Clinical findings and prognostic factors for dogs undergoing elective versus non-elective cholecystectomies for gallbladder mucoceles

by

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Abstract

Objectives: The objectives of this multiinstitutional retrospective study were to compare and contrast post-operative morbidity between elective and non-elective cholecystectomy cases in dogs with gallbladder mucoceles, as well as to determine underlying risk factors and prognostic indicators of dogs with early gallbladder mucoceles and their need for surgical intervention.

Animals: This study included 121 dogs with gallbladder mucoceles that had a cholecystectomy performed.

Procedures: Medical records were reviewed for all dogs with a gallbladder mucocele between 2004-2018 that underwent a cholecystectomy. Dogs were classified into three categories: emergent (gallbladder rupture or systemic instability), urgent (systemic clinical signs, diabetic, or biliary duct distension), or elective (no clinical signs or bloodwork abnormalities). The method of common bile duct catheterization, for example, antegrade or retrograde catheterization, was noted if performed. Post-operative complications and survival were compared between groups.

Results: Dogs that had a duodenotomy and retrograde common bile duct cannulation were 6.67 times more likely to develop a post-operative fever in comparison to dogs that had no common bile duct catheterization and 13.61 times more likely to develop post-operative fever in comparison to dogs that had normograde catheterization of the common bile duct. Dogs that had either urgent or emergent surgery were 3.45 and 4.3 times more likely, respectively, to die prior to discharge in comparison to those that had elective cholecystectomy.

Conclusions and Clinical Relevance: Dogs undergoing elective cholecystectomies have a lower mortality rate than those undergoing urgent or emergent procedures. Duodenotomy with retrograde bile duct catheterization is associated with post-operative fever development.

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Dedication

I would like to thank my husband, Randy, for his loving support during my residency and master's career. His support and love have been an unwavering source of strength throughout this difficult journey. I would like to thank my parents and siblings for always being so supportive of my career journey. Finally, I would like to dedicate my thesis and ultimately my career to my Lord Jesus Christ, who is my savior and ultimate source of strength.

Chapter 1 - Canine Gallbladder Mucocele Literature Review

Anatomy and Physiology of the Extrahepatic Biliary System

The extrahepatic biliary system in a dog consists of the hepatic ducts, cystic duct, gallbladder, and common bile duct.[1,2] The hepatic ducts converge into the common bile duct, which connects to the duodenum via the major duodenal papilla through the Sphincter of Oddi.[1,2] Position of the major duodenal papilla is variable between dogs and is approximately 3 to 6 cm aborad to the pyloric sphincter.[2] The gallbladder sits between the right medial liver lobe and the quadrate lobe and is connected via the cystic duct to the common bile duct.[1,2] Bile flows from the liver into the interlobular ducts, into the lobar ducts, and then into the hepatic ducts. At this point, it flows either up the cystic duct to be stored in the gallbladder or down the common bile duct to be secreted into the duodenum.[1]

Bile, which is created and secreted from hepatocytes, consists mainly of bile acids, bilirubin, cholesterol, phospholipids, water, and bicarbonate.[2] Bilirubin is the breakdown product of the heme molecule in hemoglobin.[1] Metabolism of myoglobin can also produce bilirubin.[2] As hemoglobin is broken down in the reticuloendothelial system, the heme molecule becomes unconjugated bilirubin and is transported with albumin to the liver.[1] In the liver, the bilirubin is conjugated via the enzyme UDP-GT and is able to be released into the extrahepatic biliary system.[1] Once conjugated, bile is released and stored in the gallbladder until food enters the duodenum.[2] Once this occurs, cholecystokinin is released from the small intestinal mucosal surface, stimulating gallbladder contraction and Sphincter of Oddi relaxation.[2]

Bile salts allow for fat emulsification and absorption, including the fat-soluble vitamins, within the intestine.[1] Bile salts also act as a detergent in the small intestine by binding and neutralizing

endotoxin in the intestines.[1] Bilirubin is excreted largely as stercobilin in feces and to a smaller extent as urobilinin in urine.[1]

Pathogenesis of Gallbladder Mucocele

Gallbladder mucoceles are a common disease process of the hepatobiliary system of canine patients and can cause biliary tract obstruction.[3-5] Gallbladder mucoceles are defined as a mucus-filled distension of the gallbladder characterized by green-black, semisolid to immobile mucin.[5,6] The etiopathogenesis is unknown and likely multifactorial.[3]

One hypothesis includes hyperplasia of the mucus secreting glands of the gallbladder promoting increased mucus accumulation in the gallbladder.[3,7,8] One study that evaluated this abnormal gallbladder mucus secretion revealed an abnormal increase in a gel-forming mucin protein Muc5ac in comparison to Muc5b protein in normal gallbladders, along with defective mucin un-packaging and defective mucin-interacting innate defense proteins.[9] This finding is suggestive of an abnormal mechanism for maintenance of gallbladder epithelial hydration and thus for gallbladder mucocele formation.[9]

Alternatively, bile acid composition may play a role in gallbladder mucocele formation. One experimental study in normal canine gallbladders found that the bile acids taurochenodeoxycholic acid and deoxycholic acid stimulate gallbladder epithelial cells to accelerate mucin secretion.[10] Other studies discovered that hydrophobic bile acids have cytotoxic effects which can contribute to gallbladder hypomotility and mucin hypersecretion.[11-13] A study found gallbladder bile content compositional changes in dogs with gallbladder mucocele and biliary sludge, namely lower levels of taurodeoxycholic acid, one of the major bile acids in dogs.[14] The effect of the lower bile acid concentration on formation of gallbladder mucoceles remains unclear, however, the decrease might be associated with cholestasis.[14] Also, gallbladder

concentrations of unconjugated deoxycholic acid and chenodeoxycholic acid are elevated in dogs with hyperadrenocorticism, a risk factor of gallbladder mucocele formation.[15]

Lastly, leptin has also been reported to play a role in gallbladder mucocele pathogenesis.[16] Leptin, derived from mature adipose tissue, is important in energy metabolism by acting as the fasting hormone to regulate food intake and energy consumption.[16] Significantly higher serum leptin concentrations were found for dogs with gallbladder mucocele formation requiring surgical intervention.[16,17] Also, a study found an increase in leptin and leptin receptor mRNA expression in gallbladder mucoceles compared to controls.[16] More studies need to be performed to evaluate the relationship between leptin concentration and the development of gallbladder mucoceles.[16]

Predisposing Factors of Gallbladder Mucocele Formation

There are several predisposing factors for gallbladder mucocele formation reported in the literature. Pre-disposed breeds reported include Shetland sheepdogs, cocker spaniels, and miniature schnauzers.[6,18,19] Other potential breed predispositions include Pomeranians and Border terriers.[20,21] Concurrent endocrinopathies, such as hyperadrenocorticism, hypothyroidism, diabetes mellitus, and hypertriglyceridemia, have been reported as a risk factor for development of a gallbladder mucocele.[8,15,18,19,22,23]

One study revealed that dogs with a gallbladder mucocele were 2.2 times as likely to have been treated for hypothyroidism and 2.6 times as likely to have been treated for hyperadrenocorticism compared to control dogs.[19] Another study revealed that dogs with hyperadrenocorticism were 29 times more likely to develop a gallbladder mucocele compared to dogs without hyperadrenocorticism.[24] Dogs with hyperadrenocorticism and hypothyroidism were reported to have greater than 25% sludge in their gallbladders in one study.[25] Another case report documented the progression of biliary sludge into a gallbladder mucocele in a miniature schnauzer with diabetes mellitus.[22] Hypercholesterolemia and hypertriglyceridemia are reported to have a 2.92 times and 3.55 times increased risk, respectively, of developing a gallbladder mucocele.[26] Therefore, treatment of these underlying endocrinopathies is recommended to minimize the odds of development of a gallbladder mucocele.[27]

Another study found an association between cholestatic disease and pituitary-dependent hyperadrenocorticism.[23] The association was proposed to be caused by hypercholesterolemia, unidentified genetic factors, or the hydrophobic nature of trilostane.[23] Dosages of trilostane, a lipid soluble drug used in treatment of hyperadrenocorticism, should be adjusted for dogs with

gallbladder mucoceles due to the decreased ability of bile secretion induced by concurrent cholestatic disease.[23]

Drug administration has been associated with gallbladder mucocele formation. Administration of corticosteroids lead to the development of biliary sludge by day 56 in all patients in one study, however, none of these patients developed a gallbladder mucocele.[15] Another study revealed that Shetland sheepdogs receiving imidacloprid were more likely to develop a gallbladder mucocele in comparison to ivermectin or milbemycin.[19] That study did not suggest that imidacloprid was the primary cause of the gallbladder mucocele formation, however, it may be an exacerbating factor for Shetland sheepdogs.[19]

Gene mutations have been studied in their role in gallbladder mucocele pathogenesis with varying results. A study found a significant association between the gene mutation ABCB4 1583_1584G found in Shetland sheepdogs and a predisposition to hepatobiliary disease and gallbladder mucocele formation, although the mechanism is unknown.[28] In contrast, other researchers did not find a significant association between the gene mutation ABCB4 1583_1584G and gallbladder mucocele formation.[29]

Sequela of Extra-hepatic Biliary Obstruction

Bile duct obstruction is a life-threatening emergency and can cause a number of sequelae that affects many body systems, including but not limited to gall bladder rupture, bile peritonitis, acute renal failure, multisystem organ failure, and death.[2] Differentials for extrahepatic biliary obstruction include gallbladder mucocele, pancreatitis, neoplasia, cholelithiasis, and inflammatory biliary tract disease.[3,30] As bile accumulates within the hepatobiliary system and blood, a downregulation of the reticuloendothelial system will soon occur.[1] Coagulopathies can occur due to either lack of vitamin K absorption within the ileum or endotoxin mediated binding of coagulation factors XI and XII.[1]

Bacterial sepsis can develop due to lack of bile in the intestinal lumen, as bile salts normally act to bind bacterial endotoxin, thus disabling its toxic effects.[1] Without bile salts, the endotoxin is freely absorbed into the portal bloodstream, causing a multitude of systemic effects, including but not limited to, hypotension, tachycardia, coagulopathy, acute tubular necrosis, gastrointestinal hemorrhage, systemic inflammatory response syndrome, myocardial damage, acute respiratory distress syndrome, and disseminated intravascular coagulation.[1]

Gallbladder mucocele can lead to gallbladder or common bile duct wall necrosis and subsequent rupture.[6,7,31] In the absence of bacterial infection, bile normally causes only a mild chemical peritonitis, however, septic bile peritonitis can lead to severe life-threatening disease .[32]

Clinical signs and Diagnosis of Gallbladder Mucocele

Dogs with gallbladder mucoceles have nonspecific clinical signs, including lethargy, inappetence, vomiting, regurgitation, weight loss, abdominal pain, fever, polyuria, polydipsia, etc.[1,2,4,5] Common clinicopathologic changes seen for dogs with extrahepatic biliary obstruction include elevated alkaline phosphatase, alanine aminotransferase, and aspartate aminotransferase, along with hyperbilirubinemia and a leukocytosis.[2] Dogs with gallbladder rupture reportedly have higher alanine aminotransferase, alkaline phosphatase, bilirubin, and total white blood cell concentrations, according to one study.[7] Amylase and lipase can also be elevated.[6,7]

Serial measurements of bilirubin can be helpful to determine the presence of extrahepatic biliary obstruction.[1] Bile acids will be elevated early in the obstructive process; however, this test will not be helpful as the disease progresses, as it rises in the early disease process then returns back to normal.[1] Dogs should be evaluated for bilirubinuria on urinalysis; due to the low renal threshold for excretion of conjugated bilirubin, this finding often will precede the production of icterus.[1]

Imaging for gallbladder mucoceles includes abdominal ultrasound and computed tomography (CT). Gallbladder mucoceles are categorized into six different ultrasonographic categories: type 1, echogenic immobile bile occuping the gallbladder; type 2, incomplete stellate pattern; type 3, typical stellate pattern; type 4, kiwi fruit like pattern and stellate combination; type 5, kiwi fruit like pattern with residual central echogenic bile; and type 6, kiwi fruit like pattern without central echogenic bile.[33] One study found no significant difference between bile patterns of asymptomatic dogs and symptomatic dogs.[33] In this study, the most common bile pattern

associated with bile rupture was type 2, although this pattern is not suggested to be pathognomonic for rupture.[33]

Ultrasound was shown in one study to have a low sensitivity for detection of gallbladder rupture at 56%.[32] An ultrasonographic diagnosis of gallbladder rupture has a sensitivity of 78.6%, specificity of 100%, and accuracy of 84%, according to another study.[33] Accuracy of gallbladder rupture can be increased when a contrast-enhanced ultrasound is performed to a 100% sensitivity and specificity.[34,35] There is variable correlation between ultrasonographic and surgical findings found in the literature and must be used as a part of the entire clinical picture when making treatment recommendations.[3,32,33,36] Rarely, extruded gallbladder mucoceles are evident on ultrasound throughout the abdominal cavity and have similar characteristics seen in non-extruded gallbladder mucoceles.[37]

As there is often question on the presence or absence of a gallbladder mucocele, multiphase CT angiography can be performed to more accurately classify the gallbladder abnormalities. One study grouped cases that had both an abdominal ultrasound and a CT angiography into 3 categories: no sludge, sludge occupying greater than 25% of the gallbladder lumen, and gallbladder mucoceles.[38] The most notable finding was that dogs with gallbladder mucoceles had mineral located centrally in 67% of cases.[38] The centrally located mineral was not visualized on dogs without a gallbladder mucocele.[38] Therefore, it was concluded that a hyperattenuating gallbladder on pre-contrast CT images and centrally distributed mineral is consistent with a gallbladder mucocele.[38] CT cholangiography can also be used to delineate the structural characteristics of the biliary system and to estimate the patency of the common bile duct in dogs with gallbladder mucoceles and thus can be used as a pre-operative screening tool.[39]

Gallbladder mucocele rupture should be considered in dogs with acute signs of abdominal pain, ultrasonographic evidence of a gallbladder mucocele, and the presence of peritoneal effusion.[31] Peritoneal fluid cytologic examination is performed in suspected cases of gallbladder mucocele rupture. Bile is typically dark green to yellow and appears on fluid cytology as a golden to green pigment either seen free on the slides or within the macrophages.[40] Bile peritonitis can also manifest as white bile, demonstrated by a fibrillar, acellular mucinous material.[41] Also, bile peritonitis is considered to be present when peritoneal bilirubin is twice the serum bilirubin measurement.[42,43] One study revealed that elevated peritoneal fluid bile acid concentrations can be compared to serum bile acids to help confirm gallbladder rupture.[31] Bile acids, unlike bilirubin, are not freely diffusible across the peritoneum and would then subsequently accumulate in the abdominal fluid of dogs with gallbladder rupture.[31]

Treatment of Gallbladder Mucocele

Medical management of incidental gallbladder mucoceles consists of the administration of S-adenosyl-methionine, omega-3 fatty acids, famotidine, ursodiol, and treatment of any underlying endocrinopathy.[44] One case series revealed resolution of a gallbladder mucocele in two dogs undergoing medical management, indicating that surgical intervention may not be necessary for all dogs with gallbladder mucoceles.[44] However, this is the only paper in the literature detailing resolution of gallbladder mucocele with medical management.

The timing of surgical removal of the gallbladder is controversial. Although a different mechanism, patients in human medicine often undergo early interventional cholecystectomy prior to the development of clinical signs and bloodwork abnormalities for biliary disease.[45] This has been shown to significantly lower post-operative morbidity and mortality in comparison to medical management or later surgical management after development of clinical signs or hematologic abnormalities.[45] Cholelithiasis, rather than gallbladder mucoceles, is present in 15% of the human population.[46] Risk factors include ethnicity, age, female gender, genetics, pregnancy, obesity, high calorie diet, metabolic syndrome, dyslipidemia, type 2 diabetes, weight loss cycling, etc.[46] The pathophysiology of cholelithiasis in people is multifactorial and often due to an imbalance of biliary constituents.[46] They are either is classified as either cholesterol or pigment choleliths.[46]

One study comparing medical and surgical management of canine gallbladder mucoceles shows a median survival time of 1340 days versus 1802 days, respectively.[47] Cholecystectomy is significantly associated with a longer survival time compared to medical management of gallbladder mucoceles.[47] A recent veterinary study compared outcomes between 45 elective and

25 nonelective cholecystectomies.[3] Dogs in the nonelective group had a significantly higher mortality rate of 20% compared to a 2% mortality rate of the dogs in the elective group.[3] Another study revealed a 4.2 times greater odds of death in dogs showing clinical disease compared to subclinical dogs.[20]

The goal of treatment of any biliary tract obstruction is to relieve the underlying cause of obstruction.[4] Cholecystectomy is the treatment modality of choice, often coupled with either normograde or retrograde flushing of the common bile duct to determine and establish patency.[4,6] After isolating the gallbladder from within the hepatic fossa, the cystic duct is ligated with hemoclips or suture and the gallbladder is removed from the peritoneal cavity.[2] A novel biodegradable magnesium alloy clip has been developed that is reported to demonstrate fewer metallic artifacts on CT scan in comparison to the conventional titanium hemoclips.[48]

Laparoscopic cholecystectomy is a treatment option for dogs that do not have evidence of extrahepatic biliary obstruction.[49-52] Potential complications of laparoscopic cholecystectomy are bile spillage, inadequate cystic duct ligation, gallbladder rupture, and conversion to an open procedure.[50] Mayhew et al recommends a double ligation of the cystic duct to minimize the risk of inadequate cystic duct ligation.[50] Disadvantages of a laparoscopic approach include limited feasibility in smaller patients and the prolonged surgical time in comparison to open procedures.[50] Reasons for conversion to an open procedure include evidence of gallbladder rupture, inability to ligate the cystic duct, and leakage of bile during dissection.[52] Iatrogenic injury of the biliary duct while undergoing dissection during laparoscopic cholecystectomy in human medicine occurs in 0.3-2.7%, while these numbers are not reported in the veterinary literature.[52,53] The risk of conversion in veterinary medicine is as high as 30%, according to one study.[52] The risk of conversion decreases significantly as the surgeon experience increases.[51]

Natural orifice transluminal endoscopic surgery is a novel minimally invasive technique demonstrated on cadavers utilizing transvaginal endoscopic removal of the gallbladder.[54] This procedure is in early stages of research design, requires new surgical instrumentation to be manufactured, and is very technically challenging to perform. It does, however, show promise for minimally invasive surgery in the future for human medicine.[54] More study needs to be performed to evaluate the feasibility and for veterinary medicine.

Intra-operative complications include hemorrhage, bile duct injury, and inability to catheterize the common bile duct.[1] Perioperative hemorrhage can occur either through technical error, ligature slippage, coagulopathy, etc.[1] Through use of electrocautery, hemostatic agents and proper surgical technique, hemorrhage can be minimized.[1] Iatrogenic damage to the hepatic, cystic, or common bile duct can occur and can lead to bile peritonitis, if unrecognized.[1] Primary repair of a damaged duct can be performed; it is, however, technically challenging and has a high rate of failure and stricture.[1] Other materials have been reported in the literature to repair a damaged bile duct, including porcine submucosa, an autologous vein graft and stent, scleroprotein patch, and a compressed human collagen patch.[7,55-57] One study evaluated closure of the common bile duct with three different methods: the scleroprotein patch in 4 dogs, compressed human collagen patch reinforced with three stitches in 6 dogs, and compressed human collagen patch without reinforcement in 6 dogs.[57] The scleroprotein patch was resorbed too quickly resulting in bile peritonitis in all 4/4 dogs.[57] This study also demonstrated that the collagen patch became unglued in 2/12, resulting in bile peritonitis.[57] The other 10/12 dogs treated with the compressed human collagen patch demonstrated successful bile duct healing 1 month after application with 1/10 dogs developing a common bile duct stricture.[57]

Dogs with gallbladder mucoceles have varying levels of hemodynamic stability which has several anesthetic implications. Patients with hepatic disease have changes associated with glucose metabolism, neurologic function, coagulation status, hypoproteinemia, and blood pressure abnormalities.[58] The latter may be due to gallbladder manipulation and subsequent vagus nerve stimulation or bacterial endotoxemia.[58] One study demonstrated that there was no significant difference in the number of anesthetic complications between dogs that underwent a cholecystectomy versus dogs that had other hepatic surgery without cholecystectomy.[58]

Bacterial infection is uncommonly reported with gallbladder mucoceles.[5,59] A retrospective analysis evaluated the relationship between gallbladder contents and bacterial infection in correlation to gallbladder mucocele development.[59] The rate of positive bacterial cultures was 10% for biliary sludge and 14.3% for gallbladder mucoceles in this study.[59] Similarly, another study revealed a positive culture in 20% of gallbladder mucoceles.[5] Therefore, a culture and sensitivity is recommended for patients undergoing cholecystectomy.[5,59]

Post-operative complications can be significant after cholecystectomy. Bile duct obstruction can occur immediately post-operatively or can be latent for a few days to weeks following the procedure.[1] Differentials for immediate biliary duct obstruction include failure to establish common bile duct patency intra-operatively, migrating sludge or choleliths, inadvertent common bile duct ligation, or post-operative pancreatitis.[1,2] Differentials for latent obstructions include common bile duct stricture formation, pancreatitis, recurrence of cholelithiasis, or obstructive mass.[1,2]

Post-operative bile peritonitis is characterized by a severe inflammatory response due to the presence of bile in the peritoneal cavity.[1] While the biliary tree holds mostly conjugated bile, the low systemic pH causes bile acid deconjugation.[1] Unconjugated bile is inflammatory to the

tissues of the peritoneal cavity, causing permeability changes, inflammation, and, in severe cases, necrosis of the tissues.[1] The increased permeability allows for transudation of fluid into the peritoneal cavity.[1] These changes predispose the tissues to harbor and encourage bacterial growth.[1] Differentials for bile peritonitis include dehiscence of the duodenotomy incision, failure of the cystic duct ligature, and common bile duct necrosis or rupture.[1] Onset of clinical signs in patients with bile peritonitis depend on the degree of bile within the abdomen and presence of bacteria.[1] One reported rare complication includes bilothorax, although the mechanisms are not clearly understood.[60]

Prognostic Features of Gallbladder Mucoceles

Prognosis following a cholecystectomy varies. Mortality rates range from 21.7-40%, however, the prognosis improves if the patient survives the immediate post-operative period.[5,6,8,30] Sterile bile peritonitis is associated with a good prognosis in the literature, but septic bile peritonitis has been reported to lead to 80-100% mortality.[4,6,8,30,32]

There is limited information in the literature comparing underlying risk factors and surgical timing. Pre-operative factors that are reported to negatively impact prognosis include elevated ALT and GGT, hyperbilirubinemia, hypoalbuminemia, icterus, age, leukocytosis, elevated preanesthetic heart rate, azotemia, and hyperphosphatemia.[3,30,36] Post-operative hypotension, dyspnea, pancreatitis, hypoalbuminemia, anemia, hyperlactatemia, and presence of band neutrophils are significantly associated with post-operative mortality.[3,6,30] Serum leptin concentrations have been proposed as a prognostic indicator.[17] One study demonstrated that higher serum leptin concentrations were more likely to be associated with a poorer prognosis in comparison to dogs that were asymptomatic for gallbladder mucoceles.[17] Another study found a significant difference in mean post-operative serum lactate, PCV, and hypotension between those that survived and died.[6]

One study revealed that cholecystitis is a common comorbidity with gallbladder mucoceles, however, the presence of cholecystitis did not affect mortality rates.[61] Another study confirmed the common correlation between cholecystitis and gallbladder mucoceles utilizing eubacterial fluorescence in situ hybridization.[62] This study also found a higher bacterial count than previously reported studies, however, the clinical significance is unknown.[62]

Hyperadrenocorticism was associated with poor prognosis in one study, while these authors did not find a lower prognosis for dogs with hypothyroidism and diabetes mellitus.[20]

Gallbladder rupture carries a variable prognosis in the literature. Several studies reveal a lack of correlation between gallbladder rupture and mortality rate.[4,6,7,63] On the other hand, another study revealed a 2.7 times increased risk of death for dogs that had a gallbladder rupture at time of surgery.[32]

Conclusions

Gallbladder mucocele is a common disease process of the hepatobiliary system of canine patients and can cause biliary tract obstruction and subsequent life-threatening signs.[3-5] The etiopathogenesis is unknown, although a widely accepted hypothesis is that hyperplasia of the mucus secreting glands leads to accumulation of excess mucus in the gallbladder. Pre-disposing factors include breed, genetics, specific pharmaceuticals, and endocrinopathies. Clinical signs are nonspecific and diagnostics include baseline bloodwork and ultrasonography. Cholecystectomy is the treatment of choice based on the current literature. Prognosis varies from 2-40%, depending on the overall systemic status of the patient.[3,5,6,8,30] Prognosis of elective gallbladder mucoceles appears to be better than dogs with systemic clinical signs or bloodwork parameters consistent with extrahepatic biliary obstruction.[3] Further controlled and randomized prospective clinical trials is warranted to fully evaluate the timing of cholecystectomy for gallbladder mucoceles.

References

1. Mehler SJ. Complications of the extrahepatic biliary surgery in companion animals. Vet Clin North Am Small Anim Pract. 2011;41(5):949-967, vi.

2. Johnston SA, Tobias KM. Veterinary Surgery: Small Animal. Vol 2. 2 ed: Elsevier, Inc.; 2018.

3. Youn G, Waschak MJ, Kunkel KAR, Gerard PD. Outcome of elective cholecystectomy for the treatment of gallbladder disease in dogs. J Am Vet Med Assoc. 2018;252(8):970-975.

4. Mehler SJ, Mayhew PD, Drobatz KJ, Holt DE. Variables associated with outcome in dogs undergoing extrahepatic biliary surgery: 60 cases (1988-2002). Vet Surg. 2004;33(6):644-649.

5. Smalle TM, Cahalane AK, Köster LS. Gallbladder mucocoele: A review. J S Afr Vet Assoc. 2015;86(1):1318.

6. Malek S, Sinclair E, Hosgood G, Moens NM, Baily T, Boston SE. Clinical findings and prognostic factors for dogs undergoing cholecystectomy for gall bladder mucocele. Vet Surg. 2013;42(4):418-426.

7. Worley DR, Hottinger HA, Lawrence HJ. Surgical management of gallbladder mucoceles in dogs: 22 cases (1999-2003). J Am Vet Med Assoc. 2004;225(9):1418-1422.

8. Pike FS, Berg J, King NW, Penninck DG, Webster CR. Gallbladder mucocele in dogs: 30 cases (2000-2002). J Am Vet Med Assoc. 2004;224(10):1615-1622.

9. Kesimer M, Cullen J, Cao R, et al. Excess Secretion of Gel-Forming Mucins and Associated Innate Defense Proteins with Defective Mucin Un-Packaging Underpin Gallbladder Mucocele Formation in Dogs. PLoS One. 2015;10(9):e0138988.

10. Klinkspoor JH, Kuver R, Savard CE, et al. Model bile and bile salts accelerate mucin secretion by cultured dog gallbladder epithelial cells. Gastroenterology. 1995;109(1):264-274.

11. Dray-Charier N, Paul A, Combettes L, et al. Regulation of mucin secretion in human gallbladder epithelial cells: predominant role of calcium and protein kinase C. Gastroenterology. 1997;112(3):978-990.

12. Klinkspoor JH, Yoshida T, Lee SP. Bile salts stimulate mucin secretion by cultured dog gallbladder epithelial cells independent of their detergent effect. Biochem J. 1998;332 (Pt 1):257-262.

13. Lavoie B, Balemba OB, Godfrey C, et al. Hydrophobic bile salts inhibit gallbladder smooth muscle function via stimulation of GPBAR1 receptors and activation of KATP channels. J Physiol. 2010;588(Pt 17):3295-3305.

14. Kakimoto T, Kanemoto H, Fukushima K, et al. Bile acid composition of gallbladder contents in dogs with gallbladder mucocele and biliary sludge. Am J Vet Res. 2017;78(2):223-229.

15. Kook PH, Schellenberg S, Rentsch KM, et al. Effect of twice-daily oral administration of hydrocortisone on the bile acids composition of gallbladder bile in dogs. Am J Vet Res. 2011;72(12):1607-1612.

16. Lee S, Kweon OK, Kim WH. Increased Leptin and Leptin Receptor Expression in Dogs With Gallbladder Mucocele. J Vet Intern Med. 2017;31(1):36-42.

17. Lee S, Lee A, Kweon OK, et al. Changes in pre- and postoperative serum leptin concentrations in dogs with gallbladder mucocele and cholelithiasis. BMC Vet Res. 2019;15(1):215.

18. Aguirre AL, Center SA, Randolph JF, et al. Gallbladder disease in Shetland Sheepdogs: 38 cases (1995-2005). J Am Vet Med Assoc. 2007;231(1):79-88.

19. Gookin JL, Correa MT, Peters A, et al. Association of Gallbladder Mucocele Histologic Diagnosis with Selected Drug Use in Dogs: A Matched Case-Control Study. J Vet Intern Med. 2015;29(6):1464-1472.

20. Jaffey JA, Pavlick M, Webster CR, et al. Effect of clinical signs, endocrinopathies, timing of surgery, hyperlipidemia, and hyperbilirubinemia on outcome in dogs with gallbladder mucocele. Vet J. 2019;251:105350.

21. Allerton F, Swinbourne F, Barker L, et al. Gall bladder mucoceles in Border terriers. J Vet Intern Med. 2018;32(5):1618-1628.

22. Saunders H, Thornton LA, Burchell R. Medical and surgical management of gallbladder sludge and mucocoele development in a Miniature Schnauzer. Int J Vet Sci Med. 2017;5(1):75-80.

23. Kim KH, Han SM, Jeon KO, et al. Clinical Relationship between Cholestatic Disease and Pituitary-Dependent Hyperadrenocorticism in Dogs: A Retrospective Case Series. J Vet Intern Med. 2017;31(2):335-342.

24. Mesich ML, Mayhew PD, Paek M, et al. Gall bladder mucoceles and their association with endocrinopathies in dogs: a retrospective case-control study. J Small Anim Pract. 2009;50(12):630-635.

25. Cook AK, Jambhekar AV, Dylewski AM. Gallbladder Sludge in Dogs: Ultrasonographic and Clinical Findings in 200 Patients. J Am Anim Hosp Assoc. 2016;52(3):125-131.

26. Kutsunai M, Kanemoto H, Fukushima K, et al. The association between gall bladder mucoceles and hyperlipidaemia in dogs: a retrospective case control study. Vet J. 2014;199(1):76-79.

27. Xenoulis PG. The association between hyperlipidemia and gall bladder mucoceles in dogs. Vet J. 2014;200(3):353-354.

28. Mealey KL, Minch JD, White SN, et al. An insertion mutation in ABCB4 is associated with gallbladder mucocele formation in dogs. Comp Hepatol. 2010;9:6.

29. Cullen JM, Willson CJ, Minch JD, et al. Lack of association of ABCB4 insertion mutation with gallbladder mucoceles in dogs. J Vet Diagn Invest. 2014;26(3):434-436.

30. Amsellem PM, Seim HB, MacPhail CM, et al. Long-term survival and risk factors associated with biliary surgery in dogs: 34 cases (1994-2004). J Am Vet Med Assoc. 2006;229(9):1451-1457.

31. Guess SC, Harkin KR, Biller DS. Anicteric gallbladder rupture in dogs: 5 cases (2007-2013). J Am Vet Med Assoc. 2015;247(12):1412-1414.

32. Jaffey JA, Graham A, VanEerde E, et al. Gallbladder Mucocele: Variables Associated with Outcome and the Utility of Ultrasonography to Identify Gallbladder Rupture in 219 Dogs (2007-2016). J Vet Intern Med. 2018;32(1):195-200.

33. Choi J, Kim A, Keh S, et al. Comparison between ultrasonographic and clinical findings in 43 dogs with gallbladder mucoceles. Vet Radiol Ultrasound. 2014;55(2):202-207.

34. Bargellini P, Orlandi R, Paloni C, et al. Contrast-enhanced ultrasound complements two-dimensional ultrasonography in diagnosing gallbladder diseases in dogs. Vet Radiol Ultrasound. 2018;59(3):345-356.

35. Bargellini P, Orlandi R, Paloni C, et al. Evaluation of contrast-enhanced ultrasonography as a method for detecting gallbladder necrosis or rupture in dogs. Vet Radiol Ultrasound. 2016;57(6):611-620.

36. Uno T, Okamoto K, Onaka T, et al. Correlation between ultrasonographic imaging of the gallbladder and gallbladder content in eleven cholecystectomised dogs and their prognoses. J Vet Med Sci. 2009;71(10):1295-1300.

37. Soppet J, Young BD, Griffin JF, et al. Extruded gallbladder mucoceles have characteristic ultrasonographic features and extensive migratory capacity in dogs. Vet Radiol Ultrasound. 2018;59(6):744-748.

38. Fuerst JA, Hostnik ET. CT attenuation values and mineral distribution can be used to differentiate dogs with and without gallbladder mucoceles. Vet Radiol Ultrasound. 2019;60(6):689-695.

39. Hayakawa S, Sato K, Sakai M, et al. CT cholangiography in dogs with gallbladder mucocoele. J Small Anim Pract. 2018.

40. Shelly S. Body cavity fluids. Atlas of Canine and Feline Cytology. Philadelphia, PA: Saunders; 2001.

41. Owens SD, Gossett R, McElhaney MR, et al. Three cases of canine bile peritonitis with mucinous material in abdominal fluid as the prominent cytologic finding. Vet Clin Pathol. 2003;32(3):114-120.

42. Fahie MA, Martin RA. Extrahepatic biliary tract obstruction: a retrospective study of 45 cases (1983-1993). J Am Anim Hosp Assoc. 1995;31(6):478-482.

43. Ludwig LL, McLoughlin MA, Graves TK, et al. Surgical treatment of bile peritonitis in 24 dogs and 2 cats: a retrospective study (1987-1994). Vet Surg. 1997;26(2):90-98.

44. Walter R, Dunn ME, d'Anjou MA, et al. Nonsurgical resolution of gallbladder mucocele in two dogs. J Am Vet Med Assoc. 2008;232(11):1688-1693.

45. Pitt HA, Murray KP, Bowman HM, et al. Clinical pathway implementation improves outcomes for complex biliary surgery. Surgery. 1999;126(4):751-756; discussion 756-758.

46. Littlefield A, Lenahan C. Cholelithiasis: Presentation and Management. J Midwifery Womens Health. 2019;64(3):289-297.

47. Parkanzky M, Grimes J, Schmiedt C, et al. Long-term survival of dogs treated for gallbladder mucocele by cholecystectomy, medical management, or both. J Vet Intern Med. 2019;33(5):2057-2066.

48. Yoshida T, Fukumoto T, Urade T, et al. Development of a new biodegradable operative clip made of a magnesium alloy: Evaluation of its safety and tolerability for canine cholecystectomy. Surgery. 2017;161(6):1553-1560.

49. Kanai H, Hagiwara K, Nukaya A, et al. Short-term outcome of laparoscopic cholecystectomy for benign gall bladder diseases in 76 dogs. J Vet Med Sci. 2018;80(11):1747-1753.

50. Mayhew PD, Mehler SJ, Radhakrishnan A. Laparoscopic cholecystectomy for management of uncomplicated gall bladder mucocele in six dogs. Vet Surg. 2008;37(7):625-630.

51. Simon A, Monnet E. Laparoscopic Cholecystectomy with Single Port Access System in 15 Dogs. Vet Surg. 2019.

52. Scott J, Singh A, Mayhew PD, et al. Perioperative Complications and Outcome of Laparoscopic Cholecystectomy in 20 Dogs. Vet Surg. 2016;45(S1):O49-O59.

53. Ooi LL, Goh YC, Chew SP, et al. Bile duct injuries during laparoscopic cholecystectomy: a collective experience of four teaching hospitals and results of repair. Aust N Z J Surg. 1999;69(12):844-846.

54. Sugimoto M, Yasuda H, Koda K, et al. Evaluation for transvaginal and transgastric NOTES cholecystectomy in human and animal natural orifice translumenal endoscopic surgery. J Hepatobiliary Pancreat Surg. 2009;16(3):255-260.

55. Li JY, Zhang F, Moon W, et al. Biliary tract reconstruction using an autologous vein graft in rats. J Reconstr Microsurg. 2000;16(1):51-55.

56. Karaayvaz M, Uğraş S, Güler O, et al. Use of an autologous vein graft and stent in the repair of common bile defects: an experimental study. Surg Today. 1998;28(8):830-833.

57. Mutter D, Aprahamian M, Evrard S, et al. Biomaterials for primary closure of a choledochotomy in dogs. Eur Surg Res. 1996;28(1):32-38.

58. Burns BR, Hofmeister EH, Brainard BM. Anesthetic complications in dogs undergoing hepatic surgery: cholecystectomy versus non-cholecystectomy. Vet Anaesth Analg. 2014;41(2):186-190.

59. Mizutani S, Torisu S, Kaneko Y, et al. Retrospective analysis of canine gallbladder contents in biliary sludge and gallbladder mucoceles. J Vet Med Sci. 2017;79(2):366-374.

60. Guillaumin J, Chanoit G, Decosne-Junot C, et al. Bilothorax following cholecystectomy in a dog. J Small Anim Pract. 2006;47(12):733-736.

61. Rogers E, Jaffey JA, Graham A, et al. Prevalence and impact of cholecystitis on outcome in dogs with gallbladder mucocele. J Vet Emerg Crit Care (San Antonio). 2020;30(1):97-101.

62. Wennogle SA, Randall EK, Priestnall SL, et al. Eubacterial fluorescence in situ hybridisation and histologic features in 25 dogs with gallbladder mucocele. J Small Anim Pract. 2019;60(5):291-297.

63. Crews LJ, Feeney DA, Jessen CR, et al. Clinical, ultrasonographic, and laboratory findings associated with gallbladder disease and rupture in dogs : 45 cases (1997-2007). Journal of American Veterinary Medical Association. 2009;1234(3):359-366.

Chapter 2 - Clinical findings and prognostic factors for dogs undergoing elective versus non-elective cholecystectomies for gallbladder mucoceles.

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Clinical findings and prognostic factors for dogs undergoing elective versus non-elective cholecystectomies for gallbladder mucoceles

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Abstract

Objectives: The objectives of this multiinstitutional retrospective study were to compare and contrast post-operative morbidity between elective and non-elective cholecystectomy cases in dogs with gallbladder mucoceles, as well as to determine underlying risk factors and prognostic indicators of dogs with early gallbladder mucoceles and their need for surgical intervention.

Animals: This study included 121 dogs with gallbladder mucoceles that had a cholecystectomy performed.

Procedures: Medical records were reviewed for all dogs with a gallbladder mucocele between 2004-2018 that underwent a cholecystectomy. Dogs were classified into three categories: emergent (gallbladder rupture or systemic instability), urgent (systemic clinical signs, diabetic, or biliary duct distension), or elective (no clinical signs or bloodwork abnormalities). The method of common bile duct catheterization, for example, antegrade or retrograde catheterization, was noted if performed. Post-operative complications and survival were compared between groups.

Results: Dogs that had a duodenotomy and retrograde common bile duct cannulation were 6.67 times more likely to develop a post-operative fever in comparison to dogs that had no common bile duct catheterization and 13.61 times more likely to develop post-operative fever in comparison to dogs that had normograde catheterization of the common bile duct. Dogs that had either urgent or emergent surgery were 3.45 and 4.3 times more likely, respectively, to die prior to discharge in comparison to those that had elective cholecystectomy.

Conclusions and Clinical Relevance: Dogs undergoing elective cholecystectomies have a lower mortality rate than those undergoing urgent or emergent procedures. Duodenotomy with retrograde bile duct catheterization is associated with post-operative fever development.

Introduction

Bile duct obstruction is a life-threatening emergency and can cause a number of sequelae affecting many body systems, including but not limited to gall bladder rupture, bile peritonitis, acute renal failure, multisystem organ failure, and death. Differentials for extrahepatic biliary obstruction include gallbladder mucocele, pancreatitis, neoplasia, cholelithiasis, and inflammatory biliary tract disease.[1,2] Gallbladder mucoceles are a common underlying cause of biliary tract obstruction in dogs.[1,3,4] Gallbladder mucoceles are defined as a mucus-filled distension of the gallbladder characterized by green-black, semisolid to immobile mucin.[4,5] This can lead to gallbladder or common bile duct wall necrosis and subsequent rupture.[5,6] In the absence of bacterial infection, bile normally causes only a mild chemical peritonitis, however, septic bile peritonitis can lead to severe life-threatening disease.[7]

The etiopathogenesis of gallbladder mucoceles is unknown and multifactorial.[1] One hypothesis includes hyperplasia of the mucus secreting glands of the gallbladder promoting increased mucus accumulation in the gallbladder.[1,6,8] It is unknown whether cystic hyperplasia of the gallbladder wall represents an inherited or acquired disease process. One study proposes that biliary sludge can lead to cholestasis, contributing to the pathogenesis of gallbladder mucocele disease progression.[9] Predisposed breeds reported include Shetland sheepdogs, cocker spaniels, and miniature schnauzers.[5,10]

The goal of treatment of biliary tract obstruction is to relieve the underlying cause of obstruction.[3] Cholecystectomy is the treatment of choice for gallbladder mucoceles, often coupled with either normograde or retrograde catheterization of the common bile duct to determine and establish patency.[3,5] Prognosis after a cholecystectomy varies. Mortality rates

range from 21.7-40%, however, the prognosis improves if the patient survives the immediate post-operative period.[2,4,5,8] Sterile bile peritonitis is associated with a good prognosis in the literature, but septic bile peritonitis has been reported to lead to 80-100% mortality.[2,5,7,8,11]

There are limited reports comparing underlying risk factors and surgical timing. Pre-operative factors that negatively impact prognosis include elevated ALT and GGT, hyperbilirubinemia, hypoalbuminemia, increasing age, elevated preanesthetic heart rate, azotemia, and hyperphosphatemia.[1,2] Post-operative hypotension, dyspnea, pancreatitis, hypoalbuminemia, anemia, hyperlactatemia, and presence of band neutrophils are significantly associated with post-operative mortality.[1,2,5]

The timing of surgical removal of the gallbladder is controversial. In human medicine, early interventional cholecystectomy for gallstones prior to the development of clinical signs and bloodwork abnormalities has been shown to significantly lower post-operative morbidity and mortality in comparison to medical management or later surgical management after development of clinical signs or bloodwork abnormalities.[12] A recent veterinary study compared survival between 45 elective and 25 nonelective cholecystectomies.[1] Dogs in the nonelective group had a significantly higher mortality rate of 20% compared to a 2% mortality rate of the dogs in the elective group.[1]

The purpose of this study was to compare and contrast post-operative morbidity between elective versus non-elective cholecystectomy cases in dogs with gallbladder mucoceles. A secondary purpose was to determine underlying risk factors and prognostic indicators of dogs with early gallbladder mucoceles and to determine their need for surgical intervention. Our hypothesis was that cases with early gallbladder mucoceles removed via elective cholecystectomy would have

less post-operative morbidity, a shorter hospitalization stay, and lower post-operative mortality rates compared to nonelective cholecystectomy cases.

Materials and Methods

Medical records from Kansas State University College of Veterinary Medicine and VCA California Veterinary Specialists from 2004-2018 were reviewed for cases of dogs with a diagnosis of a gallbladder mucocele. Inclusion criteria consisted of dogs with ultrasonographic evidence of a gallbladder mucocele that underwent a cholecystectomy via laparotomy. Exclusion criteria for this study included dogs that did not have a cholecystectomy, had a biliary surgical procedure other than a cholecystectomy with or without bile duct catheterization, or did not have either ultrasonographic or histologic confirmation of a gallbladder mucocele.

Data recorded included age, sex, clinical signs, and medical management prior to presentation to the hospital. Physical examination parameters, such as mucous membrane color, and pre-operative bloodwork parameters, such albumin, alkaline phosphatase (ALP), alanine aminotransferase (ALT), and total bilirubin, were collected. Abdominal ultrasound reports were reviewed and the following parameters were recorded as present or absent: effusion, evidence of gallbladder rupture, common bile duct distension, intrahepatic bile duct distension, and hyperechogenic liver. If there was peritoneal effusion present, fluid analysis and cytology were recorded. Pre-operative medical management was recorded. Surgical procedures were noted, specifically if a duodenotomy and retrograde common bile duct cannulation was performed or if a normograde catheterization of the common bile duct was performed.

Each cholecystectomy was classified into 3 categories of urgency: elective, urgent, or emergent, as summarized in table 1. A procedure was classified as emergent if the patient had a gallbladder mucocele with evidence of rupture or impending rupture (bile peritonitis, free floating mucocele, hyperechoic mesentery around the gallbladder) or systemic instability (hypotension, tachycardia,

or high lactate unresponsive to fluid resuscitation). A procedure was classified as urgent in patients that had a gallbladder mucocele that had none of the above criteria, however, the patient was hyperbilirubinemic, diabetic, or had biliary duct (common bile duct or intrahepatic duct) distension. A procedure was classified as elective if there was a gallbladder mucocele present with none of the criteria in the emergent or urgent categories.

Pre-, peri-, and post-operative antibiotics were recorded, as well as surgical and anesthetic time and complications. Where applicable, culture and biopsy results were noted. Placement of an esophagostomy tube was noted, as well as the feeding protocol. Post-operative medications and complications, including pre- and post-operative fever greater than 102.5 degrees Fahrenheit, regurgitation, vomiting, and respiratory distress, were recorded. Post-operative bloodwork was evaluated and the same variables as noted above were recorded, specifically noting at what point in time they were performed. Cause of death, time of death, and long-term follow-up were recorded, when applicable.

Statistical Analysis:

Analysis was performed on a commercial statistical program (WINKS SDA 7.0.9, Texasoft, Cedar Hill, TX www.texasoft.com). Urgency of surgery, presence of gallbladder rupture, and whether or not duodenotomy or normograde bile duct catheterization was performed were compared with the survival and length of stay by Independent group t-test. The findings of common bile duct distension, intrahepatic bile duct distension, concurrent disease, low or normal pre-operative albumin, normal or elevated pre-operative ALP, ALT, or bilirubin, low or normal post-operative albumin, and normal or elevated post-operative ALP, ALT, or bilirubin were compared with length of stay by independent group t-test. The pre-

operative and post-operative absolute values of albumin, ALP, ALT, and total bilirubin were compared with the length of stay by Pearson's Correlation.

Urgency of surgery, presence of gallbladder rupture, and whether or not duodenotomy or normograde bile duct catheterization was performed were compared with the overall survival, as well as the presence of regurgitation or fever by Chi-Square Analysis. The findings of common bile duct distension, intrahepatic bile duct distension, concurrent disease, low or normal pre-operative albumin, normal or elevated pre-operative ALP, ALT, bilirubin, low or normal post-operative albumin, and normal or elevated post-operative ALP, ALT, or bilirubin were compared with the overall survival as well as the presence of regurgitation or fever. The pre-operative and post-operative absolute values of albumin, ALP, ALT, and total bilirubin were compared with the overall outcome by analysis of variance with Newman-Keuls posthoc.

The pre-operative and post-operative absolute values of albumin, ALP, ALT, and total bilirubin were compared with the fever outcome and with the regurgitation outcome by Independent group t-test.

The presence of fever or regurgitation, and overall survival were compared with length of stay by independent group T-test. The presence of fever and regurgitation were compared with the overall outcome by Chi-Square analysis. The outcomes of fever and regurgitation were compared by Chi-Square analysis. A $p<0.05$ was considered significant for all comparisons. Normality of absolute blood values, age, and length of stay were performed by Modified Kolmogorov-Smirnov test; Pre-operative bloodwork values were not normally distributed. Patient age was not normally distributed.

Results

One hundred twenty-one dogs met the inclusion criteria for this study, 67 from Kansas State University College of Veterinary Medicine and 54 from VCA California Veterinary Specialists. Breeds included mixed-breed dogs (18), cocker spaniel (15), Shetland sheepdog (9), miniature schnauzer (8), bichon frise (8), beagle (7), dachshund (6), Pomeranian (6), chihuahua (6), toy poodle (6), Yorkshire terrier (5), shih tzu (4), miniature pinscher (3), Cairn terrier (2), Scottish terrier (2), and 1 each of a border collie, rat terrier, American eskimo, German shorthaired pointer, Labrador retriever, English bulldog, border terrier, wheaten terrier, toy fox terrier, Maltese, great Dane, Havanese, Jack Russell terrier, Australian cattle dog, pit bull, and a German shepherd. There were 58 spayed females, 1 intact female, 53 neutered males, and 9 intact males.

The most frequent clinical signs were lethargy (84 dogs, 71%), vomiting (82 dogs, 68%), and inappetence (77 dogs, 65%). The most common presenting complaint was elevated liver enzymes (100 dogs, 83%). Forty dogs had icteric mucous membranes (33%). Forty-five dogs had concurrent disease including diabetes mellitus (7), hyperadrenocorticism (7), hypothyroidism (4), pancreatitis (3), and a urinary tract infection (2).

Pre-operative bloodwork was available in most patients. The most common abnormalities were elevated alkaline phosphatase (97 dogs, 96%), elevated alanine aminotransferase (94 dogs, 91%), elevated total bilirubin (77 dogs, 72%), and hypoalbuminemia (32 dogs, 34%). One hundred and fifteen dogs had an abdominal ultrasound performed (115/121, 95%). One hundred and eight dogs had ultrasonographic evidence of a gallbladder mucocele (108/115, 94%). Of the dogs that had a gallbladder mucocele, scant effusion was noted surrounding the gallbladder in 46 dogs (46/108, 43%). Gall bladder rupture was suspected in 21 dogs (21/108, 19%). Choleliths were

reported in 6 (6/108, 5%). 111 dogs had biliary tree diameter reported on the ultrasound report and common bile duct distension was noted in 34/111 (31%). 112 dogs had intrahepatic duct diameter reported and distension was noted in 7/112 (6%). 112 dogs had enough information on the ultrasound report to record the echotexture of the mesentery; hyperechoic mesentery was noted in 51/112 (46%). Peritoneal fluid analysis was performed in 12 dogs and septic bile peritonitis was diagnosed in 6 (50%) of these dogs.

A cholecystectomy was performed in all cases. Elective procedures were performed in 31 dogs (26%), urgent procedures were performed in 52 dogs (43%), and emergent procedures were performed in 38 dogs (31%). Twenty-four cases had a ruptured gallbladder mucocele (20%). Duodenotomy and retrograde common bile duct catheterization was performed in 43 dogs (36%), while normograde common bile duct catheterization was performed in 51 dogs (42%). The common bile duct was not catheterized in 27 dogs (22%). Jackson-Pratt abdominal drainage was established in 9 dogs (7%). Concurrent surgical procedures included a liver biopsy (113), esophagostomy tube (19), cystotomy (8), splenectomy (7), gastrointestinal biopsies (5), choledochotomy with stent placement (3), liver lobectomy (2), kidney biopsy (2), pancreatic biopsy (2), neuter (2), gastric tube placement (2), prostatic biopsy (1), and gastropexy (1). Surgical complications were noted in 15 dogs and most commonly included gallbladder rupture (5) and common bile duct rupture (4). Mean anesthesia and surgical times were 176 and 118 minutes, respectively. Seventy-one dogs experienced anesthetic complications, most commonly including hypotension (62 dogs, 87%) and arrhythmias (5 dogs, 7%).

Bile culture was positive in 28/115 (24%). The most common bacteria were Escherichia coli (6), Enterococcus faecalis (6), and Staphylococcus epidermidis (5). Gallbladder histopathology was consistent with gallbladder mucoceles in 86 cases and not available in 35 cases. Post-operative

care included intravenous fluid therapy, injectable pain medication, gastroprotectants, anti-emetics, and other supportive care, as needed.

Post-operative bloodwork was available within the first 48 hours of surgery in 89 dogs with albumin reported in 69 dogs, alkaline phosphatase and alanine aminotransferase reported in 70 dogs, and total bilirubin reported in 89 dogs. The most common abnormalities demonstrated were elevated alkaline phosphatase (68/70 dogs, 97%), elevated alanine aminotransferase (65/70 dogs, 93%), elevated total bilirubin (64/89 dogs, 72%), and hypoalbuminemia (59/69 dogs, 86%).

Fifty-nine dogs experienced post-operative complications (50%), which are listed in table 2. Nineteen dogs developed post-operative fever (16%). Post-operative fever was significantly associated ($p<0.001$) with duodenotomy and retrograde common bile duct catheterization. Dogs that had a duodenotomy and retrograde common bile duct catheterization were 6.67 times more likely to develop a post-operative fever in comparison to dogs that had no common bile duct catheterization and 13.61 times more likely to develop post-operative fever in comparison to dogs that had normograde catheterization of the common bile duct (p-value,<0.001).

Post-operative fever was significantly associated with pre-operative hypoalbuminemia (p-value, 0.011) and post-operative regurgitation (p value, 0.012). Pre-operative hypoalbuminemia was 4.06 times more likely and post-operative regurgitation was 3.52 times more likely to be associated with post-operative fever. Post-operative fever was not significantly associated with urgency, gall bladder rupture, common bile duct distension, and other pre- and post-operative bloodwork values.

Post-operative regurgitation developed in 34 dogs (29%), while vomiting was present in 6 dogs (5%). Post-operative regurgitation was significantly (p-value, 0.004) affected by the urgency category. Dogs in the urgent category were 1.42 times more likely to develop regurgitation than those in the elective category. There was no significant difference found for dogs in the emergent category in comparison to either the elective or the urgent category in regards to the development of post-operative regurgitation.

Respiratory distress developed in 7 dogs (6%). Septic abdomen was diagnosed post-operatively in 6 dogs based on fluid cytology (5%). Another 4 dogs had a presumptive diagnosis of septic abdomen after their death, however, this was not confirmed. Other complications that occurred in 30 dogs included persistent hyperbilirubinemia (6), vomiting (6), hypertension (5), seizures (5), hypoglycemia (4), hypotension (4), thrombocytopenia and disseminated intravascular coagulation (2), diarrhea (2), cough (2), aspiration pneumonia (1), dyspnea of unknown cause (1), azotemia (1), oliguria (2), anuria (1), collapsing episode (1), and anorexia (2). These parameters were not compared statistically due to low numbers.

The average hospital stay of all patients was 4.88 days. The length of stay was significantly associated with post-operative fever (p value, 0.003), post-operative regurgitation (p value <0.001), and survival (p value 0.002. Dogs with a post-operative fever were hospitalized a mean of 6.42 days (+/- 2.61) versus 4.49 days (+/- 2.51) for dogs without a fever. Dogs with fever were 5.07 times more likely to be hospitalized longer than 7 days. Dogs that developed post-operative regurgitation had a significantly longer hospital stay; 6.76 days (+/- 3.48) versus 4.01 days (+/- 1.63) for those that did not regurgitate post-operatively. Dogs with post-operative regurgitation were 25.85 times more likely to be hospitalized longer than 7 days (p-value, <0.001). Length of stay for dogs that survived to discharge, were euthanized, or had cardiopulmonary arrest

included 4.61 days (+/- 2.02), 6.60 days (+/- 5), and 1.67 days (+/- 0.58), respectively. Dogs that survived to discharge had a 13.85 times shorter hospital stay in comparison to those that were euthanized or died. The length of stay was not associated with urgency, presence of gallbladder rupture, method of bile duct cannulation, amount of common bile duct distension, or pre- and post-operative bloodwork values.

Ninety-eight dogs survived to discharge (81%). Twenty-three dogs died prior to discharge (19%), 20 dogs (17%) were euthanized and 3 dogs (2%) experienced cardiopulmonary arrest. Reasons for death or euthanasia included suspected septic peritonitis (7), cardiac arrest (2), aspiration pneumonia (2), DIC (1), hepatic failure (1), pancreatitis (1), persistent hyperbilirubinemia (1), acute renal failure (1), respiratory distress (1), and unknown causes (5). Another patient required emergency surgery for common bile duct stent placement 14 days post-cholecystectomy. This dog was euthanized on the table after the common bile duct ruptured. Three out of the 6 confirmed septic peritonitis cases died (50%), along with four other dogs with presumptive diagnosis of septic peritonitis.

Survival was significantly (p value, 0.038) associated with the urgency category assigned. For the elective procedures, 29/31 dogs lived, 2/31 were euthanized, and 0/31 experienced cardiopulmonary arrest. 42/52 dogs that underwent an urgent cholecystectomy lived, 10/52 were euthanized, and 0/52 died of cardiopulmonary arrest. For the emergent procedures, 27/38 dogs lived, 8/38 were euthanized, and 3/38 died of cardiopulmonary arrest. Animals that had either urgent (p-value, 0.038) or emergent (p-value, 0.038) surgery were 3.45 and 4.3 times more likely, respectively, to die prior to discharge in comparison to those that had elective surgery. There was no difference in survival when comparing the urgent versus emergent surgeries. Survival was not significantly affected by type of common bile duct catheterization.

Discussion

The present study compared survival and prognostic factors between elective versus non-elective cholecystectomy cases in dogs with gallbladder mucoceles. In this study, elective procedures carried a 6% mortality rate in comparison to a 20% mortality rate for urgent procedures and a 29% mortality rate for emergent procedures, supporting the original hypothesis. Animals with either an urgent or emergent classification were significantly more likely to die prior to discharge in comparison to those in the elective category. These results are comparable to the results of Youn et al demonstrating an elective mortality rate of 2% in comparison to a nonelective mortality rate of 20%.[1] Another study reported that dogs that were clinical for gallbladder mucocele had significantly greater odds of death in comparison to those dogs that were subclinical.[13] Parkanzky et al demonstrated that patients undergoing medical management for gallbladder mucocele had a lower median survival time than those undergoing cholecystectomy, further supporting the need for early surgical intervention.[14]

This study classified patients into one of three categories of urgency. This was modified from the classification system used by Youn et al, which classified patients into elective versus nonelective cholecystectomies, with the latter category representing patients that are icteric or had questionable bile duct patency.[1] The modified classification scheme seen in this study was implemented to more closely match clinical decision making and prognosis by separating not only elective from nonelective, but also comparing elective, urgent, and emergent cases. However, there was no difference in survival when comparing the urgent versus emergent surgeries. Therefore, the elective versus nonelective classification may be sufficient for decision making.

Presence of gallbladder rupture was not associated with survival in this study. Survival of patients with gallbladder rupture has been variable in the literature. Several previously published studies have demonstrated a lack of correlation between gallbladder rupture and overall survival.[5,6,11,15] However, these results may be due to lack of statistical power, low case numbers, and the retrospective nature of these studies. One retrospective study with 219 dogs reported that dogs with intraoperative evidence of gallbladder rupture and bile peritonitis had 2.7 times higher risk of death than those that did not have evidence of rupture.[7]

The present study demonstrated a high complication rate of 50%. Nineteen dogs developed post-operative fever (16%). Pre-operative hypoalbuminemia and post-operative regurgitation were also significantly associated with development of post-operative fever. These complications are likely associated with the patient's overall poorer clinical status, as well as the development of possible aspiration pneumonia. Post-operative regurgitation developed in 29% of patients. Dogs in the urgent category are 1.42 times more likely to develop regurgitation than those in the elective category. There was not a significant difference between patients in the emergent category and the development of post-operative fever, likely due to low patient numbers in this study. Patients in the elective category are less likely to have post-operative clinical signs, which is consistent to the previously reported literature.[1,7] The length of stay was significantly associated with post-operative fever, post-operative regurgitation, and survival in this study.

Post-operative fever was significantly associated with duodenotomy and retrograde common bile duct catheterization. It is suspected that retrograde flushing of the bile duct allows enteric bacteria to be flushed into the hepatobiliary system, although further prospective studies are needed for fully evaluate this finding. Methods and overall requirement of determining common bile duct patency are controversial. In the present study, dogs that had a duodenotomy and

retrograde common bile duct catheterization were significantly more likely to develop a post-operative fever in comparison to either dogs that had no common bile duct catheterization or dogs that had normograde common bile duct catheterization. Dogs that undergo elective cholecystectomy often do not require retrograde common bile duct catherization and thus are associated with less complications and better survival. In one study, 38 dogs did not have catherization of the common bile duct performed and none had to return to surgery to reestablish patency.[1] Another study found a similar association although claimed that this could be a biased result due to surgeon preference.[5] More studies need to be performed in order to further elucidate recommendations on method of common bile duct catheterization.

Breed findings, clinical signs, ultrasonographic findings, and bloodwork parameters in this study are consistent with previously reported findings.[2,5,10] Survival was significantly affected by pre- and post-operative albumin levels, pre- and post-operative ALP levels, post-operative total bilirubin levels, post-operative regurgitation, and post-operative fever. These are consistent with previously reported findings.[2,5,11,14] Elevated lactate and immediate post-operative hypotension has also been reported to be associated with poor clinical outcome.[5,11] Other studies have demonstrated that hyperglobulinemia, presence of band neutrophils, elevated lymphocyte counts, blood urea nitrogen and creatinine levels, and prolonged partial thromboplastin times have been shown as risk factors.[2,5,11,13] These parameters were not found to be significant in this study, however, this may be due to the retrospective nature, lack of complete medical records, and low numbers with these abnormalities.

There are several limitations of this study. The retrospective nature led to lack of randomization and controls, which may have caused an incorrect classification of some of the patients, skewing the data and overall recommendations. Another limitation is that multiple surgeons participated

in the study and whether or not an elective procedure was performed was based on surgeon preference. Therefore, some animals with similar disease are not included in the study because they never received a cholecystectomy.

In conclusion, elective cholecystectomy for gallbladder mucocele carries a lower overall mortality rate in comparison to urgent and emergent procedures. Pre- and post-operative hypoalbuminemia, elevated pre-operative and post-operative ALP levels, elevated post-operative total bilirubin levels, presence of post-operative regurgitation, and post-operative fever were identified as risk factors. Duodenotomy and retrograde common bile duct catherization is associated with post-operative fever and also negatively affects survival. Given the lower mortality rate, our recommendation is to remove gallbladder mucoceles via elective cholecystectomy prior to the development of clinical signs due to the improved outcome.

References

1. Youn G, Waschak MJ, Kunkel KAR, et al. Outcome of elective cholecystectomy for the treatment of gallbladder disease in dogs. J Am Vet Med Assoc. 2018;252(8):970-975.

2. Amsellem PM, Seim HB, MacPhail CM, et al. Long-term survival and risk factors associated with biliary surgery in dogs: 34 cases (1994-2004). J Am Vet Med Assoc. 2006;229(9):1451-1457.

3. Mehler SJ. Complications of the extrahepatic biliary surgery in companion animals. Vet Clin North Am Small Anim Pract. 2011;41(5):949-967, vi.

4. Smalle TM, Cahalane AK, Köster LS. Gallbladder mucocoele: A review. J S Afr Vet Assoc. 2015;86(1):1318.

5. Malek S, Sinclair E, Hosgood G, et al. Clinical findings and prognostic factors for dogs undergoing cholecystectomy for gall bladder mucocele. Vet Surg. 2013;42(4):418-426.

6. Worley DR, Hottinger HA, Lawrence HJ. Surgical management of gallbladder mucoceles in dogs: 22 cases (1999-2003). J Am Vet Med Assoc. 2004;225(9):1418-1422.

7. Jaffey JA, Graham A, VanEerde E, et al. Gallbladder Mucocele: Variables Associated with Outcome and the Utility of Ultrasonography to Identify Gallbladder Rupture in 219 Dogs (2007-2016). J Vet Intern Med. 2018;32(1):195-200.

8. Pike FS, Berg J, King NW, et al. Gallbladder mucocele in dogs: 30 cases (2000-2002). J Am Vet Med Assoc. 2004;224(10):1615-1622.

9. Tsukagoshi T, Ohno K, Tsukamoto A, et al. Decreased gallbladder emptying in dogs with biliary sludge or gallbladder mucocele. Vet Radiol Ultrasound. 2012;53(1):84-91.

10. Aguirre AL, Center SA, Randolph JF, et al. Gallbladder disease in Shetland Sheepdogs: 38 cases (1995-2005). J Am Vet Med Assoc. 2007;231(1):79-88.

11. Mehler SJ, Mayhew PD, Drobatz KJ, et al. Variables associated with outcome in dogs undergoing extrahepatic biliary surgery: 60 cases (1988-2002). Vet Surg. 2004;33(6):644-649.

12. Pitt HA, Murray KP, Bowman HM, et al. Clinical pathway implementation improves outcomes for complex biliary surgery. Surgery. 1999;126(4):751-756; discussion 756-758.

13. Jaffey JA, Pavlick M, Webster CR, et al. Effect of clinical signs, endocrinopathies, timing of surgery, hyperlipidemia, and hyperbilirubinemia on outcome in dogs with gallbladder mucocele. Vet J. 2019;251:105350.

14. Parkanzky M, Grimes J, Schmiedt C, et al. Long-term survival of dogs treated for gallbladder mucocele by cholecystectomy, medical management, or both. J Vet Intern Med. 2019;33(5):2057-2066.

15. Crews LJ, Feeney DA, Jessen CR, et al. Clinical, ultrasonographic, and laboratory findings associated with gallbladder disease and rupture in dogs : 45 cases (1997-2007). Journal of American Veterinary Medical Association. 2009;1234(3):359-366.

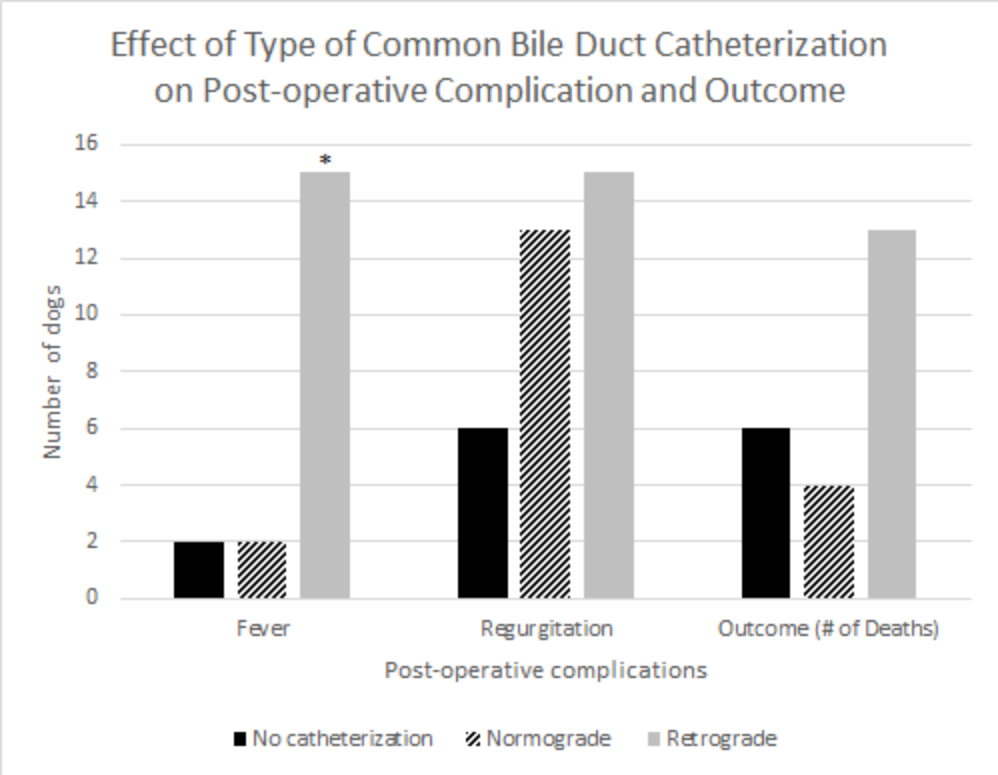

Figure 2-1: Columns marked with the asterisks are significantly different ($p<0.05$).

Table 2-1: Urgency Categories	
Emergent	• Definitive or impending ruptured gallbladder mucocele determined by the presence of: – Bile peritonitis: elevated bilirubin within the abdominal fluid sample that is two times higher than that of the serum bilirubin – Free floating mucocele – Hyperechoic mesentery around the gallbladder • Systemic instability – Hypotension (systolic blood pressure less than 90mmHg or mean arterial pressure less than 70mmHg) – Tachycardia (heart rate greater than 160bpm) – Hyperlactatemia unresponsive to fluid resuscitation (Greater than 2mmol/L)
Urgent	• Gallbladder mucocele that had none of the above criteria but one or more of the following: – Hyperbilirubinemia (Elevated above 0.2mg/dL) – Diabetes mellitus (persistent hyperglycemia due to relative or absolute insulin deficiency, treated or untreated with insulin) – Biliary duct (common bile duct or intrahepatic duct) distension (greater than 3mm)
Elective	• Gallbladder mucocele present with none of the above-mentioned criteria

Table 2-2: Post-operative Complications				
	Total	Elective	Urgent	Emergent
Fever	19	5	6	8
Regurgitation	34	10*	21*	3
Septic abdomen	6	1	4	1
Vomiting	6	1	3	2
Respiratory distress, unclassified	7	1	3	3
Persistent hyperbilirubinemia	6	0	6	0
Hypertension	5	3	2	0
Seizures	5	2	1	2
Hypoglycemia	4	1	2	1
Hypotension	4	0	0	4
Thrombocytopenia /Disseminated intravascular coagulation	2	1	0	1
Diarrhea	2	0	1	1
Cough	2	1	1	0
Aspiration pneumonia	1	1	0	0
Dyspnea of unknown cause	1	0	0	1
Azotemia	1	0	1	0
Oliguria	2	0	0	1
Anuria	1	0	0	1
Collapsing episode	1	0	0	1
Anorexia	2	2	0	0

Table 2-2: Items in the same row marked with an asterisk are significantly different ($p<0.001$)