



Risk Factors for Acute Cholangitis Caused by *Enterococcus faecalis* and *Enterococcus faecium*

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Background/Aims: Acute cholangitis (AC) is a potentially life-threatening bacterial infection, and timely antimicrobial treatment, faster than that achieved with bacterial cultures, is recommended. Although the current guidelines refer to empirical antimicrobial treatment, various kinds of antimicrobial agents have been cited because of insufficient analyses on the spectrum of pathogens in AC. *Enterococcus* spp. is one of the most frequently isolated Gram-positive bacteria from the bile of patients with AC, but its risk factors have not been extensively studied. This study aimed to analyze the risk factors of AC caused by *Enterococcus faecalis* and *Enterococcus faecium*.

Methods: Patients with AC who were hospitalized in a Japanese tertiary center between 2010 and 2015 were retrospectively analyzed. Patients' first AC episodes in the hospital were evaluated.

Results: A total of 266 patients with AC were identified. *E. faecalis* and/or *E. faecium* was isolated in 56 (21%) episodes of AC. Prior endoscopic sphincterotomy (EST), the presence of a biliary stent, prior cholecystectomy, and past intensive care unit admission were more frequently observed in AC patients with *E. faecalis* and/or *E. faecium* than in those without such bacteria. Prior EST was identified as an independent risk factor for AC caused by *E. faecalis* and/or *E. faecium* in the multivariate analysis.

Conclusions: Given the intrinsic resistance of *E. faecalis* and *E. faecium* to antibiotics, clinicians should consider empirical therapy with anti-enterococcal antibiotics for patients with prior EST. (Gut Liver 2021;15:616-624)

Key Words: Cholangitis; *Enterococcus*; Sphincterotomy; Anti-microbial agents; Therapeutics

INTRODUCTION

Acute cholangitis (AC) is a potentially life-threatening bacterial infection of the intra- and/or extrahepatic biliary system caused by obstruction of the bile ducts, with stasis and subsequent infection of the bile.¹ The common causes of AC include gallstones, bile duct stones, and bile duct stenosis in cases of chronic pancreatitis, malignant neoplasm, and sclerosing cholangitis.^{2,3} The typical symptoms of AC are fever, jaundice, and abdominal pain (Charcot's triad).⁴ The current treatment strategies support a risk-stratified approach based on the revised Tokyo Guidelines 2018

(TG18) and generally comprise a combination of antibiotic therapy and early endoscopic resolution of the obstruction,^{5,6} because delaying the endoscopic treatment often results in persistent organ failure.⁷

Pathogens most frequently isolated from the bile of patients with AC are Gram-negative Enterobacteriaceae, such as *Escherichia coli* and *Klebsiella* spp., as well as gram-positive *Enterococcus* spp., with a high proportion of polymicrobial cultures observed.^{8,9} Given that microbiological identification of the pathogens requires time, antibiotic therapy is generally initiated as an empirical therapy. The current guidelines, such as TG18, classify antimicrobial



recommendations according to the route of infection (community-acquired or healthcare-associated) and grade of AC. Many types of antibiotics, including any generation of cephalosporins, are recommended in the guidelines, and no priority is indicated.¹⁰

Enterococci, which accounts for a considerable portion of pathogens causing AC, are potentially resistant to cephalosporins. According to the Clinical and Laboratory Standards Institute guidelines, there are no breakpoints of ceftriaxone for *Enterococcus* spp. available. Susceptibility to fourth-generation cephalosporins (cefpime and ceftazidime) was reported to be 60% for *Enterococcus faecalis* and 15% for *Enterococcus faecium*.

In addition, vancomycin-resistant enterococci (VRE) are becoming an increasingly important cause of invasive infections in the United States.^{11,12} The most common type of vancomycin resistance in enterococci is associated with acquisition of the van A and van B genes, typically observed in *E. faecalis* and *E. faecium* isolates.¹³ These species comprise the majority of VRE isolated from blood stream infections and are associated with significant mortality.¹⁴

In earlier studies, *Enterococcus* spp. accounted for 40.5% of all identified pathogens.¹⁵ Moreover, *Enterococcus* spp. constituted 43% of all pathogens isolated from patients with AC.¹⁶ However, knowledge of the risk factors of AC with *Enterococcus* spp. is limited. The aims of this retrospective study were to characterize the contemporary microbial patterns in patients with AC using a pathogen-based approach and to identify the risk factors for AC caused by *E. faecalis* and/or *E. faecium*.

MATERIALS AND METHODS

1. Study design

The medical records of patients who were admitted to Mitsui Memorial Hospital, a Japanese tertiary center, from January 2010 to December 2015 were reviewed to identify patients with AC. All patients, regardless of age, whose blood and/or bile cultures had been obtained were included in our analysis. We defined AC based on TG18. Even if neither blood nor bile cultures were positive, the episodes that met the criteria of TG18 and were clinically consistent with AC were enrolled in this study. We excluded patients with other infectious diseases.

The following variables were collected from the medical records: age; sex; microbiological results of blood and/or bile cultures; underlying biliary disease (common bile duct calculus, gallstone or biliary sludge, etc.); medications, including any antibiotic used within 14 days prior to the occurrence of cholangitis; admission to the intensive care

unit; previous endoscopic diagnostic procedures or interventions; and history of biliary or upper gastrointestinal surgery. We also classified the severity of each case according to TG18.⁵ As the outcomes of cholangitis, length of hospital stay and mortality were also examined. In patients who had two or more episodes of AC during the study period, the first episode alone was included in the analysis. The development of cholangitis during the hospital stay for ≥ 48 hours was regarded as nosocomial AC.

This study was approved by the Ethical Committee of Mitsui Memorial Hospital (IRB number: MEC2018-C12). The requirement for obtaining informed consent from the patients was waived due to the retrospective nature of the study.

2. Microbiological sampling

Blood culture samples were collected before or immediately after the initiation of antibiotic treatment, followed by bedside inoculation of 10 mL of blood into the blood culture bottles (BacT/Alert; bioMerieux, Durham, NC, USA). The bottles were incubated at 37°C until microbial growth was detected, or for at least 7 days. Bile cultures were obtained by endoscopic retrograde cholangiopancreatography via catheter aspiration. Bile cultures were performed using standard solid media, e.g., sheep blood agar (Kyokuto Pharmaceutical Industrial Co., Ltd., Tokyo, Japan) for aerobic bacteria and Brucella agar (Kyokuto Pharmaceutical Industrial Co., Ltd.) for anaerobic bacteria. The cultivated microorganisms were identified, and antibiotic susceptibility testing was performed using the MicroScan WalkAway (Beckman Coulter, Brea, CA, USA), with the results interpreted according to the Clinical and Laboratory Standards Institute guidelines.

3. Statistical analysis

Statistical analysis was performed using JMP 11.0.0 (SAS Institute Inc., Cary, NC, USA). Significant differences were assessed using the Pearson chi-square test. The risk factors for cholangitis caused by *E. faecalis* and/or *E. faecium* were determined by performing a multivariate binary logistic regression analysis, including the use of significant univariate predictors through stepwise backward elimination. The significance level in two-sided testing was $p < 0.05$, without correction for multiple testing. We also compared the sensitivity of bile and blood cultures with Wilcoxon signed-rank test.

RESULTS

1. Baseline characteristics

During the study period, 266 patients who developed

one or more episodes of AC were identified. The analysis in this study was based on the first AC episode of each patient. Of the 266 patients, 174 (65%) were males. The median length of hospital stay was 16 days, and the 30-day mortality rate was 2.8%. Ten patients (4%) were in septic shock, and eight patients (3%) required inotropes. In 42 patients (16%), the onset was >48 hours after hospital admission and was regarded as nosocomial AC. Common bile duct calculus was observed in 143 patients (54%), whereas 150 patients (56%) had gallstone or biliary sludge. We investigated the patients' comorbidities that are known to suppress immunity, such as renal failure treated with hemodialysis (nine patients, 3%), diabetes mellitus (60 patients, 23%), and liver cirrhosis (10 patients, 4%). Prior biliary interventions, including endoscopic sphincterotomy (EST) (60 patients, 23%) and endoscopic papillary balloon dilation (EPBD) (15 patients, 6%), were evaluated (Table 1).

2. Overall pathogen spectra

From the 266 AC cases, 230 bile cultures and 239 blood cultures were obtained, and positive results were noted in 131 (57%) and 155 (65%) episodes, respectively. Blood and/or bile cultures were positive in 183 episodes (69%) and were negative in the remaining 83 episodes (31%). *E. faecalis* and/or *E. faecium* were identified in 56 cases (21%). *Enterococcus* spp. other than *E. faecium* or *E. faecalis* were observed in 19 cases (7%), and bacteria other than enterococci alone were isolated in 108 cases (41%) (Fig. 1).

The overall bacterial spectrum revealed that, among 75 cases with *Enterococcus* spp., the most frequently identified species was *E. faecalis* (33/266, 12.4%), followed by *E. faecium* (26/266, 9.8%) and *E. casseliflavus* (18/266, 6.8%) (Table 2, Fig. 2). Among species other than the *Enterococcus* spp., *E. coli* was the most frequently isolated pathogen (72/266, 27.2%), followed by *Klebsiella pneumoniae* (47/266, 17.7%). The isolated bacterial spectrum did not differ significantly according to severity of AC (Table 3). In addition, there was no significant difference in sensitivity between bile and blood cultures ($p=0.235$).

3. Comparison of characteristics between AC patients with and without *E. faecalis* and/or *E. faecium*

To identify the risk factors of AC caused by *E. faecalis* and/or *E. faecium*, univariate comparisons were performed between patients with and without these bacteria (Table 4). There was a significant difference in the frequency of prior EST between patients with AC with and without *E. faecalis* and/or *E. faecium* (24/56 [43%] vs 36/210 [17%]; $p<0.001$). In addition, a statistical difference was observed in the frequency of the presence of biliary stent (16/56 [29%] vs 28/210 [13%]; $p=0.006$), prior cholecystectomy (14/56 [25%

Table 1. Characteristics of the Studied Patients

Characteristics	No. (%)
Patients	
Number	266
Age at onset, median (range), yr	74 (29–97)
Age ≥ 75 yr	131 (49)
Sex	
Male	174 (65)
Female	92 (34)
Length of hospital stay, median (range), day	16 (2–184)
30-Day mortality rates, %	2.8
Septic shock	10 (4)
Usage of inotropes	8 (3)
Onset of >48 hours after admission	42 (16)
Underlying biliary disease	
Common bile duct calculus	143 (54)
Gallstone or biliary sludge	150 (56)
Malignant neoplasm of biliary tract	95 (36)
Cholecystitis	22 (8)
Hemobilia	3 (1)
Comorbidity	
Renal failure treated with hemodialysis	9 (3)
Diabetes mellitus	60 (23)
Liver cirrhosis	10 (4)
Prior biliary intervention	
Endoscopic sphincterotomy	60 (23)
Endoscopic papillary balloon dilation	15 (6)
Biliary stent	44 (17)
Percutaneous transhepatic cholangiodrainage	14 (5)
PTGBA/PTGBD	7 (3)
Biliary tract reconstruction	25 (9)
Cholecystectomy	42 (16)
Prior anastomosis in upper GI	
Billroth II	5 (2)
Roux-en-Y anastomosis	10 (4)
ICU admission	
None	109 (41)
Past	61 (23)
Current	123 (46)
Antibiotic use within 14 days prior to occurrence of cholangitis	
Cephem	30 (11)
1st generation	1 (0.4)
2nd generation	19 (29)
3rd generation	10 (4)
4th generation	0
Penicillin	3 (1)
Carbapenem	5 (2)

PTGBA/PTGBD, percutaneous transhepatic gallbladder aspiration/drainage; GI, gastrointestinal tract; ICU, intensive care unit.

vs 28/210 [13%]; $p=0.03$), and past intensive care unit admission (19/56 [34%] vs 42/210 [20%]; $p=0.03$) between the two groups (Table 4). No statistical difference was observed in age, sex, septic shock, inotrope use, malignant neoplasm of the biliary tract, cholecystitis, comorbidity (e.g., renal failure, diabetes mellitus, and liver cirrhosis), biliary tract reconstruction, prior anastomosis in the upper gastrointestinal

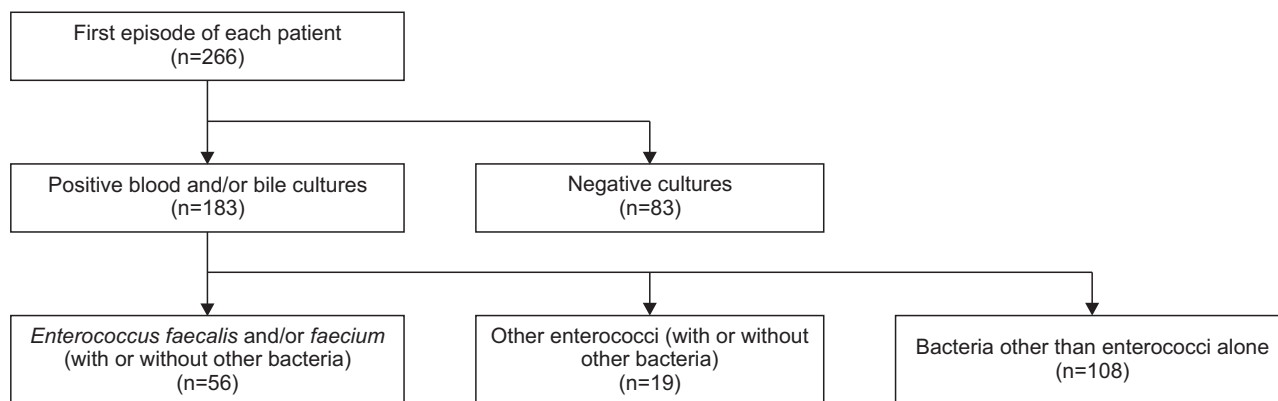


Fig. 1. Study flowchart.

Table 2. Bacteria Isolated from Bile and/or Blood Cultures Obtained from Patients with Cholangitis

Bacterial species	Total (n=266)	Bile culture (n=230), positive (n=131, 57%)	Blood culture (n=239), positive (n=155, 65%)
<i>Enterococcus</i> spp.			
<i>Enterococcus faecalis</i>	33 [12]	27 [12]	16 [7]
<i>Enterococcus faecium</i>	26 [10]	17 [7]	18 [8]
<i>Enterococcus casseliflavus</i>	18 [7]	11 [5]	11 [5]
<i>Enterococcus avium</i>	5 [2]	5 [2]	3 [1]
<i>Enterococcus raffinosus</i>	2 [1]	2 [1]	1 [0]
<i>Enterococcus gallinarum</i>	4 [2]	2 [1]	2 [1]
Others			
<i>Escherichia coli</i>	72 [27]	44 [19]	49 [21]
<i>Klebsiella pneumoniae</i>	47 [18]	36 [17]	26 [11]
<i>Klebsiella oxytoca</i>	19 [7]	13 [6]	12 [5]
<i>Enterobacter cloacae</i>	12 [5]	9 [4]	6 [3]
<i>Clostridium perfringens</i>	17 [6]	15 [7]	10 [4]
<i>Aeromonas hydrophila</i>	17 [6]	13 [6]	6 [3]
<i>Bacteroides fragilis</i>	8 [3]	7 [3]	4 [2]
<i>Pseudomonas aeruginosa</i>	24 [9]	22 [10]	11 [5]
<i>Staphylococcus epidermidis</i>	9 [3]	4 [2]	6 [3]

Data are presented as the number (%). Cases which were caused by multiple microorganisms were counted separately.

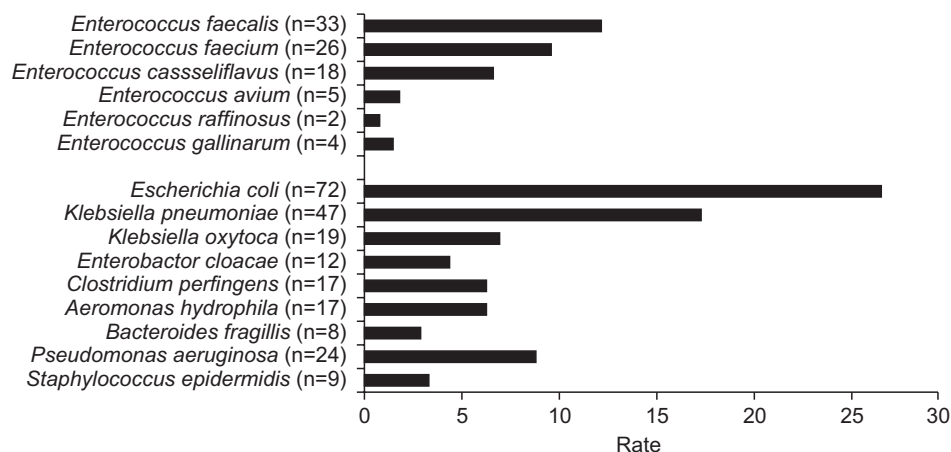


Fig. 2. Pathogen spectra in the blood and/or bile cultures of the included patients.

tract, or antibiotic use within 14 days prior to the occurrence of cholangitis in the univariate logistic regression analysis (all $p > 0.05$).

The multivariate analysis, including prior EST, biliary stent, cholecystectomy, and past ICU admission as variables, revealed that prior EST was the only independent

Table 3. Comparison of Pathogens Obtained from Blood and/or Bile Cultures and 30-Day Mortality Rates among Each Severity Grade

Bacterial species	Total (n=266)	Severity*		
		Grade I (n=129, 48%)	Grade II (n=58, 22%)	Grade III (n=79, 30%)
<i>Enterococcus</i> spp.				
<i>Enterococcus faecalis</i>	33 (12)	15 (12)	8 (14)	10 (13)
<i>Enterococcus faecium</i>	26 (10)	11 (9)	5 (9)	10 (13)
<i>Enterococcus casseliflavus</i>	18 (7)	10 (8)	2 (3)	6 (8)
<i>Enterococcus avium</i>	5 (2)	1 (1)	2 (3)	2 (3)
<i>Enterococcus raffinosus</i>	2 (1)	2 (2)	0	0
<i>Enterococcus gallinarum</i>	4 (2)	2 (2)	2 (3)	0
Others				
<i>Escherichia coli</i>	72 (27)	28 (22)	16 (28)	28 (35)
<i>Klebsiella pneumoniae</i>	47 (18)	18 (14)	13 (22)	16 (20)
<i>Klebsiella oxytoca</i>	19 (7)	8 (6)	5 (9)	5 (6)
<i>Enterobacter cloacae</i>	12 (5)	7 (5)	2 (3)	3 (4)
<i>Clostridium perfringens</i>	17 (6)	8 (6)	2 (3)	7 (9)
<i>Aeromonas hydrophila</i>	17 (6)	5 (4)	6 (10)	6 (8)
<i>Bacteroides fragilis</i>	8 (3)	1 (1)	3 (5)	4 (5)
<i>Pseudomonas aeruginosa</i>	24 (9)	14 (11)	3 (5)	7 (9)
<i>Staphylococcus epidermidis</i>	9 (3)	8 (6)	0	1 (1)
30-Day mortality rates, %	2.8	2.5	1.9	3.9

Data are presented as the number (%). Cases that were caused by multiple microorganisms were counted separately.

*Severity of acute cholangitis was defined according to the Tokyo Guidelines 2018.

predictor of AC caused by *E. faecalis* and/or *E. faecium* (odds ratio, 3.53; 95% confidence interval, 1.84 to 6.78; $p < 0.001$) (Table 4).

DISCUSSION

The current guidelines, such as TG18, recommend the combination of antimicrobial agents, including vancomycin, to cover treatment of *Enterococcus* spp. in grade III community-acquired AC and healthcare-associated acute biliary infections. In contrast, they indicated that cephalosporin-based therapy (each generation of cephalosporin± metronidazole) can also be chosen for grade I or II community-acquired AC. Although TG18 determined whether *Enterococcus* spp. should be covered or not according to the grade of AC, the current study revealed that prior EST is the only independent risk factor of AC caused by *E. faecalis* and/or *E. faecium*, regardless of the grade.

Enterococcus spp. show intrinsic resistance to any generation of cephalosporins. *E. faecalis* is usually the most frequently isolated *Enterococcus* spp. from human clinical specimens, representing 80% to 90% of the isolates in *Enterococcus* spp., followed by *E. faecium*, which accounts for 5% to 10% of all enterococcal infections.¹⁷⁻²⁰ A previous study reported that, of the 826 unique episodes of bacteremia with *Enterococcus* spp., 94.6% were caused by either *E. faecalis* (56.1%) or *E. faecium* (38.5%).²¹

Moreover, VRE currently account for >30% of entero-

coccal infections,²² and more than 90% of VRE isolates in the United States are *E. faecium*.²² In the present study, VRE were not isolated since VRE infections are less frequent in Japan than in the United States. However, *E. faecalis* and *E. faecium* are potentially important in terms of antibiotic resistance. The susceptibility rates of *E. faecalis* were <60% for levofloxacin and <25% for minocycline in Japan, whereas in Latin American regions, these were 6.1% to 69.4% and 34.9% to 51.5%, respectively.²³ The susceptibility rates of *E. faecium* were <30% for both levofloxacin and minocycline in Japan. Consequently, the 30-day mortality due to bacteremia was reportedly 21.4% and 34.6% in patients with *E. faecalis* and *E. faecium*, respectively.²⁴ In contrast, previous studies showed that the mortality associated with non-*faecium*, non-*faecalis* enterococcal bloodstream infections was relatively low.^{25,26} Treatment failure for non-*faecium/faecalis* enterococcal bacteremia also occurred less frequently, suggesting their lower virulence relative to *E. faecium* and *E. faecalis* strains.²⁷

We have identified EST as an independent risk factor for AC caused by *E. faecalis* and/or *E. faecium*. In this study, all patients who had undergone EST had a history of AC with antibiotic use (mostly cephalosporins). Consequently, *E. faecalis* and *E. faecium*, which have intrinsic resistance to cephalosporins, were likely to be detected as the pathogen for recurrent AC after EST.

Weber *et al.*¹⁶ reported that the isolation rates of *Enterococcus* spp. were significantly higher in cholangitis episodes with biliary endoprosthesis than in cholangitis

Table 4. Univariate and Multivariate Analyses of Risk Factors for Enterococcal Cholangitis

Variable	Enterococcus faecium and/or faecalis (n=56)		Others (n=210)		Univariate analysis		Multivariate analysis	
	Enterococcus faecium and/or faecalis (n=56)	Others (n=210)	OR (95% CI)	p-value	OR (95% CI)	p-value		
Patient								
Age ≥75 yr	30 (53.6)	101 (48.1)	1.25 (0.69–2.25)	0.47				
Male sex	38 (67.9)	136 (64.8)	1.15 (0.61–2.15)	0.67				
Septic shock	3 (5.4)	7 (3.3)	1.64 (0.41–6.56)	0.48				
Usage of inotropes	2 (3.6)	6 (2.9)	1.25 (0.25–6.42)	0.78				
Onset of >48 hr after admission	13 (23.2)	29 (13.8)	1.88 (0.91–3.93)	0.09				
Underlying biliary disease								
Common bile duct calculus	30 (53.6)	113 (53.8)	0.97 (0.54–1.75)	0.92				
Gallstone or biliary sludge	27 (48.2)	123 (58.6)	0.66 (0.36–1.19)	1.98				
Hemobilia	1 (1.8)	2 (1.0)	1.89 (0.17–21.24)	0.28				
Malignant neoplasm of biliary tract	25 (44.6)	70 (33.3)	1.61 (0.89–2.94)	0.12				
Cholecystitis	7 (12.5)	15 (7.1)	1.86 (0.72–4.80)	0.2				
Comorbidity								
Renal failure treated with hemodialysis	2 (3.6)	7 (3.3)	1.07 (0.22–5.32)	0.93				
Diabetes mellitus	16 (28.6)	44 (21.0)	1.51 (0.77–2.94)	0.23				
Liver cirrhosis	2 (3.5)	8 (3.8)	0.94 (0.19–4.53)	0.93				
Prior biliary intervention								
Endoscopic sphincterotomy	24 (42.9)	36 (17.1)	3.63 (1.91–6.87)	<0.001	2.81 (1.37–5.77)	0.005		
Endoscopic papillary balloon dilation	2 (3.6)	13 (6.2)	0.56 (0.12–2.56)	0.45				
Biliary stent	16 (28.5)	28 (13.3)	2.60 (1.29–5.25)	0.006	1.85 (0.81–4.13)	0.145		
Percutaneous transhepatic cholangiodrainage	3 (5.4)	11 (5.2)	1.02 (0.28–3.80)	0.97				
PTGBD/PTGBA	3 (5.4)	4 (1.9)	2.92 (0.63–13.42)	0.15				
Biliary tract reconstruction	7 (12.5)	18 (8.6)	1.52 (0.60–3.85)	0.37				
Cholecystectomy	14 (25.0)	28 (13.3)	2.17 (1.05–4.47)	0.03	2.17 (0.98–4.66)	0.056		
Prior anastomosis in upper GI								
Billroth II	2 (3.6)	3 (1.4)	2.59 (0.42–15.90)	0.29				
Roux-en-Y anastomosis	1 (1.8)	9 (4.3)	0.41 (0.05–3.32)	0.39				
ICU admission								
Current	26 (46.4)	97 (46.2)	1.01 (0.56–1.82)	0.97				
Past	19 (33.9)	42 (20.0)	2.05 (1.07–3.93)	0.03	1.83 (0.91–3.60)	0.089		
Antibiotic use within 14 days prior to occurrence of cholangitis								
Cephem	9 (16.1)	25 (11.9)	1.75 (0.75–4.08)	0.19				
Penicillin	2 (3.6)	1 (0.5)	3.91 (0.54–28.37)	0.15				
Carbapenem	2 (3.6)	3 (1.4)	2.59 (0.42–15.90)	0.29				

Data are presented as number (%).
 OR, odds ratio; CI, confidence interval; PTGBA/PTGBD, percutaneous transhepatic gallbladder aspiration/drainage; GI, gastrointestinal tract; ICU, intensive care unit.

episodes without biliary endoprosthesis. They also reported that *Pseudomonas aeruginosa* and *E. faecium* were more frequently isolated from patients with a biliary endoprosthesis. In their study, most patients with biliary endoprosthesis may have been in a post-EST state, and our findings are comparable to their results. As EST and biliary stent are similar predictors, the multivariate analysis was recalculated excluding either variable. The odds ratio for EST was then superior to that of biliary stent (3.53 vs 3.01). In addition, most patients with biliary stent in our study had malignant neoplasm of the biliary tract. Therefore, they regularly consulted with physicians and were admitted to hospitals, and their AC was thus considered to be healthcare-associated. Overall, in our study, prior EST was considered as a common predictor. Moreover, *E. faecalis* is reportedly the second most predominant strain isolated from urinary catheter biofilms, following *P. aeruginosa*.²⁸ Thus, these species are likely to colonize artificial objects and/or objects that developed after interventions took place, including micro-biliary sludge, or on the surface of the biliary tract.

In contrast to the post-EST state, no significant difference was observed between patients with and without EPBD. Both EST and EPBD are performed to dilate the duodenal papilla, but the function of the papilla after an intervention may be different because of the presence or absence of the cleavage of the sphincter muscle of the papilla. A previous study showed that the recurrence rate of common bile duct stones was higher in post-EST patients than in post-EPBD patients (26.3% vs 6.3%),²⁹ suggesting that AC is less likely to recur after EPBD than after EST. The dysfunction of Oddi may lead to the invasion of bacteria, including *Enterococcus* spp., into the bile duct.

Our study revealed that AC with prior EST was more likely to be caused by *E. faecalis* and/or *E. faecium*. Therefore, antibiotics with an activity against *Enterococcus* spp. have to be selected as an empirical therapy for AC patients after EST. Ampicillin is one of the candidates suitable for this purpose. Among cases caused by *E. faecalis*, susceptibility to ampicillin was 100% in Japan,³⁰ the United States, and Europe,³¹ and 78% to 99% in Latin American regions.²³ In contrast, among cases caused by *E. faecium*, susceptibility to ampicillin was lower, with >60% in Japan,³⁰ 0% to 7.9% in the United States and Europe,³¹ and 0.9% to 26% in Latin American regions.²³ Therefore, we cannot treat patients with AC with EST or biliary endoprosthesis using third-generation cephalosporin alone, but should consider adding ampicillin treatment for patients with prior EST. In regions where ampicillin-resistant *E. faecium* is dominant, vancomycin, linezolid, or tigecycline should be considered for empirical antimicrobial treatment.

There are limitations to our current study. First, although we analyzed the first episodes of AC observed in our hospital in each patient, the episodes prior to their hospital admission could not be examined. Second, our research is a retrospective analysis of data obtained from a single center. Third, despite the fact that VRE is a serious problem worldwide, we were unable to investigate the risk factors of AC caused by VRE. Although our institution has access to equipment for isolating VRE, no such cases were encountered in our hospital owing to the rarity of VRE cases in Japan. Therefore, the generalizability of our results to regions with an increased prevalence of AC caused by VRE is limited. Future studies conducted in other regions where AC caused by VRE is dominant are warranted. Further, we detected causative pathogens in bile and/or blood cultures. Previously, patients with AC were significantly more likely to have positive bile cultures.³² Previous research has revealed that only 1.6% of blood cultures were impacted on management of patients in the emergency department.³³ However, in the current study, for cases in which both bile and blood cultures were obtained, no significant difference in sensitivity between bile and blood cultures was observed.

In conclusion, the present study revealed that prior EST is an independent risk factor of AC caused by *E. faecalis* and/or *E. faecium*. To date, the therapeutic guidelines do not consider the existence of different bacterial spectra in patients with and without prior EST. Thus, clinicians should confirm the medical history of patients with AC for appropriate empirical therapy.

CONFLICTS OF INTEREST

No potential conflict of interest relevant to this article was reported.

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AUTHOR CONTRIBUTIONS

Study concept and design: Y.K., J.K., N.T. Data acquisition: Y.K., S.K., K.K., T.O., M.S., K.T. Data analysis and interpretation: Y.K., J.K., N.T. Drafting of the manuscript: Y.K. Critical revision of the manuscript for important intellectual content: J.K., N.T. Study supervision: J.K., N.T.

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