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The clinical significance of intraoperative lavage fluid culture during pancreaticoduodenectomy on organ/space surgical site infection

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Organ/space surgical site infection (SSI) are common after pancreaticoduodenectomy (PD). There is limited research on the clinical impact of intraoperative lavage fluid contamination in patients undergoing PD. One hundred five patients who underwent PD between August 2022 and July 2023 were retrospectively enrolled. The intraoperative bile and peritoneal lavage were collected for bacterial culture. Postoperative drainage bacterial cultures were performed every 2–3 days thereafter until drains were all removed. The bacteria isolated from intraoperative lavage fluid, intraoperative bile, and postoperative drainage fluid were examined in detail. The risk factors associated with positive intraoperative lavage fluid culture were analyzed through both univariate and multivariate analyses. Organ/space SSI occurred in 59(56.2%) of the 105 patients. The positivity rates of cultures in intraoperative lavage fluid, intraoperative bile, and postoperative drainage fluid were found to be 41.0%, 67.6%, and 84.8%, respectively. Patients with positive intraoperative lavage fluid culture had a significantly higher occurrence of organ/space SSI compared to the negative group (69.0% vs. 29.4%, $P < 0.001$). Preoperative biliary drainage (PBD) was identified as the only independent risk factor for the contamination of intraoperative lavage fluid (OR = 7.687, 95% CI: 2.164–27.300, $P = 0.002$). *K. pneumoniae* was the most common isolates both in the intraoperative lavage fluid and postoperative drainage fluid. Intraoperative lavage fluid contamination closely correlated with organ/space SSI after PD. Meanwhile, PBD was the only risk factor for the contamination of intraoperative lavage fluid.

Keywords Pancreaticoduodenectomy, Surgical site infection, Intraoperative lavage

Over the past decade, dramatic advances in surgical technique and perioperative management have transformed pancreaticoduodenectomy (PD) into a relatively safe surgery in high-volume centers. However, the incidence of postoperative complications, including clinically relevant pancreatic fistula (CR-POPF) and organ/space surgical site infection (SSI), remains high, ranging from 25–65%[1,2]. As reported in several previous studies, intraoperative bile contamination or postoperative drainage fluid contamination has an adverse impact on postoperative complications after PD including organ/space SSI[1,3,4]. As the final step of PD, intraoperative lavage was designed to isolate and eliminate potential bacterial contamination as well as other substances that may facilitate microorganism proliferation[5]. Current studies on the clinical significance of intraoperative lavage fluid culture have focused on appendicitis, peritonitis and colorectal resection[6–9]. Nevertherless, there is still a lack of research on the clinical implications of intraoperative lavage fluid culture in patients undergoing PD.

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Given the significant prevalence of organ/space SSI following PD and the paucity of studies on intraoperative lavage fluid contamination, the objective of this study was to investigate the significance of intraoperative lavage fluid contamination on postoperative complications following PD.

Methods
Patients
All patients who underwent PD between August 2022 and July 2023 were enrolled retrospectively in the Department of Pancreatic and Metabolic Surgery, Nanjing Drum Tower Hospital, Affiliated Hospital of Medical School, Nanjing University. The inclusion criteria were as follows: (a) patients who underwent PD; (b) no history of chemotherapy and/or radiotherapy, and (c) > 18 years of age. The exclusion criteria were as follows: (a) underwent simultaneous hepatic or colon resection; (b) received emergency surgery for trauma; and (c) clinical data were incomplete. The study was approved by the Health Research Ethics Board of Drum Tower Hospital of Nanjing University Medical School (2021-271-01) and conducted in accordance with the Declaration of Helsinki.

Surgical procedures and perioperative management
The modified Child's method was applied to the standard procedure for PD. It was common practice to use manual duct-to-mucosal, end-to-side, and double-layer interrupted anastomosis. The details of hepaticojejunostomy, gastrojejunostomy and the indications of preoperative biliary drainage (PBD) were described previously[10]. The intraoperative bile was obtained for culture when the bile duct was transected. After the reconstruction of the digestive tract, surgeons changed the sterile gloves as well as the suction device. Intraoperative peritoneal lavage was conducted using 3000 ml saline, and the final fluid from the lavage was collected for bacterial culture.

Prophylactic antibiotics were routinely prescribed as third-generation cephalosporins routinely, such as ceftriaxone, and administered until postoperative day (POD) 2. In patients with positive bile cultures from PBD and resistance to ceftriaxone, the prophylactic antibiotics were selected based on the antimicrobial susceptibility. Postoperative drainage fluid tests, including amylase and bacterial culture, were performed on POD 1, 3 and 5 and every 2–3 days thereafter until drains were all removed.

Clinical data collection and definition of complication
Demographic data, preoperative laboratory data, intraoperative variables, pathological diagnosis and postoperative complications were all collected. The occurrence of SSI, including incisional and organ/space SSI, was diagnosed according to the Centers for Disease Control and Prevention (CDC) guidelines[11,12]. Postoperative complications' severity was assessed by the Clavien–Dindo classification, with major complications defined as grade ≥ III[13]. CR-POPF, biliary leakage (BL), delayed gastric emptying (DGE), chyle leakage (CL) and post-pancreatectomy hemorrhage (PPH) were diagnosed according to the International Study Group of Pancreatic Surgery (ISGPS)[14–18].

Statistical analysis
Clinical data analysis was conducted by SPSS 26.0 software for Windows (SPSS Inc.). Categorical variables were expressed as absolute numbers and percentages and analyzed by the χ^2 test or Fisher's exact test. Continuous variables, which distributed normally, were expressed by mean and standard deviation (SD) and analyzed by the independent *t*-test. Continuous variables that were distributed non-normally were shown as median (interquartile range, IQR) and compared by the Mann-Whitney *U* test. Variables with $P < 0.1$ in univariate analysis entered the multivariate logistic regression model. Odds ratio (OR) and 95% confidence intervals (95% CI) were obtained. A *P* value of < 0.05, two sides, was considered statistically significant.

Results
Patient characteristics
105 patients were enrolled into the study consist of 60(57.1%) men and 45(42.9%) women with a median age of 63 years. The clinical and baseline characteristics were shown in Table 1. 102(97.1%) patients were treated by open technique and PBD was performed in 40(38.1%) patients. There were 32(30.5%) patients diagnosed with pancreatic ductal adenocarcinoma (PDAC), 41(39.0%) with Vater's ampullary carcinoma (VAC), and 8(7.6%) with distal cholangiocarcinoma(DCC). The positivity rates of cultures in intraoperative lavage fluid, intraoperative bile, and postoperative drainage fluid were found to be 41.0%, 67.6%, and 84.8%, respectively.

Surgical outcomes and risk factors for positive lavage fluid culture
All patients were classified into two groups as negative lavage fluid culture group ($n = 34$, 32.4%) and positive lavage fluid culture group ($n = 71$, 67.6%). The most prevalent and perilous postoperative complications were listed in Table 2. Organ/space SSI occurred in 59 (56.2%) of the 105 patients. Patients with positive intraoperative lavage fluid culture had a significantly higher occurrence of organ/space SSI compared to the negative group (69.0% vs. 29.4%, $P < 0.001$). Meanwhile, the rates of CR-POPF (35.2% vs. 14.7%, $P = 0.030$) and major complications (21.7% vs. 0.0%, $P = 0.004$) were significantly higher in the group with positive intraoperative lavage fluid culture.

The univariate and multivariate analysis of risk factors for positive intraoperative lavage fluid culture were shown in Table 3. PBD was identified as the only independent risk factor for the contamination of intraoperative lavage fluid (OR = 7.687, 95% CI: 2.164–27.300, $P = 0.002$).

Characteristics	All patients (*n* = 105)
Age (median, IQR), years	63.0 (54.0–71.0)
BMI (mean ± SD), kg/m^2	22.4 ± 2.6
Gender, n (%)	
Male	60 (57.1)
Female	45 (42.9)
Hypertension, n (%)	34 (32.4)
DM, n (%)	17 (16.2)
Smoking, n (%)	34 (32.4)
Drinking, n (%)	26 (24.8)
Preoperative jaundice, n (%)	47 (44.8)
PBD, n (%)	40 (38.1)
TB (median, IQR), µmol/L	15.7 (8.0–79.1)
Alb (mean ± SD), g/L	38.3 ± 3.1
CRP (median, IQR), mg/L	3.9 (2.6–7.7)
Hemoglobin (mean ± SD), g/L	119.3 ± 20.1
Pathology, n (%)	
PDAC	32 (30.5)
DCC	8 (7.6)
VAC	41 (39.0)
IPMN	4 (3.8)
SPN	2 (1.9)
SCN	2 (1.9)
pNET	3 (2.9)
Others	13 (11.4)
Type of operation, n (%)	
Open	102 (97.1)
Laparoscopic	3 (2.9)
Vessel resection, n (%)	
Yes	8 (7.6)
No	97 (92.4)
Pancreatic consistency, n (%)	
Hard	17 (83.8)
Soft	88 (16.2)
Diameter of MPD (median, IQR), mm	3.0 (2.0–3.0)
Operation time (median, IQR), min	300.0 (240.0-352.5)
Blood loss volume (median, IQR), ml	300.0 (200.0-400.0)
Blood transfusion (median, IQR), ml	0.0 (0.0-400.0)
Positive intraoperative bile culture, n (%)	43 (41.0)
Positive intraoperative lavage fluid culture, n (%)	71 (67.6)
Positive postoperative fluid culture, n (%)	89 (84.8)

Table 1. Patients characteristics. IQR: interquartile range; SD: standard deviation; BMI: body mass index; DM: diabetes mellitus; PBD: preoperative biliary drainage; TB: total bilirubin, Alb: albumin; PD: pancreaticoduodenectomy; PPPD: pylorus-preserving pancreaticoduodenectomy; MPD: main pancreatic duct; PDAC: pancreatic ductal adenocarcinoma; DCC: distal cholangiocarcinoma; VAC: Vater's ampullary carcinoma; IPMN: intraductal papillary mucinous neoplasm; SPN: solid pseudopapillary neoplasm of the pancreas; SCN: pancreatic serous cystadenoma; pNET: pancreatic neuroendocrine tumor; CRP: C-reactive protein.

Detail of bacterial isolates

The bacteria isolated from intraoperative lavage fluid, intraoperative bile and postoperative drainage fluid were shown in Table 4. The most common cultured microorganisms of intraoperative bile were *E. faecalis* (*n* = 9, 8.6%), followed by *K. pneumoniae* (*n* = 7, 6.7%), *E. faecium* (*n* = 5, 4.8%) and *E. coli* (*n* = 5, 4.8%). *K. pneumoniae* was the most common isolates both in the intraoperative lavage fluid and postoperative drainage fluid. Other common bacteria identified from intraoperative lavage fluid were *E. faecalis* (*n* = 8, 7.6%), *E. coli* (*n* = 8, 7.6%) and *E. cloacae* (*n* = 6, 5.7%). Meanwhile, in addition to *K. pneumoniae* (*n* = 14, 14.3%), *E. faecalis* (*n* = 13, 12.4%), *S. haemolyticus* (*n* = 13, 12.4%), *Fungus* (*n* = 13, 12.4%) and *E. cloacae* (*n* = 8, 7.6%) are more prevalent in postoperative drainage fluid. There were 47(44.8%) patients showing the same result in both intraoperative

Complications	All patients ($n=105$)	Lavage fluid culture negative ($n=34$)	Lavage fluid culture positive ($n=71$)	*P* value
Severe complications				
Pancreatic fistula, n (%)				0.030
Non-PF/biochemical fistula	75 (71.4)	22 (85.3)	53 (64.8)	
CR-POPF	30 (28.6)	5 (14.7)	25 (35.2)	
Major complication, n (%)	15 (14.3)	0 (0.0)	15 (21.7)	0.004
PPH, n (%)	1 0(9.5)	1 (2.9)	9 (12.7)	0.161
BL, n (%)	12 (11.4)	4 (11.8)	8 (11.3)	0.940
Organ/space SSI, n (%)	59 (56.2)	10 (29.4)	49 (69.0)	< 0.001
Non-severe complication				
Incisional SSI, n (%)	3 (2.9)	1 (2.9)	2 (2.8)	1.000
DGE, n (%)	29 (27.6)	13 (38.2)	16 (22.5)	0.092
CL, n (%)	18 (17.1)	5 (14.7)	13 (18.3)	0.647
Hospital stay (days)	31.0 (23.0–27.0)	26.0 (20.8–35.5)	32.0 (24.0–37.0)	0.052

Table 2. Surgical outcomes according to the bacterial culture from intraoperative lavage fluid. PF: pancreatic fistula; CR-POPF: Clinically relevant postoperative pancreatic fistula (Grade B/ C); BL: biliary leakage; CL: chyle leakage; PPH: post-pancreatectomy hemorrhage; DGE: delayed gastric emptying; SSI: surgical site infection.

bile culture and intraoperative lavage fluid. Meanwhile, the concordant rate between intraoperative lavage fluid and postoperative fluid was significantly higher than that between intraoperative bile culture and postoperative fluid (56.2% vs. 32.4%, $P<0.001$) (Fig. 1). Among the 71 patients with positive intraoperative lavage fluid, SSI occurred in 49(69.0%) patients. The concordance between intraoperative lavage fluid and positive fluid culture was 69.4% among these 49 patients. 28(65.1%) patients underwent organ/space SSI in the 43 patients with contaminated intraoperative bile. 53.6% of these 28 patients had similar microorganisms between intraoperative and postoperative drainage fluid. The concordant rate between intraoperative lavage fluid and postoperative fluid in patients suffered from organ/space SSI was significantly higher than that between intraoperative bile and postoperative drain fluid (69.4% vs. 53.6%, $P=0.002$) (Fig. 2).

Discussion

The current study yielded 3 primary findings. First, there was a significant correlation between the contamination of intraoperative lavage fluid and organ/space SSI, as well as the incidence of CR-POPF and major complications after PD. Second, the microorganisms isolated from lavage fluid matched the bacteria responsible for organ/space SSI in most cases. Third, PBD was the independent risk factor for positive lavage culture.

Recent studies indicated that positive culture of intraoperative ascitic fluid may correlate with an increased risk of postoperative complications following PD. Matsuki et al. demonstrated that the incidence of positive lavage culture was 44.9%, which correlates with the occurrence of CR-POPF and organ/space SSI[19]. Sugiura et al. found that the contamination of intraoperative lavage fluid increased the occurrence of organ/space SSI and CR-POPF after PD[5]. In the present study, the incidence of CR-POPF was significantly higher in patients with positive lavage culture, implying that bacterial contamination may induced CR-POPF, although the specific mechanism by which bacteria influences the development of CR-POPF. Our institution conducted peritoneal lavage with 3000 ml saline at the end of operation routinely in the attempt to eliminate the bacteria inoculation and sources that may serve as potential culture medium for microorganisms. However, it was noteworthy that the positive rate of intraoperative lavage fluid culture was remain as high as 65.7% in current study, suggesting a correlation between positive lavage fluid and postoperative organ/space SSI, which was consistent with previous reports. Consequently, in light of the aforementioned findings, it is imperative to minimized intraoperative bacterial contamination and obtain lavage fluid for bacterial culture. This approach aims to alter the treating surgeon to be more vigilant for postoperative complications and modify the antibiotic administration in the early postoperative phase based on intraoperative fluid microbiologic profile and their sensitivity patterns when clinically indicated.

In several previous studies[3,20,21], bacterobilia has frequently been identified as the origin of the causative organism responsible for organ/space SSI following PD. Nevertheless, few report the concordance between bile culture and SSI, as well as intraoperative lavage culture and SSI. In a prospective study of hepato-pancreato-biliary surgery, Yasukawa et al.[22] identified that individuals with positive lavage fluid culture exhibited a significantly higher incidence of both incisional SSI and organ/space SSI than patients with a negative lavage fluid culture. Meanwhile, they found that 32.7% of patients were consistent with the isolates detected in the lavage fluid and in organ/space SSI, as well as 10.1% were consistent with the isolates detected in the preoperative bile and in organ/space SSI. In the present study, the microorganisms cultured from intraoperative lavage fluid were consistent with postoperative drain fluid in 69.4% of patients with organ/space SSI. On the contrary, only 53.6% of the isolates cultured from intraoperative bile corresponded to those isolated from postoperative drain fluid. These results indicated that not all of the bacteria contributing to organ/space SSI were derived from biliary infection. Other potential sources of bacteria contributing to organ/space SSI could be leakage during gastrointestinal reconstruction or retrograde infection from the postoperative drainage tube. In our study, the concordance

Variables	Lavage fluid culture negative (*n* = 34)	Lavage fluid culture positive (*n* = 71)	*P*	OR (95%CI)	*P*
Age (median, IQR), years	59.5 (52.0-68.5)	65.0 (55.0–71.0)	0.024	1.001 (0.957–1.046)	0.979
BMI (mean ± SD), kg/m^2	22.3 ± 2.6	22.5 ± 2.7	0.651		
Gender, n (%)			0.857		
Male	19 (55.9)	41 (57.7)			
Female	15 (44.1)	30 (42.3)			
Hypertension, n (%)	10 (29.4)	24 (33.8)	0.653		
DM, n (%)	6 (17.6)	11 (15.5)	0.779		
Smoking, n (%)	9 (26.5)	25 (35.2)	0.370		
Drinking, n (%)	8 (23.5)	18 (25.4)	0.840		
Preoperative jaundice, n (%)	14 (41.2)	33 (46.5)	0.609		
PBD, n (%)	5 (14.7)	35 (49.3)	0.001	7.687 (2.164–27.300)	0.002
TB (median, IQR), μmol/L	15.7 (9.1–94.1)	15.7 (8.3–72.9)	0.007	0.992 (0.983–1.087)	0.095
Alb (mean ± SD), g/L	38.5 ± 3.0	38.3 ± 3.2	0.719		
CRP (median, IQR), mg/L	3.6 (2.5–5.1)	4.1 (2.8–10.2)	0.002	1.083 (0.986–1.189)	0.094
Hb (mean ± SD), g/L	122.6 ± 17.9	117.7 ± 20.9	0.242		
Pathology, n (%)			0.232		
PDAC	13 (38.2)	19 (26.8)			
Others	21 (61.8)	52 (73.2)			
Type of operation, n (%)			0.549		
Open	34 (100.0)	68 (95.8)			
Laparoscopic	0 (0.0)	3 (4.2)			
Vessel resection, n (%)			0.643		
Yes	2 (5.9)	6 (8.5)			
No	32 (94.1)	65 (91.5)			
Pancreatic consistency, n (%)			0.647		
Hard	5 (14.7)	13 (18.3)			
Soft	29 (85.3)	58 (81.7)			
Diameter of MPD (median, IQR), mm	2.0 (2.0–3.0)	3.0 (2.0–4.0)	0.841		
Operation time (median, IQR), min	292.5 (203.8–342.5)	300.0(250.0–365.0)	0.618		
Blood loss volume (median, IQR), ml	300.0 (200.0–400.0)	300.0 (200.0–500.0)	0.388		
Blood transfusion (median, IQR), ml	300.0 (200.0–400.0)	300.0 (200.0–500.0)	0.323		

Table 3. Univariate and multivariate analysis of the contamination of intraoperative lavage fluid. POD: postoperative day; IQR: interquartile range; SD: standard deviation; OR: odds ratio; CI: confidence interval; BMI: body mass index; DM: diabetes mellitus; PBD: preoperative biliary drainage; TB: total bilirubin, Hb: hemoglobin; Alb: albumin; PD: pancreaticoduodenectomy; PPPD: pylorus-preserving pancreaticoduodenectomy; MPD: main pancreatic duct; PDAC: pancreatic ductal adenocarcinoma; CRP: C-reactive protein.

Microorganisms	Intraoperative bile culture (*n* = 105)	Intraoperative lavage fluid culture (*n* = 105)	Postoperative fluid culture (*n* = 105)
Polymicrobial mixed flora, n (%)	14 (13.3)	18 (17.1)	58 (55.2)
K. pneumoniae, n (%)	7 (6.7)	10 (9.5)	14 (14.3)
E. faecalis, n (%)	9 (8.6)	8 (7.6)	13 (12.4)
E. faecium, n (%)	5 (4.8)	3 (2.9)	7 (6.7)
S. haemolyticus, n (%)	2 (1.9)	1 (1.0)	13 (12.4)
E. coli, n (%)	5 (4.8)	8 (7.6)	10 (9.5)
Fungus, n (%)	4 (3.8)	1 (1.0)	12 (11.4)
E. cloacae, n (%)	4 (3.8)	6 (5.7)	8 (7.6)
P. aeruginosa, n (%)	1 (1.0)	1 (1.0)	1 (1.0)
A. baumannii, n (%)	2 (1.9)	2 (1.9)	5 (4.8)
S. aureus, n (%)	1 (1.0)	3 (2.9)	8 (7.6)

Table 4. Details of bacterial cultures.

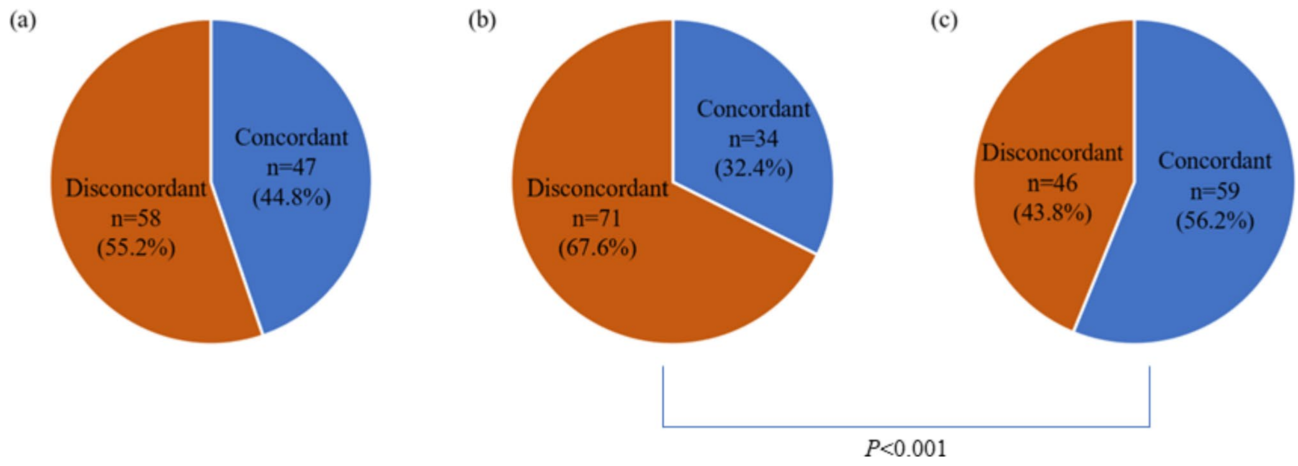

Fig. 1. (a) Intraoperative bile vs. intraoperative lavage fluid; (b) intraoperative bile vs. postoperative fluid; (c) intraoperative lavage fluid vs. postoperative fluid.

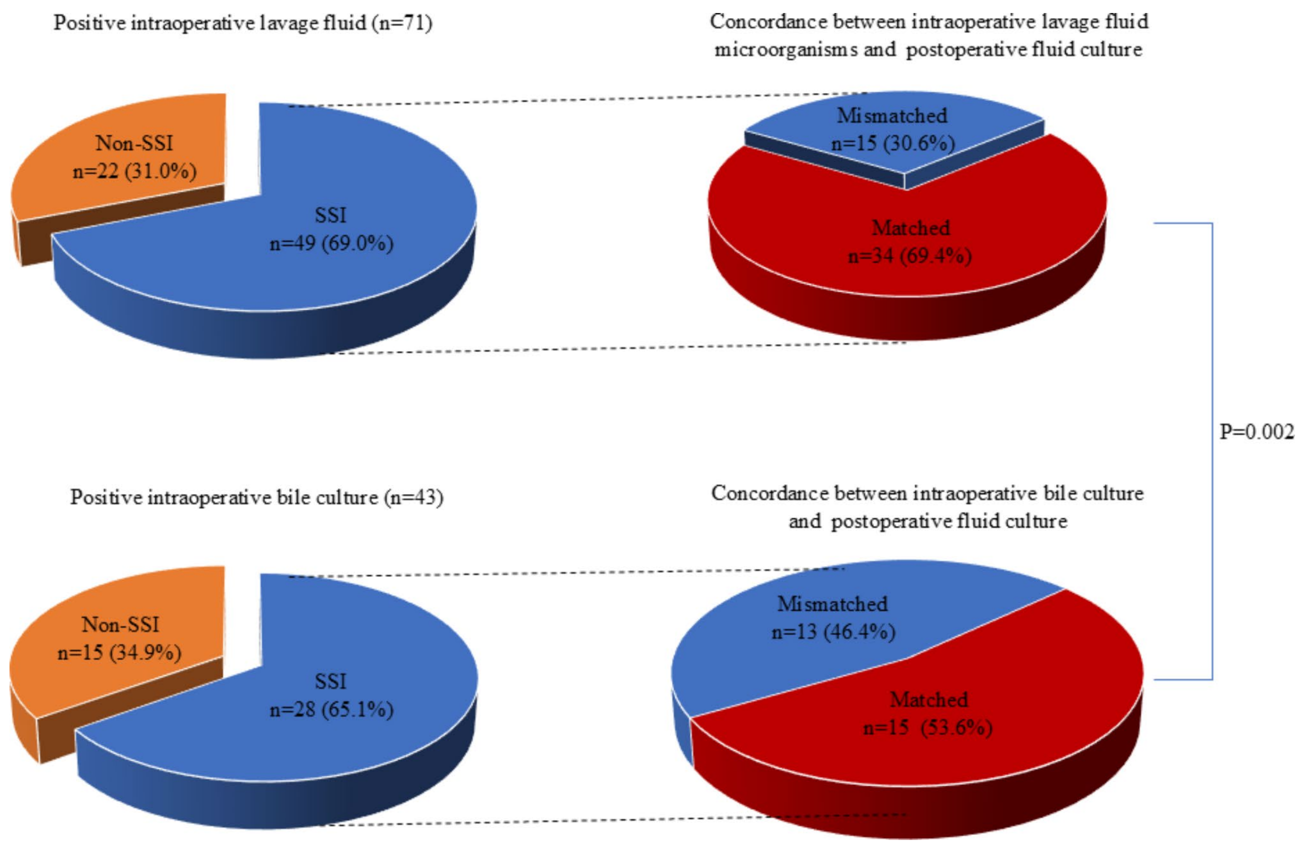

Fig. 2. (a) Positive intraoperative lavage fluid ($n = 71$). Concordance between intraoperative lavage fluid microorganisms and postoperative fluid culture; (b) positive intraoperative bile culture ($n = 43$). Concordance between intraoperative bile culture and postoperative fluid culture.

between intraoperative bile and postoperative ascites culture was only 32.4%, whereas the concordance between intraoperative peritoneal lavage fluid and postoperative ascites culture results was 56.2%. This indicated that intraoperative bile culture may not be sufficient for surveillance of organ/space SSI after PD. Given the high concordance between intraoperative and postoperative ascites culture, as well as the association between intraoperative ascites contamination and subsequent postoperative complications, including organ/space SSI, we deemed it essential to retain intraoperative lavage fluid for bacterial culture in order to adjust the appropriate administration of antibiotics during the early postoperative period.

PBD was performed as a classical method to decompress biliary obstruction and improve hepatic function for patients with jaundice before PD[23]. According to our prior studies, PBD had a significant negative effect on several postoperative complications after PD, especially organ/space SSI[10,24,25]. In current, PBD was identified as the only independent risk factor for the contamination of lavage fluid. It was an invasive procedure that induced retrograde infection of the biliary tract and pancreas from the gastrointestinal tract[26]. On the other hand, PBD may also contribute to a state of inflammation and edema around the surgical region, which escalates the complexity and extends the duration of the surgery[27,28]. These might indirectly contribute to intraoperative bacterial contamination, which in turn leads to organ/space SSI. Presently, there is still a contentious debate on whether PBD routinely improves surgical outcomes. A few studies have suggested that PBD decreases the occurrence of morbidity and mortality after PD[29,30]. Nevertheless, various retrospective analyses and meta-analyses of randomized trials have indicated that routine PBD elevates the risk of overall postoperative complications, including organ/space SSI[28,31,32]. In conjunction with the results of our study, the indication for PBD should be carefully evaluated and performed in selected patients undergoing PD. Simultaneously, routine intraoperative bile cultures help to some extent in monitoring organ/space SSI following PD.

The current study has several limitations. First, it was a single-center retrospective study, which was accompanied by selection bias. Therefore, further multicenter and randomized controlled trials are essential to confirm the significance of intraoperative lavage fluid on organ/space SSI. Second, the sample was small which may result in insufficient statistical efficiency. Then, the study suffered from bacteriological susceptibility which was more valuable for clinical management of organ/space SSI.

Conclusions

In conclusion, the outcomes of our study showed that intraoperative lavage fluid contamination closely correlated with organ/space SSI after PD. Further, limiting the indication of PBD may decrease the occurrence of positive intraoperative ascites culture. At the same time, intraoperative lavage fluid culture may be a useful adjunct to modify the antibiotic agents in the early postoperative period when clinically indicated.

Data availability

The datasets generated and/or analyzed during the present study are not publicly available due to patient privacy concerns, but are available from the corresponding author upon reasonable request.

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Author contributions
L.M. and X.F. designed this study. X.F. and C.L. supervised this study. Y.Y., J.S., C.L., H.C., G.L. and C.C. collected the data. Y.Y., X.F. and J.S. performed statistical analyses in this study. Y.Y. and J.S. prepared the manuscript. L.M. and Y.Q. revised the manuscript. All the authors read and approved the final version of this study.

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Competing interests
The authors declare no competing interests.

Ethics approval and consent to participate
The study was approved by the Health Research Ethics Board of Drum Tower Hospital of Nanjing University Medical School (2021-271-01). Informed consent was obtained from all subjects and/or their legal guardian(s).

Consent for publication
Written informed consent for publication was obtained from all participants.

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