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IS OXIDATIVE STRESS ASSOCIATED WITH ACTIVATION AND PATHOGENESIS OF INFLAMMATORY BOWEL DISEASE?

DA LI JE OKSIDATIVNI STRES POVEZAN SA AKTIVACIJOM I PATOGENEZOM ZAPALJENSKE BOLESTI CREVA?

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Summary

Background: We aimed to determine the levels of total antioxidant status (TAS), total oxidant status (TOS), oxidative stress index (OSI) and paraoxonase1/arylesterase levels in inflammatory bowel disease (IBD), and the relation between these molecules and the activity index of the disease.
Methods: Eighty IBD patients (ulcerative colitis (UC)/Crohn disease (CD) 40/40) and 80 control group participants were included in the study. Oxidative stress parameters were measured using the colorimetric method. As disease activity indexes, the endoscopic activity index (EAI) was used for UC and the CD activity index (CDAI) was used for CD.
Results: In IBD patients, mean TAS (1.3±0.2 vs 1.9±0.2, respectively; $p<0.001$) and arylesterase (963.9±232.2 vs 1252.9±275, respectively; $p<0.001$) levels were found to be lower and TOS level (5.6±1.6 vs 4.0±1.0, respectively; $p<0.001$) and OSI rate (4.5±1.6 vs 2.2±0.8, respectively; $p<0.001$) were found to be higher compared to the control group. A strong positive correlation was found between EAI and TOS levels ($r=0.948$, $p<0.001$) and OSI rate ($r=0.894$, $p<0.001$) for UC patients. A very strong positive correlation was found between EAI and TOS levels ($r=0.964$, $p<0.001$) and OSI rate ($r=0.917$, $p<0.001$) for CD patients. It was found in a stepwise regression model that C-reactive protein, OSI and arylesterase risk factors were predictors of IBD compared to the control group.

Kratak sadržaj

Uvod: Naš cilj bio je da odredimo nivoe ukupnog antioksidativnog statusa (TAS), ukupnog oksidativnog statusa (TOS), indeks oksidativnog stresa (OSI) i nivoe paraoksonaze 1/arilesteraze u zapaljenskoj bolesti creva (IBD), kao i odnos između ovih molekula i indeksa aktivnosti bolesti.
Metode: U studiju je uključeno 80 pacijenata sa IBD (ulcerozni kolitis (UC) / Kronova bolest (CD) 40/40) i 80 subjekata kao kontrolna grupa. Parametri oksidativnog stresa mereni su kolorimetrijskom metodom. Kao indeksi aktivnosti bolesti, indeks endoskopske aktivnosti (EAI) korišćen je za UC, dok je za CD korišćen indeks aktivnosti CD (CDAI).
Rezultati: Kod obolelih od IBD, nađeno je da su srednji TAS (1,3±0,2 vs 1,9±0,2; p<0,001) i nivoi arilesteraze (963,9±232,2 vs 1252,9±275; p<0,001) bili niži, a nivo TOS (5,6±1,6 vs 4,0±1,0; p<0,001) i stopa OSI 4,5±1,6 vs 2,2±0,8; p<0,001) viši u poređenju s kontrolnom grupom. Otkrivena je snažna pozitivna korelacija između nivoa EAI i TOS (r=0,948, p<0,001) i stope OSI (r=0,894, p<0,001) za obolele od UC. Veoma jaka pozitivna korelacija nađena je između nivoa EAI i TOS (r=0,964, p<0,001) i stope OSI (r=0,917, p<0,001) za obolele od CD. U modelu stepwise regresije uočeno je da su C-reaktivni protein, OSI i faktori rizika za arilesterazu bili prediktori za IBD u poređenju s kontrolnom grupom.

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Conclusions: Increased oxidative stress level in IBD patients and the detection of OSI rate as an independent predictor for disease activity indexes lead to the idea that oxidative stress might be related to the pathogenesis of IBD.

Keywords: Crohn's disease activity index, endoscopic activity index, total antioxidant status, total oxidant status, ulcerative colitis

Zaključak: Povišen nivo oksidativnog stresa kod pacijenata sa IBD i detekcija stope OSI kao nezavisnog prediktora za indekse aktivnosti bolesti navode na ideju da oksidativni stres može biti povezan sa patogenezom IBD.

Ključne reči: indeks aktivnosti Kronove bolesti, indeks endoskopske aktivnosti, ukupni antioksidativni status, ukupni oksidativni status, ulcerozni kolitis

Introduction

Inflammatory bowel disease (IBD) has two major forms: ulcerative colitis (UC) and Crohn's disease (1). UC is a chronic relapsing condition characterized by superficial mucosal ulceration, rectal bleeding, diarrhea, and abdominal pain (2) and CD is characterized by a discontinuous and transmural inflammation potentially affecting any portion of the gastrointestinal tract from the mouth to the anus and presents typically with abdominal pain, fever, bloody or non-bloody diarrhea, and weight loss (3).

Since inflammatory bowel disease is a chronic inflammatory and idiopathic disease, a great deal of factors is blamed in its etiopathogenesis. While the most commonly blamed factors are genetic and environmental risk factors, lately oxidative stress has been considered to be one of the serious risk factors (4, 5). In IBD, excessive immune response due to chronic inflammation and impaired tissue perfusion due to mucosal damage lead to excessive production of reactive oxygen and nitrogen species (ROS/RNS) (6). ROS, generated by inflamed bowel tissue and inflammatory cells, increase oxidative stress and contribute to development and progress of chronic bowel infection (7). In fact, it was shown in recent studies that increased oxidized molecules due to excessive oxidative stress in IBD are correlated with severity of mucosal inflammation (8, 9).

For whatever reason, oxidative stress arises due to disturbed equilibrium between oxidant and antioxidant mechanisms in the body in favor of oxidant radicals (10). Since there is a large number of increased oxidant radicals and decreased antioxidant molecules in the body as a result of oxidative stress, it is very time-consuming and expensive to measure all of these separately and also not really possible because the measurement is open to interaction between substances. For this reason, markers such as total oxidant status (TOS) and total antioxidant status (TAS) have been developed in order to determine the overall oxidant and antioxidant status in the body. Oxidative stress index (OSI) is also used as a general indicator of oxidative stress (11, 12). In addition, anti-inflammatory serum arylesterase/paraoxonase 1 (PON1) enzymes are among the antioxidant molecules that indicate oxidative stress in the body. These molecules are responsible for removing oxidized lipids, also known as arylesterase enzymes, from the body (13).

In our literature review, we have not found a study which investigates the relation between disease activity indexes and overall oxidant and antioxidant status and the arylesterase enzymes, which have a protective role in lipid peroxidation, in subgroups of IBD such as UC and CD. For this reason, we aimed to determine the levels of TAS, TOS and OSI, which indicate overall oxidative status in UC and CD patients, and the arylesterase enzymes, which have a protective role in lipid peroxidation, and the relation between these molecules and the activity index of the disease.

Materials and Methods

Study population

This study was performed between June 2015 and September 2015 in the Gastroenterology Clinic of Turkish High Specialty Training and Research Hospital and General Internal Medicine Clinic of Ankara Numune Training and Research Hospital. Eighty IBD patients (UC/CD: 40/40) and 80 healthy control group participants were included in the study. The IBD group consisted of patients over the age of 18 who were under regular follow-up in the IBD polyclinic and did not use immunosuppressive drugs. The control group participants were selected from healthy individuals with similar demographic characteristics to the patient group and no known chronic disease and medication use, who applied to our hospital for check-up. Participants who have documented cardiovascular and cerebrovascular diseases, inflammatory diseases such as rheumatic, autoimmune and infectious diseases, thyroid dysfunction, liver and kidney failure, vitamin and antioxidant drug use and tobacco and alcohol use were excluded from the study. Clinical and endoscopic parameters were used in order to determine disease activity. The Rachmilewitz scoring system (endoscopic activity index, EAI) was used for UC activity determination (14) and CD activity index (CDAI) was used for CD activity determination (15).

The present study was designed in accordance with the Helsinki Declaration and approved by the local Ethics Board and Research Committee. Written consent was taken from all participants included in the study.

Endoscopic examination

Colonoscopy process was carried out by a single experienced endoscopist using FUJINON XL-4450 (Fujifilm Medical Co., Tokyo, Japan).

Biochemical parameters

Venous blood samples of all participants included in the study were taken after midnight fasting in order to measure oxidative stress markers. The blood samples were centrifuged for ten minutes at 1,500 rpm and then plasma and serum samples were separated and kept at –80 °C. TAS, TOS, PON1 and arylesterase parameters were run on the same serum sample in the same session. Hemogram, biochemistry, C-reactive protein (CRP), erythrocyte sedimentation rate (ESR) levels of participants were obtained from patient files.

Measurement of oxidative stress parameters

Serum TAS level was measured with a commercial kit (Rel Assay Diagnostics, Turkey, REF. No: RL0017, LOT No: JE 14042A) via a colorimetric method. CV%: 10. Linearity: 0–2.75 mmol/L. The results are expressed in mmol Trolox equivalents /L.

Serum TOS level was measured with a commercial kit (Rel Assay Diagnostics, Turkey, REF. No: RL0024, LOT No: JE 14048O g) via a colorimetric method. CV%: 10. Linearity: 0–33.5 μmol/L. The results are expressed in micromolar H_2O_2 equivalents per liter.

Serum PON1 level was measured with a commercial kit (Rel Assay Diagnostics, Turkey, REF. No: RL0031, LOT No: JE14028P) via a colorimetric method. CV%: 5. Linearity: 0–750 U/L. Serum arylesterase level was measured with a commercial kit (Rel Assay Diagnostics, Turkey, REF. No: RL0055, LOT No: JR13017AR) via a colorimetric method. The PON1 and arylesterase results are expressed in terms of U/L.

The ratio of TOS to TAS provided the OSI, an indicator of the degree of oxidative stress (16).

Statistical analysis

The Statistical Package for Social Sciences (SPSS) for Windows 20 (IBM SPSS Inc., Chicago, USA) program was used for statistical assessments. Kolmogorov-Smirnov test was utilized to determine the distribution of data. Continuous variables with normal distribution were expressed as mean ± standard deviation, and continuous variables without normal distribution were expressed as median [interquartile range (IQR)]. Categorical variables were presented as numbers and percentage. Continuous variables were compared with independent sample t-test, ANOVA or Mann Whitney U test, Kruskall Wallis H test where appropriate. The relationship between the numeric parameters was analyzed by Pearson and Spearman correlation analysis. Stepwise multi-variable logistic regression analysis was used in order to determine independent predictors of IBD risk. Stepwise multivariable linear regression analysis was used in order to determine independent predictors of EAI and CDAI levels. Logarithmic conversion was applied to CDAI, EAI, CRP, ESR levels that did not meet the normal distribution condition in linear regression analysis. A $p<0.05$ was considered significant for statistical analyses (17).

Results

Table I summarizes the demographic characteristics and laboratory findings of the study population. The research population consisted of 80 IBD (UC: 40 patients, CD: 40 patients) and 80 control group participants. There was no significant difference between the groups included in the study in terms of age, gender and body mass index. The mean albumin level was lower in the IBD group compared to the control group (4.1±0.6 g/L vs 4.3±0.3 g/L, respectively; $p=0.029$). The median ESR and CRP levels were higher in the IBD group compared to the control group (19 mm/h vs 10 mm/h; $p<0.001$, 5 mg/L vs 2 mg/L; $p<0.001$; respectively). In IBD patients, mean TAS (1.3±0.2 mmol Trolox equivalent/L vs 1.9±0.2 mmol Trolox equivalent/L, respectively; $p<0.001$) and arylesterase (963.9±232.2 U/L vs 1252.9±275 U/L, respectively; $p<0.001$) levels were found to be lower and TOS level (5.6±1.6 lmol H_2O_2 Eq/L vs 4.0±1.0 lmol H_2O_2 Eq/L, respectively; $p<0.001$) and OSI rate (4.5±1.6 vs 2.2±0.8, respectively; $p<0.001$) were found to be higher compared to the control group.

Subgroup analyses are given in detail in *Table II*. Accordingly there was no significant difference in terms of age, BMI and gender distribution. The mean albumin level was lower in CD patients and similar in UC patients and the control group (4.0±0.7 g/L vs 4.3±0.5 g/L vs 4.3±0.3 g/L, respectively; $p<0.05$). The CD group had the highest median ESR and CRP level, while these levels were higher in the UC group compared to the control group (19 mm/h vs 13 mm/h vs 10 mm/h; $p<0.05$; 7.2 vs 3.6 vs 2; $p<0.05$; respectively). The mean TAS and arylesterase levels were similar in CD and UC patients and lower than in the control group, whereas TOS level and OSI rate were similar in CD and UC patients and higher than in the control group.

No parameters related to oxidative stress levels were found in the control group. In the IBD group, there was a negative correlation between CRP levels and TAS level ($r=-0.315$, $p=0.019$) and a positive

Table I Demographic characteristics and laboratory findings of the study population.

Variables	IBD	Control	p
	n (80)	n (80)	
Gender (female), n (%)	52 (65)	55 (68.8)	0.737
Age (years)	44.9±12.6	45.7±13.5	0.712
BMI (kg/m^2)	26±4.7	27.1±4.5	0.122
Total protein (g/L)	7.4±0.7	7.5±0.5	0.404
Albumin (g/L)	4.1±0.6	4.3±0.3	0.029*
ESR (mm/h)	19 (22)	10 (8.5)	<0.001*
CRP (mg/L)	5 (7.6)	2 (4)	<0.001*
TAS (mmol Trolox equivalent/L)	1.3±0.2	1.9±0.2	<0.001*
TOS (Imol H_2O_2 Eq/L)	5.6±1.6	4.0±1.0	<0.001*
OSI (arbitrary unit)	4.5±1.6	2.2±0.8	<0.001*
PON1 (U/L)	121.3 (94.5)	122.5 (144)	0.118
Arylesterase (U/L)	963.9±232.2	1252.9±275	<0.001*

*p<0.05 statistical significance

Abbreviations: IBD: inflammatory bowel disease, BMI: body mass index, ESR: erythrocyte sedimentation rate, CRP: C-reactive protein, TAS: total antioxidant status, TOS: total oxidant status, OSI: oxidative stress index, PON1: paraoxonase 1.

Table II Demographic characteristics and laboratory parametres of the study population.

Variables	CD	UC	Control	p^a	p^b	p^c
	n (40)	*n* (40)	*n* (80)			
Gender (female), *n*(%)	25 (62.5)	27 (67.5)	55 (68.8)	0.815	0.541	0.907
Age (years)	43.4±13.5	46.5±11.5	45.7±13.5	0.277	0.382	0.757
BMI (kg/m^2)	25.5±5	26.5±4.4	27.1±4.5	0.383	0.080	0.443
Total protein (g/L)	7.3±0.8	7.4±0.7	7.5±0.5	0.399	0.188	0.915
Albumin (g/L)	4.0±0.7	4.3±0.5	4.3±0.3	0.038*	0.002*	0.443
ESR (mm/h)	19 (22)	13 (17.5)	10 (8.5)	0.045*	<0.001*	0.078
CRP (mg/L)	7.2 (12.4)	3.6 (4.3)	2 (4)	0.035*	<0.001*	0.006*
EAI	-	7±1.9	-	-	-	-
CDAI	263.8±80.8	-	-	-	-	-
TAS (mmol Trolox equivalent/L)	1.3±0.2	1.3±0.2	1.9±0.2	0.586	<0.001*	<0.001*
TOS (Imol H_2O_2 Eq/L)	5.8±1.9	5.5±1.4	4.0±1.0	0.419	<0.001*	<0.001*
OSI (arbitrary unit)	4.5±1.6	4.5±1.6	2.2±0.8	0.926	<0.001*	<0.001*
PON (U/L)	128.6 (95)	118.4 (111.3)	122.5 (144)	0.912	0.108	0.348
Arylesterase (U/L)	994.5±213.6	933.4±248.4	1252.9±275	0.242	<0.001*	<0.001*

p^a: CD vs UC, p^b: CD vs Control, p^c: UC vs Control *p<0.05 statistical significance

Abbreviations: IBD: inflammatory bowel disease, BMI: body mass index, ESR: erythrocyte sedimentation rate, CRP: C-reactive protein, EAI: endoscopic activity index, CDAI: Crohn disease activity index, TAS: total antioxidant status, TOS: total oxidant status, OSI: oxidative stress index, PON1: paraoxonase 1.

Table III Findings related to oxidative stress parameters in the patient group and the control group.

Groups	Variables	TAS		TOS		OSI		PON1		Arylesterase	
		r	*p*	*r*	*p*	*r*	*p*	*r*	*p*	*r*	*p*
CD	Age	-0.023	0.887	0.189	0.243	0.160	0.323	0.205	0.205	0.271	0.090
	BMI	0.038	0.816	0.040	0.809	-0.014	0.933	-0.001	0.993	-0.074	0.651
	ESR	-0.198	0.220	0.170	0.296	0.256	0.111	0.399	0.011*	0.112	0.492
	CRP	-0.032	0.843	0.034	0.837	0.385	0.012*	0.265	0.099	-0.037	0.823
	CDAİ	-0.129	0.427	0.962	<0.001*	0.867	<0.001*	0.163	0.316	-0.225	0.163
UC	Age	-0.003	0.983	-0.296	0.064	-0.218	0.176	-0.266	0.097	-0.059	0.717
	BMI	-0.358	0.023*	-0.108	0.508	0.039	0.812	0.000	0.998	0.114	0.483
	ESR	-0.211	0.190	-0.015	0.927	0.005	0.974	-0.297	0.062	0.006	0.972
	CRP	-0.015	0.927	-0.097	0.553	0.395	0.010*	-0.087	0.593	0.026	0.873
	EA	-0.142	0.381	0.925	<0.001*	0.827	<0.001*	0.067	0.682	0.067	0.680
Control	Age	-0.200	0.139	-0.045	0.692	0.046	0.683	-0.033	0.769	-0.006	0.960
	BMI	-0.126	0.266	-0.069	0.544	0.046	0.684	-0.039	0.730	0.093	0.411
	ESR	-0.006	0.955	-0.126	0.265	-0.126	0.264	-0.082	0.469	-0.023	0.837
	CRP	0.173	0.125	-0.060	0.598	-0.157	0.164	-0.015	0.893	0.182	0.106

*p<0.05 statistical significance

Abbreviations: TAS: total antioxidant status, TOS: total oxidant status, OSI: oxidative stress index, PON1: paraoxonase 1, CD: Crohn disease, UC: ulcerative colitis, BMI: body mass index, ESR: erythrocyte sedimentation rate, CRP: C-reactive protein, CDAI: Crohn disease activity index, EAI: endoscopic activity index.

Table IV Regression analysis and independent risk factors predicting IBD and disease activity indexes.

Variables	OR	95% C.I.		*p*
		Lower	Upper	
IBD*	1.071	1.031	1.132	0.016
CRP	0.995	0.992	0.997	<0.001*
OSI	4.596	2.740	7.708	<0.001*
Arylesterase				
Nagelkerke R^2=0.728. *p*<0.001				
Variables	B±SE	95% C.I.		*p*
		Lower	Upper	
Log (CDAI)†				
OSI	0.710±0.060	0.590	0.830	<0.001*
R^2=0.684. *p*<0.001				
Log (EAI)†				
OSI	0.600±0.070	0.470	0.740	<0.001*
R^2=0.675; *p*<0.001				

Φ: Age, gender, BMI, and laboratory findings were included in the stepwise logistic regression model.
†: Age, gender, BMI, and laboratory findings were included in the linear regression model.
Logarithmic conversion was applied to CDAI, EAI, CRP, ESR levels.
TOS and TAS were excluded from the model due to their high correlation with OSI (multicolinearity)

*p<0.05 statistical significance
Abbreviations: IBD: inflammatory bowel disease, TAS: total antioxidant status, TOS: total oxidant status, OSI: oxidative stress index, CD: Crohn disease, UC: ulcerative colitis, BMI: body mass index, ESR: erythrocyte sedimentation rate, CRP: C-reactive protein, CDAI: Crohn disease activity index, EAI: endoscopic activity index.

correlation between TOS level (r=0.324, p=0.022) and OSI rate (r=0.405, p=0.006). A very strong positive correlation was found between EAI level and TOS level (r=0.964, p<0.001) and OSI rate (r=0.917, p<0.001) for CD patients. In addition, a positive correlation was found between CRP level and OSI rate (r=0.385; p=0.012). A very strong positive correlation was found between EAI level and TOS level (r=0.948, p<0.001) and OSI rate (r=0.894, p<0.001) for UC patients. In addition, a positive correlation was found between CRP level and OSI rate (r=0.395; p=0.010) and between ESR level and PON1 (r=0.399, p=0.011) in CD patients. A negative correlation was found between BMI level and TAS (r=-0.358, p=0.023) in UC patients (*Table III*).

In a stepwise regression model formed using age, gender, BMI, total protein, albumin, ESR, CRP, OSI, PON1 and arylesterase risk factors, CRP (OR=1.071; p=0.016), OSI (OR=4.596; p=0.007) and arylesterase (OR=0.995, p<0.001) risk factors were found to be predictors of IBD compared to the control group (*Table IV*).

Risk factors included in the logistic regression model were also used in the linear regression model employed to determine risk factors that predict log (CDAI) and log (17) levels. Accordingly, it was found that an increase of 1 unit in OSI rate increased log (CDAI) level by 0.710 (B ± SE=0.710 ± 0.060; p<0.001), and the same increase of 1 unit increased log level by 0.600 (B ± SE=0.600 ± 0.070; p<0.001). Both the logistic regression model and the linear regression model explained more than 65% of the dependent variable (Nagelkerke R^2=0.728, $R^2_{Log\ (CDAI)}$=0.684, $R^2_{Log\ (17)}$=0.675) (*Table IV*).

Discussion

In the present study, we found overall oxidative stress to be higher and arylesterase enzyme level to be lower in IBD patients compared to the control group. A positive correlation was found between EAI and OSI rate in the UC group and between CDAI and OSI in the CD group. It was determined as a result of the stepwise logistic regression analysis that OSI rate was an independent risk factor for IBD. To the best of our knowledge, this is the first study that attempts to determine the overall oxidative stress level in major IBD subgroups such as UC and CD and investigate the relation between oxidative stress and disease activity.

Oxidative stress arises as a result of excessive generation of oxidant radicals or lack of molecules involved in antioxidant defense. Proinflammatory T lymphocytes are known to play an active role in IBD, which is a chronic inflammatory disease (18). These proinflammatory cytokines may cause an increase in ROS via the nicotinamide adenine dinucleotide phosphate-oxidase enzyme (19, 20). Excessively increased ROS levels lead to changes in the structure of lipids, proteins, carbohydrates and deoxyribonucleic acids in tissues and cause oxidative damage (21). The second group of free radicals produced in IBD is RNS which is produced by selected cells in intestinal mucosa and submucosa via nitric oxide synthase (22, 23). Inducible NOS (22) level increases in inflamed tissue in diseases such as IBD. Increased iNOS levels cause activated macrophages, leukocytes and epithelial cells in intestinal mucosa to produce excessive amounts of RNS (24). As noted above, an increase in both ROS and RNS may be present in intestinal mucosa secondary to inflammation in IBD.

In the present study, we found the oxidative stress level to be higher in IBD patients compared to the control group. As mentioned above, we believe that this is associated with increased inflammation in IBD patients, because CRP levels were found to be higher in both the whole patient population and UC and CD compared to the control group. The fact that a positive correlation was found between CRP levels and OSI rate in UC and CD patients as a result of correlation analysis strongly supports our hypothesis. In a study conducted with CD patients, Sido et al. found that oxidative stress level was higher in inflamed ileum mucosa compared to non-inflamed tissue (25). It was found in other studies as well that glutathione (GSH) levels decreased and glutathione disulphide (GSSG) levels increased in inflamed tissues in UC and CD patients (4, 8, 9). Another strong piece of evidence which supports our hypothesis is that low GSH/GSSG redox status in intestinal mucosa was shown to be associated with chronic inflammation in a study conducted with rats with UC (26).

A strong positive correlation was found between EAI and OSI rate in the UC group and between CDAI and OSI in the CD group in our study. This result leads to the idea that oxidative stress is markedly high in IBD activation and increased oxidative stress might be an indirect indicator of disease activation. The fact that OSI was found to be an independent risk factor for both EAI and CDAI as a result of the linear regression analysis strongly supports our idea. It was found in a study conducted by Kruidenier et al. (27) that there was a correlation between ROS and disease severity both clinically and endoscopically in IBD patients, which supports our hypothesis as well.

Arylesterase enzyme level, which is an antioxidant enzyme, was found to be lower in IBD patients compared to the control group in our study. Arylesterase enzyme is a serum enzyme which protects low density lipoprotein against oxidative damage and is firmly bound to high density lipoprotein. We believe that arylesterase enzyme levels decreased due to high consumption in case of increased oxidative stress in IBD patients, since it has a role in antioxidant defense (13).

Our main limitations are the cross-sectional design of the study and lack of sufficient observation,

follow-up and recurrent measurements necessary to use OSI rate as a disease activity marker.

In conclusion, increased oxidative stress level in IBD patients and the detection of OSI rate as an independent predictor for disease activity indexes due to their positive correlation lead to the idea that oxidative stress might be related to the pathogenesis of IBD. In previous studies, it was shown that antioxidant therapies (oral or intraperitoneal GSH and N-acetylcysteine supplement) decreased oxidative stress level and repaired bowel mucosa structurally and histologically at the same time, which supports our idea (28–30). The fact that OSI rate was found to be among the independent predictors of IBD together with chronic inflammation markers as a result of stepwise logistic analysis enhances the hypothesis that oxidative stress might be related to the etiopathogenesis of IBD.

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Conflict of interest statement

The authors stated that they have no conflicts of interest regarding the publication of this article.

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