A High C-Reactive Protein Level on Postoperative Day 7 is Associated with Poor Survival of Patients with Pancreatic Ductal Adenocarcinoma after Resection.

The American Surgeon

A high C-reactive protein level on postoperative day 7 is associated with poor survival of patients with pancreatic ductal adenocarcinoma after resection

Journal:	*The American Surgeon*
Manuscript ID	ASU-21-0132.R2
Manuscript Type:	Original Manuscript
Date Submitted by the Author:	n/a
Complete List of Authors:	Tani, Masaji; Shiga University of Medical Science Iida, Hiroya; Shiga University of Medical Science Maehira, Hiromitsu; Shiga University of Medical Science Mori, Haruki; Shiga University of Medical Science Miyake, Toru; Shiga University of Medical Science, Kaida, Sachiko; Shiga University of Medical Science
Keywords:	C-reactive protein, pancreatic ductal adenocarcinoma, survival, adjuvant chemotherapy, postoperative inflammation
Abstract:	Introduction Pancreatic ductal adenocarcinoma (PDAC) is a common malignancy. While inflammation-related biomarkers influence patient survival after resection, it has not been known whether postoperative inflammations affects the survival of PDAC patients or not. Methods It was investigated whether the universal biomarkers on postoperative day (POD) 7 affect the survival of PDAC patients in the retrospective view, and univariate and multivariate analyses were performed via the Cox regression method. Results Overall, 108 consecutive patients underwent resection; 98 (90.7%) had T3 disease and 73 (67.6%) had lymph node metastases. Thirty-four patients (31.5%) experienced postoperative complications. Compared with preoperative values, the white blood cell count and C-reactive protein (CRP) level on POD 7 were significantly elevated ($p<0.001$ for both); conversely, the lymphocyte count was significantly reduced ($p<0.001$). Among 108 patients, 72 received adjuvant chemotherapy. The median overall survival was 21.0 months; the 5-year survival rate was 22.3%. On multivariate analysis, receiving adjuvant chemotherapy and low CRP levels on POD 7 (<7.6 mg/dL) were prognosticators of better survival. However, the CD classification was not a prognosticator of survival after resection. Conclusions Adjuvant chemotherapy and postoperative low CRP levels on POD 7 were prognosticators of better survival of PDAC patients after resection. Surgeons should be aware of managing postoperative infections, because a high postoperative CRP level is related with unfavorable

	survival.

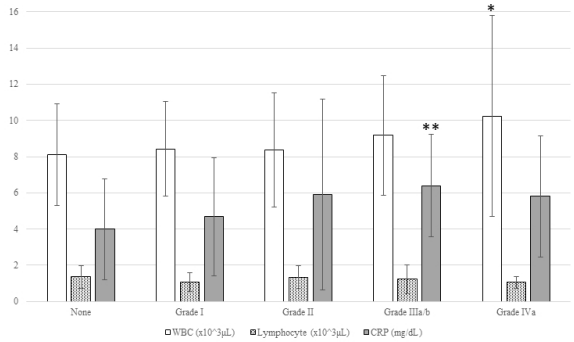

Figure 1.

338x190mm (96 x 96 DPI)

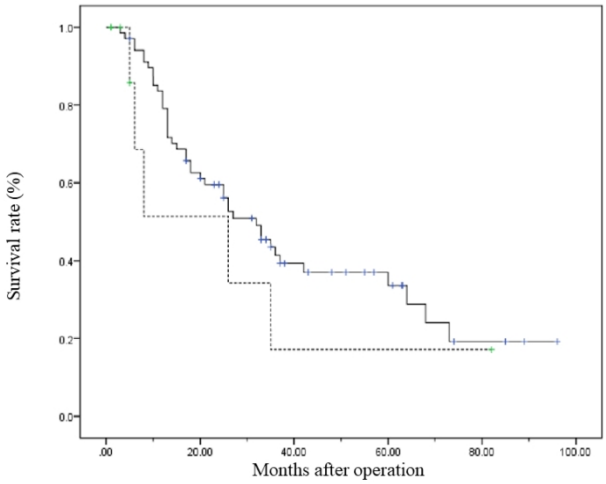

Figure 2.

338x190mm (96 x 96 DPI)

Introduction

Pancreatic ductal adenocarcinoma (PDAC) is a common malignancy, and the survival of patients with PDAC is poor worldwide. Although resection is the only treatment that results in prolonged cancer-free survival, pancreatic resection sometimes causes postoperative complications; in particular, pancreaticoduodenectomy has a high incidence of postoperative complications [1, 2]. Postoperative complications influence the survival of patients who undergo resection for PDAC [3, 4]. As the damage caused by postoperative complications varies, it is not possible to evaluate the damage caused by each complication.

The Clavien-Dindo (CD) classification is commonly used to describe the severity of postoperative complications in various surgical areas; as it is a grading system developed via consensus, it is easy to evaluate the degree of postoperative complications using this classification system [5]. The degree of severity of postoperative complications is associated with the survival of cancer patients after resection because postoperative adjuvant chemotherapy can be postponed owing to immunosuppression due to inflammatory cytokinemia, cancer-cell progression due to growth factors, and immunomodulation due to alteration of the intestinal microbiome induced by antibiotic usage for a long period, among other causes.

However, the CD classification cannot be used to evaluate the exact changes in the severity of systemic inflammation. For example, grade II complications per the CD classification indicate the need for pharmacological treatment with drugs other than those allowed for grade I complications, and grade III complications are classified according to the need for surgical, endoscopic, or radiological intervention. However, it is unclear which treatment is less invasive for patients with intra-abdominal abscess after pancreatic resection: whether long-term antimicrobial treatment should be performed for grade II complications or immediate surgical or radiological intervention is needed for grade III complications. Therefore, the CD classification does not necessarily indicate the degree of systemic inflammation, and the severity of postoperative complications cannot be evaluated, considering the number of postoperative factors.

Postoperative inflammatory biomarkers, observed within 7 postoperative days (POD), aid in the early diagnosis of complications after pancreatic surgery [6], and the median day on which inflammatory postoperative complications are diagnosed is POD 9 [7]. Postoperative systemic inflammation is thought to promote tumor-cell progression because of immunosuppression via inflammatory cytokines, cancer-cell progression via growth factors, and immunomodulation induced by antibiotic usage for a long period,

thereby resulting in postoperative adjuvant chemotherapy being postponed. Therefore, our main hypothesis is that high C-reactive protein (CRP) on POD 7 would be associated with worse survival on multivariate analysis.

Methods

We retrospectively analyzed the data of patients who underwent resection for PDAC at the Shiga University of Medical Science Hospital, Japan, between January 2011 and July 2019. The protocol of this study was approved by the ethics committee of the Shiga University of Medical Science (registration No. 29-171). We provided patients with the opportunity to opt out; however, the need for informed consent was waived because of the retrospective design of the study.

Patients

Patients with pancreatic adenocarcinoma who were resected at the Shiga University of Medical Science Hospital (SUMSH) between January 2011 and July 2019 were included. Resectability was determined by preoperative computed tomography, and the

patients with distant metastases or major arterial invasion were not indicated resection. All the patients underwent en bloc resection with lymph node dissection. Portal vein resection was performed only when the portal vein adhered to and could not be freed from the tumor. Patients who underwent residual resection (R2 resection) were excluded. Pathological tumor staging was performed using the Union for International Cancer Control TNM classification system for malignant tumors (8th edition). The CD classification system consists of 7 grades (I, II, IIIa, IIIb, IVa, IVb, and V). On the basis of the size of the population or the focus of a study, the classification can consist of 5 grades (I, II, III, IV, and V), as the "a" and "b" sub-classification can be removed [5].

Postoperative chemotherapy

Patients who underwent PDAC were informed about both the benefits and adverse effects of postoperative adjuvant chemotherapy after resection. Gemcitabine (1000 mg/m2 × 3 × 6 courses) [8] was administered to patients in the first half of the study. In the latter half of the study, after the results of the JASPAC-1 study proved the superiority of S-1 compared to gemcitabine [9], S-1 (40 mg, 50 mg, or 60 mg according to the body surface area) was orally administered twice a day for 28 days, followed by a 14-day rest, every 6 weeks [9].

Statistical analysis

Continuous data are reported as the mean (standard deviation); normally distributed data were evaluated using Student's t-test. The two-tailed $\chi 2$ test and Fisher's exact test were used to analyze of categorical data. To assess the prognostic significance of individual variables and to identify independent predictors of survival, univariate and multivariate analyses were performed via the Cox regression method. The cut-off values for white blood cell (WBC) and lymphocyte counts and CRP and albumin levels were estimated using 25% percentiles. The analyses were performed with IBM SPSS®, version 22.0 (IBM, Armonk, NY, USA). The Cox proportional hazards regression analysis was performed via a multivariable stepwise selection procedure. The survival analysis was performed according to the Kaplan-Meier method, and survival was compared using the log-rank test and generalized Wilcoxon test. Statistical significance was defined as a P-value of <0.05.

Results

A total of 108 consecutive patients underwent radical resection for PDAC, and all

patients were included in the current study. Table 1 shows the characteristics of the patients with PDAC. Advanced PDAC was the most commonly observed PDAC type; 98 patients (90.7%) had T3 tumors, and 73 patients (67.6%) had lymph node metastases. In addition, 35 patients (32.4%) required biliary drainage for obstructive jaundice. Among the 108 patients, only 8 patients (7.4%) had grade 2 disease per the modified Glasgow prognostic score (GPS). A total of 21 patients underwent neoadjuvant therapy. Table 2 shows the perioperative and postoperative characteristics of the patients. Pancreaticoduodenectomy was performed in 64 patients, distal pancreatectomy in 33, and total pancreatectomy in 11. All the patients underwent lymph node dissection. Thirty-four patients (31.5%) experienced postoperative complications by POD 30. However, no patients died or required re-admission within POD 30.

Compared to the preoperative values, the WBC count and CRP level on POD 7 were significantly elevated ($p<0.001$ for both); in contrast, the lymphocyte count was significantly reduced ($p<0.001$). Table 3 shows the comparison between the CD classification and inflammatory biomarkers. The WBC count was significantly higher in patients with grade IV complications than in those with grade I complications ($p=0.027$). There was no relationship between CRP level and the CD classification. The CRP level of patients with grade III complications was significantly higher than patients with

grade II complications ($p<0.002$; Fig. 1). When patients were classified into those with grade I/II complications and those with grade III/IV complications, the WBC count tended to be higher in patients with grade III/IV complications ($p=0.067$); however, the lymphocyte count, CRP level, and increasing WBC and CRP level were not significantly different (Table 3).

Among the 108 patients, 72 patients received adjuvant chemotherapy using gemcitabine or S-1. Adjuvant chemotherapy was initiated on a median of POD 49 (range, 22–233).

The median follow-up duration was 15 months. A total of 76 recurrence events and 69 deaths were observed. The median recurrence-free survival and overall survival were 12.0 months and 21.0 months, respectively. The 2-year recurrence-free survival rate and the 5-year survival rate were 29.5% and 22.3%, respectively.

Preoperative laboratory examinations were not associated with overall survival after surgery. To assess whether the survival of patients with PDAC, who underwent resection, was affected by the postoperative systemic factors reflecting the inflammatory status and the severity of postoperative complications distinguished using the CD classification, univariate and multivariate analyses were performed using the following factors: T factor (T0/1 or T2/3), lymph node metastasis (present or absent),

neoadjuvant chemotherapy (performed or not performed), adjuvant chemotherapy (performed or not performed), the CD classification (grade I/II or grade III/IV), the WBC count on POD 7, the lymphocyte count on POD 7, and the albumin level on POD 7. Table 4 shows the hazard ratio (HR) and 95% confidence interval (CI) of the factors associated with survival after resection of PDAC. The following factors were indicators of better survival on multivariate analysis using the Cox proportional hazard model: having received adjuvant chemotherapy and a low CRP level on POD 7 (<7.6 mg/dL). Moreover, T0/1 and a high lymphocyte count on POD 7 (>8.5 ×103 μL) tended to improve the survival of patients with PDAC. However, the CD classification was not a predictor of survival after PDAC resection.

Figure 2 demonstrates the survival curve using Kaplan-Meyer method divided into 2 groups (CRP-low and presence of an adjuvant chemotherapy group *versus* CRP-high and no adjuvant chemotherapy group) based on the result of this multivariate analysis. The survival curve were divergent between the two groups, and the 5-year survival rates were 29.3% and 19.4%, respectively (log-Rank; P=0.349, generalized Wilcoxon; P=0.143).

Discussion

Surgical resection is the only treatment method for curing PDAC, besides pancreatectomy is an aggressive procedure with a high incidence of postoperative complications [2, 10, 11]. The results of the current study demonstrate that a higher CRP level on POD 7 was associated with unfavorable survival in patients with PDAC who underwent pancreatectomy. Hence, postoperative systemic inflammation is thought to promote tumor-cell progression because of immunosuppression via inflammatory cytokines, cancer-cell progression via growth factors, and immunomodulation induced by antibiotic usage for a long period, thereby resulting in postoperative adjuvant chemotherapy being postponed.

Inflammatory biomarkers have prognostic value for patients with PDAC, including CRP and albumin levels; lymphocyte, monocyte, neutrophil, and platelet counts; and their derivatives, including the GPS, modified GPS, lymphocyte-to-monocyte ratio, neutrophil-to-lymphocyte ratio, and platelet-to-lymphocyte ratio [12-15]. CRP is useful in the evaluation of systemic inflammation and is an indicator of the postoperative course. CRP is the reactant synthesized in hepatocytes in the acute phase, and it is up-regulated by cytokines, such as interleukin-6, interleukin-8 [16], and tumor necrosis factor-α [17]. In addition, a

preoperative elevated CRP level is associated with poor survival of patients with colorectal cancer [18]. Moreover, preoperative elevated levels of serum CRP are significantly associated with a reduction in lymphocyte percentages in peripheral blood, and it can be an indicator of impaired immunity in patients with colorectal cancer [18].

The lymphocyte count is widely used as an index of immunocompetence, and lymphocytes play a role in tumor immunity during the suppression of carcinogenesis. In the current study, multivariate analysis demonstrated that the lymphocyte count, not the WBC count on POD 7, tended to contribute to better survival. These results suggest that the lymphocyte count is a surrogate marker for subclinical anti-tumor activities of the host. However, future studies should clarify the mechanism of how the lymphocytes improve the survival of patients.

Albumin is used to evaluate the nutritional and immunological status of the host. Hypoalbuminemia is an independent predictor of severe adverse postoperative outcomes [10]. Correlation analysis showed that postoperative albumin levels are only negatively correlated with the WBC and neutrophil counts as well as the CRP level [19]. However, the albumin level can be replenished intentionally via the infusion of exogenous albumin; in addition, the incidence of complications and the 30-day mortality rate do not differ significantly according to the albumin level [19]. In the

current study, the postoperative albumin level did not affect the postoperative survival of patients with PDAC.

The occurrence of postoperative complications might inhibit the performance of adjuvant chemotherapy, which is a favorable factor for survival [3, 8, 9]. The results of multivariate analysis in the current study also revealed that postoperative adjuvant chemotherapy improved patient survival after resection.

The limitations of this study include the small sample size as well as the single-institute, retrospective study design; however, postoperative adjuvant chemotherapy and management of postoperative inflammation were identified as predictors of survival of patients with PDAC who underwent resection. We evaluated the postoperative levels of biomarkers on POD 7; however, it is unknown if POD7 is the optimal timepoint. Unfortunately, the number of patients with high CRP and no adjuvant chemotherapy was too small to prove our hypothesis statistically. However, the Kaplan-Meier curve were divergent between groups (CRP-low and presence of adjuvant chemotherapy *versus* CRP-high and no adjuvant chemotherapy), and we think our hypothesis could be clarified with a larger sample size. In conclusion, surgeons should be aware of managing postoperative infections, because a high postoperative CRP level is related with unfavorable survival.

References

1. McPhee JT, Hill JS, Whalen GF, et al.: Perioperative mortality for pancreatectomy: a national perspective. *Ann Surg* 2007; 246(2): 246-53.
2. Iida H, Tani M, Maehira H, et al.: Postoperative Pancreatic Swelling Predicts Pancreatic Fistula after Pancreaticoduodenectomy. *Am Surg* 2019; 85(4): 321-326.
3. Watanabe Y, Nishihara K, Matsumoto S, et al.: Effect of postoperative major complications on prognosis after pancreatectomy for pancreatic cancer: a retrospective review. *Surg Today* 2017; 47(5): 555-567.
4. Kawai M, Murakami Y, Motoi F, et al.: Grade B pancreatic fistulas do not affect survival after pancreatectomy for pancreatic cancer: A multicenter observational study. *Surgery* 2016; 160(2): 293-305.
5. Dindo D, Demartines N, Clavien PA: Classification of surgical complications: a new proposal with evaluation in a cohort of 6336 patients and results of a survey. *Ann Surg* 2004; 240(2): 205-13.
6. John BJ, Wijeyekoon S, Warnaar N, et al.: Biochemical indicators of in-hospital complications following pancreatic surgery. *Int Surg* 2010; 95(3): 215-20.

7. Welsch T, Frommhold K, Hinz U, et al.: Persisting elevation of C-reactive protein after pancreatic resections can indicate developing inflammatory complications. *Surgery* 2008; 143(1): 20-8.

8. Oettle H, Neuhaus P, Hochhaus A, et al.: Adjuvant chemotherapy with gemcitabine and long-term outcomes among patients with resected pancreatic cancer: the CONKO-001 randomized trial. *JAMA* 2013; 310(14): 1473-81.

9. Uesaka K, Boku N, Fukutomi A, et al.: Adjuvant chemotherapy of S-1 versus gemcitabine for resected pancreatic cancer: a phase 3, open-label, randomised, non-inferiority trial (JASPAC 01). *Lancet* 2016; 388(10041): 248-57.

10. Kantor O, Talamonti MS, Stocker SJ, et al.: A Graded Evaluation of Outcomes Following Pancreaticoduodenectomy with Major Vascular Resection in Pancreatic Cancer. *J Gastrointest Surg* 2016; 20(3): 284-92.

11. DeOliveira ML, Winter JM, Schafer M, et al.: Assessment of complications after pancreatic surgery: A novel grading system applied to 633 patients undergoing pancreaticoduodenectomy. *Ann Surg* 2006; 244(6): 931-7.

12. Forrest LM, McMillan DC, McArdle CS, et al.: Evaluation of cumulative prognostic scores based on the systemic inflammatory response in patients with inoperable non-small-cell lung cancer. *Br J Cancer* 2003; 89(6): 1028-30.

13. Toiyama Y, Miki C, Inoue Y, et al.: Evaluation of an inflammation-based prognostic score for the identification of patients requiring postoperative adjuvant chemotherapy for stage II colorectal cancer. *Exp Ther Med* 2011; 2(1): 95-101.

14. Schlick K, Magnes T, Huemer F, et al.: C-Reactive Protein and Neutrophil/Lymphocytes Ratio: Prognostic Indicator for Doubling overall survival Prediction in Pancreatic Cancer Patients. *J Clin Med* 2019; 8(11): 1791

15. Onoe S, Maeda A, Takayama Y, et al.: The Prognostic Impact of the Lymphocyte-to-Monocyte Ratio in Resected Pancreatic Head Adenocarcinoma. *Med Princ Pract* 2019; 28(6): 517-525.

16. Nakazaki H: Preoperative and postoperative cytokines in patients with cancer. *Cancer* 1992; 70(3): 709-13.

17. Wakuda R, Miki C,Kusunoki M: Autoreactivity against interleukin 6 as a risk factor in elderly patients with colorectal carcinoma. *Arch Surg* 2001; 136(11): 1274-9.

18. Nozoe T, Matsumata T, Sugimachi K: Preoperative elevation of serum C-reactive protein is related to impaired immunity in patients with colorectal cancer. *Am J Clin Oncol* 2000; 23(3): 263-6.

19. Xu W, Peng X, Jiang B: Hypoalbuminemia after pancreaticoduodenectomy does

not predict or affect short-term postoperative prognosis. *BMC Surg* 2020; 20(1): 72.

Figure Legends

Figure 1. Relationship between the grade on the Clavien-Dindo classification and the white blood cell and lymphocyte counts as well as C-reactive protein level on postoperative day 7. *; p=0.027 (none vs. grade IVa), **; p=0.002 (none vs. grade IIIa/b).

Figure 2. The Kaplan-Meyer curve was shown divided into 2 groups (CRP-low and presence of adjuvant chemotherapy; solid line *versus* CRP-high and no adjuvant chemotherapy; break line), and 2 curve is divergent, suggesting that the patients with CRP-high and no adjuvant chemotherapy had poor survival factors .

Table 1. The characteristics of patients

Parameter	
Age (years old) *	68.5 (9.47)
Gender (male/female)	63/45
Body mass index (kg/m^2) *	21.47 (3.85)
Diabetes mellitus	46 (42.6%)
Biliary drainage	35 (32.9%)
T factor (0/1/2/3)	1/4/5/98
N factor (0/1/2)	35/71/2
Hemoglobin (g/dL) *	12.2 (1.68)
WBC (x10^{3}μL) *	5.4 (1.59)
Lymphocyte (x10^{3}μL) *	1.26 (1.508)
Albumin (g/dL) *	3.6 (0.44)
C-reactive protein (mg/dL) *	0.58 (1.11)
modified Glasgow prognostic score (0/1/2)	93/7/8
CEA (ng/mL)**	4.00 (0.9, 30.4)
CA19-9 (U/mL)**	79.00 (1, 3315)
Preoperative chemo(radio)therapy	21 (19.4%)

*; mean (standard deviation), **; median (range)
WBC ;white blood cells, CEA; carcinoembryonic antigen, CA19-9; carbohydrate antigen 19-9,

Table 2. The perioperative and postoperative characteristics of patients

Parameter	
Operative procedures (PD/DP/TP)	64/33/11
Portal vein resection (yes)	32 (29.6%)
Operative time (min.) **	486.5 (199, 853)
Estimate blood loss (mL) **	855.5 (20, 10208)
Blood transfusion (yes)	32 (29.6%)
WBC on POD7 ($x10^3 \mu L$) *	8.7 (3.15)
Lymphocyte on POD7 ($x10^3 \mu L$) *	1.03 (0.43)
Albumin on POD7 (g/dL) *	2.7 (0.40)
C-reactive protein on POD7 (mg/dL) *	5.36 (3.90)
The Clavien-Dindo classification (grade I/II/IIIa/IIIb/IVa/V)	6/35/24/2/8/0
Adjuvant chemotherapy (yes)	77 (71.3%)

*; mean (standard deviation), **; median (range)

PD;pancreaticoduodenectomy, DP; distalpancreatectomy, TP; total pancreatectomy,

WBC ;white blood cells, POD; postoperative days

Table 3. The comparison inflammatory biomarkers divided into the categorized CD classification.

	The CD classification		p-value
	Grade 0/I/II	Grade III/IV	
Preoperative examinations			
WBC (x10^{3}μL)	5.38 (1.59)	5.50 (1.57)	.719
Lymphocyte (x10^{3}μL)	1.55 (0.58)	1.62 (0.68)	.569
C-reactive protein (mg/dL)	.51 (1.14)	.63 (1.06)	.622
Postoperative examinations on postoperative day 7			
WBC (x10^{3}μL)	8.27 (2.93)	9.55 (3.48)	.067
Lymphocyte (x10^{3}μL)	1.05 (0.44)	0.99 (0.41)	.554
C-reactive protein (mg/dL)	4.95 (4.23)	6.26 (2.92)	.107

CD; Clavien-Dindo
WBC; white blood cell,

Table 4. The univariate and multivariate analysis on the survival after pancreatic resection.

	Univariate analysis			Multivariate analysis		
Predictor	HR	95% CI	p-value	HR	95% CI	p-value
T factor (0/1)	.185	.026-1.353	0.094	.165	.023-1.208	.076
LN metastases	1.309	.781-2.193	.307	1.30	.747-2.252	.356
NAC	1.073	.574-2.006	.825	1.119	.591-2.119	.730
Adjuvant chemotherapy	.429	.259-.71	.001	.389	.227-.667	<.001
CD classification (0/I/II)	.770	.482-1.285	.317	.966	.555-1.681	.903
WBC on POD7 (>9.9)	1.230	.724-2.089	.444	.990	.558-1.756	.972
Lymphocyte on POD7 (>8.5)	.777	.396-1.526	.464	.526	.255-1.086	.083
CRP on POD7 (>7.6)	1.020	.962-1.083	.504	1.095	1.023-1.174	.009
Albumin on POD7 (>2.5)	.618	.358-1.065	.083	.610	.333-1.115	.108

HR; hazard ratio (HR), CI; confidence interval,
LN; lymph node, NAC; neoadjuvant chemotherapy, CD; Clavien-Dindo, WBC; white blood cell, POD; postoperative day, CRP; C-reactive protein