Original Article / Özgün Araştırma

Evaluation of Dynamic Thiol/Disulphide Homeostasis in Patients with Periodontitis

Periodontitis Hastalarında Dinamik Tiyol/Disülfid Homeostazının Değerlendirilmesi

Mahmure Ayşe Tayman[1], Ceylan Bal[2], Cemil Nural[2], Meral Günhan[3]

[1]Ankara Yıldırım Beyazıt University Faculty of Dentistry, Department of Periodontology, Ankara, Turkey
[2]Ankara Yıldırım Beyazıt University Faculty of Medicine, Department of Biochemistry, Ankara, Turkey
[3]Ankara University Faculty of Dentistry, Department of Periodontiogy, Ankara, Turkey

Abstract

Objective: Thiols are antioxidant organic compounds with sulfhydryl group. Dynamic thiol/disulphide homeostasis is considered a marker of oxidative stress. This study aimed to determine disulphide and thiol levels in patients with generalised stage III grade C periodontitis and to investigate the relationship between biochemical and clinical periodontal parameters, namely, bleeding on probing (BOP), clinical attachment level (CAL) and probing pocket depth (PPD).

Materials and Methods: Forty-eight individuals participated in the study (control, n=23; periodontitis, n=25). BOP, CAL and PPD values of the groups were recorded before and after non-surgical periodontal treatment. Thiol disulphide homeostasis was assessed by an automatic and a new spectrophotometric method. Percentages of disulphide/total thiol, disulphide/native thiol and native thiol/total thiol were calculated.

Results: The clinical periodontal parameters of the periodontitis group were higher (p<0.01) before treatment and lowered significantly after the treatment (p<0.05). Thiol levels were significantly lower (p<0.05) and disulphide levels were significantly higher in the periodontitis group (p<0.05). A significant negative correlation was found between native thiol and total thiol with all clinical periodontal parameters (p<0.05). A significant positive correlation was noted between % disulphide/thiol ratios with all clinical periodontal parameters before treatment (p<0.05).

Conclusion: A significant correlation between the severity of periodontitis and serum total thiol and disulphide levels supported the hypothesis of oxidative stress in the etiopathogenesis of periodontitis.

Öz

Amaç: Tiyoller, sülfhidril grubuna sahip antioksidan organik bileşiklerdir. Dinamik tiyol/disülfid homeostazı, oksidatif stresin bir belirteci olarak kabul edilmektedir. Bu çalışmanın amacı, evre III-derece C jeneralize periodontitisli hastalarda tiyol ve disülfid düzeylerini belirlemek ve klinik periodontal parametrelerle [sondalamada kanama (SK), klinik ataçman kaybı (KAK), sondalamada cep derinliği (CD)] biyokimyasal parametreler arasındaki ilişkiyi araştırmaktır.

Gereç ve Yöntemler: Çalışmaya 48 kişi dahil edildi (Kontrol n+23, periodontitis n=25). Grupların SK, KAK ve CD değerleri cerrahi olmayan periodontal tedaviden

önce ve sonra kaydedildi. Tiyol disülfid homeostazı otomatik ve yeni bir spektrofotometrik yöntemle değerlendirildi. Disülfid/toplam tiyol, disülfür/doğal tiyol ve doğal tiyol/toplam tiyol yüzdeleri hesaplandı.

Bulgular: Periodontitis grubunun klinik periodontal parametreleri tedaviden önce yüksekti (p<0,01) ve tedaviden sonra anlamlı olarak azaldı (p<0,05). Periodontitis grubunda tiyol düzeyleri anlamlı olarak düşüktü (p<0,05) ve disülfid düzeyleri anlamlı olarak yüksekti (p<0,05). Doğal tiyol ve toplam tiyol ile tüm klinik periodontal parametreler arasında anlamlı bir negatif korelasyon vardı (p<0,05). Tedaviden önce % disülfid/tiyol oranları ile tüm klinik periodontal parametreler arasında anlamlı pozitif korelasyon vardı (p<0,05).

Sonuç: Periodontitisin şiddeti ile toplam serum tiyol ve disülfid düzeyleri arasında bulunan anlamlı korelasyon, periodontitisin etyopatogenezinde oksidatif stresin varlığı hipotezini desteklemektedir.

Introduction

During many normal metabolic processes, reactive species are continuously generated. These products give rise to the formation of disulphide bonds in various physiological pathways, including signal transmission, enzyme activation, immune system management, and gene expression. The excessive release of oxidative products contributes to the etiology of cancer, chronic renal failure, type-2 diabetes, cardiovascular diseases, neurodegenerative diseases, ischemic reperfusion injury, immune system diseases (1-5), and periodontitis (6-8). There are antioxidant defense mechanisms that can be employed to prevent the overproduction of oxidant products (9). The imbalance between the oxidant and antioxidant systems is identified as oxidative stress. In the case of oxidative stress, the oxidant products damage numerous biological molecules, particularly proteins, lipids, and nucleic acids (10).

Periodontitis is an inflammatory and multifactorial disease causing tissue damage and loss. During the occurrence of periodontitis in the host, a complex interaction is known to occur between pathogenic bacteria and the immune system (11). Previous data have demonstrated that oxidative stress is a significant factor in the etiopathogenesis of periodontal disease. Periodontal diseases caused by bacterial infections may be related to an increase in oxygen free radicals (8,12) and defective antioxidant mechanisms (13). The excessive production of oxidative products due to a defective antioxidant defense mechanism causes chronic oxidative stress in periodontal tissues. Subsequently, an exaggerated inflammation occurs, which is considered to be an important contributing factor in the development of periodontitis, in addition to the destructive effects of free oxygen radicals associated with the pathogenesis of periodontal disease (6-8).

Compounds that contain thiol groups are organic substances. They play a significant role in resisting oxidative stress owing to their reductive properties. Thiols are considered to be double-acting effective antioxidants that protect cells mostly against free radical injury. However, an excessive increase in free thiols in the circulation is associated with toxic effects caused by an exaggerated oxidation course producing reactive oxygen species (ROS). Low-molecular-weight thiols, albumin thiols, and protein thiols are known to be the main thiols in plasma (14,15) which contain-thiol (sulfhydryl) groups (16). Oxidative products, such as ROS, produced by organisms are reduced by transferring the excess electrons to the thiol-containing compounds, with thiol groups being oxidized (17). Disulphide bonds are formed by the oxidation of thiol groups. However, after this reversible reaction, the disulphide bonds can be reduced back to thiol groups. Thus, the dynamic balance of thiol/disulphide contributes to the antioxidant effect (18,19). In general, the presence of reversible disulphide bonds is the first sign of protein oxidation (20). Dynamic thiol/disulphide homeostasis plays a significant role in antioxidant defense, apoptosis, detoxification, enzymatic activity regulation, and cellular signal transduction (21).

Thiol/disulphide stability has been studied in a number of disorders; however, up until 2014, this balance has been measured as a single-sided thiol/disulphide homeostasis, in which only the increase and decrease in the thiol form could be determined (14). Currently, the levels of both thiol and disulphide can be determined bilaterally using a new method (14,22). In recent studies, homeostasis deterioration has been reported to cause chronic kidney disease, diabetes mellitus, cardiovascular disease, cancer, chronic inflammatory joint disease, and various neurodegenerative diseases. Abnormal

thiol/disulphide homeostasis is the cause of some diseases that involve prominent chronic inflammation (23,24). By measuring the dynamic thiol/disulphide homeostasis, information on numerous normal or abnormal biochemical processes can be obtained (14). However, the role of thiol/disulphide homeostasis is not clearly elucidated in the pathogenesis of periodontal disease, which is known to be a chronic inflammatory disease.

In this study, the total thiol, native thiol, and disulphide levels related to thiol/disulphide homeostasis were evaluated in the study groups, including generalized stage III grade C periodontitis patients and healthy control individuals. Moreover, the relationship between these parameters and the clinical periodontal parameters before and after the non-surgical periodontal treatment (NSPT) has been evaluated.

Materials and Methods

This study was conducted in accordance with the Good Clinical Practices Guideline (2002 Declaration of Helsinki). The study objective and the experiments to be conducted throughout the study were explained in detail to all participants. The study participants were selected from volunteer individuals who applied for periodontal treatment at the Ankara University Faculty of Dentistry, Department of Periodontology, from June 2017 to January 2019. An informed consent form was signed by all the participants. The study protocol has been approved by the Ankara University Faculty of Dentistry Clinical Research Ethics Committee (decision no: 36290600/60-08/01, date: 28.06.2016).

Study Population

Participants who were not diagnosed with diabetes, cardiovascular diseases, respiratory diseases, and osteoporosis; those who did not routinely use medication and antibiotics or anti-inflammatory drugs in the last 3 months; and those who had not been formerly exposed to periodontal treatment were eligible. Participants who were pregnant and breastfeeding and who are currently smokers or former smokers were excluded from the study.

Clinical diagnosis was considered according to the "2017 World Workshop on the Classification of Periodontal and Peri-implant Diseases and Conditions" (25). In this study, the periodontitis group (n=25) included patients with generalized stage III grade C periodontitis. Patients who had interdental radiographic bone loss at ≥5 mm in the non-adjacent teeth or buccal or oral radiographic bone loss that extends to the middle or apical third of the root with probing depth of >3 mm were diagnosed with stage III periodontitis. Extension of periodontal disease was in more than 30% of all sites so described as generalized. Patients were also diagnosed as grade C based on the bone loss/age index (>1.00= indirect evidence of progression), smoking, and/or diabetes. The control group (n=23) included patients with no history of periodontal disease, clinical or radiographic findings of attachment loss, and clinical signs of gingivitis. Periodontal health (control group) was defined as a probing pocket depth (PPD) of ≤3 mm and bleeding on probing (BOP) of (+) ≤10% (26).

Non-surgical Periodontal Treatment

Individuals were treated with full-mouth scaling and root planing (SCRP). The treatment procedure was performed using an ultrasonic scaler (Cavitron DENTSPLY, York, PA) and hand instruments (Gracey curets, Hu-Friedy, Chicago, IL) and was divided into two sessions with a 24-h interval with the aim of ending all SCRP, in accordance with the original study of Quirynen et al. (27). No time limit was. Thus, the periodontist finished the procedure as soon as the root surfaces were satisfactorily cleaned and flattened. The patients were provided with standard oral hygiene training immediately after the first SCRP procedure (tooth brushing, interdental brushing, flossing, and tongue dorsum brushing) (27). The patients were warned not to use any medication or mouthwash products. One month after the treatment, the patients were followed up two to three times and checked for the instructions given.

Periodontal Examination

BOP (%), PPD (mm), and clinical attachment level (CAL, mm) values were recorded by an experienced periodontist (M.A.T) before the NSPT. Prior to the study, the examiners were calibrated (M.G and M.A.T). A reference examiner (M.G.), with more than 20 years of experience in periodontology, has calibrated our periodontist (M.A.T.). The scores of probing depth exhibited good reproducibility, as evaluated by an inter-examiner. A total of five volunteers were assessed twice, with a 1-h interval between assessments. The reproducibility assessment resulted in 85% of sites

for which the repeat probing mean measurements were within ±1 mm. The Williams periodontal probe (the University of Michigan) was used for the measurements, and the cementoenamel junction was set as the reference point. The examination and recording of the clinical periodontal parameters were repeated by the same periodontist 1 month after the end of the treatment period. A total of six regions of each tooth (mesio-buccal, distobuccal, mid-buccal, mesio-lingual, disto-lingual, and mid-lingual) were examined to obtain the PPD, CAL, and BOP values. The PPD measurements were recorded by rounding off to the nearest millimeter. The CAL measurements were calculated by overlapping gingival recession and PPD. Also, the BOP measurements were expressed as percentage. The average of all the CAL, PDD, and BOP values was then calculated for each patient.

Collection of Serum Samples

Blood samples were obtained from the antecubital vein just before and 1 month after the NSPT. Subsequently, the blood samples were left to stand for 30 min (minutes) prior to centrifugation. All samples were centrifuged at 4,000 g for 10 min to separate the serum and stored at -80 °C until the day of the experiment.

Serum Disulphide/Thiol Homeostasis

A spectrophotometric method was employed, as defined by Erel and Neselioğlu (14), to measure disulphide/thiol homeostasis. Specifically, using this method, free functional thiol groups were obtained by reducing the reducible disulphide bonds, and formaldehyde was applied to remove the remaining sodium borohydride used as a reductant. After the reaction with 5.5'-dithiobis-(2-nitrobenzoic) acid, all thiol groups containing native and total thiols were measured. To calculate the dynamic disulphide amount, half of the difference between the total thiols and native thiols was utilized. Native thiols and total thiols were measured; then, the native thiol/total thiol percentage ratios, disulphide amounts, disulphide/native thiol percentage ratios, and disulphide/total thiol percentage ratios were calculated.

Statistical Analysis

For the statistical analysis, the Statistical Package for Social Sciences (SPSS) (version 20 for Windows, SPSS, Inc., St. Louis, MO) was used. Normal data distribution was controlled using the Shapiro-Wilk test. The parameters with a normal distribution were analyzed via the parametric tests. Intergroup comparisons were performed using the independent-samples t-test. Moreover, the Bonferroni correction was employed to evaluate the statistical significance. Pearson's correlation coefficient (Pearson's r) was employed for the correlation analysis of variables. For each group, 20 patients were collected according to the 80% power calculation to identify the least clinically significant difference with 5% type-I error. $p < 0.05$ was considered to be statistically significant.

Results

Demographic Data and Clinical Periodontal Parameters

A total of 48 individuals participated in the study; they were divided into the control group (female/male=12/11, 41±4.6 mean age, years) and the periodontitis group (female/male=13/12, 43±5.2 mean age, years). The patients were able to complete the 1-month recovery and follow-up periods without complications. No statistically significant difference was observed between the study groups in terms of age and gender distribution. The clinical periodontal parameter (CAL, BOP, and PPD) values of the measurements before and after periodontal treatment are presented in Table 1. The clinical periodontal parameter values of the periodontitis group before treatment were statistically higher than those of the control group ($p < 0.01$). It was observed that all the clinical parameters significantly decreased at the first month after treatment compared with the pre-treatment baseline measurements ($p < 0.05$). However, the results of the periodontitis group after treatment were significantly higher than those of the control group ($p < 0.01$).

Serum Parameters

The pre-treatment and post-treatment values of the periodontitis group were compared with those of the control group (Table 2). Before treatment, the levels of native and total thiols were lower in the periodontitis group ($p=0.007$, $p=0.001$, respectively) than in the control group. The disulphide, % disulphide/native thiol, and % disulphide/total thiol ratio levels were significantly higher in the periodontitis group than in the control group ($p=0.038$, $p=0.014$, $p=0.002$, respectively) prior to treatment. There was no significant biochemical parameter between the

control group and the periodontitis post-treatment group (p>0.05).

In the pre-treatment and post-treatment periodontitis groups, the levels of native and total thiols increased after treatment; however, the increase was not significant (p=0.141, p=0.248, respectively). However, after treatment, the disulphide, % disulphide/native thiol ratio, and % disulphide/total thiol ratio levels significantly decreased (p=0.007, p=0.006, p=0.001, respectively). The native and total thiol ratio levels did not change after treatment (p=0.647).

Correlation

Prior to treatment, a significant and positive correlation was observed between all the clinical periodontal parameters (BOP-CAL, BOP-PPD, PPD-CAL) in the periodontitis group (r=0.929, p<0.01,

r=0.969, p<0.01, r=0.940, p<0.01, respectively). A significant negative correlation was observed between native and total thiols and all the clinical periodontal parameters (p<0.05). Moreover, a significant positive correlation was observed between % disulphide/native thiol ratio and % disulphide/total thiol ratio and all the clinical periodontal parameters (p<0.05).

Before and after treatment, a significant correlation was observed between the total and native thiol levels (r=0.964/0.962, p<0.001, respectively). In addition, a significant negative correlation was observed between the native thiol level (mmol/L) and % disulphide/native thiol ratio levels (r=-0.737, p<0.01, r=-0.504, p<0.05). There was also a significant positive correlation between disulphide (mmol/L) and % disulphide/total thiol ratio; % disulphide and % disulphide/native thiol ratio (r=0.893/0.834, p<0.001;

Table 1. Demographics of the study population and comparisons of full mouth clinical periodontal parameters between the groups

Variables	Control (n=23)	Generalized stage III grade C periodontitis (n=25)		
		Pre-Treatment	Post-Treatment	Pre-Treatment/Post-Treatment (p)
PPD (mm)	1.12±0.23	3.85±0.56**	2.94±0.32**	p<0.01
BOP (%)	8.74±2.51	79.25±7.35**	30.93±4.74**	p<0.01
CAL (mm)	1.25±0.25	4.96±1.09**	4.34±0.80*	p<0.05
Female/male	12/11	13/12		
Age (year)	41±4.6	43±5.2		

PPD: Probing pocket depth, BOP: Bleeding on probing, CAL: Clinical attachment level. Data are experessed as mean±SD. **p<0.01, *p<0.05 statistically significant difference from control group

Table 2. Serum levels of thiol/disulphide in control and generalized stage III grade C periodontitis groups

Biochemical parameters	Control (n=23)	Periodontitis (n=25) (Pre-Treatment/Post-Treatment)	Control/periodontitis (Pre-Treatment) p	Control/periodontitis (Post-Treatment) p	Pre-Treatment/Post-Treatment (p)
Native thiol (mmol/L)	363.88±34.39	332.82±34.05/348.17±9.53	0.007*	0.113	0.141
Total thiol (mmol/L)	404.65±29.77	367.99±35.42/381.80±40.68	0.001**	0.052	0.248
Disulphide (mmol/L)	20.38±4.62	23.16±3.30/19.68±4.10	0.038*	0.635	0.007*
% Disulphide/ native thiol ratio	5.73±1.74	6.96±1.18/5.73±1.41	0.014*	0.897	0.006*
% Disulphide/ total thiol ratio	5.09±1.38	6.32±0.91/5.19±1.14	0.002*	0.526	0.001**
% Native thiol/ total thiol ratio	89.80±2.76	90.42±2.44/90.83±3.11	0.462	0.515	0.647

**p<0.01, *p<0.05, Data are expressed as mean ± standard deviation (median)

Table 3. Correlation between clinical periodontal parameters and thiol/disulphide serum levels in the study groups for pre- and post-treatment

Generalized stage III grade C periodontitis (Pre-Treatment/Post-Treatment)

Parameters		CAL	PPD	Native thiol (mmol/L)	Total thiol (mmol/L)	Disulphide (mmol/L)	% Disulphide/native thiol ratio	% Disulphide/total thio ratio	% Native thiol/total thiol ratio
BOP	r=	0.929/-0.014	0.969/0.171	-0.462/0.977	-0.540/0.025	0.304/0.084	0.398/0.086	0.467/0.082	0.12/-0.175
	p=	0.00/0.948 **/¥	0.00/0.425 **/¥	0.004/0.977 ¥/¥	0.00/0.92 */¥	0.063/0.733 ¥/¥	0.013/0.726 */¥	0.003/0.740 */¥	0.502/0.475 ¥/¥
CAL	r=	-	0,940/0.580	-0.403/0.321	-0.497/0.261	0.340/0.087	0.379/0.066	0.489/-0.032	0.191/0.165
	p=	-	0.00/0.003 **/*	0.012/0.180 */*	0.001/0.280 **/*	0.037/0.723 */*	0.019/0.787 */*	0.002/0.898 */*	0.251/0.501 ¥/¥
PPD	r=	-	-	-0.402/0.104	-0.501/0.101	0.322/0.127	0.379/0.083	0.469/0.091	0.206/0.027
	p=	-	-	0.012/0.671 */*	0.001/0.680 ***/*	0.049/0.603 */*	0.019/0.735 */*	0.003/0.712 */*	0.251/0.912 ¥/¥
Native thiol (mmol/L)	r=	-	-	-	0.967/0.962	-0.338/0.067	-0.737/-0.504	-0.702/-0.478	0.489/0.320
	p=	-	-	-	0.00/0.000 **/**	0.035/0.786 */¥	0.00/0.028 **/*	0.00/0.039 **/*	0.002/0.181 ¥/¥
Total thiol (mmol/L)	r=	-	-	-	-	-0.202/0.234	-0.621/-0.331	-0.610/-0.328	0.250/0.069
	p=	-	-	-	-	0.219/0.335 ¥/¥	0.00/0.167 **/¥	0.00/0.170 **/¥	0.125/0.778 ¥/¥
Disulphide (mmol/L)	r=	-	-	-	-	-	0.814/0.816	0.893/0.834	-0.561/-0.592
	p=	-	-	-	-	-	0.00/0.00 **/**	0.00/0.00 **/**	0.00/0.008 **/*
% Disulphide/ native thiol ratio	r=	-	-	-	-	-	-	0.939/0.992	-0.655/-0.727
	p=	-	-	-	-	-	-	0.00/0.00 **/**	0.00/0.00 **/**
% Disulphide/ total thiol ratio	r=	-	-	-	-	-	-	-	-0.557/-0.653
	p=	-	-	-	-	-	-	-	0.00/0.002 **/*

**p<0.01, *p<0.05, ¥>0.05, PPD: Probing pocket depth, BOP: Bleeding on probing, CAL: Clinical attachment level

r=0.814/0.816, p<0.001). The correlation levels between the parameters before and after treatment are presented in Table 3.

Discussion

Dynamic thiol/disulphide stability is important for humans. When this balance is disturbed, stabilization of protein structures, enzyme transcription and regulation, cellular signal transduction, and receptor and transporter functions could be affected and damaged. Thus, detoxification, antioxidant preservation, enzymatic activity regulation, and components of the cellular signaling mechanism could also be affected (28,29). In this study, we evaluated the dynamic thiol/disulphide homeostasis of the periodontitis and control groups using a new colorimetric method (14). Based on our results, levels of native and total thiols, which provide information on antioxidant conditions, significantly decreased in the periodontitis group compared to the control group. Contrarily, the disulphide levels significantly increased in the periodontitis group. A few studies obtained results similar to those of our study in different chronic diseases (24,30-32). The plasma thiol levels decrease as oxidative stress increases (33). Various studies have demonstrated the relationship between oxidative stress and chronic inflammation in the development and progression of periodontitis (7,8,12). Additionally, a good correlation was observed between the total and native thiol levels and the clinical parameters in the patient group.

To the best of our knowledge, our study is the first to evaluate the dynamic thiol/disulphide homeostasis in patients diagnosed with periodontitis. Periodontitis is a chronic inflammatory disease caused by the interaction between the host immune response and pathogenic bacteria. The immunological activity and cytokine expression increase in the gingival tissue during the activity of predominant inflammatory cells. As a result, prolonged inflammatory reaction produces a large amount of oxidants, thus causing oxidative damage. In periodontitis, oxidative radicals are known to increase more than the antioxidant molecules and cause oxidative stress (34). These oxidant radicals oxidize the thiol groups present in the side chains of sulfur-containing amino acids of proteins to form disulphide bonds (35). Subsequently, the increased disulphide bonds cause

the thiol/disulphide homeostasis to shift toward the disulphide, which results in an abnormal thiol/disulphide homeostasis (14). During abnormal thiol/disulphide homeostasis, vital cellular functions are damaged. Due to oxidative stress, pathologies occur in numerous organelles and cause imbalances (14). Moreover, oxidant radicals formed by other factors, such as inflammation, chemicals, and radiation, can also damage the mechanism of thiol/disulphide homeostasis. In both cases previously described, thiol/disulphide homeostasis is expected to weaken in the periodontitis group than in the control group.

In the present study, the levels of thiol/disulphide were lower in the periodontitis group than in the control group prior to treatment. The thiol levels may be decreased due to increased oxidation of thiol groups by oxidant radicals in periodontitis, which is a disease dominated by chronic inflammation. However, in our study, the radicals that cause oxidative stress were not investigated, which may be considered as a limitation of our study. Previous studies have revealed that oxidant radical concentrations are higher in the periodontitis group than in the control group. It has also been emphasized that periodontitis is a minor local inflammatory condition and is associated with the systemic oxidative stress level in numerous studies conducted on human subjects (36-38). It is believed that inflammation is responsible for the production of oxidant radicals; however, it is also possible for oxidant radicals to stimulate inflammation. According to the results of our study, we suggest that increased oxidant radical levels associated with periodontal inflammation shifted the thiol/disulphide balance in favor of disulphide. As a result, thiol/disulphide homeostasis weakened, thiol levels decreased, and disulphide levels increased in the periodontitis group. The present study aimed to measure systemic markers in patients with periodontitis. It would have been valuable if thiol/disulphide homeostasis was measured in the saliva samples or gingival crevicular fluid samples. This can also be considered as a limitation of our study.

One month after the treatment, the NSPT, BOP, PPD, and CAL values significantly decreased in the periodontitis group. The decrease in periodontal inflammation indicates the clinical effects of the treatment. The clinical results we obtained with the NSPT were consistent with those of the clinical study

conducted by Badersten et al. (39), Haffajee et al. (40), and Pinho et al. (41). Through the NSPT, periodontal pathogens were removed from the patients with periodontitis, systemic inflammatory mediators were reduced, and oxidative stress and disease activity were decreased. Based on the results of this study, it was important to determine a significant decrease in the serum disulphide levels and some increase in the thiol levels after periodontal treatment. This has been confirmed by the significant improvement in the values of the clinical periodontal parameters. However, the clinical periodontal parameters measured in the periodontitis group after the NSPT were still significantly higher than the periodontal parameters measured in the control group. Although the debridement with NSPT provides an improvement in patients with advanced periodontitis, there are still some regions with deep PPD and BOP+. In addition, the CAL cannot be reached on the same basis as in healthy individuals by performing NSPT alone.

Conclusion

In this study, the serum levels of native thiol, total thiol, disulphide, disulphide/native thiol, disulphide/total thiol, and native thiol/total thiol were investigated in patients with periodontitis. Dynamic thiol/disulphide homeostasis provides significant information on various normal or abnormal biochemical processes as a new marker of oxidative stress. Numerous articles on oxidative stress in the literature have demonstrated contradictory data. The results of this study confirmed the efficacy of periodontal therapy and provided valuable information on the thiol/disulphide balance. In this study, the significant relationship between the severity of periodontal disease and the serum total thiol and disulphide levels supported the hypothesis of oxidative stress, which plays a significant role in the etiopathogenesis of periodontitis.

Ethics

Ethics Committee Approval: The study protocol has been approved by the Ankara University Faculty of Dentistry Clinical Research Ethics Committee (decision no: 36290600/60-08/01, date: 28.06.2016).

Informed Consent: An informed consent form was signed by all the participants.

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Authorship Contributions

Concept: M.A.T., C.B., M.G., Design: M.A.T., C.B., C.N., M.G., Supervision: M.A.T., C.B., C.N., M.G., Fundings: M.A.T., C.B., Data Collection or Processing: M.A.T., C.B., C.N., M.G., Analysis or Interpretation: M.A.T., C.B., C.N., Literature Search: M.A.T., C.B., C.N., M.G., Critical Review: M.A.T., C.B., C.N., M.G., Writing: M.A.T., M.G.

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