

Volume 5 Issue 1 January 2023

# Correlation of Clinicopathological and Ultrasonographic Findings for Diagnosis and Prognosis of Liver Affections in Dogs

# Sameeksha Koundal<sup>1</sup>, Kuldip Gupta<sup>1\*</sup>, Jitender Mohindroo<sup>2</sup>, Shashi Kant Mahajan<sup>2</sup>, Swaran Singh Randhawa<sup>3</sup> and Amarjit Singh<sup>1</sup>

<sup>1</sup>Department of Veterinary Pathology, Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, Punjab, India

<sup>2</sup>Department of Veterinary Surgery and Radiology, Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, Punjab, India

<sup>3</sup>Department of Teaching Veterinary Clinical Complex, Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, Punjab, India

\*Corresponding Author: Kuldip Gupta, Department of Veterinary Pathology, Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, Punjab, India.

DOI: 10.31080/ASVS.2022.05.0587

# Abstract

The present study was undertaken on 83 dogs suffering from abdominal affections out of which 22 dogs having lesions in liver were included. Detailed history and signalment of each dog were recorded. Blood was collected from each dog for haematobiochemical analysis and ultrasonography of each dog was performed and ultrasound was done to detect lesions in liver. Ultrasonography guided FNAB and core biopsy samples were collected. Maximum incidence was recorded in Labrador retriever, male dogs and median age was recorded as 6 years in cases of liver affections. The lesions were classified as neoplastic and non- neoplastic lesions on the basis of cytology and histopathological findings. Neoplastic lesions were more common which included nine cases of hepatocellular carcinoma, four cases of cholangiocellular carcinoma, one case each of metastatic sarcoma and cystadenoma, whereas, non- neoplastic lesions included four cases of fatty change and two cases of hepatitis. Correlation between haematobiochemical alterations, cytological, histopathological and ultrasonographic findings was carried out for diagnosis and prognosis of liver affections. High correlation (95.21%), specificity (85.71%) and positive predictive value (93.8%) were observed between ultrasonographic and cytological and/ or histopathological diagnosis, whereas moderate correlation (42.86%), specificity (60%) and predictive value (60%) were observed between cytological and histopathological diagnosis of liver affections. In addition, median survival with and without censoring was recorded as 39.00 days and 109.00 days, respectively. Ultrasound guided core biopsy was found to be more effective in diagnosing liver affections as compared to ultrasound guided FNAB. From the study it was concluded that although ultrasonography can detect the lesions in liver, but for confirmation of diagnosis we need ultrasound FNAB and core biopsy.

Keywords: Core Biopsy; Cytology; Dog; Hepatocellular Carcinoma; Histopathology; Liver; Liver Tumors; Ultrasonography

## Abbreviations

HCC: Hepatocellular Carcinoma; AST: Aspartate Aminotransferase; ALT: Alanine Transaminase; ALKP: Alkaline Phosphatase; GGT: *Gamma-Glutamyl* Transferase; USG: Ultrasonography; IHC: Immunohistochemistry and FNAB: Fine Needle Aspiration Biopsy

#### Introduction

Dogs suffering from liver affections usually show non-specific clinical signs so they pose a diagnostic challenge for veterinary clinicians. Liver affections are difficult to diagnose on the basis of clinical signs, hematology or abdominal X-rays, but can be easily detected using abdominal ultrasound [1,2]. Cytology and histopathology have been used for differentiating neoplastic and non-neoplastic liver affections and differentiating type of lesions [3,4]. In dogs and humans liver affections are classified on basis of histopathological findings. Hepatic neoplasms in dogs account for 1.5% of all canine tumors [5]. Tumors derived from hepatocytes are either hepatocellular adenomas or carcinomas (HCC), tumors of bile duct epithelium are classified as biliary adenoma or cholangiocarcinoma, whereas, stromal cells tumors as sarcomas and neuroendocrine cells tumors as neuroendocrine carcinomas [6]. The

**Citation:** Kuldip Gupta, *et al.* "Correlation of Clinicopathological and Ultrasonographic Findings for Diagnosis and Prognosis of Liver Affections in Dogs". *Acta Scientific Veterinary Sciences* 5.1 (2023): 58-68.

Received: September 20, 2022 Published: December 19, 2022 © All rights are reserved by Kuldip Gupta., *et al.*  present work was undertaken to study clinicopathological features of liver affections in dogs and to correlate the findings for proper diagnosis and to access their prognosis.

#### **Material and Methods**

This communication is a part of research work on 83 dogs suffering from abdominal affections presented to the Department of Teaching Veterinary Clinical Complex, Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, out of these 83 cases, 22 dogs were having lesions in liver and were included in this communication. All the dogs having lesions in liver showed non-specific signs but showed lesions during ultrasonography. The study was conducted under the guidelines of the Institutional Animal Ethics Committee at Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana.

Clinical examination included signalment, palpation and hemato-biochemical analysis including complete blood count and serum biochemical analysis. Cases were followed up to one year and median survival time was calculated to ascertain the prognosis.

The standard abdominal B-mode ultrasonography was performed by surgeon in all 22 cases to analyze liver echotexture and heterogeneity. In addition, other parameters noted were whether the lesions were single or multiple, had regular or irregular margins and size of the tumor (Table 1).

|                                | Hyperechoic<br>lesions | Hypoechoic<br>lesions | Mixed<br>echotexture | Free<br>fluid |
|--------------------------------|------------------------|-----------------------|----------------------|---------------|
| Hepatocellular<br>carcinoma    | 3                      | 1                     | 5                    | 2             |
| Cholangiocellular<br>carcinoma | 2                      | -                     | 2                    | -             |
| Cystadenoma                    | -                      | 1                     | -                    | -             |
| Metastatic<br>sarcoma          | -                      | 1                     | -                    | 1             |
| Fatty changes                  | 1                      | 2                     | 1                    | -             |
| Hepatitis                      | 1                      | 1                     | -                    | 1             |
| Inappropriate<br>sample        | -                      | 1                     | -                    | -             |

| Table 1: Ul | trasonograpl | hic features | of different t | ypes |
|-------------|--------------|--------------|----------------|------|
|             | of liver aff | ections in d | ogs.           |      |

For cytological examination, sixteen samples were collected by ultrasound guided FNAB and in two cases impression smears were prepared from liver and other organs showing lesions after postmortem examination. The cytological smears were stained by Leishman's staining method and analyzed. For histopathological examination nine tissue samples were collected by core biopsy and after postmortem examination. The samples were fixed in 10% neutral buffered formalin and processed for routine paraffin sectioning and stained with hematoxylin and eosin method. The liver affections were classified into different categories on basis of cytological and histopathological findings.

The data was statistically analyzed using IBM SPSS software (version 20.0) and independent *t*-test was to analyze the changes in haemato-biochemical parameters. In addition, correlation, sensitivity and predictive values between cytological and histological diagnosis were calculated as by [7] and Kaplan-Meier method was used to determine median survival time.

# **Results and Discussion**

# **Clinical findings**

Out of twenty two cases, thirteen cases were observed in males and nine cases were observed in female dogs. According to previous workers, liver tumors were most common in males because of higher IL-6 levels compared to females and IL-6 was found to cause development of liver tumors and its production was inhibited by estrogens, therefore decreasing chances of liver tumors in cases of females [8,9]. The median age was six years with a range of 2-11 years and median weight was 28.8 kg with a range of 13 kg-51 kg. Breed- wise maximum occurrence was recorded in Labrador retriever, followed by German shepherd, Rottweiler and Pomeranian. Minimum occurrence was recorded in Golden Retriever, American Pit bull Terrier, Pug, Mongrel, Saint, St. Bernard, Dogo argentino (Daschund) and Dalmation having one case each. Earlier workers have alsoreported higher incidence of liver affections in Labrador retriever [10,11].

Clinical signs were mainly non-specific; however important clinical signs observed were anorexia, vomition, bloody faeces, abdominal distention, lethargy, weakness, icterus, ventral edema of abdomen and sudden weight loss. Similar findings have been reported in dogs suffering from liver affections by earlier workers [2,12]. On abdominal palpation masses were palpated in two cases.

**Citation:** Kuldip Gupta, *et al.* "Correlation of Clinicopathological and Ultrasonographic Findings for Diagnosis and Prognosis of Liver Affections in Dogs". *Acta Scientific Veterinary Sciences* 5.1 (2023): 58-68.

#### Haematobiochemical findings

Hematological analysis revealed anemia, low PCV and TEC along with neutrophillic leukocytosis and relative neutrophilia (Table 2). Statistically significant differences was recorded in mean values of Hb, TLC, platelets, PCV and neutrophils, eosinophils and lymphocyte count. Similar findings have been documented by earlier workers in dogs suffering from liver affections [10,11]. They were of the opinion that anemia in liver affections might be due to its dysfunction thus hampering the production of erythropoietin and extra factors needed for erythropoiesis. Other causes of anemia may be increased fragility of RBC's due to elevated levels of bile acids, decreased portal blood flow, decreased erythrocyte survival time, improper bone marrow responses, inappetence or anorexia causing decreased nutrient uptake and decreased accessibility of micronutrients to the liver, whereas, neutrophilia may be due to necrosis and inflammation of liver [13,14].

| S. No. | Hematological parameters                       | Mean ± SEM             | Range<br>observed  | Reference<br>value |
|--------|--|------------------------|--------------------|--------------------|
| 1      | Hb (g/dL)                                      | 8.6 ± 0.88*            | 3.8-18.1           | 12-18              |
| 2      | PCV (%)  | 26.1 ± 2.47**          | 15.1-47.4          | 37-55              |
| 3      | TEC ×10 <sup>6</sup> /µL                       | 4.39 ± 0.5             | 1.52-8.54          | 5.5-9.5            |
| 4      | TLC /µL  | 30622.4 ±<br>4392.21** | 1540-<br>83230     | 6000-17000         |
| 5      | Relative Neutrophil<br>count (%)               | 89.2 ± 1.9*            | 68-98              | 60-70              |
|        | Absolute Neutrophil<br>count<br>(/μL of blood) | 29321.5 ±<br>5019.1**  | 1047.2-<br>81565.4 | 3000-11500         |
| 6      | Relative Lymphocyte<br>count (%)               | 10.1 ± 1.9             | 2-32               | 12-30              |
|        | Absolute Lymphocyte<br>count<br>(/μL of blood) | 1799.5 ±<br>241.2**    | 106.2-<br>5292     | 1000-4800          |
| 7      | Relative Eosinophil<br>count (%)               | 4 ± 1.6**              | 2-10               | 3-10               |
|        | Absolute Eosinophil<br>count<br>(/μL of blood) | 706.4 ±<br>280.12**    | 260-1510           | 100-1250           |
| 8      | Platelets ×10 <sup>3</sup> /µL                 | 359.72 ±<br>72.64**    | 66-1330            | 200-500            |

# Table 2: Hematological findings in dogs suffering from liver affections.

Values with superscripts differ significantly within themselves \*\*(P < 0.01) and \* (P < 0.05) level of 'significance and (P > 0.05) level of non significance on the basis of one sample T Test. Serum chemistry analysis revealed, increased total protein, bilirubin, AST, ALT, ALKP, GGT, phosphorus and hypoalbuminemia (Table 3). Statistically significant differences were recorded in mean values of ALT and ALKP. Comparable findings have been documented by previous workers in dogs suffering from liver affections [14,15]. Increased serum levels of ALT and AST indicate hepatocellular injury and help in monitoring clinical progress [16]. In addition, sometimes systemic diseases and certain drugs lead to increase in serum levels and it becomes difficult to decide whether the liver enzyme have increased due to primary or secondary liver disease [17]. According to [18], hypoalbuminemia in liver diseases may be due to significant destruction of hepatocytes.

| S. No. | Serum chemistry<br>parameters | Mean ± SEM     | Min. and<br>max. range<br>observed | Reference<br>value |
|--------|-------------------------------|----------------|------------------------------------|--------------------|
| 1      | Total Protein<br>(g/dL)       | 17.2 ± 10.8    | 3.8-190                            | 5.4-7.1            |
| 2      | Albumin (g/dL)                | 2.5 ± 0.24     | 1.5-4.9                            | 2.6-3.3            |
| 3      | Bilirubin (mg/dL)             | 5.7 ± 3.3      | 0.2-25.1                           | 0.1-0.6            |
| 4      | SGOT/AST (U/L)                | 168.8 ± 98.3   | 34-850                             | 8.9-49             |
| 5      | SGPT/ALT (U/L)                | 206.83 ± 82.7* | 18-1400                            | 8.2-57             |
| 6      | ALKP (U/L)                    | 661.3 ± 197.1* | 63-1700                            | 10.6-101           |
| 7      | GGT (U/L)                     | 43.5 ± 26.3    | 0.4-331                            | 1-9.7              |
| 8      | BUN (mg/dL)                   | 25.7 ± 7.4     | 1.5-98                             | 8.8-26             |
| 9      | Creatinine (mg/<br>dL)        | 0.5 ± 0.5      | 0.5-7                              | 0.5-1.6            |
| 10     | Phosphorus (mg/<br>dL)        | 6.3 ± 1.3      | 2.3-15.8                           | 2.6-6.2            |
| 11     | Glucose (mg/dL)               | 82 ± 9.3       | 19-134                             | 62-108             |

# Table 3: Serum chemistry findings in dogs suffering from liver affections.

Values with superscripts differ significantly within themselves \*\*(P < 0.01) and \* (P < 0.05) level of 'significance and (P > 0.05) level of non significance on the basis of one sample t test.

# Clinicopathological findings in dogs suffering from liver affections

The different affections diagnosed in liver on the basis of cytological and histopathological findings have been presented in table 4. Out of these cases, sixteen cases showed neoplastic and six cases showed non-neoplastic lesions. Neoplastic lesions included nine cases of HCC, four cases of cholengiocellular carcinoma, one case

**Citation:** Kuldip Gupta, *et al.* "Correlation of Clinicopathological and Ultrasonographic Findings for Diagnosis and Prognosis of Liver Affections in Dogs". *Acta Scientific Veterinary Sciences* 5.1 (2023): 58-68.

each of cystadenoma and metastatic sarcoma. The findings of our study were similar to earlier workers [2,4]. Non-neoplastic lesions comprised of four cases of fatty changes and two cases of hepatitis. In one case diagnosis could not be made of the basis of cytology due to inappropriate sampling. The detailed ultrasonographic, cytologic and histopathologic findings in each type of lesions are presented below.

| Diagnosis number of cases   |   |  |  |
|-----------------------------|---|--|--|
| Neoplastic (n = 16)         |   |  |  |
| Hepatocellular carcinoma    | 9 |  |  |
| Cholangiocellular carcinoma | 4 |  |  |
| Cystadenoma                 | 1 |  |  |
| Metastatic sarcoma          | 1 |  |  |
| Inappropriate sample        | 1 |  |  |
| Non-neoplastic (n = 6)      |   |  |  |
| Fatty changes               | 4 |  |  |
| Hepatitis                   | 2 |  |  |

**Table 4:** Diagnosis of liver affections in dogs on the basis of cytology and or histopathology.

#### **Neoplastic lesions**

#### Hepatocellular carcinoma

Nine cases of hepatocellular carcinoma (HCC) were observed. Ultrasonographic examination was performed in all the nine cases of HCC, out of which four cases showed multiple hyperechoic lesions, one case showed multiple hypoechoic lesions, one case showed multiple hypoechoic and hyperechoic lesion in which only right lobe of liver was affected, two cases showed multiple heterogeneous or mixed echotexture lesions and one case showed multiple hyperechoic to anechoic lesion all over liver. Free fluid in abdomen was recorded in two cases (Figure 1). Similar type of ultrasonographic findings has been reported in dogs having HCC [15,19,20].

Cytological examination was performed in seven cases of HCC. Cytologically HCC revealed presence of neoplastic hepatocytes arranged individually or in acinar pattern along with anisocytosis and anisokaryosis, increased nuclear cytoplasmic ratio, round to ovoid nuclei, variable number of prominent nucleoli and coarse chromatin. In addition, basophilic cytoplasm with sharply demar-



**Figure 1:** Ultrasound, of liver from a case of hepatocellular carcinoma showing multiple irregular sized nodules.

cated clear lipid globules were also observed (Figure 2). Similar type of cytological findings has been reported in dogs having HCC [19,21]. Previous workers reported that impression smears from liver biopsy showed greater accuracy compared to USG guided FNAB samples [22].



**Figure 2:** USG guided FNAB of liver showing varying sized individual hepatocytes and coarse chromatin suggestive of hepatocellular carcinoma. Leishman stain x 100X.

Histopathology could be performed in one case of HCC as no tissue samples were submitted in six cases. One dog was euthanized, and postmortem examination showed diffuse, varying sized, round, yellowish to red colored, and multiple to coalescing nodules (Figure 3). Similar gross findings in cases of HCC in dogs have been reported [23].



**Figure 3:** Liver, Hepatocellular carcinoma showing varying sized raised nodules distributed throughout the liver on post-mortem examination following euthanasia.

Microscopically it was characterized by proliferation of neoplastic hepatocytes arranged in irregular trabeculae and separated by several irregular vascular channels, cavernous spaces and dilated sinusoids. Moderate anisokaryosis and anisocytosis with prominent nucleoli and pale and vacuolated cytoplasm due to lipid were recorded. Blood pools of different sizes were present within liver parenchyma causing irregular dilation of sinusoidal like blood spaces (Figure 4). On the basis of histological changes the case was diagnosed as peliotic HCC. The microscopic findings coincided with earlier reports of HCC in dogs [19,21,23]. Peliotic lesions have been reported in human livers due to hepatocellular necrosis, obstruction in outflow of blood from the liver at hepatic sinusoid-venule junction or direct injury in sinusoidal wall by toxic substances or medications [24].

#### Cholangiocellular carcinoma

Four cases of cholangiocellular carcinoma were diagnosed. ultrasonographic examination of two cases showed presence of multiple hyperechoic lesions and two cases showed multiple het-



**Figure 4:** Microphotograph of liver showing proliferation of neoplastic hepatocytes separated by varying sized blood channels devoid of endothelial limning suggestive of Peliotic hepatocellular carcinoma. H and E x 20X

erogeneous or mixed echotexture lesion on ultrasonographic examination (Figure 5). Similar type of ultrasonographic findings has been reported in dogs having cholangiocellular carcinoma [25,26].



Figure 5: Ultrasound of liver from a case of cholangiocellular carcinoma - Multiple nodules in liver with large amount of echogenic free fluid.

Cytological examination was performed in all the four cases of cholangiocellular carcinoma. Cytological examination revealed increased cellularity with round neoplastic cells arranged in acinar arrangement with moderate anisocytosis and anisokaryosis, increased N:C ratio, coarse chromatin and oval nuclei with prominent, single basophilic nucleoli along with increased cellularity and minimal blue cytoplasm (Figure 6). Similar type of cytological findings has been reported in dogs having cholangiocellular carcinoma [26].



**Figure 6:** USG guided FNAB of liver from a case of cholangiocellular carcinoma showing round to oval neoplastic cells arranged in acinar pattern showing increased N:C ratio, coarse chromatin and prominent nucleoli. Leishman stain x 100X.

One dog was euthanized and post mortem examination showed presence of dark red, multiple raised nodules all over the liver and blood was oozing out from cut section. Similar gross findings in cases of cholangiocellular carcinoma in dogs have been reported [27].

Histopathology was performed in three cases of cholangiocellular carcinoma (two samples submitted by core biopsy and one sample collected during post mortem) and no tissue samples were submitted in one case. Microscopically it was characterized by presence of neoplastic cells arranged in small, irregular gland like structures, disrupting the normal liver parenchyma and margins of tumor invading hepatic parenchyma along with hepatic necrosis



**Figure 7:** Microphotograph of the above case collected by ultrasound guided core biopsy showing proliferation of bile duct cells arranged in varying sized tubules and acini separated by fibrous stroma along with hemorrhages. H and E x 10X.

(Figure 7). These neoplastic cells were also separated by fibrous stroma or connective tissue. The microscopic findings coincided with earlier reports of cholangiocellular carcinoma in dogs [28,29].

#### Cystadenoma

One case of cystadenoma was diagnosed, and on ultrasonographic examination presence of mixed echotexture and congested liver along with multiple hypoechoic nodules seen throughout parenchyma of one hepatic lobe (Figure 8). Similar type of ultrasonographic findings has been reported in dogs having cystadenoma [30].

No cytological sample was submitted and histopathology was performed on sample by core biopsy. Microscopically it was characterized by presence of multiple cysts lined by single stratified cuboid or slightly flattened cells layers of neoplastic biliary cells. These proliferating cells looked similar to normal biliary epithelium. In addition, papillary projections were seen within lumen of cyst (Figure 9). The microscopic findings coincided with earlier reports of cystadenoma in dogs and biliary cystadenomas were reported as rare cystic tumors arising from liver and less commonly in extrahepatic biliary system in cases of dogs [30-31].



Figure 8: Ultrasound of liver from case of cystadenoma showing multiple hypoechoic nodules throughout liver parenchyma in single hepatic lobe.



**Figure 9:** Microphotograph of cystadenoma collected by ultrasound guided core biopsy of liver showing proliferation of bile duct epithelium giving rise to multiple cysts with papillary projections. H and E x 20X.

#### Metastatic sarcoma

One case of metastatic sarcoma was diagnosed and ultrasonographic examination revealed presence of enlarged liver with multiple hypoechoic lesions. Large amount of echogenic free fluid was also recorded. Similar type of ultrasonographic findings has been reported in dogs having metastatic sarcoma [32]. Cytological examination showed presence of mesenchymal cells individually or in aggregates having round nuclei and basophilic cytoplasm with tapered end. Coarse chromatin and multiple nucleoli were also noted. Similar type of cells were seen in spleen indicating metastasis (Figure 10). Similar type of cytological findings has been reported in dogs and humans having metastic hepato-splenic sarcoma [33,34]. Histopathology could not be performed as biopsy sample was not submitted. Liver has been reported as most common site for hematogenous metastases and 30% of tumors have been reported to metastasize to liver by hematogenous [20]. **Non- neoplastic affections** 



**Figure 10:** USG guided FNAB of liver showing elongated neoplastic cells having basophilic cytoplasm, coarse chromatin and neutrophilic infiltration suggestive of metastatic sarcoma as similar cells were seen in aspirates from spleen. Leishman stain x 100X.

#### **Fatty change**

Four cases of fatty changes were diagnosed and ultrasonographic examination revealed presence of multiple hyperechoic and hypoechoic lesions to mixed echotexture with irregular margin of liver. Similar type of ultrasonographic findings have been reported in dogs having fatty change in liver [35].

Cytological examination showed presence of clear vacuoles of varying size in the cytoplasm of hepatocytes. Nuclei were eccentrally placed, coarse chromatin with prominent basophilic nucle-

**Citation:** Kuldip Gupta, *et al.* "Correlation of Clinicopathological and Ultrasonographic Findings for Diagnosis and Prognosis of Liver Affections in Dogs". *Acta Scientific Veterinary Sciences* 5.1 (2023): 58-68.

oli along with neutrophil infiltration. Similar type of cytological findings have been reported in dogs having fatty changes in liver [36,37].

Histopathology was performed in three cases of fatty changes collected by core biopsy. Microscopically it was characterized by presence of diffuse intracytoplasmic vacuolation of hepatocytes with displaced nucleus at periphery. In addition, mild coagulative necrosis, fibrosis and congestion were also recorded. The microscopic findings were similar with earlier reports of fatty change in liver of dogs [37].

#### Hepatitis

One case of hepatitis with fatty change was diagnosed and ultrasonographic examination showed presence of irregular liver margin along with multiple hyperechoic nodules. Similar type of ultrasonographic findings has been reported in dogs having hepatitis [38].

Cytological examination revealed markedly distended hepatocytes having large clear vacuoles in the cytoplasm and the nucleus was pushed towards periphery. Histopathology could not be performed as biopsy sample was not submitted. Similar type of cytological findings has been reported in dogs having hepatitis with fatty changes in liver [37,39]. According to earlier workers, biopsy specimens showing necrosis and hepatitis could have been collected from that area of neoplasm which showed inflammation or necrosis however cytologic examination might detect correct diagnosis [39].

Another case of hepatitis with coagulative necrosis was diagnosed and ultrasonographic examination showed presence of multiple hypoechoic regions in liver along with large amount of free fluid. Similar type of ultrasonographic findings has been reported in dogs having hepatitis with coagulative necrosis [38].

Microscopically the architecture of the liver was maintained but the structural details were lost. There was cytoplasmic appeared eosinophilic along with varying changes in the nuclei of the hepatocytes. The nuclei were lost but few leftover nuclei showed granular debris. Sinusoids were also congested with neutrophilic infiltration in liver parenchyma. The microscopic findings coincided with earlier reports of necrotic hepatitis in dogs [40]. Previous workers reported that sometimes material aspirated around the liver tumor show reactive and proliferative changes and aspirated sample from center of large tumor show degenerative and necrotic changes [41]. Thus for proper aspiration needle should pass through the complete liver mass. So in our study it's possible that sample would have been collected from the center of the nodules.

#### Survival analysis in dogs suffering from liver affections

The overall survival time of dogs died from liver affections ranged from one day to 292 days and median survival of dogs without and with censoring was 39.00 days and 109.00 days respectively. Contrary to our study, median survival time of 1336, 200 and 707 days respectively was recorded in dogs suffering from hepatic tumors [3,42,43].

# Correlation between ultrasonographic, cytological and histopathological diagnosis

Moderate correlation (42.86%), specificity (60%) and predictive value (60%) were observed between cytological and histopathological diagnosis of liver affections, whereas, high correlation (95.21%), specificity (85.71%) and positive predictive value (93.8%) were observed between ultrasonographic and cytological and/or histopathological diagnosis of liver affections (Table 5). Earlier workers reported that although ultrasound helped in determining the incidence of neoplasm, visualize the lesion, and evaluate changes in surrounding organs but it could not distinguish the neoplastic and non-neoplastic lesions [19,44].

On the contrary, earlier workers reported high sensitivity (93%) and specificity (96%) of cytology in diagnosis of inflammatory lesions of liver of dogs [39]. Cytology has been reported to be more sensitive in diagnosing neoplasia like round cell tumor (60%), than other tumors including non-HCC reported (54.5%) and HCC (34.8%) [45], whereas, poor correlation has been between histopathologic and cytologic diagnosis of liver affections in dogs [27].

| Case No. | Ultrasonographic diagnosis        | Cytological diagnosis                        | Histopathological diagnosis |
|----------|-----------------------------------|--|-----------------------------|
| 1        | Suspected Tumor (True Positive)   | Hepatocellular carcinoma (True Positive)     | Hepatocellular carcinoma    |
| 2        | Suspected Tumor (True Positive)   | Hepatocellular carcinoma                     | Not performed               |
| 3        | Suspected Tumor (True Positive)   | Hepatocellular carcinoma                     | Not performed               |
| 4        | Suspected Tumor (True Positive)   | Hepatocellular carcinoma                     | Not performed               |
| 5        | Suspected Tumor (True Positive)   | Hepatocellular carcinoma                     | Not performed               |
| 6        | Suspected Tumor (True Positive)   | Not performed                                | Inappropriate sample        |
| 7        | Suspected Tumor (True Positive)   | Chronic active inflammation (False negative) | Cholangiocellular carcinoma |
| 8        | Suspected Tumor (True Positive)   | Hepatocellular carcinoma (True positive)     | Not performed               |
| 9        | Suspected Tumor (False Positive ) | Not performed                                | Necrosis Hepatitis          |
| 10       | Suspected Tumor (True Positive)   | Inappropriate sample (False negative)        | Cholangiocellular carcinoma |
| 11       | Suspected Tumor (True Positive)   | Not performed                                | Cholangiocellular carcinoma |
| 12       | Suspected Tumor (True Positive)   | Metastatic sarcoma                           | Not performed               |
| 13       | Suspected Tumor (True Positive)   | Hepatocellular carcinoma                     | Not performed               |
| 14       | Suspected Tumor (True Positive)   | Hepatocellular carcinoma                     | Not performed               |
| 15       | Suspected Tumor (True Positive)   | Cholangiocellular carcinoma (True Positive)  | Cholangiocellular carcinoma |
| 16       | Suspected Tumor (True Positive)   | Not performed                                | Cyst adenoma                |
| 17       | Suspected Tumor (True Negative)   | Purulent hepatitis (False positive)          | Fatty change                |
| 18       | Suspected Tumor (True Negative)   | Fatty change (True Positive)                 | Fatty change                |
| 19       | Suspected Tumor (True Negative)   | Hepatitis with fatty change                  | Not performed               |
| 20       | Suspected Tumor (True Negative)   | Fatty change                                 | Not performed               |
| 21       | Suspected Tumor (True Negative)   | Fatty change                                 | Not performed               |
| 22       | Suspected Tumor (True Negative)   | Early carcinoma (False positive)             | Fatty change                |

Table 5: Correlation between ultrasonographic, cytological and histopathological diagnosis of liver affections.

### Conclusions

From the present study it was concluded that due to non-specific clinical signs it is difficult to diagnose liver affections in dogs, so a comprehensive approach involving ultrasonography, hematology, serum chemistry, cytology and histopathology is required for final diagnosis. Although we can detect the lesions in liver on ultrasonography, but for confirmation of diagnosis we need ultrasound FNAB and core biopsy. High correlation, specificity and positive predictive value were observed between ultrasonographic and cytological and/or histopathological diagnosis, and a moderate correlation between cytology and histopathology findings was observed for the diagnosis of liver affections. Ultrasound guided core biopsy was found to be more effective in diagnosing liver affections as compared to ultrasound guided FNAB.

# **Conflict of Interest**

All authors declare that they have no conflict of interest.

# **Bibliography**

1. Kumar V., *et al.* "Diagnostic imaging of canine hepatobiliary affections: a review". *Veterinary Medicine International* 2012 (2012): 672107.

66

- 2. Leela-Arporn R., *et al.* "Predictive Factors of Malignancy in Dogs with Focal Liver Lesions Using Clinical Data and Ultrasonographic Features". *The Journal of Veterinary Medical Science* 81.5 (2019): 723-729.
- 3. Kinsey JR., *et al.* "Factors Associated with Long-Term Survival in Dogs Undergoing Liver Lobectomy as Treatment for Liver Tumors". *The Canadian veterinary Journal. La Revue Veterinaire Canadienne* 56.6 (2015): 598-604.
- Burti S., *et al.* "Contrast-Enhanced Ultrasound Features of Malignant Focal Liver Masses in Dogs". *Scientific Reports* 10.1 (2020): 6076.

- Nemoto Y., *et al.* "Identification of cancer stem cells derived from a canine lung adenocarcinoma cell line". *Veterinary Pathology* 48 (2011): 1029-1034.
- Balkman C. "Hepatobiliary Neoplasia in Dogs and Cats". The Veterinary clinics of North America. Small Animal Practice 39.3 (2009): 617-625.
- 7. Gerstman BB and Cappucci DT. "Evaluating the reliability of diagnostic test results". *Journal of the American Veterinary Medical Association* 188.3 (1986): 248-251.
- Naugler WE., et al. "Gender Disparity in Liver Cancer Due to Sex Differences in MyD88-Dependent IL-6 Production". Science (New York, N.Y.) 317.5834 (2007): 121-124.
- Yeh SH and Chen PJ. "Gender Disparity of Hepatocellular Carcinoma: The Roles of Sex Hormones". Oncology 78.1 (2010): 172-179.
- 10. Sharma MC., *et al.* "Liver- structure, disorders, diagnosis and therapeutic management". *IVRI Publication* (2001).
- Chikazawa S., *et al.* "Change in Serum Ferritin Concentration in Experimentally Induced Anemia of Chronic Inflammation in Dogs". *The Journal of Veterinary Medical Science* 75.11 (2013): 1419-1426.
- 12. Selmic LE. "Hepatobiliary Neoplasia". *The Veterinary Clinics of North America. Small Animal Practice* 47.3 (2017): 725-735.
- 13. Elhiblu MA., *et al.* "Clinico-Hemato-Biochemical Profile of Dogs with Liver Cirrhosis". *Veterinary World* 8.4 (2015): 487-491.
- Murikipudi NS., *et al.* "Prevelence of hepatic disorders among different breeds, sex, and age groups of dogs". *International Journal of Science, Environment and Technology* 6.5 (2017): 20-25.
- Liptak JM., et al. "Massive Hepatocellular Carcinoma in Dogs: 48 Cases (1992-2002)". Journal of the American Veterinary Medical Association 225.8 (2004): 1225-1230.
- 16. Tennant BC and Center SA. "Hepatic Function". *Clinical Biochemistry of Domestic Animals*. Elsevier (2008): 379-412.
- Nelson RW and Couto CG. "Diagnostic tests for the hepatobiliary system". Maryland Heights, Mo, USA: Mosby (1998): 487-509.

- Bigoniya P., *et al.* "A comprehensive review of different liver toxicants used in experimental pharmacology". *International Journal of Pharmaceutical Science and Drug Research* 1 (2009): 124-135.
- 19. Vatnikov Y., *et al.* "Size of canine hepatocellular carcinoma as an adverse prognostic factor for surgery". *Journal of Advanced Veterinary and Animal Research* 7.1 (2020): 127-132.
- Teshima T., et al. "Hepatocellular Carcinoma in a Young Dog". The Canadian Veterinary Journal. La Revue Veterinaire Canadienne 54.9 (2013): 845-848.
- 21. Masserdotti C., *et al.* "Characterization of Cytoplasmic Hyaline Bodies in a Hepatocellular Carcinoma of a Dog". *Research in Veterinary Science* 96.1 (2014): 143-146.
- Roth L. "Comparison of Liver Cytology and Biopsy Diagnoses in Dogs and Cats: 56 Cases". *Veterinary Clinical Pathology* 30.1 (2001): 35-38.
- 23. Marconato L., *et al.* "Sorafenib for the Treatment of Unresectable Hepatocellular Carcinoma: Preliminary Toxicity and Activity Data in Dogs". *Cancers* 12.5 (2020): 1272.
- 24. Zafrani ES., *et al.* "Ultrastructural Lesions of the Liver in Human Peliosis. A Report of 12 Cases". *The American Journal of Pathology* 114.3 (1984): 349-359.
- 25. Tanaka T., *et al.* "Computed Tomography Findings in Canine Cholangiocellular Carcinoma'. *Veterinary Record Case Reports* 10.e303 (2022): 1-5.
- Martínez-Sogues L., *et al.* "Hypercalcemia of Malignancy in a Dog Diagnosed with Cholangiocellular Carcinoma". *Topics in Companion Animal Medicine* 35 (2019): 1-5. S1938973618301946.
- 27. Wang KY, *et al.* "Accuracy of ultrasound-guided fine-needle aspiration of the liver and cytologic findings in dogs and cats: 97 cases (1990-2000)". *Journal of the American Veterinary Medical Association* 224.1 (2004): 75-78.
- Sharma V., et al. "Cholangiocellular carcinoma in a labrador bitch: a case report". Haryana Veterinarian 55.2 (2016): 243-244.

**Citation:** Kuldip Gupta, *et al.* "Correlation of Clinicopathological and Ultrasonographic Findings for Diagnosis and Prognosis of Liver Affections in Dogs". *Acta Scientific Veterinary Sciences* 5.1 (2023): 58-68.

- 29. Javanbakht J., *et al.* "Evaluation of metastatic cholangiocarcinoma in a Spitz dog". *Journal of Cancer Science and Therapy* 05.03 (2013): 113-114.
- Moon S-J., et al. "Biliary Cystadenoma in a Maltese Dog: Clinical and Diagnostic Findings". The Journal of Veterinary Medical Science 73.12 (2011): 1677-1679.
- Akkoç A., et al. "Multicentric Intrahepatic Biliary Cystadenoma in a Dog'. Turkish Journal of Veterinary and Animal Sciences 33.2 (2009): 161-164.
- Irausquin RA., et al. "Comparative Evaluation of the Liver in Dogs with a Splenic Mass by Using Ultrasonography and Contrast-Enhanced Computed Tomograph". The Canadian Veterinary Journal. La Revue Veterinaire Canadienne 49.1 (2008): 46-52.
- Boucher LD., *et al.* "Cytology of Angiosarcoma. Findings in Fourteen Fine-Needle Aspiration Biopsy Specimens and One Pleural Fluid Specimen". *American Journal of Clinical Pathology* 114.2 (2000): 210-219.
- Sharma S., *et al.* "Primary Splenic Angiosarcoma with Liver Metastasis: A Rare Neoplasm Diagnosed on Fine-Needle Aspiration Cytology and Cell Block Immunocytochemistry". *Journal of Cytology* 35.2 (2018): 114-116.
- Lessa AS., *et al.* "Ultrasound imaging in an experimental model of fatty liver disease and cirrhosis in rats". *BMC Veterinary Research* 6.6 (2010).
- Zeppa P., *et al.* "Fine Needle Aspiration Biopsy of Hepatic Focal Fatty Change. A Report of Two Cases". *Acta Cytologica* 46.3 (2002): 567-570.
- Soudah B., *et al.* "Overview and Evaluation of the Value of Fine Needle Aspiration Cytology in Determining the histogenesis of Liver Nodules: 14 Years of Experience at Hannover Medical School". *Oncology Reports* 33.1 (2015): 81-87.
- Tantary HA., et al. "Diagnosis and therapeutic studies in ascites in dogs". Veterinary Research 6.6 (2013): 111-114.
- Weiss DJ., *et al.* "Cytologic Evaluation of Inflammation in Canine Liver Aspirates". *Veterinary Clinical Pathology* 30.4 (2001): 193-196.

- 40. Kook PH., *et al.* "Clinical and Histologic Outcome in a Dog Surviving Massive Hepatic Necrosis". *Journal of Veterinary Internal Medicine* 33.2 (2019): 879-884.
- Balani S., *et al.* "Cytomorphological Variables of Hepatic Malignancies in Fine Needle Aspiration Smears with Special Reference to Grading of Hepatocellular Carcinoma". *Journal of Cytology* 30.2 (2013): 116-120.
- 42. Vilkovyskiy IF., *et al.* "Influence of Hepatic Neoplasia on Life Expectancy in Dogs". *Veterinary World* 13.3 (2020): 413-418.
- 43. Moyer J., *et al.* "Factors Associated with Survival in Dogs with a Histopathological Diagnosis of Hepatocellular Carcinoma: 94 Cases (2007-2018)". *Open Veterinary Journal* 11.1 (2021): 144-153.
- Meyer DJ. "The liver. In: Raskin RE, Meyer D, editors. *Canine and feline cytology: a color atlas and interpretation guide"*. 3<sup>rd</sup>. Elsevier; St. Louis, MO (2016).
- 45. Bahr KL., *et al.* "Accuracy of US-Guided FNA of Focal Liver Lesions in Dogs: 140 Cases (2005-2008)". *Journal of the American Animal Hospital Association* 49.3 (2013): 190-196.

**Citation**: Kuldip Gupta, *et al.* "Correlation of Clinicopathological and Ultrasonographic Findings for Diagnosis and Prognosis of Liver Affections in Dogs". *Acta Scientific Veterinary Sciences* 5.1 (2023): 58-68.