Relation of Antioxidant Native Thiol Level with Inflammatory Markers and Disease Activity Index in Pediatric Ulcerative Colitis

Pediatrik Ülseratif Kolit Hastalarında Antioksidan Native Tiyol Düzeyinin İnflmatauvar Markırlar ve Hastalık Aktivite İndeksi ile İlişkisi

Selim DERECİ[1], Fatih DURAN[1], Salim NEŞELİOĞLU[2], Burcu HIDIMOĞLU[1], Duygu İSKENDER MAZMAN[1], Aysel ÜNLÜSOY AKSU[1], Arzu Meltem DEMİR[1], Özcan EREL[2], Şamil HIZLI[1,3]

[1]Department of Pediatric Gastroenterology, Bilkent City Hospital, Ankara, Turkey
[2]Department of Medical Biochemistry, Ankara Yildirim Beyazit University, Ankara, Turkey
[3]Department of Pediatric Gastroenterology, Ankara Yildirim Beyazit University, Ankara, Turkey

ABSTRACT

Objective: Pediatric ulcerative colitis (PUC) is an inflammatory disease. PUC pathogenesis is associated with an imbalance between reactive oxygen species and antioxidant activity which creates oxidative stress. Native thiol (NT) level is antioxidant capacity which is practical and repeatable marker of inflammation and antioxidan level. We aimed to analyse the relation of NT level with inflammatory markers and pediatric ulcerative colitis activity index (PUCAI).

Material and Methods: Thirty-eight PUC patients (SG) and 33 control group (CG) participants were included in the study. PUC patients grouped as in remission, mild, moderate and severe activity according to disease activity according to PUCAI. NT, hemoglobin (Hb), white blood cell (WBC), platelet (PLT), mean platelet volume (MPV), albumin, C-reactive protein (CRP), erythrocyte sedimentation rate (ESR), interleukin 6 (IL-6) levels of all participants were measured at the time they were enrolled in the study and recorded.

Results: Mean age and gender ratio of groups were similar (p>0.050). NT level of SG statistically high compared to CG (p=0.001). Hb, PLT, MPV, CRP, ESR, IL-6 levels of SG were statistically different than CG (p=0.045, p=0.026, p=0.001, p=0.001, p=0.001, p=0.001). NT level of SG was positively correlated with Hb, MPV, albumin (p=0.001, for all). NT level of SG was negatively correlated with PLT, CRP, ESR, IL-6 and PUCAI (p=0.001).

Conclusion: NT level of PUC is significantly lower than CG. NT level of SG was positively correlated with albumin which is a good prognostic factor in PUC patients. NT may be repeatable, noninvasive candidate serum biomarker for PUC management.

Key Words: Activity, Antioxidant, Children, Thiol, Ulcerative colitis

0000-0001-8689-3783 : DERECİ S
0000-0001-6436-1030 : DURAN F
0000-0002-0974-5717 : NEŞELİOĞLU S
0000-0002-3498-2781 : HIDIMLIOĞLU B
0000-0001-9244-5792 : İSKENDER MAZMAN D
0000-0002-4550-0717 : ÜNLÜSOY AKSU A
0000-0002-9176-9100 : DEMİR A
0000-0002-2996-3236 : EREL Ö
0000-0001-6732-493X : HIZLI Ş

Conflict of Interest / *Çıkar Çatışması*: On behalf of all authors, the corresponding author states that there is no conflict of interest.

Ethics Committee Approval / *Etik Kurul Onayı*: This study was conducted in accordance with the Helsinki Declaration Principles. This study was approved by Ankara Bilkent City Hospital Second Ethical committee (10.10.2021/E2-20-106).

Contribution of the Authors / Yazarların katkısı*: *DERECİ S: Constructing the hypothesis or idea of research and/or article, Planning methodology to reach the Conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in necessary literature review for the study, Taking responsibility in the writing of the whole or important parts of the study, Reviewing the article before submission scientifically besides spelling and grammar. ***DURAN F:*** Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments. ***NEŞELİOĞLU S:*** Organizing, supervising the course of progress and taking the responsibility of the research/study. ***HIDIMLIOĞLU B:*** Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments. ***İSKENDER MAZMAN D:*** Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the eperiments. ***ÜNLÜSOY AKSU A:*** Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments. ***DEMİR A:*** Taking responsibility in patient follow-up, collection of relevant biological materials, data management and reporting, execution of the experiments. ***EREL Ö:*** Taking responsibility in logical interpretation and conclusion of the results. ***HIZLI Ş:*** Planning methodology to reach the Conclusions, Organizing, supervising the course of progress and taking the responsibility of the research/study, Taking responsibility in logical interpretation and conclusion of the results, Taking responsibility in necessary literature review for the study, Taking responsibility in the writing of the whole or important parts of the study, Reviewing the article before submission scientifically besides spelling and grammar.

How to cite / *Atıf yazım şekli*: Dereci S, Duran F, Neşelioğlu S, Hıdımlıoğlu B, İskender Mazman D, Ünlüsoy Aksu A, et al. Relation of Antioxidant Native Thiol Level with Inflammatory Markers and Disease Activity Index in Pediatric Ulcerative Colitis. Turkish J Pediatr Dis 2023;17:309-314.

Correspondence Address / Yazışma Adresi:
Selim DERECİ
Department of Pediatric Gastroenterology,
Bilkent City Hospital, Ankara,Turkey
E-posta: dereciselim@hotmail.com

Received / Geliş tarihi : 05.03.2023
Accepted / Kabul tarihi : 11.04.2023
Online published : 04.05.2023
Elektronik yayın tarihi
DOI: 10.12956/tchd.1260388

ÖZ

Amaç: Pediatrik ülseratif kolit (PÜK) inflamatuar bir hastalıktır. PÜK patogenezinde oksidatif stres oluşturan antioksidan aktivite ile reaktif oksijen türleri arasındaki dengesizlik ile ilişkilidir. Native tiyol (NT) seviyesi, iltihaplanma ve antioksidan seviyesinin pratik ve tekrarlanabilir belirteci olan antioksidan kapasitedir. NT düzeyinin inflamatuar belirteçler ve pediatrik ülseratif kolit aktivite indeksi (PÜKAI) ile ilişkisini incelemeyi amaçladık.

Gereç ve Yöntemler: Çalışmaya 38 PÜK hastası (ÇG) ve 33 kontrol grubu (KG) katılımcısı dahil edildi. PÜK hastaları, PUKAI'ye göre hastalık aktivitesi remisyonda, hafif, orta ve şiddetli olarak gruplandı. Tüm katılımcıların NT, hemoglobin (Hb), beyaz kan hücresi (WBC), trombosit (PLT), ortalama trombosit hacmi (MPV), albümin, C-reaktif protein (CRP), eritrosit sedimantasyon hızı (ESH), interlökin 6 (IL-6) seviyeleri ölçüldü ve kaydedildi.

Bulgular: Grupların ortalama yaş ve cinsiyet oranları benzerdi (p>0.050). ÇG'nin NT düzeyi, KG'ye göre istatistiksel olarak yüksek (p=0.001). ÇG'nin Hb, PLT, MPV, CRP, ESR, IL-6 düzeyleri istatistiksel olarak KG'den farklıydı (p=0.045, p=0.026, p=0.001, p=0.001, p=0.001, p=0.001). ÇG'nin NT düzeyi Hb, MPV, albümin ile pozitif korelasyon gösterdi (p=0.001). ÇG'nin NT düzeyi PLT, CRP, ESR, IL-6 ve PUCAI ile negatif korelasyon gösterdi (p=0.001).

Sonuç: PÜK hastalarında NT seviyesi, KG'den anlamlı olarak düşük bulundu. ÇG'nin NT düzeyi PÜK hastalarında iyi bir prognostik faktör olan albümin ile pozitif korelasyon gösterdi. NT ölçümü PÜK yönetimi için tekrarlanabilir, noninvaziv bir serum biyobelirteç olabilir.

Anahtar Sözcükler: Aktivite, Antioksidan, Çocuk, Native tiyol, Ülseratif kolit

INTRODUCTION

Pediatric ulcerative colitis (PUC) is a chronic relapsing systemic idiopathic disease. Most of the findings of PUC are similar to an adult-onset disease. PUC incidence and prevalence rate tends to increase worldwide and up to one-fourth of cases have a more severe disease course and longer duration than inflammatory bowel disease (IBD) diagnosed in adulthood. It is widely known that pathogenesis and progression of the inflammatory cascade in this disease are often attributed to genetic, environmental factors (1,2). For successful treatment of PUC, early diagnosis and follow disease activity is important but there is no well established ideal serum biomarker for PUC, yet.

Lately oxidative stress has been considered to be one of the important steps in disease pathogenesis (3,4). In ulcerative colitis (UC), 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) and amount of these oxidative stress markers in UC were found as correlated with the severity of mucosal inflammation (5-7).

In healthy state, there is equilibrium between oxidant and antioxidant mechanisms in the body. Thiols are organic antioxidants since they possess a sulfhydryl group. Thiol groups can form reversible disulfide bridges by the effect of oxidants in plasma (6). The formed disulfide bridges can be reduced to thiol groups again via the antioxidant systems in the organism. Thus, dynamic native thiol-disulfide balance is maintained. The thiol-disulfide balance plays a critical role in antioxidant defense, detoxification, apoptosis, regulation of enzyme activities, and mechanisms of transcription and cellular signal transduction, proliferation and immunity (6,7). In inflammatory diseases, oxidant radicals increases due to oxidative stress and this equilibrium is disturbed (6). To measure levels of all oxidant radicals and antioxidant molecules separately is very time-consuming and expensive and also not really possible since interaction of these substances is always ongoing during all the time in the body (3,4,6).

Native thiol (NT) is the main element of the antioxidant defense and measurement of serum NT level is a good indicator of total amount of antioxidant capacity (6-9). For this reason, a new method was developed to determine native thiol-disulfide balance. Native thiol balances oxidative stress by reducing the levels of reactive oxygen species or by accelerating their inactivation (6,8-10).

We propose that, NT level may be a possible candidate biomarker for PUC diagnosis and follow up. The aim of the study was to determine the relation between NT levels and widely used inflammatory markers and pediatric ulcerative colitis activity index (PUCAI).

MATERIAL and METHODS

This study was carried out in the Pediatric Gastroenterology, Hepatology and Nutrition Clinic of Ankara Bilkent City Hospital, between October 2021 and March 2022. This study was approved by Ankara Bilkent City Hospital Second Ethical committee (10.10.2021/E2-20-106). The written informed consent was received from the family for every child who were included into the study.

This prospective study included two groups; the study group (SG) consisted of 38 children (male 19, female 19) diagnosed and treated with a diagnosis of PUC, and the control group (CG) consisted of 33 healthy children (male 16, female 17) who were admitted for a routine check-up. The diagnosis of UC was made according to clinical, endoscopic, and histopathological criteria of ESPGHAN (11). PUC patients were grouped as in remission, mild, moderate, and severe activity according

to PUCAI score; a non-invasive multi item UC activity index to differentiate the disease activity states accurately and to assess change over time, without the need for colonoscopic assessment for pediatric patients accepted as objective by clinical authorities (12). Patients with any complications related to UC and other chronic or systemic disease and patients with any infection were excluded from the study.

Results of NT, hemoglobin (Hb), white blood cell (WBC), platelet (PLT), mean platelet volume (MPV), albumin, C-reactive protein (CRP), erythrocyte sedimentation rate (ESR), interleukin 6 (IL-6) levels of all participants were measured at the time they were enrolled in the study and recorded. Blood samples were taken to measure NT levels with a newly developed method by Erel/ Neselioglu and NT levels were measured using an automated clinical chemistry analyzer (Roche, Cobas 501, Mannheim, Germany) (8).

Hemoglobin, WBC and PLT were measured by an automatic analyzer (Sysmex XE-2100, USA) within five minutes of sampling. C-reactive protein (Siemens BN II System, Germany), ESR (Therma NE, Spain), and albumin (Roche Cobas 8000, Mannheim, Germany) levels were measured using commercial kits. IL-6 levels were measured by using an enzyme-linked immunosorbent assay kit (Affymetrix Ebioscience).

Statistical Analysis

Statistical Package for Social Sciences (SPSS) 22 for Windows (IBM SPSS Inc.,Chicago, USA) program was used for statistical analyses. Kolmogorov Smirnov test was used to determine the distribution of data. Categorical variables were expressed as numbers and percentage. Continuous variables were compared with independent sample t-test, ANOVA, Mann Whitney U test, and Kruskal-Wallis test where appropriate. Post- Hoc analysis was implemented within subgroups, including PUCAI score, NT level, MPV, PLT, IL-6. Chi-square test and Fisher's exact chi-square test were used to compare categorical variables. The relationship between the numeric parameters was analyzed by Pearson and Spearman correlation analysis. A $p<0.05$ was considered significant for statistical analyses.

RESULTS

During the study in 15 months, a total of 71 children were included. The demographic characteristics and laboratory results of the two groups are summarized in Table I. There were 38 UC patients; 19 (50 %) male and 19 (50 %) girls in SG. CG was consisted of 33 healthy children, 17 (51.5 %) girls, and 16 (48.5 %) boys. The mean ages ± SD were 13.6±3.3 years in SG and 13.3±4.1 years in CG. No statistically significant difference was observed between the SG and CG with respect to age, and gender ratio ($p> 0.050$). NT, Hb, MPV, albumin levels were significantly lower in SG compared to CG ($p=0.001$, $p=0.045$, $p=0.001$, $p=0.015$, respectively). PLT, CRP, ESR, IL-6 were significantly higher in SG compared to CG ($p=0.026$, $p=0.001$, $p=0.001$, $p=0.001$, respectively) (Table I).

Table I: Demographic and laboratory findings of Study and Control groups

	SG (mean±SD) n=38	CG (mean±SD) n=33	p
Gender (male)*	19 (50)	16 (48.5)	0.899
Age (yr)	13.6±3.3	13.3±4.1	0.351
Hb (g/dL)	12.2±1.97	13±1.4	0.045
WBC (x10^9/L)	7.18±2.23	6.96±1.65	0.602
PLT (x10^9/L)	380.9±172.4	309.4±57.3	0.026
MPV (fL)	7.9±1.1	8±1.4	0.001
Albumin g/L)	44.3±5.2	46.9±3.2	0.015
Native Thiol (µmol/l)	489±93	592.7±80.3	0.001
CRP (g/L)	8.97±10.96	1.93±1.4	0.001
ESR (mm/hour)	12.1±6.8	6.2±3.4	0.001
IL-6 (pg/mL)	8.8±8.3	3.9±1.8	0.001

***:** *n (%),* ***SG:** *Study Group,* ***CG:** *Control Group,* ***Hb:** *Hemoglobin,* ***WBC:** *White Blood Cell,* ***MPV:** *Mean Platelet Volume,* ***PLT:** *Platelet,* ***CRP:** *C-reactive protein,* ***ESR:** *Erythrocyte Sedimentation Rate,* ***IL-6:** *Interleukin 6*

In order to determine the relation between the inflammatory parameters with NT levels of PUC patients with different disease activity level, SG subdivided into groups as remission, mild attack, moderate or severe attack groups that defined according to PUCAI score. But since the number of patients in severe attack group was low, we combined moderate and severe attack cases as one group in order to make comparisons. Mean NT level in subgroups were all different than CG (remission vs control; $p=0.038$, mild vs control; $p=0.002$, moderate-severe vs control; $p=0.001$, respectively) (Table II and Figure I).

Results of correlation analysis of NT and PUCAI, Hb, WBC, PLT, MPV, albumin, CRP, ESR, IL-6 were summarized in Table III. There was a significant correlation between NT level with PUCAI and albumin level ($r=-0.336$, $p=0.039$; $r= 0.573$, $p=0.001$, respectively). Results of correlation analysis of disease severity and NT, Hb, WBC, PLT, MPV, albumin, CRP, ESR, IL-6 were summarized in Table IV.

DISCUSSION

To the best of our knowledge, this is the first case-control prospective study that investigated NT as a novel repeatable and practical biomarker of oxidative capacity in PUC patients. In our study, we demonstrated that the mean level of NT which is reflecting the antioxidant capacity of the body is significantly lower in SG as compared to CG. Our study results showed that, NT level gives fairly precise information about antioxidant capacity of the PUC patients. Measurement of all antioxidant molecules separately is very time-consuming and expensive and need some invasive techniques like biopsy. In a study

Table II: The Relationship Between Disease Severity and Inflammatory Markers

	Control mean±SD (n=33)	Remission mean±SD (n=10)	Mild mean±SD (n=18)	Moderate-Severe mean±SD (n=10)	ANOVA	Post hoc p	
Native Thiol (µmol/l)	592.7±80.3	506.3±120.9	497.9±77.9	462.6±88.7	0.001	p* p† p‡	0.00 0.002 0.000
IL-6 (pg/mL)	3.9±1.83	6.19±5.7	9.04±6.98	11.02±11.99	0.004	p* p† p‡	NS 0.029 0.003
ESR (mm/hour)	6.24±3.44	10.4±4.24	11.11±7.56	15.5±6.75	0.001	p* p† p‡	NS 0.015 0.000
Albumin (g/L)	46.92±3.2	45.2±6.43	44.76.±4.4	42.67±5.4	0.051	p* p† p‡	NS NS 0.045
CRP (g/L)	1.93±1.39	8.6±9.62	7.27±9.79	12.4±14.1	0.002	p* p† p‡	NS NS 0.003
PLT (x10^9/L)	309.4±57.3	420±130.6	330.5±123.7	432.7±257.8	0.019	p* p† p‡	NS NS 0.049
MPV (fL)	8.7±1.4	8.2±1.5	7.97±0.9	7.5±0.71	0.044	p* p† p‡	NS NS 0.051

IL-6: *Interleukin 6,* ***ESR:*** *Erythrocyte Sedimentation Rate,* ***CRP:*** *C-reactive protein,* ***PLT:*** *Platelet,* ***MPV:*** *Mean Platelet Volume,* ***NS:*** *Not Significant,* ****:*** *Remission vs control,* ***†:*** *Mild vs control,* ***‡:*** *Moderate-severe vs control*

Table III: Correlation of Native Thiol and Laboratory Parameters

	r	p
Hb (g/dL)	0.538	0.001
WBC (x10^9/L)	-0.177	0.139
PLT (x10^9/L)	-0.468	0.001
MPV (fL)	0.408	0.001
Albumin (g/L)	0.573	0.001
CRP (g/L)	-0.566	0.001
ESH (mm/hour)	-0.414	0.001
IL-6 (pg/mL)	-0.504	0.001

Hb: *Hemoglobin,* ***WBC:*** *White Blood Cell,* ***MPV:*** *Mean Platelet Volume,* ***CRP:*** *C-reactive protein,* ***PLT:*** *Platelet,* ***ESR:*** *Erythrocyte Sedimentation Rate,* ***IL-6:*** *Interleukin-6*

Table IV: Correlation of pediatric ulcerative colitis severity and laboratory parameters

	r	p
Hb (g/dL)	-0.051	0.76
WBC (x10^9/L)	-0.047	0.778
PLT (x10^9/L)	0.056	0.737
MPV (fL)	-0.254	0.124
Albumin (g/L)	-0.383	0.017
Native Thiol (µmol/l)	-0.336	0.039
CRP (g/L)	0.078	0.640
ESR (mm/hour)	0.282	0.087
IL-6 (pg/mL)	0.037	0.824

Hb: *Hemoglobin,* ***WBC:*** *White Blood Cell,* ***PLT:*** *Platelet,* ***MPV:*** *Mean Platelet Volume,* ***CRP:*** *C-reactive protein,* ***ESR:*** *Erythrocyte Sedimentation Rate,* ***IL-6:*** *Interleukin-6*

done in biopsy material, Holmes and Tsunada et al. (13,14) demonstrated that oxidized glutathione (GSSG) in inflamed mucosa from patients with active UC was increased. Holmes et al. (13) also found that the tissue GSSG content of the mucosa showed a significant positive correlation with clinical and histological indices of disease severity among UC patients. NT is used to maintain reduced glutathione level in the body. Our study reported that there is a significant decrease in NT level in PUC patients. Our study results support the results of biopsy study by Holmes et al. (13) but our technique is superior since there is no need for tissue biopsy. An adult study using our technique by Erel, demonstrated that there was a significant decrease in antioxidant capacity reflected by a decrease in NT level (15).

To our knowledge, there is only one pediatric study reported that active PUC patients and controls which are composed of children with functional bowel disease had similar antioxidant levels in gastrointestinal tract tissue biopsies. They had used a different, manual immunodiagnostic technique with 4% intraassay variability, and also the control group was not healthy children (10). Although measurement of the level of GSSG in tissue needs some special techniques and tissue biopsy,

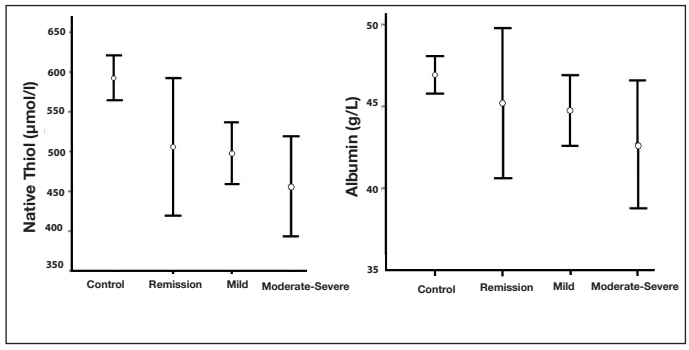

Figure I: Relation of Native Thiol Level and Albumin with Disease Activity.

measurement of serum NT level is a practical, repeatable and automated technique and could be used in PUC patients easily.

Our study results also demonstrated that there is a weak but statistically significant negative correlation between the activity of disease as scaled by PUCAI and NT (r=-0.336, p=0.039). Our study also demonstrated that NT level was also positively correlated with albumin; (r= 0.573, p=0.001). NT level of remission group was significantly lower than CG, NT level of mild activity group significantly lower than control group, means that NT level measurement is fairly precise enough to differentiate these groups (p=0.038).

Our results showed that there is also a statistically significant correlation between NT level and Hb, CRP which reflects an indirect confirmation of the NT level value in this patient group.

Our results suggested that NT level measurement by the Erel technique, it would be possible to measure antioxidant capacity more definitely by measuring NT level and help to early diagnosis of PUC and follow-up purposes as a predictor of the activation in PUC cases. We believe that larger-scale case-control studies with this new Erel technique would help to understand and clarify the progression, diagnosis of recurrence, and pathogenesis of PUC. With this technique, it would be possible to increase the diagnostic accuracy in deciding the disease severity in PUC cases.

The mean Hb level of SG was statistically lower than CG. This finding was similar to literature findings (16). Anemia in PUC patients is probably caused by inadequate intake, malabsorptive state, and chronic blood loss by mucosal ulcerations.

Although WBC was defined as a strong predictor of severe clinical disease as measured by PUCAI, we could not find a statistically important difference between the study and control group (17). This finding of our study would be related to the presence of cases with remission in our study group.

MPV is an indicator of platelet activity and aggregation capacity (16). The mean MPV result of the SG was lower than the CG. There was also positive correlation of NT level with MPV in our study. There are multiple adult studies revealed that MPV decreased in active UC patients compared to the control group (17-19). There is no pediatric study about relation of MPV and NT level. Our study was also novel in pediatric PUC literature.

The platelet count of the study group was significantly higher than the control group. This finding supports the hypothesis that in chronic diseases, number, shape, and functions of platelets are important in the amplification of the disease severity (20). According to Chen et al. (19) chronic disease activity and iron deficiency anemia lead to thrombocytosis. Our study results support the idea that iron deficiency would cause thrombocytosis. The mean level of CRP in our SG was significantly higher than the CG which is similar to the findings of Cifci et al. (21). Although CRP was accepted as a classical parameter for IBD patients but our study results showed that CRP level was not correlated with disease activity levels as similar to the literature findings (22).

As expected, the mean level of ESR in our SG was significantly higher than CG which is parallel to the finding of Croft et al. (23). But the correlation with PUCAI was not significant. This finding supports the idea that ESR could not be used for follow-up purposes efficiently.

Our study results showed that the mean IL-6 level was found significantly higher in SG. According to a study by Feng, the life span of T lymphocytes was extended with the help of IL-6 and also IL-6 was defined as a marker of recurrence development in IBD patients who were treated (24). Karaskova et al. (25) also demonstrated that in pediatric IBD patients the level of IL-6 decreased after treatment. Our study results about IL-6 were parallel to the findings of pediatric and adult studies in literature (24,25).

Limitation of our study: Since antioxidant capacity of body would be affected from dietary factors, standard diet would be recommended before measurements. Generally, our patients use grossly similar nutrients since they were given similar instructions and recommendations about diet at the diagnosis time, but we did not check the diet of the children in detail. This study was limited by relatively small sample size in severe PUC cases. And PUCAI that we used to differentiate subgroups, accepted as subjective according to some authorities.

CONCLUSION

This is the first study demonstrating antioxidant NT level of PUC is significantly lower than healthy children. NT level also showed a good correlation with known inflammatory biomarkers, albumin and MPV. There is a significant negative correlation between disease activity defined by PUCAI and NT level. NT level would be a good candidate serum biomarker for PUC patients at the diagnosis and follow-up period without the need of a tissue biopsy. But there is a need for larger and more definitive studies for making decision about the place of NT level measurement in diagnostic and follow-up workup of PUC.

REFERENCES

1. Rubalcava NS, Gadepalli SK. Inflammatory Bowel Disease in Children and Adolescents. Adv Pediatr 2021; 68:121-42.
2. Turner D. Severe acute ulcerative colitis: the pediatric perspective. Dig Dis 2009;27:322-6.
3. Karp SM, Koch TR. Oxidative stress and antioxidants in inflammatory bowel disease. Dis Mon 2006; 52: 199– 207.
4. Hamouda HE, Zakaria SS, Ismail SA, Khedr MA, Mayah WW. p53 antibodies, metallothioneins, and oxidative stress markers in chronic ulcerative colitis with dysplasia. World J Gastroenterol 2011; 17: 2417–23.
5. Pravda J. Radical induction theory of ulcerative colitis. World J Gastroenterol 2005; 11: 2371–284.
6. Winther JR, Thorpe C. Quantification of thiols and disulfides. Biochim Biophys Acta 2014;1840: 838-46.
7. Rodosskaia NK, Chernousova GM. Immune system and thiols: some peculiarities of thiol exchange. Comp Immunol Microbiol Infect Dis 2010; 33: 65-71.
8. Erel O, Neselioglu S. A novel and automated assay for thiol/disulphide homeostasis. Clin Biochem 2014; 47: 326-32.
9. Patlevič P, Vašková J, Švorc P Jr, Vaško L, Švorc P. Reactive oxygen species and antioxidant defense in human gastrointestinal diseases. Integr Med Res 2016; 5: 250-8.
10. Grzybowska-Chlebowczyk U, Wysocka-Wojakiewicz P, Jasielska M, Cukrowska B, Więcek S, Kniażewska M, et al. Oxidative and Antioxidative Stress Status in Children with Inflammatory Bowel Disease as a Result of a Chronic Inflammatory Process. Mediators Inflamm 2018:4120973.
11. Birimberg-Schwartz L, Zucker DM, Akriv A, Cucchiara S, Cameron FL, Wilson DC, et al. Development and Validation of Diagnostic Criteria for IBD Subtypes Including IBD-unclassified in Children: a Multicentre Study From the Pediatric IBD Porto Group of ESPGHAN. J Crohns Colitis 2017; 11:1078-84.
12. Turner D, Otley AR, Mack D, Hyams J, de Bruijne J, Uusoue K, et al. Development, validation, and evaluation of a pediatric ulcerative colitis activity index: a prospective multicenter study. Gastroenterology 2007; 133:423-32.
13. Holmes EW, Yong SL, Eiznhamer D, Keshavarzian A. Glutathione content of colonic mucosa: evidence for oxidative damage in active ulcerative colitis. Dig Dis Sci 1998; 43:1088-95.
14. Tsunada S, Iwakiri R, Ootani H, Aw TY, Fujimoto K. Redox imbalance in the colonic mucosa of ulcerative colitis. Scand J Gastroenterol 2003; 38:1002-3.
15. Neselioglu S, Keske PB, Senat AA, Yurekli OT, Erdogan S, Alisik M, et al. The relationship between severity of ulcerative colitis and thiol-disulphide homeostasis. Bratisl Lek Listy 2018;119: 498-502.
16. Mahadea D, Adamczewska E, Ratajczak AE, Rychter AM, Awada A, Eder P, et al. Iron Deficiency Anemia in Inflammatory Bowel Diseases-A Narrative Review. Nutrients 2021;13:4008.
17. Holtman GA, Lisman-van Leeuwen Y, Day AS, Fagerberg UL, Henderson P, Leach ST, et al. Use of Laboratory Markers in Addition to Symptoms for Diagnosis of Inflammatory Bowel Disease in Children: A Meta-analysis of Individual Patient Data. JAMA Pediatr 2017;171:984-91.
18. Kapsoritakis AN, Koukourakis MI, Sfiridaki A,Potamianos SP, Kosmadaki MG, Koutroubakis IE, et al. Mean platelet volume: a useful marker of inflammatory bowel disease activity. Am J Gastroenterol 2001; 96: 776-81.
19. Chen Z, Lu Y, Wu J, Zhang H. Clinical significance of blood platelets and mean platelet volume in patients with ulcerative colitis. J Int Med Res 2021; 49: 3000605211009715.
20. Voudoukis E, Karmiris K, Koutroubakis IE. Multipotent role of platelets in inflammatory bowel diseases: a clinical approach. World J Gastroenterol 2014; 20:3180-90.
21. Cifci S, Ekmen N. Prediction of Mucosal Health by NLR, CRP x NLR and MPV in Ulcerative Colitis: Can Their Availability Change According to Treatment Options? Cureus 2021; 13:e19942.
22. Vermeire S, Van Assche G, Rutgeerts P. Laboratory markers in IBD: useful, magic, or unnecessary toys? Gut 2006; 55: 426-31.
23. Croft A, Lord A, Radford-Smith G. Markers of systemic inflammation in acute attacks of ulcerative colitis: What level of C-reactive protein constitutes severe colitis? J Crohns Colitis 2022;11:jjac014.
24. Feng JS, Yang Z, Zhu YZ, Liu Z, Guo CC, Zheng XB. Serum IL-17 and IL-6 increased accompany with TGF-β and IL-13 respectively in ulcerative colitis patients. Int J Clin Exp Med 2014; 7:5498-04.
25. Karaskova E, Volejnikova J, Holub D, Velganova-Veghova M, Spenerova M, Mihal V, et al. Changes in serum hepcidin levels in children with inflammatory bowel disease during anti-inflammatory treatment. J Paediatr Child Health 2020; 56: 276-82.