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Perioperative Elevated Peripheral Blood Eosinophil Identifies Postoperative Surgical Site Infections in Crohn's Patients with Gastrointestinal Fistula

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Abstract

Background: Recent studies have implied peripheral blood eosinophil (PBE) can be used as a biomarker in inflammatory bowel disease (IBD). However, the role of PBE in predicting postoperative surgical site infection (SSI) in patients of Crohn's disease (CD) with gastrointestinal (GI) fistula remains unknown. The purpose of this study was to evaluate the predictive value of perioperative PBE in SSI in Crohn's patients undergoing definitive bowel resection.

Methods: One hundred and twenty-two CD patients with GI fistula undergoing definitive bowel reconstruction were enrolled in this study. Leukocyte, neutrophil, C-reactive protein (CRP), procalcitonin (PCT), peripheral blood eosinophil (PBE) was evaluated preoperatively and postoperatively. SSI was an infection occurred at the surgical site within 30 days after operation, which was identified as purulent drainage from the surgical site, organism cultured from the fluid of surgical site, and/or incisional inflammation. Multivariate logistic regression analysis was used to identify independent risk factors of SSI and receiver operating characteristic curve (ROC) analysis was used to evaluate effectiveness of PBE. Kaplan-Meier curve was used to describe the length of hospital and postoperative stay days in patients with and without high-PBE.

Results: Overall, 37 patients (30.3%) developed surgical site infection (SSI). The level of PBE in SSI group was higher than NSSI group on POD (postoperative day) -1 (p=0.002), 3 (p=0.0001) and POD 5 (p=0.026). In multivariate logistic regression analysis, preoperative high-PBE was independently associated with SSI (P=0.032, OR=3.308, 95%CI: 2.002-5.750). ROC analysis showed that the ideal cutoff value of PBE in predicting SSI was 0.2007×10^9/L and 0.2082×10^9/L on POD -1 and 3 with area under the curve (AUC) was 0.74 and 0.71, respectively. Patients with high-PBE had a longer length of hospital stay (33 *vs* 23 days, p=0.01) and longer length of postoperative stay (16 *vs* 14 days, p=0.04) compared to patients without high-PBE.

Conclusion: Peripheral blood eosinophil (PBE) can be used as a novel marker to predict SSI in CD patients with GI fistula undergoing intestinal resection. Measurement of perioperative PBE is recommended in the routine assessment of CD with GI fistula undergoing bowel resection.

Keywords: Peripheral blood eosinophil; Surgical site infection; Crohn's disease; Fistula; Biomarker

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Abbreviations: PBE: Peripheral Blood Eosinophil; SSI: Postoperative Surgical Site Infections; GI Fistula: Gastro Intestinal Fistula; CD: Crohn's Disease

Introduction

Surgical site infection (SSI) is the most important postoperative complication in patients undergoing definitive bowel resections

[1]. SSI can lead to additional financial burden, prolong the length of postoperative hospital stay and increase the total duration of hospitalization [2]. Timely recognition of SSI can significantly minimize the adverse consequence of this complication. Furthermore, Gastrointestinal (GI) fistula surgery are commonly contaminated or dirty surgery and have higher risk in developing SSI compared with general abdominal operations. SSI is especially common in patients undergoing gastrointestinal fistula resection. Therefore, earlier diagnosis and management of SSI should attract our attention.

GI fistula is a most serious complication in Crohn's disease (CD) that can increase mortality and hospitalization [3,4]. Up to two thirds of CD patients would develop stricture, intra-abdominal abscess and fistula in the clinical course of the disease [5,6]. A population-based study showed that the cumulative risk of GI fistula after diagnosed CD for 10 and 20 years was 33% and 50%, respectively [7]. Approximately 70% CD patients received at least one gastrointestinal resection during the natural course of disease due to spontaneous or postoperative complication [8]. On the one hand, bacteria existed in fistula tract was associated with SSI [9]. Contaminated or dirty wounds provided a fertile ground for intestinal or environmental bacterial colonization. Patients with GI fistula would have more opportunity to be infected by bacteria originating from the fistula tract than the skin [10]. Hence, CD patients with GI fistula were risk at SSI. On the other hand, CD patients frequently accompanied with malnutrition, anemia, or immunosuppression owing to using corticosteroids, immunosuppressant, and biologic [9]. Generally, CD patients with GI fistula were more likely to happen SSI compared with others.

Presently, there are no reports describing the association between PBE and SSI in CD with GI fistula. CD is a chronic gut inflammation disease which involves both innate and adaptive gut immune system [11]. Eosinophils grow in the bone marrow. It release into peripheral blood and intestinal lamina propria when the time is ripe. IL-33, one of Damage associated molecular pattern (DAMPs), bind to the ST receptor on the eosinophil surface, causing IL-13 to be released [12]. A previous study demonstrated that high-PBE was more prevalent in ulcerative colitis (UC) and UC patients with recurrent high-PBE had higher rates of colectomy than normal-PBE patients [13]. IBD patients with high-PBE increased healthcare utilization, and required more aggressive medical therapy compared to normal-PBE. Multivariate logistic anal showed that PBE was significantly associated with hospitalization and surgery in IBD [14]. However, the exact role of PBE in CD, especially in identifying SSI, remains unknown. The present study evaluates the predictive SSI role of PBE in CD patients with GI fistula.

Methods

Study population

We retrospectively reviewed CD patients with GI fistula who were inpatients in GI Fistula Center, Department of Surgery, and Jinling Hospital, China form January 2014 to December 2017. The inclusion criteria were: (1) patients diagnosed as CD according to clinical manifestations, radiologic and endoscopic evidences; (2) GI fistula confirmed by abdominal CT or fistulography; (3) patients undergoing definitive bowel resection for GI fistula caused by CD. A definitive bowel resection was defined as an operation that removes bowel containing CD. The exclusion criteria were: (1) patients who didn't receive an operation; (2) patients undergoing surgical treatment for perianal; (3) patients who accompanied hypereosinophilic syndrome.

Clinical data collection and laboratory measurements

We retrieved data of enrolled patients from the medical record system of Jinling hospital: baseline demographic characteristics (age, sex, body mass index and history of smoking), comorbidity (including diabetes mellitus, hypertension, Hepatitis B virus, oral ulcer and medication allergies), other characteristic of enrolled patients included duration of disease, previous abdominal surgery for CD and history of appendectomy. Blood samples were routinely taken from patients preoperatively and postoperatively at 5:00 AM. Routine laboratory measurements included leukocyte, neutrophil, eosinophil counts, C-reactive protein (CRP), procalcitonin (PCT), hemoglobin (Hb), albumin (Alb), platelet (Plt), and total bilirubin (TBi). According to the World Health Organization [15], anemia is defined as Hb level less than 120 g/L for non-pregnant females and less than 130 g/L for males; leukocytosis was defined as a white blood cell counts $\geq 11 \times 10^9$/L and serum albumin <35 g/L was considered as hypoalbuminemia. High-PBE was defined as absolute count of peripheral blood eosinophil count $\geq 0.4 \times 10^9$/L14 according to criteria of the medical record system.

SSI was defined as an infection of the incision or organ or space occurring at the surgical site within 30 days after surgery [16]. Superficial incisional SSI must meet the following criteria: involves only skin and subcutaneous tissue of the incision and patient has at least one of the following: A. purulent drainage from the superficial incision. B. organisms identified from an aseptically-obtained specimen form the superficial incision or subcutaneous tissue by a culture or non-culture based microbiologic testing method. C. patient has at least one of the following signs or symptoms: pain or tenderness; localized swelling; erythema; or heat. Deep incisional SSI must involve deep soft tissues of the incision and patient has at least one of the following: A. purulent drainage from the deep incision. B. a deep incision that spontaneously dehisces, or is deliberately opened or aspirated by a surgeon. Organ/Space SSI must involve any part of the body deeper than the fascia/muscle layers that is opened or manipulated during the operative procedure. All three subtypes of SSI including superficial incisional, deep incisional and organ/space SSI were taken into account for subsequent analysis.

Preoperative management

For all patients, preoperative management included induction and maintenance of remission using medical management for 2 months prior to operation, preoperative nutritional support by enteral and/or parenteral nutrition, as well as appropriate source control strategy including percutaneous drainage for intra-abdominal abscess. The medical management within 60 days prior to surgery included 5-aminosalicylic acid (sulfasalazine, mesalamine, balsalazide, either orally or topically), steroids, immunomodulator agents (azathioprine, methotrexate, 6-mercaptopurine), anti-TNF therapy (infliximab, adalimumab, certolizumab pegol). Financial data included all healthcare charges for both inpatient and outpatient sources within the medical record system, excluding pharmacy charges.

Statistics

All statistical analysis was performed by Graphpad Prism7 (version 7.0a; GraphPad, San Diego, CA, USA). Continuous variables were expressed as mean ± SE (standard error) and T-test. Categorical variables were presented as numbers and percentages and tested using the Chi-square test or Fisher's exact test. Binary logistic regression analysis was performed to identify significant risk factors for SSI. To better classify the independent risk factor for SSI, all variables found to be different statistically between the two groups were subjected to logistic regression analysis. Receiver operating characteristic (ROC) curves and the corresponding area under curve (AUC) were used to compare the accuracy of each biomarker as a predictor of SSI. Kaplan-Meier curve was used to describe the length of hospital and postoperative stay days in patients with and without high-PBE. A p-value ≤ 0.05 was considered statistically significant.

Results

Clinical characteristics

There were 122 patients including 37 patients in SSI group and 85 in NSSI group. The overall incident of SSI was 30.3% (37/122). Male predominance was observed in both groups (78.4% and 65.9%, respectively). The mean age of the two groups was 38.3 ± 12.2 and 36.0 ± 11.3, respectively. The average BMI of SSI group was lower than NSSI group, even though there was no statistical difference (18.9 ± 2.4 *vs* 18.6 ± 4.2). For smoking, it was similar between the two groups. 16 patients (43.2%) had received more than one abdominal surgery for CD previously in SSI group, which was higher than NSSI (43.2% *vs* 25.9%), but were no different between the two groups. 17 patients (45.9%) had received appendectomy in SSI and 28 patients (32.9%) in NSSI. Perianal lesion was no difference in the two groups. The wound class >III between the two groups reached significant difference, which was 29.7% and 14.9%, respectively (p=0.043) **(Table 1)**.

The medical management was summarized in **Table 2**. 5-amino salicylic acid (5-ASA) including sulfasalazine and mesalazine, were the most commonly used drug in both groups, and the proportion of patients who had this treatment was 65.6% and 69.9%, respectively. Immunosuppressant, azathioprine (AZA) and 6-mercaptopurine (6-MP), were used by 4 patients (10.8%) in SSI and 7 patients (8.2%) in NSSI. Corticosteroids and anti-TNF agents were less used in both groups (13.5% *vs* 9.4%, 10.8% *vs* 8.2%, respectively). Notably, most of patients received enteral nutrition and/or parenteral nutrition before bowel resection (70.3% *vs* 83.5%, 75.7% *vs* 76.5%, respectively).

Table 1 Demographics and disease characteristics of 122 patients.

Variable	SSI (n=37)	NSSI (n=85)	p-value
%	30.3	69.7	-
Gender (male)	29 (78.4)	56 (65.9)	0.168
BMI, mean ± SD, kg/m^2	18.9 ± 2.4	18.6 ± 4.2	0.587
Age, mean ± SD, y	38.3 ± 12.2	36.0 ± 11.3	0.313
Smoking (%)			0.874
Never	29 (78.4)	70 (82.4)	
Former	6 (16.2)	11 (12.9)	
Current	2 (5.4)	4 (4.7)	
Comorbidity (%)			0.893
Diabetes mellitus	1 (2.7)	2 (2.3)	
Hypertension	2 (5.4)	2 (2.3)	
HBV	3 (8.1)	5 (5.9)	
Oral ulcer	3 (8.1)	2 (2.3)	
Medication allergies	2 (5.4)	4 (4.7)	
Perianal disease (%)			0.571
Yes	5 (13.5)	15 (17.6)	
No	32 (86.5)	70 (82.4)	
Duration of disease, mean ± SD, y	6.0 ± 5.4	6.0 ± 4.8	0.975
Wound class (%)			0.043
Contaminated	26 (70.3)	15 (85.1)	
dirty	11 (29.7)	10 (14.9)	
Previous abdominal surgery for CD			0.041
0-1	21 (56.8)	64 (75.3)	
>1	16 (43.2)	21 (24.7)	
History of appendectomy (%)			0.171
Yes	17 (45.9)	28 (32.9)	
No	20 (54.1)	65 (67.1)	

Inflammatory markers and SSI

We compared the laboratory features between the two groups before and after surgery. The level of preoperative leukocyte (7.41 ± 3.38 *vs* 6.28 ± 2.22, p=0.031), eosinophil counts (0.27 ± 0.13 *vs* 0.17 ± 0.17, p=0.002) and neutrophil (5.29 ± 2.95 *vs* 4.08 ± 1.90, p=0.008) showed statistically significant difference between the two groups. 8 patients (21.6%) existed hyperbilirubinemia in the SSI group and 6 patients (7.1%) was observed in the NSSI group (P=0.044), which was higher in SSI group than NSSI group (7.1%) **(Table 3)**.

Postoperatively, the level of leukocyte count was similar on POD 1 but differed on POD 3, 5, 7 in the two groups: 10.40 ± 5.40 *vs* 6.98 ± 2.95 × 10^9/L (p=0.0001) on POD 3; 9.05 ± 3.55 *vs* 5.96 ± 2.12 × 10^9/L (p=0.0001) on POD 5; 10.03 ± 4.53 *vs* 7.29 ± 3.18 × 10^9/L (p=0.0001) on POD 7 **(Figure 1A)**. The level of neutrophil count was different on POD 1, 3, 5 and 7: 12.74 ± 5.32 *vs* 10.65 ± 4.71 × 10^9/L (p=0.0001) on POD 1; 8.86 ± 3.91 *vs* 5.49 ± 2.64 × 10^9/L (p=0.0001) on POD 3; 7.43 ± 3.68 *vs* 4.43 ± 1.92 × 10^9/L (p=0.0001) on POD 5; 8.38 ± 4.34 *vs* 5.54 ± 2.77 × 10^9/L (p=0.001) on POD 7 **(Figure 1B)**. The level of eosinophil count was significant difference between patients with and without SSI on POD 3 and 5: 0.33 ± 0.26 *vs* 0.18 ± 0.14 × 10^9/L (P=0.0001); 0.29 ± 0.37 *vs* 0.21 ± 0.14 × 10^9/L (p=0.026) on POD 5 **(Figure 1C)**. The CRP concentration was similar on POD 1 but different on POD 3, 5 and 7: 168.18 ± 52.22 *vs* 95.35 ± 51.04 mg/L (p=0.0001)

Table 2 Medical management of 122 patients with CD and GI fistula for within 3 months prior to definitive bowel resection.

Variable	SSI (n=37)	NSSI (n=85)	p-value
5-ASA (%)			0.856
Yes	28 (75.7)	63 (74.1)	
No	9 (24.3)	22 (25.9)	
Corticosteroid (%)			0.931
Yes	5 (13.5)	11 (9.4)	
No	32 (86.5)	74 (90.6)	
AZA/6-MP (%)			0.91
Yes	4 (10.8)	7 (8.2)	
No	33 (89.2)	78 (91.8)	
Anti-TNF antibody (%)			0.438
Yes	6 (16.2)	8 (9.4)	
No	31 (83.8)	77 (90.6)	
Preoperative enteral nutrition (%)			0.095
<3 months	11 (29.7)	14 (16.5)	
≥ 3 months	26 (70.3)	71 (83.5)	
Preoperative parenteral nutrition (%)			0.924
Yes	28 (75.7)	65 (76.5)	
No	9 (24.3)	20 (23.5)	

Table 3 Relationship between laboratory parameters and SSI in 122 patients with CD and GI fistula.

Variable	SSI (n=37)	NSSI (n=85)	p-value
Preoperative leukocyte counts, mean ± SD (10^9/L)	7.41 ± 3.38	6.28 ± 2.22	0.031
Preoperative neutrophil counts, mean ± SD (10^9/L)	5.29 ± 2.95	4.08 ± 1.90	0.008
Preoperative eosinophil counts, mean ± SD (10^9/L)	0.27 ± 0.13	0.17 ± 0.17	0.002
Preoperative CRP, mean ± SD (mg/L)	20.82 ± 26.06	13.38 ± 23.74	0.126
Preoperative PCT, mean ± SD (ng/mL)	0.23 ± 0.62	0.09 ± 0.10	0.056
Preoperative platelet count, mean ± SD (10^9/L)	248.84 ± 81.48	225.56 ± 84.38	0.16
Preoperative anima, % (g/L)			0.54
Yes	17 (45.9)	34 (40)	
No	20 (54.1)	51 (60)	
Hypoalbuminemia (%) (g/L)			0.071
Yes	12 (32.4)	15 (17.6)	
No	25 (67.6)	70 (82.4)	
Preoperative total bilirubin (%) (μmol/L)			0.044
<22			
≥ 22	29 (78.4)	79 (92.9)	
	8 (21.6)	6 (7.1)	
High-PBE (%)			0.003
<0.4 × 10^9	21 (56.8)	70 (82.4)	
≥ 0.4 × 10^9	16 (43.2)	15 (17.6)	

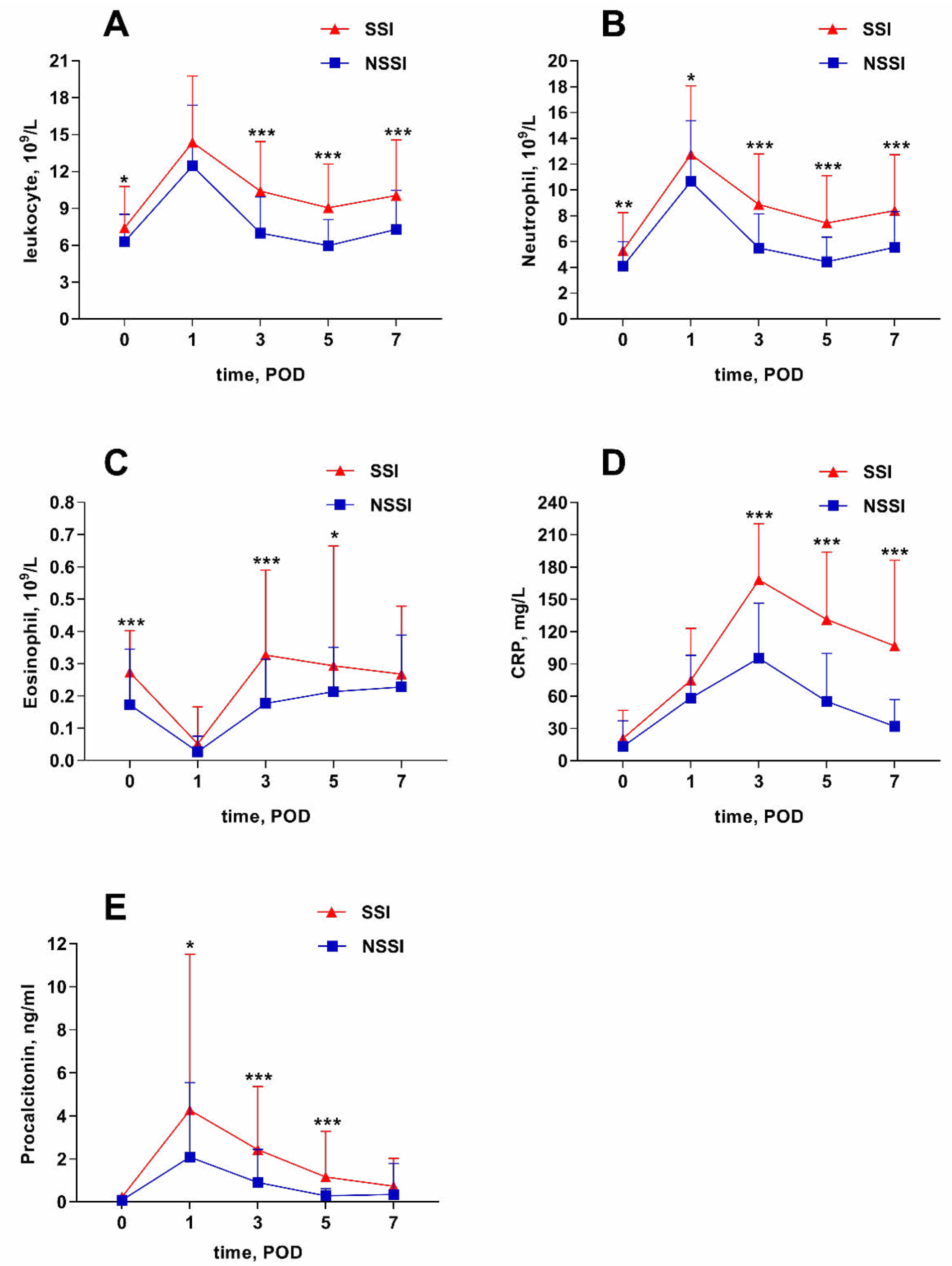

Figure 1 The level of (A) leukocyte, (B) neutrophil, (C) eosinophil, (D) CRP, (E) PCT one day before surgery and on postoperative days (POD) 1, 3, 5 and 7 (*p<0.05, **p<0.01, ***p<0.001). PCT: procalcitonin; CRP: C-Reactive Protein; POD: Post-Operative Day.

on POD 3; 131.35 ± 62.58 *vs* 55.27 ± 44.68 mg/L (p=0.0001) on POD 5; 106.86 ± 79.68 *vs* 32.03 ± 24.72 mg/L (p=0.0001) on POD 7 **(Figure 1D)**. The PCT concentration was different between the two groups on POD 1, 3 and 5: 4.27 ± 7.24 *vs* 2.09 ± 3.46 ng/L (p=0.037) on POD1; 2.43 ± 2.94 *vs* 0.91 ± 1.53 ng/L (p=0.001) on POD 3; 1.16 ± 2.13 *vs* 0.29 ± 0.33 ng/L (p=0.037) on POD 5 **(Figure 1E)**.

Fistula characteristics

10 patients (27%) and 10 (13.9%) had multifistula previously in the two groups, respectively (p=0.036) **(Table 4)**. 18 patients (48.6% *vs* 55.3%) received definitive bowel resection using laparoscopic-assisted while 19 patients (51.4% *vs* 44.7%) by laparotomy in SSI group (p=0.499) **(Table 5)**.

The association between preoperative PBE with postoperative SSI

Superficial incisional SSI were observed in 33 patients (27.0%), deep incisional SSI in 4 patients (5.9%), and organ/space SSI in 14 (11.5%) patients **(Table 6)**. According to whether had PBE elevated preoperatively, we divided patients into high-PBE and normal-PBE group. Furthermore, we found that the frequency of overall SSI (p=0.003), superficial incisional SSI (p=0.009) and organ/space SSI (p=0.025) were different between patients with and without high-PBE. However, no significant statistical

Table 4 Fistula characteristics.

Variable	SSI (n=37)	NSSI (n=85)	p-value
Duration of fistula, mean ± SD (months)	17.6 ± 10.4	15.3 ± 8.8	0.507
Site of fistula (%)			0.944
Small intestinal	17 (39.5)	25 (37.3)	
Colon	14 (32.6)	22 (32.8)	
Rectum	2 (4.7)	5 (7.5)	
Ileocolon Anastomosis	10 (23.3)	15 (22.4)	
Multifistula (%)			0.036
Yes	10 (27)	10 (13.9)	
No	27 (73)	75 (71.1)	
The flow of fistula (%) (mL/24 h)			0.571
<500	32 (86.5)	70 (82.4)	
≥ 500	5 (13.5)	15 (17.6)	

Table 5 Operative treatment of GI fistula in 122 patients[1].

Variable	SSI (n=37)	NSSI (n=85)	p-value
Operative approach (%)			0.499
Laparoscopic-assisted	18 (48.6)	47 (55.3)	
Laparotomy	19 (51.4)	38 (44.7)	
Duration of operation (%) (h)			0.931
<3	32 (86.5)	74 (87.1)	
>3	5 (13.5)	11 (12.9)	
Type of anastomosis			0.52
End-to-end	1 (2.3)	3 (3)	
End-to-side	36 (83.7)	73 (75.3)	
Side-to-side	6 (14)	21 (21.7)	
Number of anastomosis (%)			0.956
0-1	29 (78.3)	67 (78.8)	
≥ 2	8 (21.7)	18 (21.2)	
Intraoperative blood loss (%) (ml)			0.839
<300	34 (91.9)	79 (92.9)	
>300	3 (8.1)	6 (7.1)	
Total cost of hospitalization, mean ± SD (CNY, 1000)	144.8 ± 25.6	64.8 ± 31.1	<0.0001
[1]Because more than one anastomosis may be performed during the surgery, the total number of anastomosis was more than the total number of patients.			

Table 6 Correlation between preoperative PBE and SSI[2].

Frequency of High-PBE			
Incidence of SSI, %	**High-PBE**	**normal-PBE**	**p-value**
Overall SSI			0.003
Present	16 (51.6)	21 (23.1)	
Absent	15 (48.4)	70 (76.9)	
Superficial incisional SSI			0.009
Present	14 (45.2)	19 (20.9)	
Absent	17 (54.8)	72 (79.1)	
Deep incisional SSI			0.985
Present	1 (3.2)	3 (3.3)	
Absent	30 (96.8)	88 (96.7)	
Organ/Space SSI			0.025
Present	7 (22.6)	7 (7.5)	
Absent	24 (77.4)	84 (92.5)	
[2]High-PBE was defined as absolute count of peripheral blood eosinophil count ≥ 0.4 × 10^9/L; Normal-PBE was defined as absolute count of peripheral blood eosinophil count between 0.05 to 0.4 × 10^9/L.			

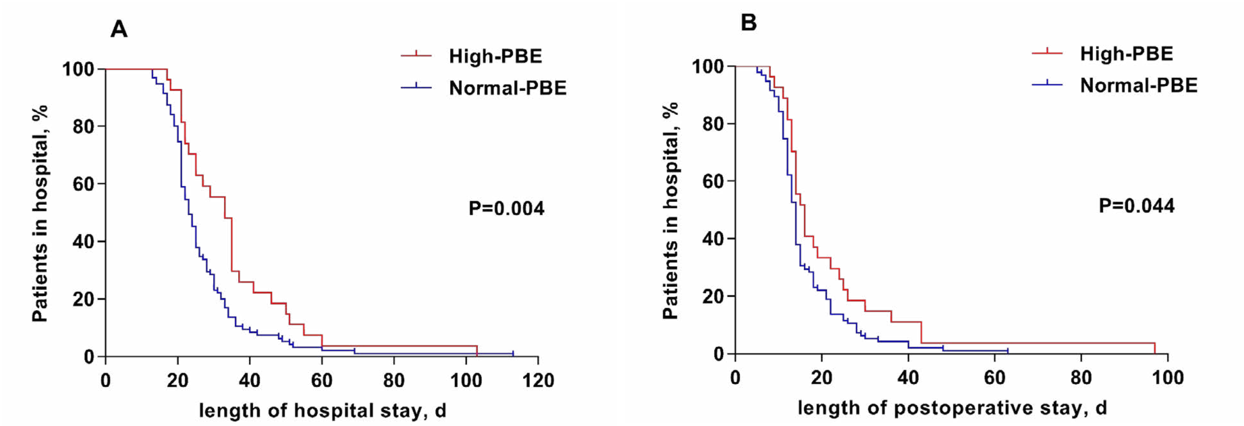

Figure 2 The length of the overall hospital stay and postoperative hospital stay between patients with and without high-PBE.

Table 7 Multiple logistic regression for risk factors for postoperative SSI in CD with GI fistula.

Variable	95%CI	OR	p-value
Preoperative PBE ≥ 0.4 × 10^9	2.002-5.750	3.038	0.032
Preoperative leukocytosis	0.623-1.933	1.097	0.748
Preoperative neutrophil elevated	1.308-3.582	1.776	0.485
Multifistula	0.204-15.406	1.774	0.043
Preoperative TBi≥22	0.027-2.692	0.269	0.264
Previous abdominal surgery for CD	0.530-1.224	0.806	0.311
Wound class>III	2.039-6.861	2.814	0.049

difference was found in deep incisional between patients with and without high-PBE. Patients with high-PBE preoperatively had a longer length of hospital stay (33 *vs* 23 days, P=0.01) and longer length of postoperative stay (16 *vs* 14 days, p=0.04) compared with patients with normal-PBE **(Figure 2)**.

All variables that reached statistically significant difference between the two groups were subjected to multivariate logistic regression analysis. These variables included the level of preoperative PBE, total bilirubin, previous abdominal surgery and type of fistula. Of these, preoperative high-PBE (P=0.032, OR=3.038, 95%CI: 2.002-5.750) and type of fistula (P=0.043, OR=1.774, 95%CI: 0.204-15.406) were independent risk factors for SSI in CD patients with GI fistula **(Table 7)**.

ROC cure analysis

ROC cure analysis showed that eosinophil count was the ideal marker for predicting SSI on POD -1. The optimal cutoff value of eosinophil was 0.2007 × 10^9/L and AUC was 0.74 with 73% sensitivity and 70% specificity on POD -1. The optimal cutoff value of eosinophil was 0.2082 × 10^9/L and the AUC was 0.71 on POD 3, which the sensitivity and specificity was 69% and 73%, respectively. PCT just showed good diagnostic value on POD 3, 5, which the AUC was 0.70 and 0.73, respectively. On the contrary, CRP had a better AUC than PCT on POD 3, 5 (0.85 *vs* 0.70, 0.85 *vs* 0.73). Preoperatively, the AUC of neutrophil was 0.62 with poor sensitivity and specificity. Postoperatively, the AUC of neutrophil was similar with leukocyte on POD 3 and 5 (0.79 *vs* 0.77, 0.77 *vs* 0.77) **(Figure 3)**.

Discussion

The role of perioperative peripheral blood eosinophil in recognizing postoperative SSI in CD with GI fistula has not been previously identified. High-PBE has been reported to be more prevalent in UC and UC with high-PBE had higher rates of colectomy [13]. SSI are one of the most common postoperative complications in patients undergoing colorectal surgery, with increased postoperative morbidity, longer hospital stay, and health care burden [17]. Therefore, it's necessary to identify and manage SSI early following GI fistula surgeries. Hence, the present study explored the predictive value of PBE in CD patients with GI fistula who undergoing definitive bowel resection. Firstly, majority of previous studies focused on colonic eosinophil in ulcerative colitis, with little or no attention to PBE, especially in CD. Furthermore, our result demonstrated that PBE was better than CRP and leukocyte in identifying SSI.

Fistula is the most serious complication in CD patients. Patients with enteroatmospheric or multifistula eventually need definitive bowel reconstruction. Bacteria originating from environment and intestinal colonized in the tract. Patients with GI fistula would have more opportunity to be infected by bacteria from the fistula tract. The present study showed that multifistula was an independent risk factor for SSI.

CRP, a very widely accepted inflammation biomarker, usually has been used to predict SSI. A meta-analysis demonstrated that the ability of CRP to predict postoperative complications was highest on POD 4 (0.84) but worse on POD 1 (0.64) [18]. Another meta-analysis revealed that the highest diagnostic accuracy of CRP was

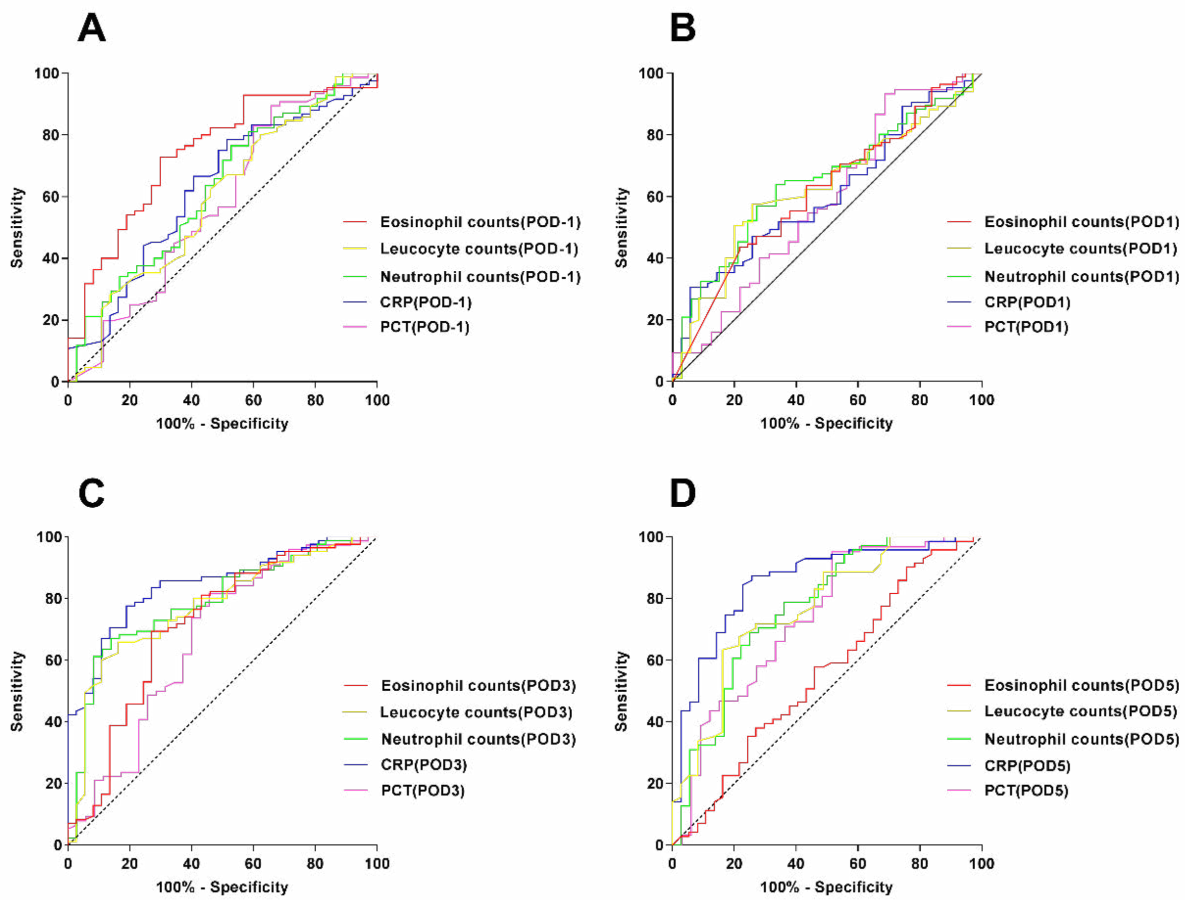

Figure 3 Receiver operating characteristic curve (ROC) for biomarkers on POD -1, 1, 3, 5. AUC: area under the curve.
(A) POD -1: The eosinophil has best AUC, which is 0.74 with 73% sensitivity and 70% specificity.
(B) POD 1: The neutrophil has best AUC, which is 0.65 with 64% sensitivity and 67% specificity.
(C) POD 3: The CRP has best AUC, which is 0.85 with 78% sensitivity and 81% specificity.
(D) POD 5: The CRP has best AUC, which is 0.85 with 85% sensitivity and 77% specificity.

detected on POD 4 with an AUC of 0.7 in the abdominal surgery [19]. Both the two meta-analysis were consistent with our result. In the present study, CRP had a better AUC on the POD 3 (AUC: 0.85) and 5 (AUC: 0.85) than other markers. A prospective study to investigate the diagnostic accuracy of CRP and PCT in early detection of SSI after elective colorectal surgery showed that CRP was superior in identifying SSI compared with PCT [20]. PCT, which was elevated in the infection, showed significant difference between the two groups on POD 1, 3, 5. However, the AUC of PCT was worse than CRP with low sensitivity and specificity on POD 3 and 5 (0.85 *vs* 0.70, 0.85 *vs* 0.73). Two prospective studies also showed that the diagnostic value of CRP was better than PCT in patients after colorectal surgery [21,22].

On the other hand, a previous study show that PCT was a more reliable marker on POD 3 for intra-abdominal infection compared with CRP and leukocyte [23]. Moreover, PCT had a better diagnostic value in anastomotic leak compared with CRP and leukocyte [20]. The prediction of PCT in abdominal infection also was better than CRP on POD 3 and 5, which the value was 0.98 ng/L and 0.83 ng/L, respectively. However, most type of SSI was incision infection in SSI group (33/37). The AUC of PCT was 0.70 and 0.73 on POD 3 and 5 with low sensitivity and specificity. So the role of PCT in identifying SSI in CD need to further research.

A previous study showed that lower preoperative albumin and higher bilirubin were observed in CD patients, making them prone to SSI [24,25]. However, we found that the preoperative albumin was similar between the patients with and without SSI. For the reason, all CD patients who are to have definitive bowel resection for GI fistula received parenteral nutrition or combined with enteral nutrition support to induce remission. Therefore, the abnormal albumin may well have been corrected preoperatively.

Few studies have utilized PBE in predicting postoperative complication, especially in SSI. In the present work, univariate analysis showed that preoperative PBE and neutrophil were significantly different between the two groups. However, only PBE was an independent factor for predicting SSI. Utilizing inflammatory cytokines is one way to improve the early recognition of SSI [26]. CD is an autoimmune disease which involved dysfunction of many inflammatory cytokines. Eosinophil can secrete several cytokines, such as TNF-α, TGF-β and IL-13 [27]. Eosinophil contributed to intestinal inflammation *via* CRTH2 receptor expressed on Th2 cell in experimental colitis [11]. Griseri et al. [28] showed that interleukin-23 released by mature DC and macrophage stimulated Th17 and Th1, what promoted eosinophil to drive chronic colitis. These evidences indicate eosinophils can secrete IL13 and TNF-α inducing intestinal damage. On the other hand, Masterson et al. [29] proposed that eosinophil-deficient mice aggravated intestinal inflammation during acute experimental models of colitis, indicating that eosinophils exert a protective effect in acute mouse colitis. Thus, we believe that perioperative eosinophil levels, which reflect both inflammatory status and immune dysfunction, could be used to predicting SSI in CD patients. Hence, physician should pay attention on PBE in CD patients who receive definitive bowel resection. High-PBE was observed in SSI group (43.2%), which was higher than NSSI group. Eosinophil accumulated in the flamed gut and influenced the healing of mucosa [13,28]. So it can explain why eosinophil had big influence on organ/space SSI. The present report has some limitations.

Conclusion

In conclusion, preoperative elevated peripheral blood eosinophil can significantly identify SSI in Crohn's patients with GI fistula undergoing bowel reconstruction. The number of patients was not large. The study was a retrospective review and detailed information on other postoperative complications was not completely available. Despite this limitation, this may be the first study to evaluate the prognostic significance of PBE in CD patients with GI fistula undergoing definitive bowel resections.

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Conflict of Interests

The authors declare that they have no conflict of interest.

References

1 Uchino M, Ikeuchi H, Tsuchida T, Nakajima K, Tomita N, et al. (2009) Surgical site infection following surgery for inflammatory bowel disease in patients with clean-contaminated wounds. World J Surg 33: 1042-1048.

2 Warren DK, Nickel KB, Wallace AE, Mines D, Tian F, et al. (2017) Risk factors for surgical site infection after cholecystectomy. Open Forum Infect Dis 4: ofx036.

3 Mawdsley JE, Hollington P, Bassett P, Windsor AJ, Forbes A, et al. (2008) An analysis of predictive factors for healing and mortality in patients with enterocutaneous fistulas. Aliment Pharmacol Ther 28: 1111-1121.

4 Huang J, Li Z, Hu Q, Chen G, Ren Y, et al. (2018) Bioinspired anti-digestive hydrogels selected by a simulated gut microfluidic chip for closing gastrointestinal fistula. IScience 8: 40-48.

5 Ren J, Liu S, Wang G, Gu G, Ren H, et al. (2016) Laparoscopy improves clinical outcome of gastrointestinal fistula caused by Crohn's disease. J Surg Res 200: 110-116.

6 Liu S, Ren J, Gu G, Wang G, Han G, et al. (2014) Comparative outcomes of trocar puncture with sump drain, percutaneous drainage, and surgical drainage in the management of intra-abdominal abscesses in Crohn's disease. Surg Innov 21: 580-589.

7 Schwartz DA, Loftus EV, Jr., Tremaine WJ, Panaccione R, Harmsen WS, et al. (2002) The natural history of fistulizing Crohn's disease in Olmsted County, Minnesota. Gastroenterol 122: 875-880.

8 Wang GF, Ren JA, Liu S, Chen J, Gu GS, et al. (2012) Clinical characteristics of non-perianal fistulating Crohn's disease in China: a single-center experience of 184 cases. Chin Med J (Engl) 125: 2405-2410.

9 Guo K, Ren J, Li G, Hu Q, Wu X, et al. (2017) Risk factors of surgical site infections in patients with Crohn's disease complicated with gastrointestinal fistula. Int J Colorectal Dis 32: 635-643.

10 Fan YP, Ren JA, Zhao YZ, Gu GS, Zhao K, et al. (2014) Fistula output microorganism-susceptible antimicrobial prophylaxis is associated with a lower risk of surgical site infection in gastrointestinal fistula patients undergoing one-stage definitive surgery. Surg Infect (Larchmt) 15: 774-780.

11 Radnai B, Sturm EM, Stancic A, Jandl K, Labocha S, et al. (2016) Eosinophils contribute to intestinal inflammation via chemo attractant receptor-homologous molecule expressed on th2 cells, crth2, in experimental crohn's disease. J Crohns Colitis 10: 1087-1095.

12 Masterson JC, Capocelli KE, Hosford L, Biette K, McNamee EN, et al. (2015) eosinophils and il-33 perpetuate chronic inflammation and fibrosis in a pediatric population with stricturing crohn's ileitis. Inflamm Bowel Dis 21: 2429-2440.

13 Barrie A, Mourabet ME, Weyant K, Clarke K, Gajendran M, et al. (2013) Recurrent blood eosinophilia in ulcerative colitis is associated with severe disease and primary sclerosing cholangitis. Dig Dis Sci 58: 222-228.

14 Click B, Anderson AM, Koutroubakis IE, Rivers CR, Babichenko D, et al. (2017) Peripheral eosinophilia in patients with inflammatory bowel disease defines an aggressive disease phenotype. Am J Gastroenterol 112: 1849-1858.

15 Allegranzi B, Zayed B, Bischoff P, Kubilay NZ, De Jonge S, et al. (2016) New WHO recommendations on intraoperative and postoperative measures for surgical site infection prevention: an evidence-based global perspective. Lancet Infect Dis 16: e288-e303.

16 Zhang D, Ren J, Arafeh MO, Sawyer RG, Hu Q, et al. (2018) The Significance of Interleukin-6 in the Early Detection of Surgical Site Infections after Definitive Operation for Gastrointestinal Fistulae. Surg Infect (Larchmt) 19: 523-528.

17 Fan Y, Wei Z, Wang W, Tan L, Jiang H, et al. (2014) The incidence and distribution of surgical site infection in mainland China: a meta-

analysis of 84 prospective observational studies. Sci Rep 4: 6783.

18 Warschkow R, Beutner U, Steffen T, Müller SA, Schmied BM, et al. (2012) Safe and early discharge after colorectal surgery due to C-reactive protein: a diagnostic meta-analysis of 1832 patients. Ann Surg 256: 245-50.

19 Adamina M, Steffen T, Tarantino I, Beutner U, Schmied BM, et al. (2015) Meta-analysis of the predictive value of C-reactive protein for infectious complications in abdominal surgery. Br J Surg 102: 590-598.

20 Garcia-Granero A, Frasson M, Flor-Lorente B, Puga R, Carratala A, et al. (2013) Procalcitonin and C-reactive protein as early predictors of anastomotic leak in colorectal surgery: a prospective observational study. Dis Colon Rectum 56: 475-483.

21 Oberhofer D, Juras J, Pavicic AM, Rancić Zuric I, Rumenjak V, et al. (2012) Comparison of C-reactive protein and procalcitonin as predictors of postoperative infectious complications after elective colorectal surgery. Croat Med J 53: 612-619.

22 Lagoutte N, Facy O, Ravoire A, Chalumeau C, Jonval L, et al. (2012) C-reactive protein and procalcitonin for the early detection of anastomotic leakage after elective colorectal surgery: pilot study in 100 patients. J Visc Surg 149: e345-e349.

23 Giaccaglia V, Salvi PF, Cunsolo GV, Sparagna A, Antonelli MS, et al. (2014) Procalcitonin, as an early biomarker of colorectal anastomotic leak, facilitates enhanced recovery after surgery. J Crit Care 29: 528-532.

24 Yamamoto T, Allan RN, Keighley MR. Pocard M, Vicaut E, et al. (2000) Risk factors for intra-abdominal sepsis after surgery in Crohn's disease. Dis Colon Rectum 43: 1141-1145.

25 Alves A, Panis Y, Bouhnik Y, Pocard M, Vicaut E, et al. (2007) Risk factors for intra-abdominal septic complications after a first ileocecal resection for Crohn's disease: A multivariate analysis in 161 consecutive patients. Dis Colon Rectum 50: 331-336.

26 Facy O, Paquette B, Orry D, Binquet C, Masson D, et al. (2016) Diagnostic accuracy of inflammatory markers as early predictors of infection after elective colorectal surgery: Results from the imacors study. Ann Surg 263: 961-966.

27 Adar T, Shteingart S, Ben Ya'acov A, Bar-Gil Shitrit A, Goldin E, et al. (2014) From airway inflammation to inflammatory bowel disease: eotaxin-1, a key regulator of intestinal inflammation. Clin Immunol 153: 199-208.

28 Griseri T, Arnold IC, Pearson C, Krausgruber T, Schiering C, et al. (2015) Granulocyte macrophage colony-stimulating factor-activated eosinophils promote interleukin-23 driven chronic colitis. Immunity 43: 187-199.

29 Masterson JC, McNamee EN, Fillon SA, Hosford L, Harris R, et al. (2015) Eosinophil-mediated signalling attenuates inflammatory responses in experimental colitis. Gut 64: 1236-1247.