Post mortem computed tomography as a complementary tool for diagnosing cholangiohepatitis in a giant panda (*Ailuropoda melanoleuca*)

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ABSTRACT. A geriatric female giant panda developed grave signs of illness and was diagnosed with suspected hepatobiliary tract obstruction or other severe hepatic disease such as advanced cholangiohepatitis. The giant panda was euthanized and post mortem computed tomography was performed prior to necropsy. Common bile duct obstruction at the major duodenal papilla by a mineral attenuating calculus causing dilatation of common bile and gallbladder with concurrent multiple areas of liver abscess were detected by postmortem computed tomography. These were confirmed with gross necropsy. This is the first case report of common bile duct obstruction by mineral calculus with concurrent severe cholangiohepatitis in a giant panda.

KEYWORDS: *Ailuropoda melanoleuca*, common bile duct obstruction, computed tomography, giant panda, necrotizing cholangiohepatitis

In the past few years, there is an increasing number of reports on the usage of post mortem computed tomography (PMCT) in the investigation of the cause of death in animals [8, 14]. This is especially pertinent for zoological species as there may be lack of complete history of the presentation, physical examination constraints due to large patient size or aggressive demeanor, or other challenges [2, 5, 9, 10, 13, 15, 16, 18]. It is documented that PMCT is superior to conventional necropsy in detecting fractures, as well as gas in body cavities, organ parenchyma, and the vascular system [14].

The complementary usage of PMCT to conventional necropsy in zoological species may help to increase the accuracy of conventional necropsy [7, 8, 10, 16]. The report describes obstruction of common bile duct by a mineral calculus leading to necrotizing cholangiohepatitis in a giant panda, with the clinical course, PMCT and pathological findings.

The patient was an approximately 38-year-old intact female giant panda. The panda was rescued from the wild as a 2–3-year-old juvenile in 1980 and remained under human care thereafter. Several chronic medical conditions were encountered during the panda’s care including significant systemic hypertension, psychogenic polydipsia, multifocal osteoarthritis, and bilateral mature cataracts as well as lens luxation. The patient also had intermittent episodes of mucoid stool and gastrointestinal upset, as is commonly seen in captive giant pandas [12]. Long-term medications for this animal included carprofen (1–2 mg/kg p.o., s.i.d.) and amantadine (2 mg/kg p.o., s.i.d.) to address osteoarthritis, and amiodipine (0.13–0.18 mg/kg p.o., s.i.d.) to treat hypertension.

Preventative care for this patient included monthly routine blood draws for general health monitoring. Venipuncture was accomplished using operant conditioning, with the animal conscious and voluntarily participating in the procedure. About six months prior to death, routine bloodwork revealed marked elevations in liver enzymes including aspartate aminotransferase (AST, 498 μl, reference range 4–116), alanine aminotransferase (ALT, 640 μl, ref. range 23–119), and gamma-glutamyltransferase (GGT, 51 μl, ref. range 0–14) [3]. At this time total white blood cell count was within normal for this individual’s in-house reference range (18.28 × 10³ cells/μl, ref. range 10.44–19.3), although above range for published standards for the species (2.41–12.43 × 10³ cells/μl) [3]. Since no clinical signs were present, the patient was monitored pending a recheck of blood values. An ultrasound of the liver six days later did not reveal any abnormalities. Recheck bloodwork on day seven revealed a worsening of liver enzyme elevations...
A moderate amount of gas and approximately 50 ml of foul-smelling, grey-brown, opaque material thinned, and the mucosal surface was lined with a creamy grey diphtheritic membrane of fibrin. Small focal hemorrhages were greyish red fluid and fibrin. The gallbladder was distended, firm, and adhered tightly to the duodenum. The gallbladder wall was necrosis. All other liver lobes contained similar but smaller foci of liquefactive necrosis. Prominent bile ducts were filled with bile, and small gas bubbles are present in the wall of the gallbladder (white chevron) (A). A small amount of fluid accumulation (white asterisk) at the dependent aspect of area of coalescing gas at the right cranial dorsal aspect of the liver (C). Gallbladder contains gas and dependent bile, and small gas bubbles are present in the wall of the gallbladder (white chevron) (A and B). A small amount of fluid is present in the stomach (white S) (C).

Fig. 1. Axial plane CT images of the liver, view in soft tissue window (WL: 40, WW: 350). Note the linear gas in the hepatic parenchyma (white $)$ (C).

There were three focal areas of coalescing gas bubbles (white notched arrows) (A and B). A small amount of fluid accumulation (white asterisk) at the dependent aspect of area of coalescing gas at the right cranial dorsal aspect of the liver (C). Gallbladder contains gas and dependent bile, and small gas bubbles are present in the wall of the gallbladder (white chevron) (A and B). A small amount of fluid is present in the stomach (white S) (C).

A very small amount of effusion was present in the cranial peritoneal cavity adjacent to the liver, and a moderate amount of effusion at the right caudal peritoneal cavity. A few gas bubbles were present in the peritoneal cavity adjacent to a few loops of intestines. There was a small mineral attenuating focus located at the left lateral wall of the caudal vena cava adjacent to the left adrenal gland. Cylindrical mineral attenuating structures were present within ventral aspect of the left adrenal gland. The right adrenal gland was normal. A few renal hypoattenuating structures were present in both kidneys (Fig. 3).

Conventional necropsy was performed the following morning, approximately 13 hr after PMCT. The mucous membranes were mildly jaundiced. The abdominal cavity contained approximately 300 ml of reddish brown, opaque fluid with fibrin. The liver was grossly abnormal with a diffuse, mottled, creamy brown to red discoloration. All liver lobes were swollen. On cut surface, bile ducts were thickened and dilated. A solitary 2.5 cm diameter creamy red abscess was present on the surface of the right central lobe, surrounded by a 1–2 mm thick band of hemorrhage. On cut surface, this lesion contained a large pocket of liquefactive necrosis. All other liver lobes contained similar but smaller foci of liquefactive necrosis. Prominent bile ducts were filled with greyish red fluid and fibrin. The gallbladder was distended, firm, and adhered tightly to the duodenum. The gallbladder wall was thickened, and the mucosal surface was lined with a creamy grey diphtheritic membrane of fibrin. Small focal hemorrhages were present under the membrane. A moderate amount of gas and approximately 50 ml of foul-smelling, grey-brown, opaque material

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A round, smooth layered approximately 12 mm × 12 mm mineral calculus was present at the distal aspect of the common bile duct at the duodenal papilla (Fig. 4). The common bile duct was dilated and filled with greyish red fluid and fibrin. Mineral foci of the left adrenal gland were present. Multiple cysts in both kidneys were observed.

On histological examination, the liver showed evidence of marked post mortem decomposition. However, there was also ample evidence of severe, acute, necrotizing cholangiohepatitis with multiple variably-sized foci of necrosis and large numbers of neutrophils. There was evidence of chronic fibrosis and bile duct proliferation in the portal areas with a more acute inflammatory reaction super-imposed on the chronic changes. In many places the more acute reaction extended into the adjacent liver lobules.

In one section there were numerous very large areas of necrosis with hemorrhage, fibrin, and neutrophils surrounding lytic hepatic lobules.

The gall bladder mucosa contained moderate numbers of mononuclear inflammatory cells and neutrophils. Areas of hemorrhage were present in the serosa. In a section contiguous with the liver, there was massive thickening of the wall with extensive areas of necrosis and edema, fibrin, and massive numbers of inflammatory cells. The correlation of PMCT findings with gross necropsy and pathology results is listed in Table 1.

The cause of severe illness in this animal was concluded to be partial and/or complete obstruction of the common bile duct by a choledocholith, leading to severe, necrotizing cholecystitis, cholangiohepatitis, and hepatic necrosis/abscessation. The findings of PMCT, conventional necropsy, and histology agreed with each other. The exact onset of the obstruction of the common bile duct could not be determined. The patient did have elevated liver enzymes six months prior to death, but this resolved with conservative...
therapy (hepatoprotectant medications) and thus was unlikely to have been reflective of ongoing bile duct obstruction. The obstruction may have been intermittent or recurrent. However, there may indeed have been cholangitis or cholangiohepatitis present at the time of the live enzyme elevations, and it may have led ultimately to stone formation in the bile duct. Ultrasonographic examination of the liver at multiple points during the six-month period did not reveal obvious abnormalities. The liver may either have indeed been normal at those times, or the limitations of conscious ultrasound in this species may have precluded accurate imaging of the entire liver. The common bile duct was not examined during the ultrasound exams due to overlying gas-filled intestine and technical challenges due to the large body size patient.

In dogs, cholelithiasis is one of the more common diseases of the gall bladder, and stones can migrate to the common bile duct or form there in cases of primary choledocholithiasis [1]. Dogs may be asymptomatic or may present with signs such as icterus, abdominal pain, vomiting, and anorexia. When severe biliary obstruction occurs, surgical intervention is usually required and may be accomplished via multiple techniques, with or without placement of stents [6]. The incidence and outcome of complications of obstructive disease such as the hepatic necrosis and abscessation seen in this giant panda is unknown. Because the animal's status deteriorated rapidly after the onset of clinical signs and the condition was considered moribund, aggressive treatment such as surgery was not considered.

The severity of the pathology as demonstrated by the accumulation of gas in the hepatic parenchyma, intrahepatic bile duct and

| PMCT findings | Gross necropsy findings | Pathology/histology results |
|---------------|-------------------------|-----------------------------|
| Intrahepatic linear gas | Foci of coalescing gas bubbles in hepatic parenchyma with dependent fluid | Abscess with liquefactive necrosis | Severe, acute necrotizing cholangiohepatitis with multiple foci of necrosis and large numbers of neutrophils |
| Distended gallbladder with dependent bile and non-dependent gas | Distended, firm and adhered to the duodenum. Contained a moderate amount gas and 50 ml of foul smelling, grey-brown, opaque material | Thickened gallbladder wall, and the mucosal surface was lined with a creamy grey diphtheritic membrane of fibrin | Massive thickening of gallbladder wall with extensive areas of necrosis and edema, fibrin, and inflammation |
| Mildly thickened, irregular and intermural gas bubbles of gallbladder wall | Thickened gallbladder wall | Peritoneal effusion (reddish brown, opaque with fibrin) |
| Layered mineral calculus at major duodenal papilla | Smooth layered mineral calculus at the duodenal papilla | Dilated common bile duct with greyish red fluid and fibrin |
| Dilated common bile duct with fluid and gas | Dilated common bile duct with dependent fluid and gas | Massive thickening of gallbladder wall with extensive areas of necrosis and edema, fibrin, and inflammation |
| Small amount of peritoneal effusion | Peritoneal effusion (reddish brown, opaque with fibrin) | Peritoneal effusion (reddish brown, opaque with fibrin) |
| Small gas bubbles in the peritoneal cavity | Mineral focus at left lateral wall of caudal vena cava | Mineral focus at left adrenal gland |
| Mineral focus at left adrenal gland | Mineral focus in left adrenal gland | Hypoattenuating structures in both kidneys | Multiple renal cysts |
common bile duct seen on the PMCT may be missed or underestimated by the conventional necropsy. This is because conventional necropsy may not accurately estimate the amount of gas in body cavities, organ parenchyma and vascular system, and thus the severity of disease [14]. The gas in the hepatic parenchyma, intrahepatic bile duct and common bile duct seen on the PMCT was not due to post mortem putrefaction as PMCT was performed soon after euthanasia [4, 11]. Thus, it is likely that this finding was indicative of a severe, gas-forming bacterial infection. This is also supported by the finding of necrotizing cholangiohepatitis on histopathology. Bacterial cultures of the liver yielded growth of Enterococcus faecalis, E. coli, Morganella morganii, and Clostridium perfringens. A clostridial infection could certainly have caused gas production in the organs, but it is difficult to ascertain whether this particular bacteria grew on post mortem cultures as a result of true infection vs. post mortem decomposition. The same bacterial species grew from multiple other organs sampled during the necropsy, so it is likely that the Clostridium was reflective of post mortem overgrowth. The underlying cause of the gas formation seen on PMCT thus is not definitively known.

To the authors’ knowledge, this is the first report of both cholelithiasis and the resulting severe hepatopathy in a giant panda. Post-mortem CT is a useful adjunct to conventional necropsy [14, 17]. In this case, the PMCT was particularly helpful in identifying the calculus in the bile duct prior to conventional necropsy, leading the prossectors to delicately dissect this area in order to confirm and demonstrate the gross lesions. In zoological species, PMCT can also be useful to help elucidate the anatomy of species that are rarely presented for necropsy [10, 13, 16], and can alert the prossectors of anatomical peculiarities of the species or individual.

POTENTIAL CONFLICTS OF INTEREST. The authors have nothing to disclose.

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