**Original Article** 





Percutaneous cholecystocentesis in cats with suspected hepatobiliary disease Journal of Feline Medicine and Surgery 2017, Vol. 19(12) 1254–1260 © The Author(s) 2017 Reprints and permissions: sagepub.co.uk/journalsPermissions.nav DOI: 10.1177/1098612X16689335 journals.sagepub.com/home/ifms

This paper was handled and processed by the American Editorial Office (AAFP) for publication in JFMS



# Victoria L Byfield<sup>1\*</sup>, Julie E Callahan Clark<sup>1†</sup>, Bradley J Turek<sup>2‡</sup>, Charles W Bradley<sup>2</sup> and Mark P Rondeau<sup>1</sup>

## Abstract

*Objectives* The objective was to evaluate the safety and diagnostic utility of percutaneous ultrasound-guided cholecystocentesis (PUC) in cats with suspected hepatobiliary disease.

*Methods* Medical records of 83 cats with suspected hepatobiliary disease that underwent PUC were retrospectively reviewed.

*Results* At the time of PUC, at least one additional procedure was performed in 79/83 cats, including hepatic aspiration and/or biopsy (n = 75) and splenic aspiration (n = 18). Complications were noted in 14/83 cases, including increased abdominal fluid (n = 11), needle-tip occlusion (n = 1), failed first attempt to penetrate the gall bladder wall (n = 1) and pneumoperitoneum (n = 1). There were no reports of gall bladder rupture, bile peritonitis or hypotension necessitating treatment with vasopressor medication. Blood products were administered to 7/83 (8%) cats. Seventy-two cats (87%) survived to discharge. Of the cats that were euthanized (9/83) or died (2/83), none were reported as a definitive consequence of PUC. Bacteria were identified cytologically in 10/71 samples (14%); all 10 had a positive aerobic bacterial culture. Bile culture was positive in 11/80 samples (14%). Of the cases with a positive bile culture, cytological description of bacteria corresponded to the organism cultured in fewer than 50% of cases. The most common cytologic diagnosis was hepatic lipidosis (49/66). The most common histopathologic diagnosis was cholangitis (10/21).

*Conclusions and relevance* PUC was safe in this group of cats with suspected hepatobiliary disease. Complications were likely associated with ancillary procedures performed at the time of PUC. Bile analysis yielded an abnormal result in nearly one-third of cats with suspected hepatobiliary disease. Complete agreement between bile cytology and culture was lacking. Further evaluation of the correlation between bile cytology and bile culture is warranted.

Accepted: 18 December 2016

## Introduction

Feline hepatobiliary disease can be challenging to diagnose. While histopathology is considered the gold standard for reaching a diagnosis, hepatic biopsies are often not obtained owing to lack of owner consent or risk to the potentially unstable patient.<sup>1,2</sup> The most common hepatobiliary diseases in cats include hepatic lipidosis and cholangitis.<sup>3–5</sup> Given the non-specific clinical presentation and commonality of standard biochemical abnormalities, advanced diagnostic evaluation of these patients becomes critical to establish an effective treatment plan. Furthermore, it is possible that the two conditions occur concurrently. Therefore, when standard evaluation fails to characterize the hepatobiliary disease, obtaining a liver and bile sample may be warranted.

Both neutrophilic and lymphocytic cholangitis have been associated with bactibilia.<sup>1,6–12</sup> Given the potential

for bacterial infection, bile culture could provide valuable diagnostic information. Culture of bile, as opposed to

#### Corresponding author:

Julie Callahan Clark DVM, DACVIM, New England Animal Medical Center, West Bridgewater, MA, USA Email: juleslpcali@gmail.com

<sup>&</sup>lt;sup>1</sup>Department of Clinical Studies, University of Pennsylvania, School of Veterinary Medicine, Philadelphia, PA, USA <sup>2</sup>Department of Anatomic Pathobiology, University of

Pennsylvania, School of Veterinary Medicine, Philadelphia, PA, USA

<sup>\*</sup>Current address: Mount Laurel Animal Hospital, Mount Laurel, NJ, USA

<sup>&</sup>lt;sup>†</sup>Current address: New England Animal Medical Center, West Bridgewater, MA, USA

<sup>&</sup>lt;sup>‡</sup>Current address: Antech Diagnostics, Irvine, CA, USA

hepatic tissue, is more likely to isolate bacteria.<sup>13,14</sup> Therefore, obtaining bile culture routinely during the diagnostic evaluation of a cat with suspected hepatobiliary disease would both aid in diagnosis and help determine effective therapy.

Cholecystocentesis can be performed percutaneously with ultrasound guidance, during laparoscopy or during an abdominal exploratory surgery. In healthy cats, percutaneous ultrasound-guided cholecystocentesis (PUC) was found to be safe and technically simple with no major complications.<sup>15</sup> PUC has also been described as a minimally invasive technique for bile collection in several species, including pigs, cows and dogs.<sup>16–19</sup> It has been used successfully in people for diagnostic and therapeutic purposes.<sup>20–27</sup>

In people it has been suggested that PUC carries an increased risk of complications such as hemorrhage, bacteremia, intraperitoneal bile leakage and vagal hypotension.<sup>25,28,29</sup> Limited information is available regarding the safety of PUC in cats with pre-existing hepatobiliary disease. In a small case series reporting the clinicopathological findings from six cats with acute neutrophilic cholangitis, the authors concluded that PUC was an effective and minimally invasive diagnostic technique to identify bactibilia in affected cats. However, one of the four cats that had PUC developed gall bladder rupture and bile peritonitis.<sup>6</sup>

The purpose of our study was to describe the frequency and type of complications associated with PUC in a population of cats with suspected hepatobiliary disease. We also aimed to determine the cytologic utility of bile evaluation and evaluate the correlation between bile cytology and bile culture. We hypothesized that performing PUC in cats with suspected hepatobiliary disease would be safe and yield useful diagnostic information.

### Materials and methods

The Matthew J Ryan Veterinary Hospital of the University of Pennsylvania radiology database was retrospectively queried to identify cats that had bile collected via PUC for analysis between January 2003 and September 2012. The clinical pathology database was searched for cholecystocentesis and/or bile cytology and the microbiology database was searched for bile culture over the same time frame.

A total of 154 records were selected for review. Of those, 41 were excluded because of missing or incomplete medical records. An additional 30 were excluded because PUC was not performed, leaving 83 records for review. Medical records were reviewed by a single investigator (VLB). Data collection included signalment, presenting complaint, vital parameters, clinicopathology, radiology and ultrasound imaging, bile and hepatic cytology, bile culture, hepatic histopathology and final diagnosis. All hepatic biopsies were reviewed by a pathology resident (BJT) and a board-certified pathologist (CWB). Hepatic biopsies were classified according to World Small Animal Veterinary Association standards.<sup>2</sup> Records were reviewed for complications that could have been associated with PUC, including comments in the radiology report from the time of PUC, pre- (within 24 h of PUC) and post- (12–24 h after PUC) procedure packed cell volume (PCV) and non-invasive blood pressure measurements, as well as the need for vasopressor or blood product administration following the procedure. Blood pressure was measured using ultrasonic Doppler flow detectors. Survival data included time to discharge and the last known visit to the hospital.

All PUC procedures were performed by a boardcertified radiologist or radiology resident. Standard protocol consisted of introducing a 22 G needle (attached to an extension set, three-way stopcock and 10 ml syringe) through the ventral or ventrolateral portion of the liver into the gall bladder. Patient positioning varied depending on access to the gall bladder and ability to aspirate through the liver, but typically the patient was in dorsal or left lateral recumbency. As much bile as possible was aspirated from the gall bladder in an attempt to minimize bile leakage. There was minimal variation in technique, depending on clinician preference and accessibility to the gall bladder. Repeat ultrasound examination of the cranial-ventral abdomen was performed immediately after completion of all procedures in each patient, to evaluate for the presence of free peritoneal fluid. Cats were either heavily sedated or placed under general anesthesia (propofol induction, maintenance with isoflurane gas) for the procedure.

Immediate complications were defined as any adverse event reported by the radiologist at the time of the procedure. Complications were categorized as major or minor. Major complications were defined as life-threatening insults requiring therapeutic intervention, or the appearance of new or increased free abdominal fluid. Minor complications were either related to a procedural obstacle (such as with the needle or ability of the radiologist to penetrate the gall bladder wall) or an insult to the patient which did not require intervention (such as minor skin bleeding from the needle puncture).

#### Results

A total of 83 PUCs were performed. Cases included 45 male castrated cats and 38 female spayed cats with a median age of 10 years (range 1–17 years) and a median body weight of 4.125 kg (range 2–9 kg). The most common breed represented was domestic shorthair (65/83; 78%). The most common presenting complaints were decreased appetite or anorexia (65/83; 78%), lethargy (52/83; 63%), vomiting (47/83; 57%), weight loss (44/83; 53%), diarrhea (11/83; 13%) and jaundice (8/83; 10%).

21

Parameter	RI	Median	Range	% Above RI
ALT (n = 81)	33–152 U/I	349	13–3352	77
AST (n = 75)	1–37 U/I	180	21–836	92
ALP (n = 80)	22–87 U/I	177.5	14–3997	65
GGT (n = 74)	5–19 U/I	7.5	5–463	20
TBILI ( $n = 79$ )	0.1–0.8 mg/dl	1.4	0.1–13.9	57

Table 1 Serum liver enzyme activity, total bilirubin and cholesterol parameters

RI = reference interval; ALT = alanine aminotransferase; AST = aspartate aminotransferase; ALP = alkaline phosphatase; GGT = gammaglutamyl transpeptidase; TBILI = total bilirubin; CHOL = cholesterol

45-410

184

A pre-procedure complete blood count was available for 68/83 cats (82%). Twenty-four of 68 (35%) were classified as anemic (hematocrit <31.7%). Ten of 68 (15%) had leukocytosis (>18.7 ×10<sup>3</sup>/µl) and 15/68 (22%) had neutrophilia (>14 × 10<sup>3</sup>/µl). Seven cats were thrombocytopenic (<175,000/µl). Liver enzyme activity, total bilirubin and cholesterol values are presented in Table 1. Elevated liver enzyme activity was common and hyperbilirubinemia was present in over half of the population.

96-248 mg/dl

Coagulation times were measured in 52/83 cats (63%). Nineteen of 52 (37%) had a prolonged activated partial thromboplastin time (aPTT; >15.7 s) and 9/52 (17%) had a prolonged prothrombin time (PT; >13.2 s). Prolongation of both PT and aPTT was present in 6/52 (12%).

A complete abdominal ultrasound report, prior to PUC, was available in 77/83 cats (93%). In sixty-five cats (84%) the liver was reported to be abnormal. The most common abnormalities included hepatomegaly (60/77; 78%), hyperechoic parenchyma (47/77; 61%), hypo-echoic parenchyma (14/77; 18%) and coarse parenchyma (11/77; 14%). The gall bladder or biliary tract appeared abnormal in 55/77 cats (71%). Abnormalities are presented in Table 2. Thirty-three cats (43%) had abdominal effusion.

PUC was performed under general anesthesia in 39/83 cats (47%), with the remainder performed under sedation. A procedure other than cholecystocentesis was

 Table 2
 Biliary tract ultrasonographic abnormalities in 77 cats

Abnormality	Number of cats (%)
Increased gall bladder sediment Distended common bile duct Thickened common bile duct wall Thickened gall bladder wall	33 (43) 32 (42) 22 (29) 19 (25)
Distended gall bladder	15 (20)
Dilated intrahepatic bile ducts	9 (12)
Presence of cholelith(s)	8 (10)
Hyperechoic common bile duct wall	6 (8)

performed concurrently in 79/83 cats (95%). Hepatic aspiration was the most common procedure (66/83; 80%) followed by esophagostomy tube placement (20/83; 24%), hepatic Tru-cut biopsy (17/83; 20%), upper gastrointestinal endoscopy with gastric and duodenal biopsies (18/83; 22%), splenic fine-needle aspiration (18/83; 22%), mesenteric lymph node aspiration (9/83; 11%), abdominocentesis (3/83; 4%), renal aspiration (2/83; 2%), abdominal mass aspiration (2/83; 2%), lung mass aspiration (1/83; 1%) and endotracheal wash (1/83; 1%). Eight cats had both a hepatic aspirate and biopsy performed.

Immediate complications were reported by the radiologist in 14/83 cats (17%) at the time of PUC. Complications were minor in 3/83 (4%), including one needle-tip occlusion, one re-aspiration of the gall bladder following a failed first attempt because the gall bladder wall was thickened and difficult to penetrate, and one pneumoperitoneum that developed following multiple unsuccessful attempts to penetrate the gall bladder. The radiology report cited inadequate sedation as being the possible cause of this technical difficulty. Major complications were reported in 11/83 (13%). All 11 cats had some increase in abdominal effusion indicative of hemorrhage. All 11 cats had concurrent procedures performed including liver fine-needle aspiration (n = 5), combined liver fine-needle aspiration and Trucut liver biopsies (n = 3), combined splenic fine-needle aspiration and Tru-cut liver biopsies (n = 1), combined liver and splenic fine-needle aspiration (n = 1) and Trucut liver biopsies (n = 1). Effusion was noted immediately after one of these concurrent procedures in 9/11 cats. In the remaining two cats, effusion was observed after all procedures were performed (one cat had combined liver fine-needle aspiration and Tru-cut liver biopsies, and one had combined liver and splenic fine-needle aspiration performed). Of these 11 cats, six had effusion noted on abdominal ultrasound prior to PUC. Two of the cats had normal coagulation times, four cats had prolonged aPTT, three had prolonged PT and aPTT, and two did not have coagulation times performed. Five of the 11 cats received vitamin K prior to procedures and three

CHOL (n = 75)

received fresh frozen plasma. There were no reports of gall bladder rupture or suspected bile leakage in any cat.

The median systolic blood pressure recorded in 24 cats immediately after PUC (<5 mins) was 105 mmHg (range 80–160 mmHg). Within a median of 1 h after the procedure (range 0.5–5 h), the median blood pressure was 120 mmHg (range 90–170 mmHg). In 13 cases a blood pressure was available before and immediately after PUC; the median change in blood pressure of these cases was -30 mmHg (range -72 to +50 mmHg). In one cat that developed hemorrhage secondary to liver and splenic aspiration, the systolic blood pressure was 50 mmHg 2 h after the aspiration procedures and prior to resuscitative efforts. No cat required vasopressor therapy.

Seventy cats had a pre-procedure PCV recorded with a median of 30% (range 18–50%). Fifty-nine cats had a PCV the morning following the procedure with a median PCV of 28% (range 15–43%). In 55 cats paired PCV measurements were available (within 24 h of each other) with a median change in PCV of -4% (range -14 to +5%). In addition to sedation and/or general anesthesia, most cats received intravenous fluid therapy between PUC and venepuncture the following morning.

Blood product transfusion was performed in 7/83 cats (8%) after PUC. One cat received fresh whole blood, four cats received packed red blood cells and four cats were given fresh frozen plasma. Two of these cats received a combination of blood and plasma. One transfusion was administered immediately following the procedures after hemorrhage was identified. Three transfusions were given the following day and one transfusion was given 2 days after the procedure, based on declining PCV. Of the four cats that received plasma, one had prolongation of both PT/aPTT and the remaining cats had prolongation of only aPTT. Three cats had a combination of anemia and coagulopathy. All seven cats requiring transfusion had a liver aspirate performed in addition to PUC.

Bile from 71/83 cats (86%) was examined cytologically. Aerobic and anaerobic culture was performed on bile from 80/83 cats (96%). Paired cytology and culture were performed in 68/83 cats (82%). No cytologic abnormalities were noted in 57/71 samples (80%). Bacteria (rods, cocci or both) were seen in 10/71 (14%). Bacterial culture was positive in 9/10 samples in which bacteria were reported cytologically. Neutrophilic inflammation was seen with bacteria in 4/71 (6%) and without bacteria in 4/71 (6%). The four cases with aseptic neutrophilic inflammation had negative bile cultures. Bile culture was positive for aerobic bacterial growth in 11/80 samples (14%). *Escherichia coli* was the most common isolate (7/11). Other organisms included alpha-hemolytic Streptococcus species (n = 2), Klebsiella pneumoniae (n = 1), Pseudomonas aerugi*nosa* (n = 1) and *Enterococcus faecalis* (n = 1). No anaerobic

bacteria were cultured. All 11 cats with positive bacterial cultures had concurrent bile cytology performed. Of those 11, bacteria were seen cytologically in 10 cats and one cat had normal bile cytology (Table 3).

Bacterial culture was performed on liver tissue from 29 cats. Sixteen of 29 (55%) were from a fine-needle aspirate, 7/29 (24%) from tissue biopsy and 6/29 (21%) in which the sample source could not be determined (both a fine-needle aspirate and tissue biopsy were performed). Only one liver culture was positive for bacterial growth. This cat also had a bile culture that was positive for the same bacterial species. Liver culture was negative in four cats that had concurrent positive bile cultures. Over half of the cats in this study (44/83; 53%) received an antibiotic within 1 week prior to the procedure.

Of the 66 liver samples examined cytologically, 49/66 (74%) were consistent with hepatic lipidosis, 24 (36%) showed evidence of inflammation and two (3%) revealed neoplasia (carcinoma and plasmacytoma). Twenty liver biopsies were available for review. There were 15/17 Tru-cut biopsies obtained at the time of PUC (one was lost and one was not submitted). Five additional biopsies included three wedge biopsies, one punch biopsy and one Tru-cut biopsy obtained later during the course of hospitalization. Eight of 20 (40%) revealed hepatic lipidosis (ranging from mild to severe), 10/20 (50%) were inflammatory and 2/20 (10%) were neoplastic (one with lymphoma and one with hepatocellular adenoma). Three cases were classified as 'other hepatopathy', which included non-specific reactive hepatitis, hepatocellular swelling and clearing, and rare hepatocellular necrosis. Of the cases diagnosed with cholangitis, five were classified as lymphocytic (one severe, two moderate, two minimal-to-moderate) and five as neutrophilic (four moderate chronic, one mild acute).

Seventy-two cats (87%) survived to discharge. Nine of 83 cats (11%) were euthanized during their hospital stay and 2/83 (2%) died. Reasons for euthanasia included poor prognosis or complications from their underlying diseases (n = 7) and financial limitations (n = 2). Four cats that were euthanized had a necropsy performed. Gall bladder rupture or bile leakage was not appreciated in any of these cases. Cause of death for the two cats that died included hemorrhage from Tru-cut liver biopsy (2 h post-PUC) and respiratory arrest secondary to underlying disease (5 days post-PUC). While neither cat that died had a necropsy performed, the deaths did not appear to be a result of PUC.

Median time from PUC to discharge was 2 days (range 0–12 days). Twelve of 72 cats (17%) were discharged the same day as the PUC. Fifty-three of 72 cats (74%) had a follow-up appointment at the veterinary hospital. The median time to follow up was 11 days (1–386 days). Thirty-five of 53 (66%) were known to be alive at 30 days. Five cats were euthanized within 30 days

Liver diagnosis	FNA vs biopsy	Bile cytology	Bile culture
Marked fatty change	FNA	Normal	<i>Bacillus</i> species, alpha-hemolytic <i>Streptococcus</i> species
Lymphocytic, neutrophilic inflammation	FNA	Mixed bacteria	Klebsiella species
Lymphoma	Biopsy	Mixed bacteria	Negative
Mixed inflammation	FNA	Suppurative inflammation	Escherichia coli
Hepatic lipidosis	Biopsy	Mixed bacteria	Escherichia coli
Hepatic lipidosis	Biopsy	Coccobacilli Suppurative inflammation	Pseudomonas species
Presumptive cholangitis	NA	Mixed bacteria	<i>Escherichia coli, Enterococcus</i> species
Neutrophilic inflammation	FNA	Mixed bacteria	Escherichia coli, unidentified cocci
Neutrophilic, lymphocytic inflammation	FNA	Rods Suppurative inflammation*	Escherichia coli
Neutrophilic inflammation	FNA	Mixed bacteria	Alpha-hemolytic <i>Streptococcus</i> species
Normal	FNA	Rods	Escherichia coli
Mixed inflammation	FNA	Coccobacilli Suppurative inflammation	Escherichia coli

 Table 3
 Cytologic and histopathologic diagnoses in 12 cats with bacteria identified in bile either cytologically or via culture

\*Two distinct populations of rods

FNA = find-needle aspiration; NA = not applicable

reportedly owing to their underlying clinical diagnoses including gastrointestinal lymphoma, inflammatory bowel disease, metastatic cholangiocellular carcinoma (necropsy diagnosis), acute onset of neurologic disease (suspected multiple myeloma) and a combination of pancreatitis, chronic kidney disease and liver disease.

## Discussion

In this group of cats with suspected hepatobiliary disease, PUC was an apparently safe and minimally invasive tool used to obtain bile. PUC has been performed successfully in a group of healthy cats.15 Speculation regarding whether PUC would carry a greater risk of complication in patients with biliary tract disease exists. This belief was supported by a prior case series in which 1/4 cats developed bile peritonitis following PUC.6 Cholecystocentesis was performed successfully in a larger group of cats with a variety of gastrointestinal, hepatobiliary and pancreatic diseases; however, the number of cats for which bile was obtained via PUC was not provided.<sup>30</sup> The current study selected for a group of cats with suspected hepatobiliary disease and the total complication rate for PUC was 17% with no reports of gall bladder rupture, bile peritonitis or hypotension necessitating vasopressor therapy. Importantly, only minor complications consisting of technical challenges could be definitively attributed to PUC. These challenges could have been attributed to ineffective sedation,

attempting aspiration with a smaller gauge needle than warranted or radiologist inexperience with the procedure. Given the potential for technical complications, careful planning of sedation protocol, equipment and technique should be reviewed prior to each procedure. A major complication, consisting of increased free abdominal effusion, occurred in 11 cats. However, in the majority of cases the increased effusion was noted following a procedure other than PUC. Furthermore, in the majority of cases the volume of effusion was reported to be scant, minimal or mild. Neither fact eliminates the possibility that delayed bleeding from the PUC site could have occurred and contributed to the fluid accumulation. We theorize that the fluid was blood, though this was only confirmed in one case via abdominocentesis. Coagulation times were prolonged in over half of these cats and likely contributed to the observed bleeding. All 11 cases had either splenic or hepatic aspiration or hepatic Tru-cut biopsy in addition to PUC.

In two cases the increase in abdominal fluid was significant enough to mandate a prompt change in plan or treatment. In both cases the radiologist ultrasonographically identified that the source of hemorrhage was arising from the liver and/or spleen. The rate of presumptive bleeding in this study is higher than what has been reported previously following intra-abdominal aspiration and/or biopsy.<sup>31</sup> In a report of 51 cats undergoing percutaneous ultrasound-guided fine-needle aspiration of intra-abdominal organs, no complications were reported. Two of the cats that underwent ultrasoundguided tissue core biopsies of the liver developed bile peritonitis secondary to punctured bile ducts. The authors theorized that the diagnosis of hepatic lipidosis rendered their liver tissue friable. In the present study, nearly 75% of cats had some component of hepatic lipidosis, which may account for the higher rate of presumptive bleeding reported.

While it was not possible to document the degree of hemorrhage following PUC, the results suggest that significant blood loss was uncommon. Median change in PCV was small, especially given that the majority of cats received a combination of sedation/anesthesia and intravenous fluids. In this study, five cats required a blood transfusion following the procedures. Review of the medical records demonstrated that blood was administered for ongoing or developing hemorrhage in only one case.

Acute vagal reaction with severe hypotension and bradycardia has been seen in human patients undergoing PUC.<sup>28,29</sup> However, a literature review of 231 cases of PUC in people showed that this was an uncommon complication with only four cases of severe vasovagal reaction.<sup>32</sup> In cats, vasovagal reaction with shock was reported in 19% of cases undergoing automatic Tru-cut liver biopsy.<sup>33</sup> All cats died despite resuscitative efforts.<sup>33</sup> In the current study, there were no reports of hypotension necessitating treatment with a vasopressor medication. One cat did become significantly hypotensive within 2 h of the procedure; however, it was suspected to be secondary to ongoing intra-abdominal hemorrhage that the radiologist noted to be coming from liver and splenic aspiration sites. Unfortunately, paired blood pressure readings were only available in a small percentage of cats. In those cases, the median decrease in blood pressure was modest; especially in the light of the sedation that all cats received. Given the retrospective nature of this study, we may have missed or not recorded episodes of hypotension.

Bile analysis (cytology and/or culture) was abnormal in nearly one-third of the population. This is comparable to a recent retrospective analysis of 78 cats that had either surgical or percutaneous cholecystocentesis in which just over 20% of cats had inflammation and/or infection.<sup>30</sup> In accordance with previous studies bile culture was more likely to yield a positive result (14%) compared with liver culture (3%).<sup>13,14</sup> A smaller percentage of cats in the current study had positive bile cultures when compared with prior reports. One study reported that 36% of bile cultures were positive in cats with hepatobiliary disease, with anaerobic organisms accounting for 26% of all positive cultures.14 Anaerobic bacteria were not identified in the present study which could account for the overall lower percentage of positive cultures. This may reflect differences in laboratory practices and

culture techniques. It is also possible that false-negative results were encountered as routine culture techniques may not be sufficient to diagnose all cases of biliary tract infection.<sup>7,8</sup> The possibility of false-negative results was most likely among the 50% of cats that received an antibiotic prior to bile obtainment. PCR has been utilized to detect bacterial species in feline bile.<sup>8</sup> Molecular methods may be beneficial to identify accurately organisms that routine culture techniques fail to identify. Finally, although the study population contained cats with cholangitis, many had a final diagnosis of hepatic lipidosis. Therefore, the percentage of cats with bactibilia may be lower than if we had investigated a group of cats with a diagnosis restricted to cholangitis in which bacterial infections may be seen in up to 41% of cases.<sup>7</sup>

Results suggest that bile cytology may not always correlate with bile culture. There was agreement between the presence of bacteria cytologically and positive bile culture, with only one false-negative cytology. In addition, in only one cat was bacteria seen cytologically with a negative bile culture. However, in multiple cases, bile cytology revealed a mixed population of bacteria while culture yielded only one bacterial organism. This is in agreement with the cytological findings of 140 bile samples from dogs and cats in which cytology and culture results did not always match.<sup>30</sup> In six cases, cytology identified a mixed bacterial population, whereas culture was positive for only one organism. Therefore, prediction of the type of bacterium present based on cytological evaluation may be inaccurate. With the concern that routine culture techniques may yield false-negative results, further evaluation of the utility of bile cytology and the correlation with culture is warranted.

### Conclusions

PUC performed in cats with suspected hepatobiliary disease was a safe technique for bile collection that yielded useful diagnostic information in approximately 30% of cases. The complications encountered were likely secondary to an ancillary procedure performed at the time of the PUC. Bile cytology may not be predictive of bile culture results. Bile analysis should be further evaluated in a prospective setting to better characterize its diagnostic utility and determine how well cytology correlates with culture.

**Acknowledgements** The authors would like to thank the Radiology, Clinical Pathology and Microbiology services at Penn Vet for their technical expertise and help in identifying cases.

**Conflict of interest** The authors declared no potential conflicts of interest with respect to the research, authorship, and/ or publication of this article.

**Funding** The authors received no financial support for the research, authorship, and/or publication of this article.

### References

- 1 Center SA. Diseases of the gall bladder and biliary tree. Vet Clin North Am Small Anim Pract 2009; 39: 543–598.
- 2 van den Ingh TSGAM, Cullen JM, Twedt DC, et al. Morphological classification of biliary disorders of the canine and feline liver. In Rothuizen J, Bunch SE, Charles JA, et al. (eds). WSAVA standards for clinical and histological diagnosis of canine and feline liver diseases. Edinburgh: Saunders Elsevier, 2006, pp 61–76.
- 3 Callahan Clark JE, Haddad JL, Brown DC, et al. Feline cholangitis: a necropsy study of 44 cats (1986–2008). *J Feline Med Surg* 2011; 13: 570–576.
- 4 Day DG. Feline cholangiohepatitis complex. Vet Clin North Am Small Anim Pract 1995; 25: 375–385.
- 5 Gagne JM, Weiss DJ and Armstrong PJ. Histopathologic evaluation of feline inflammatory liver disease. *Vet Pathol* 1996; 33: 521–526.
- 6 Brain PH, Barrs VR, Martin P, et al. Feline cholecystitis and acute neutrophilic cholangitis: clinical findings, bacterial isolates and response to treatment in six cases. J Feline Med Surg 2006; 8: 91–103
- 7 Twedt DC, Cullen J, McCord K, et al. Evaluation of fluorescence in situ hybridization for the detection of bacteria in feline inflammatory liver disease. *J Feline Med Surg* 2014; 16: 109–117.
- 8 Otte CM, Gutierrez OP, Favier RP, et al. Detection of bacterial DNA in bile of cats with lymphocytic cholangitis. *Vet Microbiol* 2012; 156: 217–221.
- 9 Warren A, Center S, McDonough S, et al. Histopathologic features, immunophenotyping, clonality, and eubacterial fluorescent in situ hybridization in cats with lymphocytic cholangitis/cholangiohepatitis. *Vet Pathol* 2011; 48: 627–641.
- 10 Day MJ. Immunohistochemical characterization of the lesions of feline progressive lymphocytic cholangitis/ cholangiohepatitis. J Comp Pathol 1998; 119: 135–147.
- 11 Prasse KW, Mahaffey EA, DeNovo R, et al. Chronic lymphocytic cholangitis in three cats. *Vet Pathol* 1982; 19: 99–108.
- 12 Weiss DJ, Armstrong PJ and Gagne J. Inflammatory liver disease. Semin Vet Med Surg Small Anim 1997; 12: 22–27.
- 13 Morgan M, Rankin S, Berent A, et al. Prospective evaluation for bacterial infection in hepatic tissue and bile of cats with diffuse hepatobiliary disease. J Vet Intern Med 2008; 22: 806.
- 14 Wagner KA, Hartmann FA and Trepanier LA. Bacterial culture results from liver, gallbladder, or bile in 248 dogs and cats evaluated for hepatobiliary disease: 1998–2003. J Vet Intern Med 2007; 21: 417–424.
- 15 Savary-Bataille KCM, Bunch SE, Spaulding KA, et al. Percutaneous ultrasound-guided cholecystocentesis in healthy cats. J Vet Intern Med 2003; 17: 298–303
- 16 Klapdor R, Scherer K, Sepehr H, et al. The ultrasonically guided puncture of the gallbladder in animals. *Endoscopy* 1997; 9: 166–169.

- 17 McGaham JP, Phillips HE, Nyland T, et al. Sonographically guided percutaneous cholecystostomy performed in dogs and pigs. *Radiology* 1983; 149: 841–843.
- 18 Voros K, Sterczer A, Manczur F, et al. Percutaneous ultrasound-guided cholecystocentesis in dogs. Acta Vet Hung 2002; 50: 385–393.
- 19 Braun U and Gerber D. Percutaneous ultrasound-guided cholecystocentesis in cows. Am J Vet Res 1992; 53: 1079– 1084.
- 20 Denning DA, Ellison EC and Carey LC. Preoperative percutaneous biliary decompression lowers operative morbidity in patients with obstructive jaundice. *Am J Surg* 1981; 141: 61–65.
- 21 Gobien RP, Stanley JH and Soucek CD. Routine preoperative biliary drainage: effect on management of obstructive jaundice. *Radiology* 1984; 152: 353–356.
- 22 McGaham JP and Walter JP. Diagnostic percutaneous aspiration of the gallbladder. Radiology 1985; 155: 619–622.
- 23 vanSonnenberg E, Wittich GR, Casola G, et al. Diagnostic and therapeutic percutanueous gallbladder procedures. *Radiology* 1986; 160: 23–26.
- 24 Vogelzang RL and Nemcek AA. Percutaneous cholecystostomy: diagnostic and therapeutic efficacy. *Radiology* 1988; 168: 29–34.
- 25 vanSonnenberg E, D'Agostino H and Casola G. Interventional gallbladder procedures. *Radiol Clin North Am* 1990; 28: 1185–1190.
- 26 Swobodnik W, Hagert N, Janowitz P, et al. Diagnostic fineneedle puncture of the gallbladder with US guidance. *Radiology* 1991; 178: 755–758.
- 27 Tudyka J, Kratzer W, Kuhn K, et al. Diagnostic value of fineneedle puncture of the gallbladder: side effects, safety, and prognostic value. *Hepatology* 1995; 21: 1303–1307.
- 28 VanSonnenberg E, Wing VW, Pollard JW, et al. Life-threatening vagal reactions associated with percutaneous cholecystostomy. *Radiology* 1984; 151: 377–380.
- 29 vanSonnenberg E, D'Agostino HB and Goodacre BW. Percutaneous gallbladder puncture and cholecystostomy: results, complications, and caveats for safety. *Radiology* 1992; 183: 167–170.
- 30 Peters LM, Glanemann B, Garden OA, et al. Cytological findings of 140 bile samples from dogs and cats and associated clinical pathological data. *J Vet Intern Med* 2016; 30: 123–131.
- 31 Leveille R, Partington BP, Biller DS, et al. Complications after ultrasound-guided biopsy of abdominal structures in dogs and cats: 246 cases (1984–1991). J Am Vet Med Assoc 1993; 203: 413–415.
- 32 Teplick SK, Brandon JC, Wolferth CC, et al. Percutaneous interventional gallbladder procedures: personal experience and literature review. *Gastrointest Radiol* 1990; 15: 133–136.
- 33 Proot SJ and Rothuizen J. High complication rate of an automatic Tru-Cut biopsy gun device for liver biopsy in cats. J Vet Intern Med 2006; 20: 1327–1333.