CYPRUS JOURNAL OF MEDICAL SCIENCES

Original Article

DOI: 10.5152/cjms.2018.548

The Relationship between Climacteric Symptoms and Thiol/Disulphide Homeostasis

Raziye Desdicioğlu[1], Melahat Yıldırım[1], Ceylan Bal[2], Almila Şenat[2], Edip Alptuğ Kır[3], Özcan Erel[2], Ayşe Filiz Yavuz[1]

[1]Department of Obstetrics and Gynecology, Ankara Yıldırım Beyazıt University School of Medicine, Ankara, Turkey
[2]Department of Biochemistry, Ankara Yıldırım Beyazıt University School of Medicine, Ankara, Turkey
[3]Department of Obstetrics and Gynecology, Ankara Training and Research Hospital, Ankara, Turkey

ORCID IDs of the authors: R.D. 0000-0002-5190-5083; M.Y. 0000-0003-4077-7490; C.B. 0000-0002-1678-1281; A.Ş. 0000-0002-5806-562X; E.A.K. 0000-0002-2293-3624; O.E. 0000-0002-2996-3236; A.F.Y. 0000-0003-3699-7757.

Cite this article as: Dediscioğlu R, Yıldırım M, Bal C, Şenat A, Kır EA, Erel Ö, et al. The Relationship between Climacteric Symptoms and Thiol/Disulphide Homeostasis. Cyprus J Med Sci 2018; 3(3): 132-6.

BACKGROUND/AIMS
The aim of this study was to examine the relationship between the serum thiol-disulfide balance and climacteric symptoms.

MATERIAL and METHODS
A total of 106 women aged 42 to 78 years (53.18±7.39) participated in the study. The patients were divided into three groups as the perimenopausal period, early postmenopausal period, and late postmenopausal period. The climacteric symptoms of patients were assessed with the Greene scale. The serum thiol/disulfide hemostasis was assessed with a new automatic method. The Mann-Whitney U test, one-way analysis of variance, and post-hoc Tukey tests were used.

RESULTS
Native thiol and total thiol levels start decreasing after menopause. Women in the late postmenopausal period have been observed to have significantly lower levels compared to women in the perimenopausal and early postmenopausal periods. In the late postmenopause, perimenopause, and early post menopause, native thiol was measured as 430.81±45.35, 474.64±47.06, 461.22±47.66 ($p<0.05$), while total thiol was 460.22±49.96, 506.74±51.74, 492.25±57.81 ($p<0.05$), respectively. There was a negative correlation observed between vasomotor symptoms and all thiol components only in the early menopause group.

CONCLUSION
When the duration of the menopause exceeds 5 years, thiol levels reduce. This variation may be considered to be related to increasing age. Especially in the early menopausal period, administering antioxidant agents as support may contribute to reducing the vasomotor symptoms of patients.

Keywords: Climacterium, menopause, thiol/disulfide homeostasis, vasomotor symptoms

INTRODUCTION

Menopause is a natural stage in a woman's life. In addition to physical problems of women in the postmenopausal period, they encounter a variety of physiologic, psychologic, and sociologic problems that disrupt the quality of life (1). One of these problems are vasomotor symptoms. The etiology of vasomotor symptoms is not clearly understood. Many studies have shown that vasomotor symptoms do not only disrupt the quality of life linked to estrogen deficiency but are also early indicators of cardiovascular diseases (2-4). As a result, the presence and severity of vasomotor symptoms are important.

Oxidative stress is a situation formed by the antioxidant systems becoming ineffective against free oxygen radicals due to being insufficient (5). Estrogen acts like a typical antioxidant enzyme and has been shown to neutralize free oxygen radicals (6). Reduced estrogen levels in the menopause, and increased age, negatively affect antioxidant capacity (7). Although there are studies showing that disruption of the balance between oxidative stress-antioxidant capacity may

This study was presented at the 7th İstanbul Obstetrics and Gynecology Congress. December 7-10, 2017. İstanbul, Turkey.
Corresponding Author: Raziye Desdicioğlu
E-mail: raziyedesdicioglu@gmail.com
Received: 02.06.2018
Accepted: 06.09.2018

be effective in determining the development and severity of climacteric symptoms, the results are not clear (8).

The thiol/disulfide (T/D) balance plays a role in many metabolic pathways. The T/D balance has been shown to be disrupted in situations related to the endothelial damage, such as diabetes and preeclampsia (9-11). Among patients with postmenopausal osteoporosis, T/D homeostasis was shown to be disrupted (12).

In this study, we planned to research the correlation between the severity of climacteric symptoms, led by vasomotor symptoms, in the menopausal transition period and postmenopausal period and the correlation of severity of symptoms with the T/D balance. Previous studies researching the correlation of serum T/D balance with climacteric symptoms only studied the thiol component. In our research, we studied all components of the T/D balance with a new automatic method described by Erel et al. (13).

MATERIAL and METHODS

Planned as a cross-sectional study, the research received the necessary permission from the ethics committee of Atatürk Training and Research Hospital. All women signed the consent form for this research. All patients were questioned in detail about demographics, medical and reproductive history, habits, and medication use. The study included 106 women aged 42 to 78 years. The grouping of patients according to menopausal situation used the STRAW staging criteria (14). The perimenopausal group included patients with 2-11 months of amenorrhea, and the early postmenopausal period group included patients with 1-5 years since last period. The late menopausal group included patients with 6 years or more since last period. Patients who did not volunteer, who did not abide by period descriptions, who used exogenous hormones, who entered the menopause surgically, and with chronic diseases that affect oxidative stress, such as diabetes and hypertension, were not included in the study. Anthropomorphic measurements like height and weight were made.

Biochemical Assessment

Patients had serum samples taken after 8 hours of fasting. Samples were stored at -80°C until study. T/D hemostasis was studied with a newly developed automatic method (Roche, cobas 501, Mannheim, Germany) (13).

Climacteric Symptoms

The Greene climacteric scale was used to assess climacteric symptoms (15). This scale comprises 21 questions separately evaluating vasomotor symptoms, anxiety state, depression, psychological state, somatic symptoms, and sexual symptoms. All questions have a 4-degree answer choice ranging from 0 to 3. If the situation does not apply to the patient, 0 points are given, with 1 given for mild severity, 2 for moderate severity, and 3 for severe symptoms. The Greene scale was used to assess climacteric symptoms with total and subgroup scale points.

Statistical Analysis

The Kolmogorov-Smirnov test was used to assess whether the sample abided by normal distribution. To assess differences between the three groups for variables with normal distribution, the one-way analysis of variance and post-hoc Tukey test were used. The Kruskall-Wallis analysis was performed to assess differences between groups for variables without normal distribution. The Mann-Whitney U test was used to show which groups were different for variables with differences. For correlation between thiol and disulfide values with climacteric symptoms, the Pearson analysis was used for parameters with normal distribution, while the Spearman correlation analysis was used for those without normal distribution. Statistical significance was accepted as $p<0.05$.

TABLE I. Comparison of parameters between groups

	Perimenopause (n=40)	Early Postmenopause (n=30)	Late Postmenopause (n=36)
Age	47.30±3.63	51.97±2.98	60.72 ±6.51*
BMI	27.94±5.15	28.67 ±5.20	29.00±5.31
T.Thiol (SH+SS)(µmol/L)	506.74±51.74	492.25±57.81	460.22±49.96*
N.Thiol (SH)(µmol/L)	474.64±47.06	461.22±47.66	430.81±45.35[a]
Disulfide (SS)(µmol/L)	16.00±8.49	17.09±10.74	14.65±8.82
SS/SH (%)	3.39±1.84	3.67±2.16	3.42±2.16
SS/SS+SH (%)	3.10±1.61	3.36±1.84	3.13±1.86
SH/SS+SH (%)	93.78±3.26	93.98±5.67	93.71±3.74
Greene vasomotor subscale	4.03±1.34[a]	2.27±1.55	2.14±1.60
Greene anxiety subscale	6.64±3.73	8.47±4.24[a]	5.97±3.85
Greene psychology subscale	10.72±6.13	14.50±7.15[a]	10.36±6.27
Greene depression subscale	4.10±3.09	6.03±3.55[a]	4.39±3.06
Greene somatic subscale	5.28±3.96	9.27±3.99[a]	6.58±4.03
Greene sexual subscale	0.92±.83[a]	1.53±1.10	1.56±0.93
Greene total score	20.79±8.52	27.23±10.65*	20.19±9.24

*Between all groups $p<0.05$ (Analysis of variance-Tukey)
[a]a group found to be statistically significant from other two groups ($p<0.05$) (Kruskall-Wallis/Mann-Whitney U)
SS: disulfide; SH: native thiol; BMI: body mass index

TABLE 2. Correlation of age and the BMI with parameters in all groups

N=106	Age		BMI	
	r*	p	r*	p
Native thiol (µmol/L)	-0.427	<0.01	-0.262	<0.01
Disulfide (µmol/L)	-0.143	0.14	-0.031	0.74
Total thiol (µmol/L)	-0.442	<0.01	-0.249	0.01

*Spearman correlation coefficient. BMI: body mass index

TABLE 3. Correlation analysis results for thiol/disulfide balance with vasomotor subscale in the early postmenopausal group

n=30	r*	p
Native thiol	-0.376	0.04
Disulfide	-0.392	0.03
Total thiol	-0.394	0.03

*Spearman correlation coefficient

RESULTS

The mean age of the 106 women participating in our study was 53.18±7.39 years. The mean age of patients was highest in the late postmenopause group (60.72±6.51), and this was significant compared to the other groups. The mean body mass index (BMI) for all groups was 28.51±5.19 with no differences between the groups.

Total thiol and native thiol levels reduced as the menopause progressed. The late postmenopausal group had significantly lower levels compared to the other two groups. Disulfide level was highest in the early postmenopausal group, but there was no significant difference observed between the groups.

On the Greene climacteric scale, only vasomotor symptoms were observed to be significantly high in the perimenopause group compared to the other groups ($p<0.05$). Anxiety, psychologic, depression, and somatic and total Greene scores were significantly higher in the early postmenopause group compared to the other groups ($p<0.05$). Comparison of the perimenopausal period with the other two groups observed the highest vasomotor score and lowest sexual score ($p<0.05$). In the late postmenopause group, the sexual subscale score was observed to be significantly higher compared to the other groups ($p<0.05$). Findings are summarized in Table 1.

An analysis of the correlation between age and BMI with T/D components observed significant negative correlations between increasing age and BMI with native thiol and total thiol levels ($p<0.01$). In our study, a correlation analysis was performed on T/D components, and their indices with the Greene scale subgroups. There was no significant correlation observed between parameters for perimenopausal and late postmenopausal women. In the early postmenopausal period, there were significant negative correlations between all components of the T/D balance with vasomotor subscale scores ($p<0.05$). In this group, the index showing D/T thiol ratios was observed to have a negative correlation with vasomotor subscale ($p<0.05$). Correlation analysis data are summarized in Tables 2 and 3.

DISCUSSION

The menopause is a significant physiological condition characterized by the cessation of the menstrual cycle and loss of ovarian functions. In addition to vasomotor symptoms disrupting the quality of life in the menopausal period, it is known to increase the tendency for osteoporosis, diabetes, hypertension, and cardiovascular diseases in the long term. Oxidative conditions constitute major risk factors for the development of a number of pathologies, such as tumor development, diabetes, and cardiovascular complications (16). An abnormal T/D homeostasis state is involved in the pathogenesis of a variety of diseases (13). The plasma thiol levels assessed in our study are a pool formed of albumin and low molecular weight thiols (like glutathione, cysteine). When oxidative stress dominates the environment, the balance shifts to disulfide due to oxidation of the thiol group proteins (17). The reduction of estrogen in the menopausal period is known to be the main factor in vasomotor symptoms. There are publications proposing an association between reduced estrogen and increased oxidative stress markers (18, 19). The estrogen replacement therapy (ERT) in menopausal women nearly completely resolves vasomotor symptoms (20). However, due to conflicting results about ERT increasing thromboembolic events and malignancies like breast cancer, not all patients use it (21). As a result, developing alternative treatments to reduce vasomotor symptoms becomes important. If oxidative stress can be shown to be an effective mechanism on vasomotor symptoms, the use of antioxidant treatment may be considered.

In all groups, there were negative correlations between the age and BMI with native thiol and total thiol levels. The negative effect of increasing age and obesity on antioxidant systems is in accordance with other studies in the literature (22-24).

A study of 245 patients that was researching the correlation between hot flush symptoms, and serum and urine levels of oxidative stress markers such as 8-iso-prostaglandin F2-alpha (PFG2 alpha), 8-hidroxy-deoxyguanosine (8OHDG), thiol, and paraoxanase-1, showed that none of the markers correlated with the severity of vasomotor symptoms. There was no difference identified for the serum and urine levels of any marker between women with and without symptoms (2). In this study, all patients in the perimenopause and early postmenopause were included in a single group, and assessments were only made for the hot flush complaint. In the T/D balance, only the thiol component was studied. In our study, patients were separated into perimenopause, early postmenopause, and late postmenopause groups, and differences were assessed. Our results found that native thiol and total thiol rates were lowest in the late postmenopause, and this difference was significant. Patients in this period were also the oldest patients and were expected to have lowest estrogen levels. As a result, the low native thiol and total

thiol levels in these patients is an expected outcome. Disulfide levels were highest in the early postmenopause; however, the difference was not significant. A correlation analysis observed a negative correlation between the vasomotor subscale and all thiol components only in the early postmenopause group. In the study mentioned above, there was no correlation between vasomotor symptoms and thiol levels. Our patient numbers, design, and methods are different, which may have led to differences in the study results.

Another study assessed the serum total thiol level and found it was lower in postmenopausal women compared to women in the premenopausal and perimenopausal periods (22). In our study, all components of the T/D balance were studied with a new method, with the native thiol, and total thiol components of the T/D balance significantly lower in the late postmenopause. Highest disulfide levels were measured in the early postmenopausal group; however, no significant correlation was observed between the groups. In the late postmenopausal period, native thiol and linked total thiol levels reduce with age and are associated reduced estrogen levels. This complies with our study results.

A study evaluating the antioxidant enzyme capacities of patients found that superoxide dismutase, catalase, and gluthathione peroxidase levels were significantly lower among women in the postmenopausal period compared to the premenopausal group (1). This decrease was emphasized to be associated with reduced estrogen levels. Similarly, in our study results, it was shown that antioxidant capacity reduced as the menopause duration increased.

A study assessing 50 women physiologically in the menopause evaluated the free oxygen radical test and free oxygen radical defense (FORD) of patients. In this study, only the vasomotor subscale was negatively correlated with FORD levels (25). In our study, a negative correlation between all thiol components and vasomotor subscale was only observed in the early premenopause. Differences may be due to differences in the marker characteristics and patient groups studied.

The highest score on the Greene climacteric scale used in the study was in the early postmenopausal period, and this is considered to accompany physiologic changes in the first 5 years of the process. The highest sexual scale was recorded in the late postmenopausal period. It is known that urogenital symptoms increase with the duration of menopause. These findings are expected changes within the menopausal process.

In this assessment, patients had the highest number of vasomotor symptoms in the perimenopausal period, with disulfide levels highest among late postmenopausal women but low in the early postmenopausal period. There may be limited benefits to administering antioxidant treatment to patients in this period. Native thiol serum levels showing antioxidant activity were lowest in the late postmenopause, and this difference was significant between the groups. As a result, administering antioxidant support in addition to hormone replacement therapy and other medical treatments to patients from the beginning of the menopausal process may be considered. Antioxidants may contribute to an increasing antioxidant capacity, even if they do not support vasomotor symptoms. Among the limitations of our study are the relatively low number of patients, the assessment of oxidative stress only with the T/D balance, and the subjective nature of responses to questions on the Greene scale. According to our research, this is the first study researching the correlation between all components of the T/D balance and climacteric symptoms.

Ethics Committee Approval: Ethics committee approval was received for this study from the Ethics Committee of Atatürk Training and Research Hospital. (Approval Date: 13.04.2016, Approval Number: 2637996/145).

Informed Consent: Informed consent was obtained from the patients who participated in this study.

Peer-review: Externally peer-reviewed.

Author contributions: Concept - R.D., M.Y.; Design - R.D., M.Y.; Supervision - Ö.E., A.F.Y.; Resource - Ö.E., R.D.; Materials - E.A.K., A.Ş.; Data Collection and/or Processing - E.A.K., A.Ş., C.B.; Analysis and/or Interpretation - R.D., C.B.; Literature Search - M.Y.; Writing - R.D., C.B.; Critical Reviews - Ö.E., A.F.Y., R.D.

Acknowledgements: The authors would like to thank to the Departments of Obstetrics and Gynecology and Biochemsitry of the Ankara Atatürk Training and Research Hospital.

Conflict of Interest: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

REFERENCES

1. Ansar S, Tayef A, Ansari MA. Status of trace elements and antioxidants in premenopausal and postmenopausal phase of life: a comparative study. Int J Clin Exp Med 2015; 8: 19486-90.
2. Bonaccorsi G, Romani A, Cremonini E, Bergamini CM, Castaldini MC, Fila E Et al. Oxidative stress and menopause-related hot flashes may be independent events. Taiwan J Obstet Gynecol 2015; 54: 290-3. [CrossRef]
3. Bechlioulis A, Naka KK, Kalantaridou SN, Kaponis A, Papanikolaou O, Vezyraki P, et al. Increased vascular inflammation in early menopausal women is associated with hot flush severity. J Clin Endocrinol Metab 2012; 97: E760-4. [CrossRef]
4. Gast GC, Grobbee DE, Pop VJ, Keyzer JJ, Wijnands-van Gent CJ, Samsioe GN, et al. Menopausal complaints are associated with cardiovascular risk factors. Hypertension 2008; 51: 1492-8. [CrossRef]
5. Sánchez-Rodríguez MA, Castrejón-Delgado L, Zacarías-Flores M, Arronte-Rosales A, Mendoza-Núñez VM. Quality of life among post-menopausal women due to oxidative stress boosted by dysthymia and anxiety. BMC Women's Health 2017; 17: 1. [CrossRef]
6. Kumar S, Lata K, Mukjopadhyay S, Mukherjee TK. Role of estrogen receptors in pro-oxidative and anti-oxidative actions of estrogens: a perspective. Biochim Biophys Acta 2010; 1800: 1127-35. [CrossRef]
7. Sánchez-Rodríguez MA, Zacarías-Flores M, Arronte-Rosales A, Correa-Muñoz E, Mendoza-Núñez VM. Menopause as risk factor for oxidative stress. Menopause 2012; 19: 361-7. [CrossRef]
8. Lee R, Margaritis M, Channon KM, Antoniades C. Evaluating oxidative stress in human cardiovascular disease: methodological aspects and considerations. Curr Med Chem 2012; 19: 2504-20. [CrossRef]
9. Ates I, Kaplan M, Yuksel M, Mese D, Alisik M, Erel Ö, et al. Determination of thiol/disulphide homeostasis in type I diabetes mellitus and the factors associated with thiol oxidation. Endocrine 2016; 51: 47-51. [CrossRef]

10. Korkmaz V, Kurdoglu Z, Alisik M, Cetin O, Korkmaz H, Surer H, et al. Impairment of thiol-disulfide homeostasis in preeclampsia. J Matern Fetal Neonatal Med 2016; 3: 1-6. [CrossRef]
11. Bektas H, Vural G, Gumusyayla S, Deniz O, Alisik M, Erel O. Dynamic thiol-disulfide homeostasis in acute ischemic stroke patients. Acta Neurol Belg 2016; 116: 489-94. [CrossRef]
12. Korkmaz V, Kurdoglu Z, Alisik M, Turgut E, Sezgın O O, Korkmaz, H et al. Thiol/disulfide homeostasis in postmenopausal osteoporosis. J Endocrinol Invest 2017; 40: 431-5. [CrossRef]
13. Erel O, Neselioglu S. A novel and automated assay for thiol/disulphide homeostasis. Clin Biochem 2014; 47: 326-32. [CrossRef]
14. Soules MR, Sherman S, Parrott E, Rebar R, Santoro N, Utian W, et al. Executive summary: stages of reproductive aging workshop (STRAW). Climacteric 2001; 4: 267-72. [CrossRef]
15. Greene JG. Constructing a standard climacteric scale. Maturitas 1998; 29: 25-31. [CrossRef]
16. Rani V, Deep G, Singh RK, Palle K, Yadav UC. Oxidative stress and metabolic disorders: Pathogenesis and therapeutic strategies. Life Sci 2016; 148: 183-93. [CrossRef]
17. Jones DP, Liang Y. Measuring the poise of thiol/disulfide couples in vivo. Free Radic Biol Med 2009; 47: 1329-38. [CrossRef]
18. Oter S, Jin S, Cucullo L, Dorman HJD. Oxidants and antioxidants: friends or foes? Oxid Antioxid Med Sci 2012; 1: 1-4. [CrossRef]
19. Pansini F, Mollica G, Bergamini CM. Management of the menopausal disturbance and oxidative stress. Curr Pharm Des 2005; 11: 2063-73. [CrossRef]
20. Jacob SS, Somashekhar SP, Jacob SS. Role of Hormone Replacement Therapy (HRT) in Gynecological Cancers: Endocrinologist's Perspective. Indian Journal of Gynecologic Oncology 2016; 14: 1-6. [CrossRef]
21. Marjoribanks J, Farquhar C, Roberts H, Lethaby A, Lee J. Long-term hormone therapy for perimenopausal and postmenopausal women. Cochrane Database Syst Rev 2017; 1: CD004143. [CrossRef]
22. Cervellati C, Pansini FS, Bonaccorsi G, Bergamini CM, Patella A, Casali F et al. 17β-estradiol levels and oxidative balance in a population of pre-, peri-, and post-menopausal women. Gynecol Endocrinol 2011; 27: 1028-32. [CrossRef]
23. Vincent HK, Innes KE, Vincent KR. Oxidative stress and potential interventions to reduce oxidative stress in overweight and obesity. Diabetes Obes Metab 2007; 9: 813-39. [CrossRef]
24. Furukawa S, Fujita T, Shimabukuro M, Iwaki M, Yamada Y, Nakajima Y et al. Increased oxidative stress in obesity and its impact on metabolic syndrome. J Clin Invest 2017; 114: 1752-61. [CrossRef]
25. Cagnacci A, Cannoletta M, Palma F, Bellafronte M, Romani C, Palmieri B. Relation between oxidative stress and climacteric symptoms in early postmenopausal women. Climacteric 2015; 18: 631-6. [CrossRef]