Primary Gallbladder Adenocarcinoma in a Cat

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ABSTRACT

Background: Biliary tumors have low incidence in cats and are more common in elderly animals. Hepatobiliary tumors have different classifications and their etiology is difficult to interpret. In most cases, the diagnosis is only possible in advanced stages, which clouds the precursor factors. The late diagnosis is explained by the absence or specificity of clinical manifestations and laboratory changes. The majority of hepatobiliary neoplasms in cats are incidental findings in surgeries or necropsies. This study aimed to report clinical, laboratory, pathological and immunohistochemical results in a feline case of gallbladder adenocarcinoma.

Case: 4-year-old male castrated mix breed cat was admitted at a veterinary clinic with a history of polyuria / polydipsia, anorexia, apathy, jaundice and emesis for 60 days. The animal had been treated in another clinic with silymarin, famotidine and cyanocobalamin, and fed by esophageal tube based on the presumptive clinical diagnosis of cholangitis. The clinical examination revealed jaundice, abdominal pain, weight loss, enlargement of the liver and gallbladder and the presence of a structure in the epigastric region. Based on clinical signs, blood tests (complete blood count and liver enzymes), abdominal ultrasound and thoracic radiography were requested. In the exam results, eosinophilia, bilirubinemia and increased alkaline phosphatase, glutamic pyruvic transaminase and gamma glutamyl transferase were observed, in addition to the presence of icteric and hemolyzed serum. Ultrasound exam revealed thickened and dilated cystic and common ducts, large and thick gallbladder, bile with bile mud, hypoechogenic liver, thickening in the duodenal papilla and enlarged pancreatic and duodenal lymph nodes. There were no alterations in thoracic radiography. Based on the findings, the diagnosis of extrahepatic biliary obstruction was suggested. In celiotomy, dilated gallbladder with a thick blackened wall was observed. Bile puncture was performed for analysis, and total cholecystectomy and removal of obstructive content was performed. The patient suffered cardiorespiratory arrest after the procedure. A fragment of gallbladder and liver were sent to evaluation with histopathology and immunohistochemistry, and results were compatible with gallbladder adenocarcinoma.

Discussion: Hepatobiliary neoplasms in cats are uncommon, apart from lymphoma. The incidence is higher in elderly cats and in males, which did not match the animal in this case. The clinical presentation of the condition is quite nonspecific, corroborating with other reports, in which changes are often only evident in advanced stages of the disease. Laboratory and imaging findings pointed to a biliary tree disorder, and cholangitis was suggested. However, the lack of clinical improvement and examination findings of exams lead to an exploratory laparotomy for better inspection of the organs and identification of possible causes. One of the differential diagnoses that should be considered in cats with extrahepatic biliary obstruction is gallbladder adenocarcinoma, especially in animals with an unsatisfactory response in clinical treatment. Tests such as histopathology and immunohistochemistry are essential for the definitive diagnosis of this neoplasm. Surgical resection is indicated in cases of malignant masses, providing longevity and better quality of life. Hypotension is a common complication in hepatobiliary surgeries, which can result in death.

Keywords: feline, immunohistochemical, liver, neoplasm, surgery.

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INTRODUCTION

Primary gallbladder neoplasms are rare in cats, in which incidence values vary from 1 to 3% among different types [11]. These conditions are more common in elderly animals and breed-related predispositions have not been reported [1].

Hepatobiliary neoplasms are classified as hepatocellular adenomas, hepatocellular adenocarcinomas, cholangiocellular adenomas, cholangiocellular adenocarcinomas or mesenchymal tumors. The cholangiocellular adenocarcinomas include intrahepatic bile duct adenocarcinomas, extrahepatic bile duct adenocarcinomas and gallbladder carcinomas [3].

The etiologies of gallbladder tumors are hard to interpret. In most cases, diagnosis is confirmed only in advance stages of the disease, which hinders the identification of precursor lesions [7]. Late diagnosis may be explained by the clinical presentation that is absent in 50% of cases and is composed by nonspecific signs, such as inappetence, weight loss, lethargy and emesis [6].

Laboratory alterations, such as the increased levels of serum liver enzymes are also unspecific [1]. Hence, half the cases of hepatobiliary tumors in cats are incidental findings of surgeries or necropsies [4].

This study aimed to report clinical, laboratory, pathological and immunohistochemical findings in a case of gallbladder adenocarcinoma in a cat.

CASE

A 4-year-old male castrated mix breed cat was admitted at a veterinary clinic for consultation. According to the owner, the individual presented a history of apathy, polyuria/polydipsia, anorexia, emesis and jaundice for 60 days. Based on a previous diagnosis of cholangitis in another clinic, the individual was being treated orally for 40 days with Silymarin¹ [40 mg/kg, twice a day BID], Famotidine² [1 mg/kg, once a day - SID], and with Cyanocobalamin³ [250 µg, via subcutaneous injections every 7 days]. Patient was being fed through an esophageal tube with hypercaloric pasty feed for 30 days. In the clinical examination, emaciation was observed with a body score of 4 (1-9 scale). In addition, the individual presented jaundice in skin and mucosa, hepatomegaly, abdominal pain and a mass was identified in the epigastric region. In view of the situation, some tests were requested, such as complete blood count, biochemical enzyme activity (direct-bilirubin, free-bilirubin, indirect-bilirubin, Alkaline

Phosphatase - AP, Glutamic Pyruvic Transaminase - GPT and Gamma-Glutamyl Transferase - GGT), total abdominal ultrasound and thoracic radiography.

Leukogram results demonstrated intense eosinophilia (2.624 cells/µL - reference: 0-1,500 cells/µL/L). Serum biochemical profile presented elevated levels of bilirubin (Direct bilirubin 11.4 mg/dL - reference: 0.30 mg/dL; Indirect bilirubin 4.6 mg/dL - reference: 0.0-0.5 mg/dL) liver enzymes (FA 255 mg/dL - reference: 25.0-93.0 mg/dL; TGP 527 mg/dL - reference: 6.0-83.00 mg/dL; GGT 40.00 mg/dL - reference: 1.3-5.10 mg/dL), icteric and hemolyzed serum.

In abdominal ultrasonography, the liver presented reduced and gross echogenicity with no evidence of nodules or cysts. Gallbladder presented severely thick echogenic walls (0.35 cm), anechoic bile with hypoechoic content in the lowest part (biliary mud). Cystic and common ducts were dilated (0.46 cm and 0.29 cm in diameter, respectively) and thick (0.09 cm in both). Duodenum was also thick in the duodenal papilla (0.30 cm) and pancreatic parenchyma was slightly hypoechoic. Lymphadenomegaly was observed in pancreatic and duodenal lymph nodes. Thoracic radiography did not present alterations.

Considering the case and the unsatisfactory therapeutic response, a possible diagnosis of extrahepatic biliary obstruction was considered. Hence, a surgical approach was suggested to perform a bilioenteric bypass. Maintenance fluid therapy was recommended, and pain was managed using Meloxicam⁴ [0.05 mg/kg, SC, SID] for 3 days. The animal was submitted to surgery, in which pre-anesthesia consisted of Methadone⁵ [0.3 mg/kg, SC] and Dexmedetomidine⁶ (3 µ/kg, SC). Isoflurane was used for induction via mask and in maintenance at 2%. In the celiotomy, gallbladder was severely dilated (Figure 1) and manual decompression was not possible. In addition, the left liver duct, cystic duct and common bile duct were ingurgitated. Gallbladder walls were thick and dark. Hyperemia was observed in proximal duodenal walls, omentum and peripancreatic fat, in addition to lymphadenomegaly. Based on these findings, total cholecystectomy was performed. At the end of surgery, a severe hypotension occurred, which resulted in cardiorespiratory arrest. Resuscitation protocol was performed with no success. A bile sample was collected for cytology and bacterial culture. The excised gallbladder and a liver fragment were sent for histopathological analysis. Bacterial culture was negative and cytology

results demonstrated squamous epithelial cells, neutrophils, macrophages and amorphous acellular material with no evidence of bacteria or parasite eggs.

In histopathological examination, marked proliferation of unencapsulated carcinoma epithelial cells which infiltrated the mucosa and submucosa of the gallbladder was observed. Neoplastic cells formed irregular acini and papillary structures, which were sustained by abundant fibrovascular stroma (marked desmoplasia) [Figure 2A]. Multifocal intramural inflammatory infiltrate of neutrophils and eosinophils was observed. Tumoral clots were seen in lymph and blood vessels, in addition to necrotizing vasculitis and fibrin thrombi in submucosal vessels. In the liver fragment adjacent to the gallbladder, discrete fibrosis and marked proliferation of periportal biliary ducts was observed. In addition, a fine diffuse hepatocellular vacuolization was identified.

According to the histopathological findings, gallbladder adenocarcinoma was diagnosed. Samples from these tissues were submitted to immunohistochemical analysis, in which four biomarkers were used in the peroxidase streptavidin-biotin technique with the monoclonal antibodies Anti-Hepatocyte Specific Antigen (Clone OCH1E5)⁷, Anti-CK Pan (Clone AE-1AE3)⁸, Anti-CK7 (Clone OV-TL12/30)⁹ and CK20 (Clone Ks20.8)⁹. Immune expressions for CK Pan and CK7 were observed, which confirmed the diagnosis (Figure 2B,D) [Figure 2C].

DISCUSSION

Hepatobiliary neoplasia in cats is relatively uncommon with the exception of lymphomas [9]. A study conducted by Patnaik [10], demonstrated that among hepatobiliary tumors in cats, those of epithelial origin and biliary system were most frequent, unlike humans and dogs, in which liver neoplasia are more common.

Primary hepatobiliary tumors occur more frequently in older and male individuals with average age between nine and ten years [12]. However, the animal in this case was much younger (4-year-old).

Clinical presentation presented by the cat in this report consisted of apathy, anorexia and emesis, which are unspecific. This finding corroborates with [9] that reported these signs in hepatobiliary cases. According to Liptak *et al.* [6], only 50% of hepatobiliary neoplasia cases in cats present clinical manifestations, which are nonspecific and mostly identified in advanced

stages of the disease. Other clinical manifestations include weight loss, jaundice, hepatomegaly, polyuria and polydipsia [4,12].

The main biochemical alterations that were identified in this case are unspecific for the tumor and could occur in several conditions affecting liver and biliary tree, including increased liver transaminases and bilirubin [9]. Other expected laboratory findings include neutrophilia and chronic anemia, which were not observed in this report [1].

Clinical and laboratory alterations suggested the initial diagnosis of cholangitis. However, the lack of improvement after the clinical treatment and the ultrasonographic findings suggestive of obstructive condition, especially due to the dilation of common and cystic ducts led to the extrahepatic biliary duct obstruction diagnosis [2]. In addition, the presence of biliary mud suggests cholestasis, which is considered a significant finding in cats [4].

This condition may occur as a consequence of chronic cholangitis, pancreatitis, gallstones, parasite infestation and neoplasms [9]. Hence, all of these conditions are considered as differential diagnoses of gallbladder adenocarcinoma. Surgical exploration for biliary decompression and inspection of liver and other organs is recommended when the cause is not identified or in the absence of clinical improvement after the treatment is initiated [2], which was the procedure adopted in this case.

Neoplasm was demonstrated by the malignant epithelial cell infiltrate in mucosa and submucosa of the gallbladder. These cells presented immune expression by CK7, which marks ductal, glandular, superficial squamous and transition epitheliums, and by CK Pan, which is a specific antigen marker of filaments of epithelial cells that characterizes adenocarcinoma [5].

Surgical resection is indicated to provide better duration and quality of life for the animal [12]. Chemotherapy does not present adequate results in the treatment of primary hepatobiliary tumors [1]. During surgery, the occurrence of hypotension is common in cat anesthesia during procedures involving gallbladder and biliary tree, which can result in death [8] as occurred in this case. A mechanism that may affect muscle tonus in these patients is the presence of increased bilirubin concentrations in blood. In addition, a possible increased liberation of nitric oxide associated with obstructive jaundice have been reported in dogs [8].

Prognosis for malignant hepatobiliary neoplasia is unfavorable with elevated metastasis rates [3], which were not observed in this case. However, the

possibility was clear, considering the tumoral clots identified in histopathological analysis.

The gallbladder adenocarcinoma should be considered as a differential diagnosis for extrahepatic biliary obstruction in cats, especially in cases that are unresponsive to clinical treatment. Histopathology and immunohistochemistry are key tests to confirm the presence of this neoplasia. Despite the risk of death during surgery, the indicated treatment is surgical resection with the aim to promote better quality of life for the animal.

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Figure 1. Gallbladder carcinoma in a cat. Marked dilation and thickening of gallbladder walls.

Figure 2. Gallbladder carcinoma in a cat. A- Marked proliferation of carcinoma cells with acini-like structures. Cell morphology range from polygonal to cuboidal with scarce to moderate eosinophilic cytoplasm. Nuclei are round to oval with vacuolated chromatin and evident nucleoli [H.E.; Obj. 20x]. B- Intense cytoplasmatic immune marking of epithelial cells for CK Pan [Obj. 10x]. C- Moderate cytoplasmatic immune marking of epithelial cells for CK7 [Obj. 40x]. D- Intense cytoplasmatic immune marking of epithelial cells for CK Pan [Obj. 20x].

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