ORIGINAL STUDY

Serum chemokine CXCL8 as a better biomarker for diagnosis and prediction of pancreatic cancer than its specific receptor CXCR2, C-reactive protein, and classic tumor markers CA 19-9 and CEA

Ala Litman-Zawadzka[1], Marta Łukaszewicz-Zając[2], Mariusz Gryko[3], Agnieszka Kulczyńska-Przybik[1], Barbara Mroczko[1,2]

1 Department of Neurodegeneration Diagnostics, Medical University of Bialystok, Białystok, Poland
2 Department of Biochemical Diagnostics, Medical University of Bialystok, Białystok, Poland
3 Second Department of General Surgery, Medical University of Bialystok, Białystok, Poland

KEY WORDS

chemokines, chemokines receptor, pancreatic cancer

ABSTRACT

INTRODUCTION Novel biomarkers are critically needed to improve the management of patients with pancreatic cancer (PC).

OBJECTIVES We aimed to evaluate the clinical usefulness of serum CXCL8 in relation to its specific receptor CXCR2 in the diagnosis and prediction of PC compared with classic tumor markers (carbohydrate antigen 19-9 [CA 19-9] and carcinoembryonic antigen [CEA]) and C-reactive protein (CRP).

PATIENTS AND METHODS The study included 76 subjects: 42 patients with PC and 34 healthy volunteers. Serum protein levels were measured by immunological methods.

RESULTS Serum CXCL8 and CXCR2 concentrations were significantly higher in PC patients compared with healthy controls, similarly to classic tumor markers and CRP. CXCL8 levels were significantly elevated in patients with lymph node metastasis compared with individuals without nodal involvement. The diagnostic sensitivity, accuracy, negative predictive value, and areas under the receiver operating characteristic curves for CXCL8 were higher than those for CXCR2, CRP, CA 19-9, and CEA. Moreover, serum CXCL8 was the only significant predictor of PC risk.

CONCLUSIONS Our findings indicate the significance of the CXCL8–CXCR2 axis in the pathogenesis of PC. Serum CXCL8 is emerging as the strongest candidate for a potential PC biomarker among all proteins tested.

Correspondence to:
Prof. Barbara Mroczko, MD, PhD, Department of Neurodegeneration Diagnostics, Medical University of Bialystok, ul. Waszyngtona 15a, 15-269 Białystok, Poland, phone: +48 85 831 87 85, email: mroczko@umb.edu.pl
Received: April 26, 2018.
Revision accepted: July 20, 2018.
Published online: July 27, 2018.
Conflict of interest: none declared.
Pol Arch Intern Med. 2018; 128 (9): 524-531
doi:10.20452/pamw.4307

INTRODUCTION Pancreatic cancer (PC), as a highly lethal disease, is the eighth leading cause of cancer-related deaths worldwide.[1] The main reason for the extremely poor prognosis is the impossibility of detecting this disease at an early stage. It has been proved that only complete resection is the potentially curative treatment, and the 5-year survival for these patients is estimated at 25% to 30% for node-negative disease and 10% for node-positive disease. Despite advances in diagnostic techniques, only 15% to 20% of PC patients are candidates for resection due to lack of disease symptoms and late diagnosis.[2,3] Approximately 85% of patients with this malignancy already have advanced stage at first diagnosis, and in these cases the 5-year survival rate is only 1% to 2%.[4]

In current clinical practice, PC detection techniques include computed tomography, endoscopic ultrasound scanning, endoscopic retrograde cholangiopancreatography, magnetic resonance imaging, and magnetic resonance cholangiopancreatography.[5] However, due to their high cost, invasive nature, and limitations

in detecting early-stage disease, novel methods of diagnosis are critically needed. Carbohydrate antigen 19-9 (CA 19-9) has been established as the most validated serum marker due to its high positive predictive value. In addition, the combined analysis of CA 19-9 with carcinoembryonic antigen (CEA) and CA 125 is useful in the prediction of patients' surgical and chemotherapy outcomes.[2] However, the usefulness of well-established serum biomarkers is unsatisfactory due to their low diagnostic sensitivity and specificity in the early stages of PC. Furthermore, there may be no correlations between imaging test results and concentrations of PC biomarkers.[2] Therefore, diagnosing PC at the early stage of the disease is crucial to improve patients' clinical outcomes.

A growing body of evidence has confirmed the significance of chronic inflammation in the development of various malignancies, including PC.[6] Chemokines are small chemotactic proteins that are involved in physiological and pathological processes, including inflammation and wound healing.[7-9] The family of chemokines has been divided into 4 groups (C, CC, CX3C, CXC), depending on the position of key cysteine residues. Some authors suggest that these proteins may promote the proliferation, migration, invasion, and angiogenesis of tumor cells.[10-13]

CXCL8, known as interleukin 8 (IL-8), belongs to the CXC chemokines, in which the 2 N-terminal cysteines are separated by one amino acid (X). CXCL8 might be produced by monocytes, T lymphocytes, neutrophils, natural killer cells, as well as fibroblasts and epithelial cells. This chemokine acts via its 2 specific receptors: CXCR1 and CXCR2. These proteins are G-protein-coupled receptors with 7 transmembrane domains comprised of 3 extracellular and 3 intracellular domains. The N-terminal domain is extracellular and is involved in chemokine binding. The most potent ligand of CXCR2 is CXCL8 as well as cleavage products of this chemokine.[14,15] CXCR2 is present on monocytes and neutrophils.[7-9,14] It has been proved that the CXCL8–CXCR2 axis is linked to inflammatory processes and might play a role in tumor progression and angiogenesis.[7,14] CXCL8 is known as a powerful promoter of tumor angiogenesis that binds to CXCR2, while its receptor, CXCR2, may regulate the response of endothelial cells to CXCL8.[8] Stromal and neoplastic cells are able to produce CXCL8, which promotes the invasion, metastasis, and, particularly, the angiogenic potential of a number of malignancies, including non–Hodgkin lymphomas as well as solid tumors such as pancreatic neoplasms.[16-20] An immunohistochemical study by Chen et al[21] revealed that the expression of CXCL8 and its specific receptor was higher in PC tissue in comparison with non–PC samples. The authors confirmed that CXCL8 was produced by PC cells.[21] Furthermore, serum CXCL8 levels in PC patients were significantly higher than in patients with other tumors of the digestive system as well as chronic and acute pancreatitis.[21] The authors concluded that PC cells have a higher capability to produce chemokines and function in an autocrine manner than inflammatory cells.

However, to the best of our knowledge, the present study is the first to demonstrate serum concentrations of CXCL8 in relation to those of its specific receptor CXCR2 in patients with PC. The aim of our study was to evaluate the diagnostic significance of serum CXCL8 and CXCR2 as potential novel tumor markers of PC in comparison with well-established tumor markers such as CA 19-9 and CEA, as well as with an inflammatory marker, C-reactive protein (CRP). In addition, correlations between serum levels of the tested proteins and the clinicopathological characteristics of the tumor were assessed. We also examined the relationship between potential risk factors (age, sex, serum levels of the proteins) and the prediction of PC. Moreover, we evaluated the diagnostic usefulness based on diagnostic sensitivity and specificity, accuracy, negative and positive predictive values (NPV and PPV), as well as the areas under the receiver operating characteristic curve (AUC) for CXCL8 and its receptor CXCR2 in comparison with other tested proteins (CRP, CA 19-9 CEA) was also evaluated. The current study is the continuation of our previous investigations in which we assessed whether serum levels of selected chemokines and their specific receptors might be used as potential tumor markers of esophageal carcinoma.[22,23] On the other hand, in our previous studies we established the diagnostic and prognostic utility of other specific proteins, such as matrix metalloproteinase 9 (MMP-9) and its tissue inhibitor 1 (TIMP-1) as well as other cytokines, including macrophage-colony stimulating factor (M-CSF) in PC.[24,25]

PATIENTS AND METHODS

The total study group included 76 patients: 42 patients with PC (17 women and 25 men, aged 35–84 years) and 34 healthy volunteers (14 women and 20 men, aged 22–76 years). Patients were diagnosed in the 2nd Department of General Surgery, Medical University of Bialystok, Poland. The clinical diagnosis of PC was based on the microscopic examination of tissue samples. PC was staged based on the TNM classification, presented by the International Union Against Cancer.[26] All patients with PC were divided into 4 subgroups, dependent on tumor stage (TNM), depth of tumor invasion (T factor), the presence of lymph node metastasis (N factor), and distant metastasis (M factor). The characteristics of PC patients are presented in TABLE 1. Informed consent was obtained from all patients and the present project was approved by the Local Ethics Committee (R-I-002/65/2017) of the Medical University of Bialystok.

Serum samples from PC patients were collected prior to the commencement of treatment and stored at –80°C until assayed. Serum concentrations of CXCL8 and CXCR2 were measured using enzyme-linked immunosorbent assay (ELISA) kits

TABLE 1 Characteristics of patients with pancreatic cancer (n = 42)

Variable		No. (%) of patients
Sex	Male	25 (59.5)
	Female	17 (40.5)
Tumor stage (TNM classification)	I+II	10 (23.8)
	III	15 (35.7)
	IV	17 (40.5)
Depth of tumor invasion	T1+2+3	14 (33.3)
	T4	28 (66.7)
Nodal involvement	N0	17 (40.5)
	N1	25 (59.5)
Distant metastases	M0	25 (59.5)
	M1	17(40.5)

TABLE 2 Differences in serum levels of proteins between patients with pancreatic cancer and healthy controls

Protein	PC (n = 42)	Control group (n = 34)	*P* value
CXCR2, ng/ml	0.93 (0.00–2.26)	0.64 (0.00–1.90)	0.003
CXCL8, pg/ml	39.78 (20.84–1083.49)	6.56 (0.00–27.01)	<0.001
CRP, mg/l	8.65 (0.30–269.40)	1.05 (0.20–5.00)	<0.001
CA 19-9, U/ml	198.52 (2.00–1200.00)	4.97 (2.00–40.97)	<0.001
CEA, ng/ml	2.80 (0.80–319.20)	1.47 (0.50–4.50)	<0.001

Data are presented as median (min–max). A *P* value of less than 0.05 was considered significant.

SI conversion factors: to convert CA 19-9 to kU/l, multiply by 1; CEA to µg/l, by 1; CRP to nmol/l, by 9.524; CXCL8 to µg/l, by 0.001; and CXCR2 to µg/l, by 1.

Abbreviations: CA 19-9, carcinoembryonic antigen 19-9; CEA, carcinoembryonic antigen; CRP, C-reactive protein; CXCL8, C-X-C motif chemokine 8; CXCR2, specific C-X-C motif chemokine receptor 2; PC, pancreatic cancer

(Quantikine ELISA Human CXCL8/IL-8 Immunoassay, Abingdon, R&D Systems, United Kingdon and Human C-X-C chemokine receptor type 2, EIAab, Wuhan, China, respectively) in accordance with the manufacturer's instructions. Serum levels of CA 19-9 and CEA were measured with a microparticle immunoassay (Abbott, Abbott Park, Illinois United States) using ARCHITECT 8200 ci. Serum CRP levels were measured using the immunoturbidimetric method (ARCHITECT 8200 ci, Abbott) in accordance with the manufacturer's instructions.

The Youden index was used to select the optimal predicted probability cutoff values. The reference cutoff values were 23.7 ng/ml for CXCL8, 0.72 ng/ml for CXCR2, 5.7 mg/l for CRP, 54.5 U/ml for CA 19-9, and 1.3 ng/ml for CEA.

Statistical analysis Serum CXCL8, CXCR2, CA 19-9, CEA, and CRP concentrations did not follow a normal distribution in the preliminary statistical analysis (χ^2 test). Therefore, the nonparametric statistical analyses were used. For the comparison between 2 groups, the Mann–Whitney test was used. The Kruskal–Wallis test was used for the analysis of 3 or more groups. The post hoc Dwass–Steele–Critchlow–Fligner test was employed if significant differences were observed.[27] The Spearman rank correlation test was used for correlation analyses. Furthermore, diagnostic parameters, including sensitivity, specificity, accuracy, NPV, and PPV for the tested proteins were also evaluated. The differences were considered as significant at a *P* value of less than 0.05. For statistical analysis, IBM SPSS Statistics 20.0 was used, whereas Microsoft Office Excel was used to calculate diagnostic parameters. Logistic regression was used to assess the strength of the association between various risk factors and PC. Univariate logistic regression models were used first to evaluate the relationship of each variable with PC risk. At the next step, variables with a *P* value of less than 0.05 were introduced into the multivariate model. Finally, the least significant variables were removed in a stepwise manner from the model based on the Wald statistic.

RESULTS Serum levels of the tested proteins (CXCL8, CXCR2, CA 19-9, CEA, and CRP) in patients with PC in comparison with healthy controls are presented in TABLE 2. CXCL8 and CXCR2 levels were higher in PC patients compared with healthy controls (P <0.001 and P = 0.003, respectively), similarly to CA 19-9 and CEA (P <0.001), as well as CRP (P <0.001) (FIGURE 1, TABLE 2).

Following the analysis of the relationship between serum protein concentrations and the clinicopathological characteristics of the tumor, we established that CXCL8 and CXCR2 levels, similarly to CRP and CEA levels, were higher in the serum of patients with lymph node metastasis (N1 subgroup) and distant metastasis (M1 subgroup) in comparison with patients without nodal involvement (N0 subjects) and the presence of distant metastasis (M0 subjects) (TABLE 3). However, a significant difference was found only between serum CXCL8 levels and nodal involvement (P = 0.046; FIGURE 2).

The results of the Spearman rank correlation analysis are presented in TABLE 4. Serum CXCL8 concentrations correlated with CRP levels (P = 0.01) and the presence of lymph node metastasis (P = 0.04) in PC patients.

To assess the diagnostic usefulness of CXCL8 and its receptor in PC patients, we calculated its sensitivity and specificity, accuracy, NPV and PPV, as well as AUC, and compared them with those of CA 19-9 and CEA. The percentage of elevated concentrations (diagnostic sensitivity) of CXCL8 was 98% and was higher than that of CXCR2, CA 19-9, CEA, and CRP (74%, 69%, 90%, and 62%, respectively) (FIGURE 3). The combined measurement of CXCL8 and CXCR2 or CXCL8 and the classic tumor markers increased the diagnostic sensitivity to 100%. Similar results were obtained for the NPV, which was also the highest for CXCL8. The diagnostic specificity of CXCL8 level measurement was 95% and was higher than that of CXCR2 and CEA, but marginally lower than that of CRP and CA 19-9 concentrations, similarly

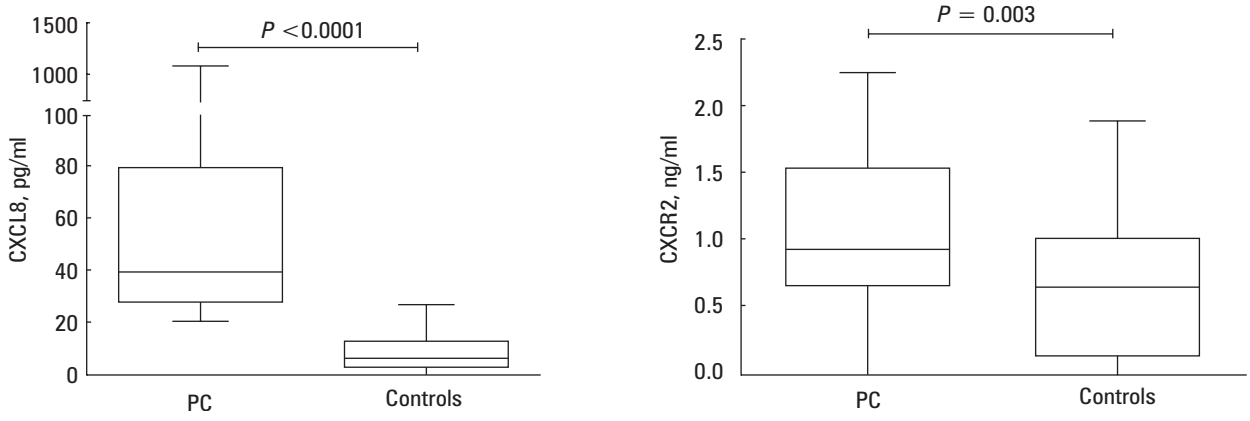


FIGURE 1 Serum concentrations of chemokine CXCL8 and its specific receptor CXCR2 in patients with pancreatic cancer (PC) and healthy controls

TABLE 3 Serum levels of protein biomarkers in patients with pancreatic cancer in relation to clinicopathological features of the tumor

Feature		CXCR2, ng/ml	CXCL8, pg/ml	CRP, mg/l	CA 19-9, U/ml	CEA, ng/ml
Tumor stage (TNM classification)	I+II (n = 10)	1.06 (0.00–1.86)	52.49 (24.56–97.17)	11.10 (0.70–161.50)	162.67 (2.00–1200.00)	1.94 (1.20–10.94)
	III (n = 15)	0.74 (0.08–2.19)	34.59 (24.56–73.75)	3.30 (0.30–157.10)	831.79 (2.00–1200.00)	3.10 (0.92–17.35)
	IV (n = 17)	1.00 (0.25–2.26)	42.55 (20.84–1083.49)	13.70 (0.60–269.40)	122.89 (2.00–1200)	2.81(0.84–319.21)
	P value[a]	–	0.20	0.08	0.46	0.51
Depth of tumor invasion	T1+2+3 (n = 14)	0.95 (0.00–1.86)	52.49 (24.56–1083.49)	11.10 (0.70–161.50)	330.69 (2.00–1200.00)	1.94 (1.20–10.94)
	T4 (n = 28)	0.88 (0.08–2.26)	38.17 (20.84–608.04)	7.75 (0.30–269.40)	198.52 (2.00–1200.00)	2.93 (0.84–319.21)
	P value[b]	0.87	0.82	0.98	0.82	0.25
Presence of lymph node metastasis	N0 (n = 17)	0.85 (0.08–2.19)	30.31 (24.56–573.66)	6.40 (0.30–161.50)	213.32 (2.00–1200.00)	2.06 (0.90–7.20)
	N1 (n = 25)	1.00 (0.00–2.26)	45.95 (20.84–1083.49)	12.90 (0.30–269.40)	122.93 (2.00–1200.00)	2.83 (0.80–319.20)
	P value[b]	0.95	0.046[a]	0.18	0.55	0.26
Presence of distant metastasis	M0 (n = 25)	0.85 (0.00–2.19)	36.08 (24.56–97.17)	6.80 (0.30–161.50)	213.32 (2.00–1200.00)	2.28 (0.90–17.40)
	M1 (n = 17)	1.00 (0.25–2.26)	42.55 (20.84–1083.49)	13.700 (0.6–269.4)	122.89 (2.00–1200.00)	2.81 (0.80–319.20)
	P value[b]	0.73	0.12	0.06	0.28	0.51

Data are presented as median (min–max). A *P* value of less than 0.05 was considered significant.

a Kruskal–Wallis test; **b** Mann–Whitney test

SI conversion factors and abbreviations: see **TABLE 2**

to the PPV. In addition, the diagnostic accuracy of serum CXCL8 levels was also higher than that of the other proteins. The highest accuracy was calculated for the combined measurement of CXCL8 and CA 19-9 (97%) (data not shown). The AUC for CXCL8 (0.9898, $P < 0.001$) was higher than that for CXCR2 (0.6989, $P = 0.001$), CA 19-9 (0.8480, $P < 0.001$), CEA (0.7370, $P < 0.001$), and CRP (0.8260, $P < 0.001$) (**FIGURE 4**). The cutoff values of all the tested proteins were estimated using the Youden index.

The relationship between various risk factors and the risk of PC was first examined in the univariate analysis to identify risk factors to be included in the multivariate model (results were presented as odd ratios [ORs] and *P* values). The concentrations of CXCL8 (OR = 1.653, $P = 0.01$), CXCR2 (OR = 3.839, $P = 0.004$), CA 19-9 (OR = 1.044, $P = 0.02$), CEA (OR = 1.743, $P = 0.01$), and CRP (OR = 1.573, $P = 0.002$), as well as age (OR = 1.125, $P < 0.001$) were significant predictors of increased PC risk. Significant variables in the univariate logistic regression analysis were included in the multivariate model. Subsequently, the least significant variables were removed from the model in a stepwise manner. Therefore, in the final model only the serum CXCL8 level was a significant predictor of PC risk (OR = 1.653, $P = 0.01$).

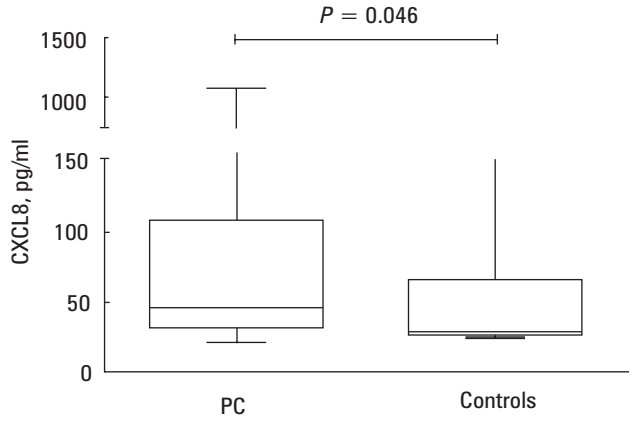


FIGURE 2 Serum concentrations of chemokine CXCL8 in patients with pancreatic cancer (PC) and lymph node metastasis in comparison with individuals without nodal involvement

TABLE 4 Correlations between clinicopathological features of the tumor and serum levels of proteins in patients with pancreatic cancer

Parameter		T	N	M	G	Age	CXCR2	CXCL8	CRP	CA 19-9	CEA
T	*r*	1.00	0.37	0.19	0.51	0.16	0.01	–0.01	0.03	–0.01	0.20
	P	–	0.02	0.24	0.001	0.32	0.94	0.95	0.85	0.93	0.21
N	*r*	0.37	1.00	0.38	0.44	0.10	–0.01	0.31	0.21	–0.09	0.18
	P	0.02	–	0.01	0.003	0.55	0.95	0.044	0.18	0.55	0.27
M	*r*	0.19	0.38	1.00	0.90	0.29	0.05	0.25	0.29	–0.17	0.10
	P	0.241	0.01	–	<0.001	0.07	0.73	0.12	0.06	0.29	0.51
G	*r*	0.51	0.44	0.90	1.00	0.29	0.00	0.17	0.19	–0.12	0.16
	P	0.001	0.003	<0.001	–	0.06	0.99	0.27	0.23	0.45	0.33
Age	*r*	0.16	0.10	0.29	0.29	1.00	0.10	0.14	0.30	–0.01	–0.01
	P	0.32	0.55	0.07	0.06	–	0.53	0.38	0.06	0.94	0.94
CXCR2	*r*	0.01	–0.01	0.05	0.00	0.10	1.00	0.28	0.12	0.00	–0.19
	P	0.94	0.95	0.73	0.999	0.54	–	0.07	0.45	0.99	0.23
CXCL8	*r*	–0.01	0.31	0.25	0.17	0.14	0.28	1.00	0.40	0.23	0.16
	P	0.95	0.044	0.12	0.27	0.38	0.07	–	0.01	0.14	0.32
CRP	*r*	0.03	0.21	0.29	0.19	0.30	0.12	0.40	1.00	–0.04	0.23
	P	0.85	0.18	0.06	0.23	0.06	0.45	0.01	–	0.81	0.14
CA 19-9	*r*	–0.01	–0.09	–0.17	–0.12	–0.01	0.00	0.23	–0.04	1.00	0.18
	P	0.93	0.55	0.29	0.45	0.94	0.99	0.14	0.81	–	0.26
CEA	*r*	0.20	0.18	0.10	0.16	–0.01	–0.19	0.16	0.23	0.18	1.00
	P	0.21	0.27	0.51	0.33	0.94	0.23	0.32	0.14	0.26	–

A *P* value of less than 0.05 was considered significant.

Abbreviations: G, differentiation grade of the tumor; others, see TABLES 1 and 2

DISCUSSION PC is a highly lethal disease, usually diagnosed at a late stage. It is estimated that more than 90% of patients with PC die from this malignancy.[28] Therefore, it is crucial to facilitate the early-stage diagnosis. It has been proved that endogenous cytokines, including selected chemokines and their specific receptors, might be produced by PC cells. In addition, these small peptides serve as autocrine growth factors as well as indicators of immune response to malignancy.[6] Moreover, CXCL8 may act as a prooncogenic effector in many malignancies, including PC.[21,29] The most critical role of this chemokine is its powerful angiogenic potential and its ability to promote the invasion and metastasis of neoplastic cells; thus this chemokine may mimic functions of vascular endothelial growth factor.[17-20]

The present study is a continuation of our previous investigations which focused on the role of specific proteins as candidates for tumor markers in gastrointestinal malignancies.[22-23] According to our knowledge, there have been no studies assessing the diagnostic and prognostic significance of both CXCL8 and its specific receptor CXCR2 in PC in comparison with the well-established tumor markers. In addition, we investigated the role of the CXCL8–CXCR2 axis in PC progression. In our present study, we demonstrated that the concentrations of CXCL8 and CXCR2, similarly to the well-established tumor markers and CRP, were significantly higher in PC patients compared with healthy controls. Our findings suggest that PC cells are able to produce CXCL8. Our results are similar to those of other authors who also observed increased levels of this chemokine

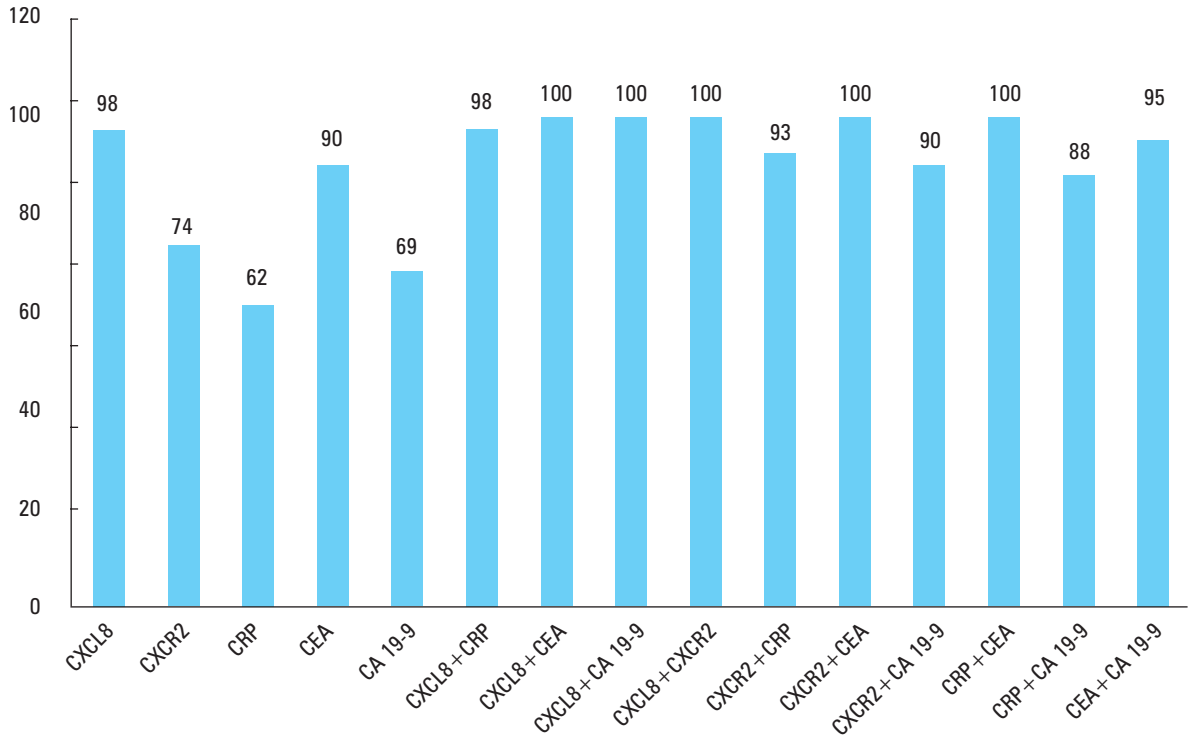
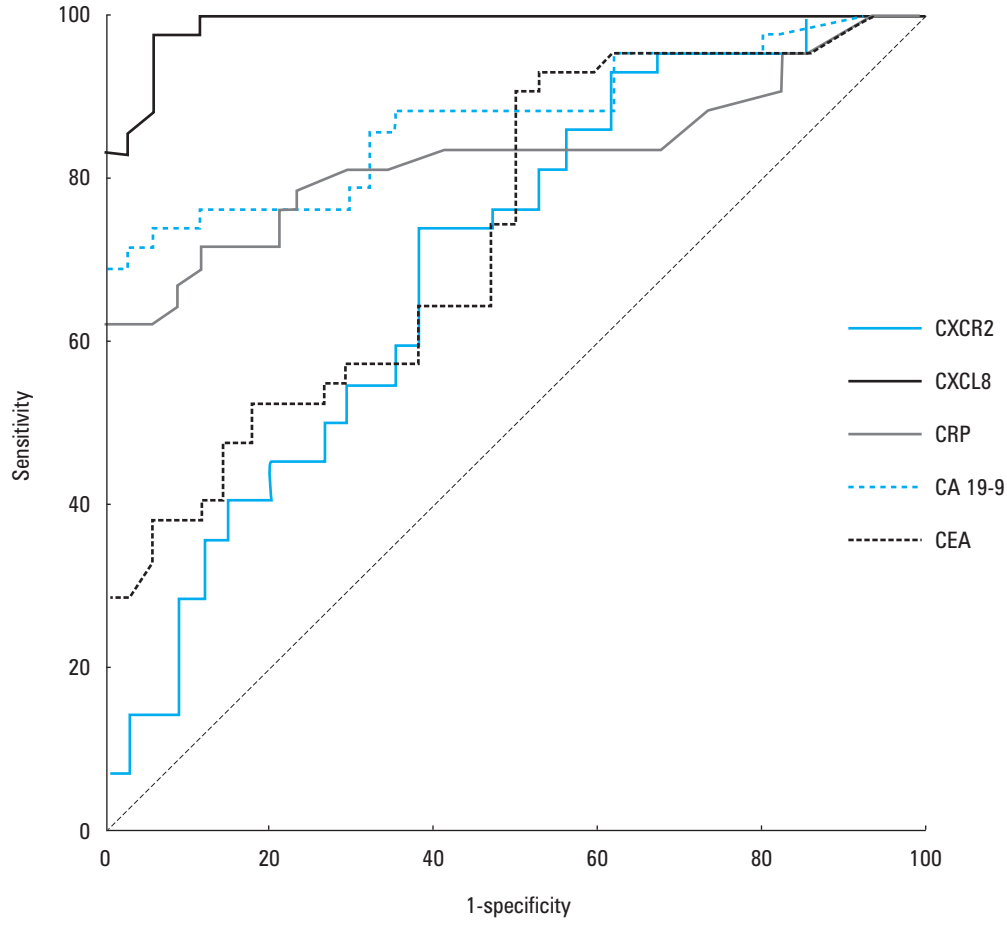


FIGURE 3 Percentage of elevated concentrations of CXCL8 and its specific receptor CXCR2 as well as classic tumor markers and C-reactive protein levels in patients with pancreatic cancer

FIGURE 4 Areas under the receiver operating characteristic curves for chemokine and its specific receptor CXCR2, classic tumor markers CA 19-9 and CEA, and C-reactive protein in patients with pancreatic cancer



in the serum of PC patients in comparison with healthy controls.[30,31] Moreover, Chen et al[21] established that CXCL8 levels were significantly higher in patients with PC than in those with other tumors of the digestive system as well as those with chronic and acute pancreatitis. In our previous study, we investigated the serum levels of MMP-9 and its inhibitor TIMP-1 as well as other

cytokines, including M-CSF, in PC patients.[24,25] Serum M-CSF, MMP-9, and TIMP-1 levels, similarly to those of the classic tumor markers CA 19-9 and CEA, were significantly higher in patients with PC than in healthy controls. Also serum CXCR2 levels were higher in cancer patients compared with the control group, although the study was performed in patients with esophageal cancer.[23]

Our current study demonstrated that CXCL8 concentrations were significantly higher in patients with lymph node metastasis in comparison with patients without nodal involvement, which was confirmed by the Spearman rank correlation test. Based on our present results, we conclude that elevated serum CXCL8 levels may serve as a predictor of nodal involvement in PC, and that they might be used as a marker of tumor progression when the results of imaging methods are inconclusive. Our findings are in line with those of Kuwada et al,[12] who also confirmed that CXCL8 might be an indicator of the invasiveness of PC although they used immunohistochemical techniques to investigate the expression of this protein in PC tissue. Contrasting results were reported by Kim et al,[32] who concluded that this chemokine is not correlated with inflammatory markers and is unable to predict the tumor progression pattern.

In our study, the sensitivity, accuracy, and NPV of serum CXCL8 concentrations were higher than those of CXCR2, CA 19-9, CEA, and CRP. Furthermore, the AUC for CXCL8 was the highest among all the tested proteins in PC patients. If we consider the diagnostic characteristics of all the proteins studied, we may conclude that serum CXCL8 is the best candidate for a tumor marker in the diagnosis of PC. However, our findings need to be confirmed in future studies due to some limitation such as the number of PC patients and control group.

The incidence rate of PC is estimated to be similar to its mortality rate; therefore, there is an urgent need to find a novel biomarker of PC.[33] Numerous papers have presented the expression of CXCL8 or its specific receptor in PC tissue using the labor-intensive immunohistochemical method. However, easy-to-perform, cost-effective, and noninvasive methods in the diagnosis of PC are essential. In conclusion, our study found that out of all the tested proteins, serum CXCL8 was the only significant predictor of PC risk in multiple logistic regression models. Based on diagnostic characteristics, serum CXCL8 seems to be a better marker in PC diagnosis than the classic tumor markers such as CA 19-9 or CEA. Our results confirm the significance of CXCL8 and its specific receptor in the pathogenesis of PC, but the role of the CXCL8–CXCR2 axis in this malignancy is very complicated. Despite the nonspecific nature of these molecules, further investigations are necessary to clarify whether this chemokine might be used as a potential tumor marker of PC.

ACKNOWLEDGMENTS The present study was supported by the Medical University of Bialystok, Poland (N/ST/ZB/17/001/1198; to BM). This study was conducted with the use of equipment purchased by the Medical University of Bialystok as part of the RPOWP 2007–2013 funding, Priority I, Axis 1.1, contract No. UDA-RPPD.01.01.0020-001/15-00.

CONTRIBUTION STATEMENT AL-Z, MŁ-Z, and BM conceived the concept of the study. AL-Z, MŁ-Z, and AK-P contributed to research design and measurement of the tested proteins. MG and BK were involved in sample collection. All authors analyzed the data. BM coordinated project funding. All authors edited and approved of the final version of the manuscript.

REFERENCES

1 Siegel RL, Miller KD, Jemal A. Cancer statistics, 2016. CA Cancer J Clin. 2016; 66: 7-30.

2 Chang JC, Kundranda M. Novel diagnostic and predictive biomarkers in pancreatic adenocarcinoma. Int J Mol Sci. 2017; 18: 667.

3 Li D, Xie K, Wolff R, et al. Pancreatic cancer. Lancet. 2004; 363: 1049-1057.

4 Zhang P, Zou M, Wen X, et al. Development of serum parameters panels for the early detection of pancreatic cancer. Int J Cancer. 2014; 134: 2646-2655.

5 Templeton AW, Brentnall TA. Screening and surgical outcomes of familial pancreatic cancer. Surg Clin North Am. 2013; 93: 629-645.

6 Balkwill F, Mantovani A. Inflammation and cancer: back to Virchow? Lancet. 2001; 357: 539-545.

7 Sui P, Hu P, Zhang T, et al. High expression of CXCR-2 correlates with lymph node metastasis and predicts unfavorable prognosis in resected esophageal carcinoma. Med Oncol. 2014; 31: 809.

8 Shrivastava MS, Hussain Z, Giricz O, et al. Targeting chemokine pathways in esophageal adenocarcinoma. Cell Cycle. 2014; 13: 3320-3327.

9 Keeley BR, Islami F, Pourshams A, et al. Prediagnostic serum levels of inflammatory biomarkers are correlated with future development of lung and esophageal cancer. Cancer Sci. 2014; 105: 1205-1211.

10 Lesina M, Kurkowski MU, Ludes K, et al. Stat3/Socs3 activation by IL-6 transsignaling promotes progression of pancreatic intraepithelial neoplasia and development of pancreatic cancer. Cancer Cell. 2011; 19: 456-469.

11 Fukuda A, Wang SC, Morris JP, et al. Stat3 and MMP7 contribute to pancreatic ductal adenocarcinoma initiation and progression. Cancer Cell. 2011; 19: 441-455.

12 Kuwada Y, Sasaki T, Morinaka K, et al. Potential involvement of IL-8 and its receptors in the invasiveness of pancreatic cancer cells. Int J Oncol. 2003; 22: 765-771.

13 Ikeda O, Egami H, Ishiko T, et al. Signal of proteinase-activated receptor-2 contributes to highly malignant potential of human pancreatic cancer by up-regulation of interleukin-8 release. Int J Oncol. 2006; 28: 939-946.

14 Ogura M, Takeuchi H, Kawakubo H, et al. Clinical significance of CXCL-8/CXCR-2 network in esophageal squamous cell carcinoma. Surgery. 2013; 154: 512-520.

15 Van Damme J, Decock B, Conings R, et al. The chemotactic activity for granulocytes produced by virally infected fibroblasts is identical to monocyte-derived interleukin 8. Eur J Immunol. 1989; 19: 1189-1194.

16 Mazur G, Jaskuła E, Kryczek I, et al. Proinflammatory chemokine gene expression influences survival of patients with non-Hodgkin's lymphoma. Folia Histochem Cytobiol. 2011; 49: 240-247.

17 Matsuo Y, Ochi N, Sawai H, et al. CXCL8/IL-8 and CXCL12/SDF-1alpha co-operatively promote invasiveness and angiogenesis in pancreatic cancer. Int J Cancer. 2009; 124: 853-861.

18 Chen L, Fan J, Chen H, et al. The IL-8/CXCR1 axis is associated with cancer stem cell-like properties and correlates with clinical prognosis in human pancreatic cancer cases. Sci Rep. 2014; 4: 5911.

19 Miyamoto M, Shimizu Y, Okada K, et al. Effect of interleukin-8 on production of tumor-associated substances and autocrine growth of human liver and pancreatic cancer cells. Cancer Immunol Immunother. 1998; 47: 47-57.

20 Li M, Zhang Y, Feurino LW, et al. Interleukin-8 increases vascular endothelial growth factor and neuropilin expression and stimulates ERK activation in human pancreatic cancer. Cancer Sci. 2008; 99: 733-737.

21 Chen Y, Shi M, Yu GZ, et al. Interleukin-8, a promising predictor for prognosis of pancreatic cancer. World J Gastroenterol. 2012; 18: 1123-1129.

22 Łukaszewicz-Zając M, Mroczko B, Kozłowski M, et al. Serum concentrations of chemokine CXCL12 and its specific receptor CXCR4 in patients with esophageal cancer. Dis Markers. 2016; 2016: 7963895.

23 Łukaszewicz-Zając M, Kulczyńska-Przybik A, Muszyński P, et al. Serum concentrations of receptor for interleukin 8 in patients with esophageal cancer. Pol Arch Med Wewn. 2016; 126: 854-861.

24 Łukaszewicz-Zając M, Mroczko B, Wereszczyńska-Siemiątkowska U, et al. Clinical significance of serum macrophage: colony stimulating factor (M-CSF) in pancreatic cancer patients. Gastroenterol Pol. 2009; 16: 29-33.

25 Mroczko B, Lukaszewicz-Zajac M, Wereszczynska-Siemiatkowska U, et al. Clinical significance of the measurements of serum matrix metalloproteinase-9 and its inhibitor (tissue inhibitor of metalloproteinase-1) in patients with pancreatic cancer: metalloproteinase-9 as an independent prognostic factor. Pancreas. 2009; 38: 613-618.

26 Jass JR, Sobin LH. Histological Typing of Intestinal Tumors. Berlin, Germany: Springer-Verlag; 1989.

27 Hollander M, Wolfe DA. Distribution-free two-sided all-treatments multiple comparisons based on pairwise rankings: general configuration. In: Hollander M, Wolfe DA, eds. Nonparametric Statistical Methods. New York, NY: John Wiley & Sons, Inc.; 1999: 240-249.

28 Balasenthil S, Huang Y, Liu S, et al. A plasma biomarker panel to identify surgically resectable early-stage pancreatic cancer. J Natl Cancer Inst. 2017; 109: djw341.

29 Xie K. Interleukin-8 and human cancer biology. Cytokine Growth Factor Rev. 2001; 12: 375-391.

30 Shaw VE, Lane B, Jenkinson C, et al. Serum cytokine biomarker panels for discriminating pancreatic cancer from benign pancreatic disease. Mol Cancer. 2014; 13: 114.

31 Ebrahimi B, Tucker SL, Li D, et al. Cytokines in pancreatic carcinoma: correlation with phenotypic characteristics and prognosis. Cancer. 2004; 101: 2727-2736.

32 Kim HW, Lee JC, Paik KH, et al. Serum interleukin-6 is associated with pancreatic ductal adenocarcinoma progression pattern. Medicine (Baltimore). 2017; 96: e5926.

33 Torre LA, Bray F, Siegel RL, et al. Global cancer statistics, 2012. CA Cancer J Clin. 2015; 65: 87-108.