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A new biological marker in inflammatory bowel disease: Pentraxin 3

İnflamatuvar barsak hastalığında yeni biyolojik göstergeç: Pentraxin 3

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Abstract

Aim: Pentraxin 3 is known to show inflammation, however, few related studies with contradictory results have been conducted in Inflammatory Bowel Disease patients. We aimed to investigate whether PTX3 can be used in the discrimination between Crohn's disease and Ulcerative Colitis (UC) patients and the correlation of PTX3 with disease activity, involved intestinal segments and their length.

Methods: Ethical committee approval was obtained for this prospective case-control study. IBD patients who were older than 18 years of age, not pregnant, and did not have any malignancies, acute/chronic infections or inflammatory diseases were included. Healthy volunteers were included in the control group. PTX3 levels were measured in the blood. The results were statistically analyzed with the NCSS 2007 program.

Results: The median age of 56 male and 42 female participants was 37 years (25 CD, 43 UK and 30 controls). In the UC group, PTX3, which was not related to the Trulove Witts or Mayo scores, was significantly higher in patients with pancolitis (P<0.05 and P=0.046, respectively). There was no relationship between PTX3 levels and involvement, behavior pattern, CDAI scores and activity of the disease in the CD group (P<0.05 for all).

Conclusion: PTX3 is not an appropriate biomarker for monitoring the activation of IBD and it can't distinguish two subtypes from each other. PTX3 is insufficient to indicate both the behavior pattern and the location of involvement in CD patients. Until the control endoscopy times of the patients with UC, it can be monitored whether the disease affects the entire colon by following PTX3 levels.

Keywords: PTX3, IBD, Crohn, Ulcerative colitis, Inflammation

Öz

Amaç: Pentraxin **3**, inflamasyonu gösterdiği bilinen, ancak İBH hastalarında güncel literatürde birbirleri ile çelişkili ve az sayıda çalışma yapılan moleküldür. Bizde bu çalışmamız da PTX3'ün İBH hastalarında Crohn ve Ülseratif kolit hastalarının ayırımında kullanılıp kullanılamayacağını, PTX3 ile hastalığın aktivitesi, tutulan barsak segmenti ve uzunluğu ile korelasyonu olup olmadığını araştırmayı amaçladık.

Yöntemler: Etik kurul onayı bu prospektif vaka kontrol çalışması için alındı. 18 yaşından büyük, gebeliği, malignitesi, akut/kronik enfeksiyöz veya inflamatuvar hastalığı olmayan İBH tanılı hastalar alındı. Kontrol grubu olarak ta sağlıklı gönüllüler çalışmaya dahil edildi. Kanda PTX3 seviyesi bakıldı. Sonuçlar NCSS 2007 programı ile istatistiksel olarak analiz edildi.

Bulgular: 56 kadın, 42 erkek katılımcının ortalama yaşı 37 idi (25 CH, 43 ÜK, 30 kontrol). Ülseratif kolit grubunda Trulove Witts skoru, Mayo skoru ile ilişkiye sahip olmayan PTX3, pankolit tutulumlu hastalarda istatistiksel olarak anlamlı daha yüksekti (P<0.05, P=0.046 sırasıyla). Crohn hastalığı grubunda tutulum yeri, davranış paterni, CDAI skoru ve aktivite ile PTX3 arasında istatistiksel bir ilişki saptanmadı (P<0.05).

Sonuç: PTX3 İBH aktivasyonunun takibinde uygun bir biomarker değildir ve iki alt tipi birbirinden ayıramaz. Crohn hastalarında gerek davranış paterni gerekse tutulum yerini belirtmede yetersizdir. Ancak Ülseratif kolit hastalarının takibinde endoskopik inceleme olmadan tutulan kolon alanının yaygınlığını gösterebilir, dolayısıyla Ülseratif kolit hastalarının kontrol endoskopi zamanları gelene kadar ki ara dönemde PTX3 düzeyleri takip edilerek tutulan barsak alanının tüm kolonu etkileyip etkilemediği izlenebilir.

Anahtar kelimeler: PTX3, İBH, Crohn, Ülseratif kolit, İnflamasyon

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Introduction

Crohn's disease and ulcerative colitis are the subclasses of inflammatory bowel diseases and can affect the digestive system to result in diarrhea, constipation, rectal bleeding, abdominal pain, and malnutrition. Today, the diagnosis is made with the combination of anamnesis, physical examination, laboratory data, and radiology, endoscopy and biopsy results [1].

Histological diagnosis of ulcerative colitis is made by incessant epithelial damage, crypt abscess, and detection of goblet cell decline. Although neutrophils play a significant role in pathogenesis, there is still no treatment in which they are targeted [2-4].

Pentraxin-3 (PTX3) is from the same pentraxin family with short pentraxins such as CRP and serum amyloid A, but it is a long pentraxin. As a multimeric inflammation mediator, it is an acute phase reactant secreted especially from neutrophils [5-7].

Unlike short pentraxins released from the liver, PTX3 is released by the stimulation of cytokines such as IL-1B from different cell types such as mononuclear phagocytes, dendritic cells, endothelial cells, and smooth muscle cells. Unlike CRP, its blood level rises in response to inflammation and returns to normal in a shorter time [8,9].

As neutrophils play a central role in the pathogenesis of Crohn's disease and Ulcerative colitis, we aimed to determine if PTX3 increases at different levels in the blood according to the subtypes of these diseases, their activity status, and their involvement.

Materials and methods

This prospective case-control study was conducted after the Prof. Dr. Cemil Taşcıoğlu City Hospital Ethics Committee's approval (4/16/2019, 1175) was obtained. Blood samples were taken after obtaining the informed consent of the patients who were older than 18 years of age and previously followed up by the gastroenterology outpatient clinic with the diagnosis of inflammatory bowel diseases based on endoscopic evaluations and biopsies. PTX3 levels were measured by the ELISA method. The criteria for exclusion from the study were being under the age of 18, presence of an acute or chronic infection, having any other inflammatory disease that would raise the acute phase reactants, pregnancy, malignancy, inability to participate in the study, chronic or acute kidney failure, or chronic liver disease. The patient group was divided into two as ulcerative colitis and Crohn's disease groups. Smoking history of the patients was also recorded. In all patients, simultaneously with PTX levels, ESR and CRP values were also recorded.

In the ulcerative colitis group, the patients were divided into 4 groups according to the disease location (proctitis, distal colon involvement, left colon involvement and pancolitis). Ulcerative colitis disease activity was defined according to Truelove Witts, endoscopic activity index and Mayo scoring.

In the Crohn's disease group, patients were evaluated according to involvement (ileal, ileocolonic, colonic involvement) and behavior (non-penetrating, stricturing, penetrating) of the disease, and clinical activities were assessed with Crohn's disease activity index (CDAI). Those with CDAI <150 were classified as having mild disease, and those >150 were classified as severe.

Healthy volunteers with no history of disease or medication were included as the control group in the study.

Statistical analysis

Power analysis was performed with the G*power (version 3.1.9.7) program. The minimum calculated sample size was 29 (effect size 0.87, alpha error 0.05, power 0.95).

NCSS (Number Cruncher Statistical System) 2007 (Kaysville, Utah, USA) program was used for statistical analysis. Descriptive statistical methods (mean, standard deviation, median, frequency, and percentage, minimum, maximum) were used when evaluating the study data. The suitability of quantitative data for normal distribution was tested by Shapiro-Wilk test and graphical examinations. Mann-Whitney U test was used to compare the quantitative variables between the two groups that did not show normal distribution. Kruskal Wallis test was used for comparisons of three or more groups that did not show normal distribution, and Bonferroni-Dunn test was used for binary comparisons. Spearman correlation analysis was used to evaluate the relationships between quantitative variables. Statistical significance was set at $P<0.05$.

Results

This study was carried out on 68 patients who volunteered to participate and were under follow-up with the diagnosis of inflammatory bowel disease by the gastroenterology outpatient clinic and 30 healthy volunteers. Among the participants, 57.1% (n=56) were female and 42.9% (n=42) were male; their ages ranged between 18 and 69 years, with a mean age of 37.2 (11.9) years. While 30.6% (n=30) of the cases were in the healthy control group, 43.9% (n=43) were in ulcerative colitis group, 25.5% (n=25) were in Crohn's disease group. PTX3 measurements ranged from 91 to 1691, with a mean of 408.56 (87.03). The rate of smokers was 43.0% (n=37) (Table 1).

There was no statistically significant difference regarding age, gender distribution and smoking rates between the groups ($P=0.198$, $P=0.096$, $P=0.129$). There was a statistically significant difference regarding the PTX3 measurements between the groups ($P=0.021$). The binary comparisons made to determine which group the significant difference originated from yielded that Crohn's disease group's measurements were lower than that of the control group's ($P=0.028$). No statistically significant difference was found in other binary comparisons ($P>0.05$).

Sedimentation and CRP levels were significantly higher in the Crohn's disease group than the ulcerative colitis group ($P<0.05$).

In the ulcerative colitis group, colonic involvement was reported as proctitis in 18.6% (n=8), distal colon in 16.3% (n=7), left colon in 25.6% (n=11) and pancolitis in 39.5% (n=17) of the patients. According to Truelove Witts scoring, 27.9% (n=12) were in remission, 20.9% (n=9) had mildly active, 18.6% (n=8) had moderately active and 32.6% (n=14) had severely active disease. In addition, as patients were analyzed according to the Truelove-Witts clinical activity index, while 27.9% (n=12) of the cases were in remission, 72.1% (n=31) were in their active period. The Mayo score was zero in 25.6% (n=11), one in 11.6%

(n=5), two in 14.0% (n=6), and three in 48.8% (n=21) of the patients.

In the Crohn's disease group, the involvement was ileal in 20.0% (n=5), ileocolonic in 60.0% (n=15) and colonic in 20.0% (n=5) of the patients. The disease was non-penetrating (B1) in 60.0% (n=15), structuring (B2) in 16.0% (n=4) and penetrating (B3) in 24.0% (n=6). According to the CDAI index, 20.0% (n=5) of the cases were in remission while 80.0% (n=20) were in their active period. CDAI measurements ranged from 62 to 511 with a mean of 295.44 (136.21); the disease was mild in 32.0% (n=8) and severe in 68.0% (n=17). Disease duration of all IBD patients varied between 0.01 and 260 months, with a mean of 37.50 (60.29) months. ESR measurements ranged from 2 to 92 with a mean of 33.66 (22.15), and CRP measurements ranged from 1.2 to 446, with a mean of 36.03 (70.87) (Table 2).

There was a statistically significant difference regarding PTX3 measurements between ulcerative colitis, Crohn's disease and control groups (P=0.021). The binary comparisons made to determine which group the significant difference originated from showed that Crohn's disease group's measurements were significantly lower than that of the control group's (P=0.028). No statistically significant difference was found in other binary comparisons (P>0.05) (Table 3).

Table 1: Demographic data

		Control (n=30)	Ulcerative Colitis (n=43)	Crohn's Disease (n=25)	*P*-value
Age (years)	Min-Max (Median)	19-60 (31.5)	18-69 (40)	20-55 (32)	[a]0.198
	Mean (SD)	37.29 (13.64)	39.33 (12.20)	34.04 (9.19)	
Gender	Female	22 (73.3)	22 (51.2)	12 (48.0)	[b]0.096
	Male	8 (26.7)	21 (48.8)	13 (52.0)	
PTX3	Min-Max (Median)	111-848 (453)	142-1138 (296)	91-1691 (245)	[a]0.020*
	Mean (SD)	485.03 (241.90)	378.93 (265.73)	367.76 (357.90)	
Sedimentation	Min-Max (Median)	-	2-92 (25)	6-84 (46)	[c]0.049*
	Mean (SD)	-	28.60 (18.25)	42.36 (25.72)	
CRP	Min-Max (Median)	-	1.16-446 (6.12)	2.1-320 (25)	[c]0.024*
	Mean (SD)	-	29.34 (70.72)	47.52 (71.07)	
Smoking	No	15 (62.5)	25 (64.1)	9 (39.1)	[b]0.129
	Yes	9 (37.5)	14 (35.9)	14 (60.9)	

[a] Kruskal Wallis Test, [b] Pearson Chi-Square test, [c] Mann Whitney U test, * P<0.05

Table 2: The features of Ulcerative Colitis and Crohn's disease groups

*The features of Ulcerative Colitis group (n=43)		
Involvement; n (%)	Proctitis	8 (18.6)
	Distal	7 (16.3)
	Left colon	11 (25.6)
	Pancolitis	17 (39.5)
True love Witts; n (%)	Remission	12 (27.9)
	Mild-active	9 (20.9)
	Moderate-active	8 (18.6)
	Severe-active	14 (32.6)
Activity; n (%)	Remission	12 (27.9)
	Active	31 (72.1)
Mayo; n (%)	0	11 (25.6)
	1	5 (11.6)
	2	6 (14.0)
	3	21 (48.8)
* The features of Crohn's disease group (n=25)		
Involvement; n (%)	İleal	5 (20.0)
	İleocolonic	15 (60.0)
	Colonic	5 (20.0)
Behavior; n (%)	B1 Non-penetrating	15 (60.0)
	B2 Stricturing	4 (16.0)
	B3 Penetrating	6 (24.0)
Activity; n (%)	Remission	5 (20.0)
	Active	20 (80.0)
CDAI; n (%)	Min-Max (Median)	62-511 (313)
	Mean(SD)	295.44 (136.21)
	Mild	8 (32.0)
	Severe	17 (68.0)

Table 3: Evaluation of PTX3 Measurements by Groups

	PTX3			*P*-value
	n	Min-Max (Median)	Mean (SD)	
Control group	30	111-848 (453)	485.03 (241.90)	[a]0.020*
Ulcerative colitis group	43	142-1138 (296)	378.93 (265.73)	
Crohn's disease group	25	91-1691 (245)	367.76 (357.90)	

[a] Kruskal Wallis Test, * P<0.05

In the ulcerative colitis group, a significant relationship was detected between involvement and PTX3 measurements (P=0.049). Binary comparisons to determine which group the significance originated from showed that the PTX3 value of the pancolitis group was higher than the distal involvement group (P=0.046). No statistically significant difference was found in other binary comparisons (P>0.05). There was no statistically significant relationship between Truelove Witts activity or Mayo score and PTX3 measurements (P>0.05).

There was no significant relationship between involvement, behavior, activity and CDAI level and PTX3 measurements in the Crohn's Disease group (P>0.05). PTX3 measurements did not show statistically significant differences among any IBD groups with regards to smoking status (P>0.05) (Table 4).

There was no significant relationship between ESR, CRP levels and PTX3 measurements in the whole IBD group (P>0.05) (Table 5).

There was no statistically significant difference between PTX3 measurements with regards to CDAI levels (P=0.682) (Table 6).

Table 4: Evaluation of PTX3 Measurements According to Descriptive Features

			PTX3		*P*-value
		n	Min-Max (Median)	Mean (SD)	
Smoking status (n=86)	No	49	111-1135 (296)	408.94 (267.69)	[b]0.547
	Yes	37	117-1691 (322)	431.35 (325.64)	
*The features of Ulcerative Colitis group (n=43)					
Involvement; n (%)	Proctitis	8	142-449 (232)	260.50 (111.03)	[a]0.049*
	Distal	7	142-678 (194)	248.14 (191.68)	
	Left colon	11	194-1138 (296)	391.09 (283.9)	
	Pancolitis	17	142-1135 (398)	480.65 (300.37)	
True love Witts	Remission	12	142-678 (296)	344.58 (170.28)	[a]0.568
	Mild-active	9	194-1138 (322)	471.67 (320.56)	
	Moderate-active	8	142-1135 (245)	372 (331.32)	
	Severe-active	14	142-882 (232)	352.71 (271)	
True love Witts Activity	Remission	12	142-678 (296)	344.58 (170.28)	[b]0.800
	Active	31	142-1138 (245)	392.23 (295.92)	
Mayo	0	11	142-678 (296)	363 (165.58)	[a]0.204
	1	5	194-1138 (219)	398 (414.21)	
	2	6	219-1135 (602)	580.17 (345.42)	
	3	21	142-882 (245)	325.24 (234.58)	
* The features of Crohn's disease group (n=25)					
Involvement; n (%)	İleal	5	117-1009 (373)	438.6(347.36)	[a]0.745
	Ileocolonic	15	91-1691 (245)	349.6 (390.68)	
	Colonic	5	168-932 (219)	351.4 (325.32)	
Behavior; n (%)	B1 Non-penetrating	15	142-1009 (270)	384.07 (264.47)	[a]0.204
	B2 Stricturing	4	91-347 (219)	219 (104.51)	
	B3 Penetrating	6	117-1691 (155)	426.17 (624.38)	
Activity	Remission	5	219-475 (270)	316.4 (106.18)	[b]0.375
	Active	20	91-1691 (219)	380.6 (398.2)	
CDAI	Mild	8	142-475 (257.5)	286.25 (106.01)	[b]0.682
	Severe	17	91-1691 (219)	406.12 (427.01)	

[a] Kruskal Wallis Test, [b] Mann Whitney U Test, * P<0.05

Table 5: Relationship between ESR and CRP and PTX3 levels

		PTX3
ESR	r	-0.181
	P-value	0.139
CRP	R	-0.128
	P-value	0.299

r: Spearman's Correlation Coefficient, * $P<0.05$

Table 6: Evaluation of PTX3 Measurements by CDAI Levels

		PTX3			*P*-value
		n	Min-Max (Median)	Mean (SD)	
CDAI	Mild	8	142-475 (257.5)	286.25 (106.01)	[a]0.682*
	Severe	17	91-1691 (219)	406.12 (427.01)	

[a] Mann Whitney U test

Discussion

PTX3 is an indicator which increases in inflammatory events. Previous studies have shown that plasma PTX3 levels increase in acute myocardial infarction, sepsis, acute pancreatitis, and autoimmune diseases such as rheumatoid arthritis, psoriasis, Churg-Strauss syndrome, Wegener granulomatosis, and microscopic polyangiitis. Unlike CRP, which is another inflammation indicator produced from the liver through IL-6, PTX3 is produced from the inflamed tissue through IL-1, IL-8, TNF-alpha and LPS. It reaches peak and decreases to normal levels in a brief time. PTX3 is normally produced from the lamina propria of colon cells and its secretion from inflammatory tissue increases in intestinal inflammation.

Although there are many studies showing the relationship of PTX3 with inflammation, the number of studies conducted with IBD in the literature is very few and their results are contradictory. In their in vitro studies, Savchenko et al. [9] showed the relationship between neutrophil activation and PTX3 levels in colon tissues of ulcerative colitis patients. Contrary to the results of this study, Chen et al. [1] reported that PTX3 was superior to CRP in showing disease activity in Crohn's disease patients, although they could not detect any relationship between ulcerative colitis and PTX3, like us. In the study of Kato et al. [10], the presence of PTX3 in cryptic abscesses containing neutrophils was also shown. They also found that PTX3 levels correlated with CRP and were associated with the activation of the disease. However, in our study, there was no relationship between PTX3 and neither CRP and ESR nor the activity of ulcerative colitis. In our Crohn's disease group, we think that the reason for lower PTX3 levels was the immunosuppressive treatment our patients received. There was a statistically significant relationship between PTX3 levels only when ulcerative colitis manifested as pancolitis.

Limitation

This study has two potential limitations, one being the small number of patients. The second one is that it is based on data from a single center. Multicenter studies with more IBD patients could yield more valid results.

Conclusions

In the light of all these studies, which are very few in the IBD patient group with PTX3 and contradict with our study, PTX3 blood level is not more sensitive, more specific, or effective in determining the type of disease, and not strong in understanding the disease activity in existing IBD patients. Therefore, PTX3 is not suitable for use in the IBD patient group except for follow-up of ulcerative colitis patients with pancolitis in daily practice, without studies and meta-analyses involving more patients.

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