



Case report

Cholangiocarcinoma in Backyard Chicken

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Submitted June 26th 2013, Accepted November 24th 2013

Abstract

Tumors arising from intrahepatic bile ducts and gall bladder have been described in several species of domestic animals. Cholangiocarcinoma is a neoplastic proliferation of the intrahepatic biliary system commonly reported in dogs, cats, sheep, cattle, horses, goat, birds kept in captivity, and occasionally in wildlife animals. A few cases have been described in broiler chicken. This is the first report of cholangiocarcinoma in poultry, species *Gallus gallus domesticus*, in Brazil. The diagnosis was based in histology lesions and immunohistochemistry findings.

Key Words: backyard chickens, cholangiocarcinoma, *Gallus gallus*, neoplasia.

Introduction

Tumors of the liver and the intrahepatic biliary system are common in most domestic animals and infrequent in wildlife animals. Primary tumors of the liver are nodular hyperplasia, hepatocellular adenoma, hepatocellular carcinoma, and hepatoblastoma. Tumors of the biliary system are cholangioma, biliary cystadenoma, cholangiocarcinoma, adenomas and carcinomas of the gall bladder. These tumours of the liver and the intrahepatic biliary system are fairly common in most domestic animals, although absolute prevalence figures are generally not available (5,16).

Cholangiocarcinoma or intrahepatic bile duct carcinoma is a malignant neoplasm composed of epithelium resembling biliary tract epithelium. It commonly spreads to the liver capsule, resulting in widespread serosal dissemination, as well as metastasizes to the lymphatic system and lungs (16). This neoplasm is reported in dogs, cats, sheep, cattle, horses, and goats (5). Cholangiocarcinoma is rare in wildlife animals. So far it has been described in Dwarf Galagos (3), Capuchin Monkey (4), Ferret (7), and Margay (14). In birds kept in

captivity this neoplasm is frequent (12) and described in flamingo (20), peach-fronted conure (8), captive Adélie penguin (17), Amazon parrot (2), Florida sandhill crane (1), and psittacine *Amazona aestiva* (9). Cases of cholangiocarcinoma in poultry were reported by Peyghambari et al., (2007) in broiler chicken and by Wight (1961) in fowl *Gallus domesticus*. The present study is the first reported case of a cholangiocarcinoma in the poultry species *Gallus gallus domesticus* in Brazil.

Case report

An adult male chicken (*Gallus gallus domesticus*) of indeterminate lineage, reared in extensive system, was forwarded to the Department of Veterinary Pathology for routine necropsy. At macroscopic evaluation was observed moderate hepatomegaly, the liver parenchyma was diffusely firm and with multiple white-yellowish areas of size ranging between 0.1–0.3 cm in diameter (Fig. 1). Neoplastic areas deepened the surface affecting the whole liver tissue. No significant changes were observed in other tissues. Liver samples were submitted for histopathology, fixed in 10% buffered formalin, processed for routine

paraffin embedding, sectioned with 3-5 μm width and stained with hemotoxylin and eosin (HE).

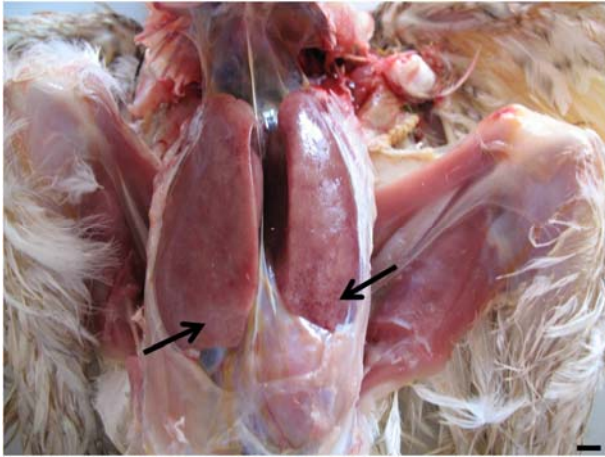


Figure 1. Backyard chicken: Coelomic cavity: Marked hepatomegaly with multiple white-yellowish areas (arrow).

Histopathology revealed proliferation of epithelial malignant cells of the intrahepatic biliary system. The cells were cuboidal with less granular basophilic cytoplasm, and arranged in a poorly defined microacinar pattern (Fig. 2A). Cell proliferation was observed in the portal triads, and invading the liver lobules (Fig. 2B). These cells were organized in ducts comprising cuboidal columnar epithelial cells, arranged in glandular complex arrangements, with or without lumen, separated from each other by small septum tissue. Associated with the neoplastic proliferation moderate inflammatory heterophilic infiltrate was observed.

Neoplastic fragments were submitted to immunohistochemistry (IHC) assay for the determination of the tumoral cell histogenesis. Three- μm sections were stained by standard streptavidin-biotin immunoperoxidase method (LSAB+ Kit, Dako Corp., Carpinteira, CA) with

the following primary antibodies: monoclonal mouse anti-human cytokeratin (CKs) AE1/AE3 (Dako, dilution 1:50). Negative control (goat serum) and positive control tissue (the epithelium of the bile duct cell) sections were included for each antibody. All neoplastic cells were marked by the IHC reaction (Fig. 3A,B).

Discussions

Hepatic neoplasms are characterized by abnormal, uncontrolled and progressive growth from hepatocytes or bile duct cells (18). Among these hepatic neoplasms, cholangiocarcinoma is characterized by proliferation of epithelial cells originating in the intrahepatic biliary system (5). Although this neoplasm is reported in captive birds, there are only two cases in the literature of this tumor in chicken (15, 21). This appears to be the first reported case of cholangiocarcinoma in fowl in Brazil.

Gross lesions of this case, differently than the described in the literature for birds (2,9) and dogs (5,16), included mild and nonspecific macroscopic lesions associated with the hepatic neoplasm, and the first pathology suspicion was hepatitis. Histological changes were characterized by proliferation of epithelial malignant cells consistent with cholangiocarcinoma, which was confirmed with IHC. According with Ponomarkov & Markey (1976), this proliferation was classified as moderately differentiated by the production of poorly defined acini and solid cords of cells with occasional lumen formation. This histology changes observed were similar to the described in the literature on cholangiocarcinoma in chicken (15,21), exotic birds (2,4) and domestic animals (9). Although metastases have been commonly reported in the lymphatic system and lungs (15,16), in this case the tumor did not either spread to the liver capsule or disseminate to other tissues.

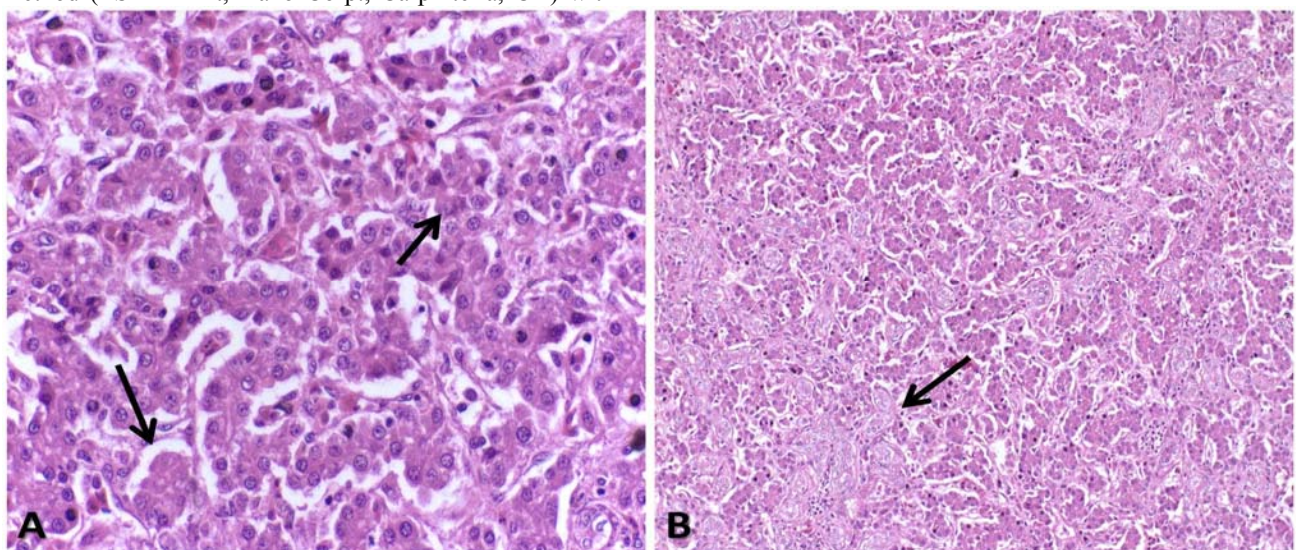


Figure 2. Backyard chicken: Liver. HE: (A) Region of liver lobules with invasion of neoplastic epithelium cells organized in acinar arrangement (arrows). 60X. (B) Neoplastic proliferation is observed predominantly located in the periportal zone (arrow) with invasion of almost every hepatic lobe. 20X.

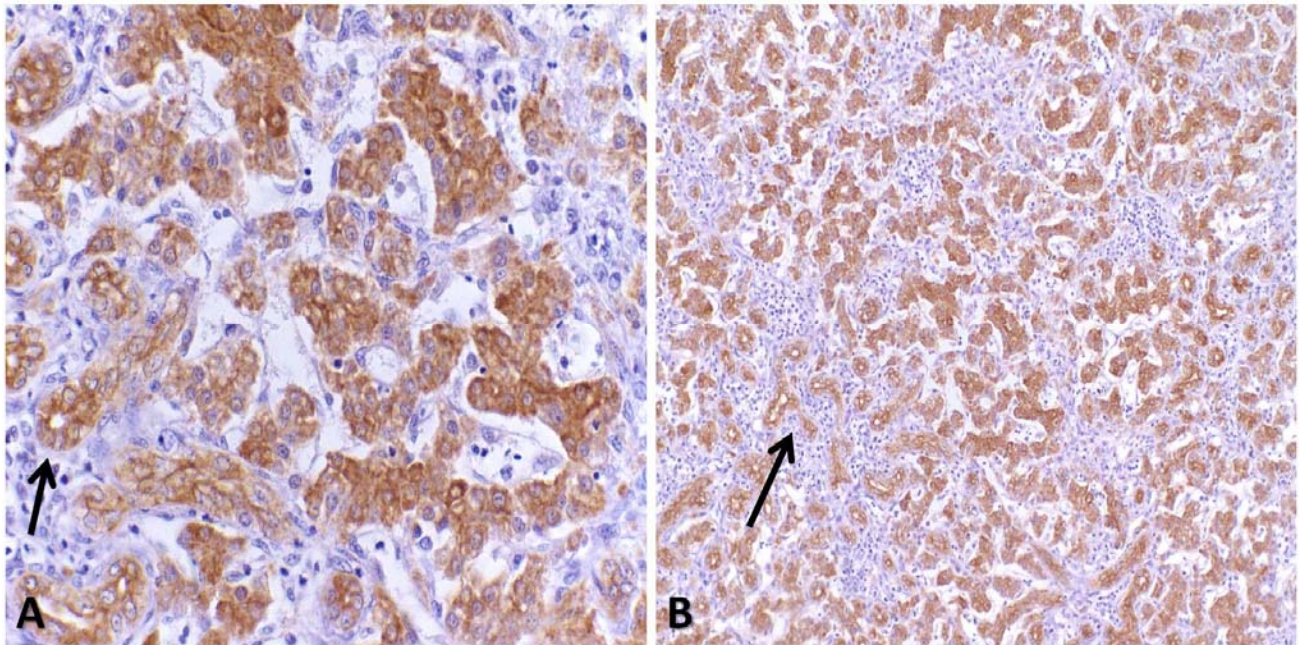


Figure 3. Backyard chicken. Liver. Anticytokeratin (AE1/AE3) IHC, Harris' hematoxylin counterstain (A) Note the proliferation in acinar organization (arrow) and also in solid strands. 60X (B) Proliferation of epithelial cells invading the liver lobules with loss of tissue architecture (arrow). 20X.

The diagnosis was based on histological changes and the positive staining in IHC analysis. Although some studies demonstrate that the markers AE1/AE3 are not as effective as other cytokeratin markers such as MAK-6 and CAM-5.2 (13), other authors have reported that these markers are effective for biliary normal and neoplastic cells, with a negative reaction of normal hepatocytes (19). The marker used in the reaction (AE1/AE3) also has proven to be effective for marking cells of the bile ducts of birds (9).

Several cases of cholangiocarcinoma are reported in wild animals, but its pathophysiology remains unclear in these species (5,16,18), although chronic inflammation may be associated (6,7). In poultry, liver neoplastic processes may be associated with chronic ingestion of mycotoxins, which are metabolites produced by fungal chemical carcinogens mainly found in seeds and grain contaminated by aflatoxin produced by *Aspergillus flavus* and *A. parasiticus* (10). In the present case lesions suggestive of primary pathology were not observed, only an associated moderate heterophilic inflammatory infiltrate.

Differential diagnosis was based on comparative histology among hepatocellular carcinoma, cholangioma, biliary cytadenoma, and inflammatory conditions associated with biliary ducts proliferation. The conclusive diagnosis of cholangiocarcinoma in birds and other animals was confirmed by the IHC reaction against cytokeratin of the epithelial cells (AE1/AE3) (9,11).

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