes. Parenchymal changes were again subdivided into focal and diffuse changes.

Clinical signs suggestive of liver diseases were ascites and icterus. Non-specific signs observed were inappetence, polydypsia, polyuria, vomiting, diarrhoea and dysuria. Important findings on physical examination were abdominal distension with perceivable

fluid thrill, yellowish discolouration of mucous membrane and enlargement of liver, palpable at caudal abdomen.

Transverse and sagittal scan of the animal was taken both in dorsal recumbency and on right lateral intercostal approach.

Changes in the echotexture of the hepatic parenchyma, liver lobe margins, hepatic vasculature and gall bladder were studied.

Among primary liver diseases, focal parenchymal change detected were haematoma and lymphosarcoma. Disease conditions that caused diffuse parenchymal changes were leptospirosis, cirrhosis, fibrosis, fatty infiltration and chronic active hepatitis.

Vascular changes observed in this study were portal hypertension with ascites and porto-systemic shunts. Ascitic fluid was detected in cirrhosis, chronic active hepatitis and portal hypertension.

Diffuse parenchymal changes were noticed in liver, secondary to pyometra, proctitis and cholecystitis. Dystrophic hepatic calcification caused focal parenchymal change secondary to chronic renal failure. Vascular changes identified were passive venous congestion in liver secondary to right sided heart failure.

Varying degrees of leucocytosis with shift to left was a consistent finding in hepatopathies.

Level of total protein and albumin in serum were markedly reduced in cases of cirrhosis. In all other cases studied, total protein, albumin and globulin levels were near normal. Blood urea nitrogen and serum creatinine level were elevated in case of leptospirosis suggested renal damage.

Serum activities of Alkaline phosphatase and Alanine amino transferase showed mild to marked elevation in all liver affections.

Protein content of ascitic fluid varied from 0.2 g/dl in cirrhosis to 4.2 g/dl in the case of portal hypertension.

Ultrasound guided biopsy helped in the confirmation of lymphosarcoma, fatty infiltration and chronic active hepatitis. Histological changes were studied in case of leptospirosis and cirrhosis.

Important histological changes noticed in lymphosarcoma were disruption of normal hepatic architecture and diffuse infiltration with hyperchromatic cells suggestive of lymphosarcoma. In leptospirosis, ultrasound revealed hypoechoic liver with histological changes suggestive of acute hepatitis, that is venous and sinusoidal congestion and atrophy of hepatocytes. Liver was diffusely hyperechoic in both cirrhosis and fatty infiltration. In cirrhosis, fibrous tissue proliferation and pseudolobulation were the prominent histological changes whereas vacuolation and kupffer cell reaction were the major histological changes in latter case. Normal to hypoechoic liver in chronic active hepatitis corresponded to hepatic necrosis with dense infltrate of lymphocytes.

The most important limitation of ultrasonography noticed in this study was its inability to identify the diffuse changes noticed in hepatic diseases for which biopsy was essential.

From this study it was concluded that

1. Ultrasonography could be used for identification and classification of lesions in liver.

2. Ultrasonography was more reliable in identifying focal changes in hepatic parenchyma.

3. Diffuse lesions identified in ultrasonography should be confirmed with ultrasound guided biopsy.

4. Correlation existed between ultrasound appearance and histological changes.

5. Ultrasonography alone was an imperfect tool in diagnosing hepatic disorders but, provided accurate diagnosis in conjunction with clinico-pathological and biopsy findings.

6. Presentation of hepatopathies at a more advanced stage of disease and non co-operation from part of owners were the major constraints in the clinical management of hepatic disorders.

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ULTRASONOGRAPHIC EVALUATION OF CANINE HEPATIC DISORDERS

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ABSTRACT

Study on 'Ultrasonographic evaluation of canine hepatic disorders' was conducted on forty eight dogs to evaluate the utility of ultrasonography as a tool for early and better diagnosis of liver diseases and classify liver diseases based on ultrasonographic findings.

Parameters studied were history and clinical signs, physical examination, ultrasonography of liver, ultrasound guided biopsy, haematology and serum biochemistry, post-mortem examination and histopathology.

Specific clinical signs noticed were ascites and icterus. Physical examination revealed abdominal distension, yellowish discolouration of skin and hepatomegaly. Primary liver disorders identified by ultrasound scans were haematoma, lymphosarcoma, hepatitis due to leptospirosis, cirrhosis, fibrosis, fatty infiltration, chronic active hepatitis, portal hypertension and portosystemic shunt. Secondary changes noticed in liver were due to pyometra, proctitis and cholecystitis, hepatic calcification in chronic renal failure and passive venous congestion in right sided heart failure.

Ultrasound guided biopsy confirmed lymphosarcoma, fatty infiltration and chronic active hepatitis. Histological changes in leptospirosis were chronic venous and sinusoidal congestion whereas in cirrhosis, fibrosis and pseudolobulation were the prominent histological change.

Alkaline phosphatase and Alanine aminotransferase levels were above normal values in all the cases studied.

From the present study it was inferred that ultrasonography was a valuable tool for diagnosing hepatopathies along with clinico-pathological and ultrasound guided biopsy.