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Evaluation of Oxidative Stress with "Dynamic Thiol/Disulfide Homeostasis" in Cases with Endometrioma

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ABSTRACT

Objective: This preliminary study aims to use "dynamic thiol/disulfide homeostasis" with the new Erel method to evaluate the effect of oxidative stress in patients with endometrioma.

Material and Method: The study group consisted of 40 cases with histologically confirmed endometrioma, and 40 women with no laparoscopically confirmed endometriosis were taken as the control group. Plasma thiol, total thiol (TT), and disulfide (SS) levels were measured with the new, fully automatic method described by Erel and Neselioglu. Serum Ca-125, sedimentation (Sed), C-reactive Protein (CRP), and thiol/disulfide levels were measured. The two groups' plasma thiol, total thiol, and disulfide levels were compared, and the relation between thiol/disulfide homeostasis and stage of the endometriosis, Ca-125, Sed, and CRP was evaluated.

Results: In cases with endometrioma, disulfide/native thiol (3,12±2,02, 2,05±1,21, p=0,005) and disulfide/total thiol ratios (3,50±2,52, 2,22±1,36, p=0,006) were significantly increased, native thiol (469,30±126,52, 571,72±125,32 p=0,00) total thiol levels (505,17 ± 133,88, 603,0±134,22 p=0,02) were significantly decreased when compared with the control group. There was a positive correlation between native thiol level and revised American Society for Reproductive Medicine Classification (r-ASRM) (p=0,041).

Conclusion: As expected, "dynamic thiol/disulfide homeostasis" with the new Erel method, the significant decrease in total thiol and native thiol levels, which are used as oxidative stress markers, and the increase in disulfide values demonstrated by this study support the hypothesis that oxidative stress plays a role in endometriosis and these markers can be used in the management of endometriosis.

Keywords: Disulfides, endometrioma, oxidative stress, thiols

INTRODUCTION

The presence of functional endometrium outside the uterine cavity is defined as endometriosis. Endometriosis is an estrogen-dependent, benign gynecological disease that affects an important percentage of women of reproductive age. Endometriosis is associated with an altered inflammatory response in the peritoneal cavity (1,2). The prevalence of endometriosis is estimated at around 20-50% in the infertile patient group (3,4). Almost 17 to 44 % of patients with endometriosis will have endometrioma in the future (5,6) . The most common symptoms are dysmenorrhea, dyspareunia, dyschezia, and dysuria according to the localization of the endometriotic implants (2).

Although the etiology and pathogenesis of endometriosis are not well known, many studies are investigating the relationship between endometriosis and oxidative stress as oxidative stress is considered to play an essential role in the formation and progression of the disease (8–11). The imbalance between the reactive oxygen species (ROS) products and the antioxidant capacity of the system in the organism is called oxidative stress (11,12). Oxidative stress is blamed for having an adverse effect on folliculogenesis, oocyte maturation, ovulation, and embryogenesis (13,14). Moreover, it causes oxidative damage of the proteins, lipids, carbohydrates, and DNA leading to cellular damage and dysfunction. These changes may affect folliculogenesis. In patients with endometriosis, impaired follicular development may lead to decreased oocyte and embryo quality, reduced fertilization, and implantation rate (15).

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Reactive sulfur derivatives are produced from thiols. Thiols are organic sulfur derivatives defined by the presence of sulfhydryl groups (-SH) in their active site. The thiol-redox system is very important for the normal function of the protein in the human body (16). When the sulfhydryl group of thiols undergoes oxidation reactions by oxygen radicals, disülfide bonds are formed. Disulfide bonds can be reduced to thiols again; thus, a dynamic thiol/disulfide homeostasis is maintained. A dynamic thiol/disulfide homeostasis status plays an important role in detoxification, apoptosis, regulation of enzymatic activity, and cellular signaling mechanisms (16,17). Abnormal thiol/disulfide homeostasis has been demonstrated to play a role in the pathogenesis of many conditions such as cardiovascular disease (CVD), menopause, osteoporosis, atopic dermatitis, cancer, rheumatoid arthritis, diabetes mellitus (18–23). In recent studies, disulfide and thiol levels of low molecular weight disulfide compounds in plasma were determined by using high performance liquid chromatography (HPLC) (24,25) fluorescent capillary electrophoresis (26) and bioluminescence systems (27). However, in these complex systems, removal of the remaining reductants for separation processes and precipitation of proteins is necessary (28). These pretreatment applications and measurement procedures are time-consuming and costly. However, to the best of our knowledge, there is no automatic colorimetric method for the measurement of plasma/serum dynamic disulfide levels, (28). While only a part of this bilateral dynamic thiol/disulfide balance was measured in the late seventies, today it's possible to measure the arms of the balance both separately and in combination with a new method developed by Erel and Neselioğlu (29).

Many diseases have been investigated in this new method in which oxidative stress balance is evaluated in the literature. We aimed to evaluate this balance in patients with endometrioma. The primary aim of the study is to compare the serum oxidative stress and the dynamic thiol/disulfide homeostasis status in patients with endometrioma. The secondary aim is to evaluate the correlation of the dynamic thiol/disulfide homeostasis with the severity of the disease and sedimentation, C-reactive protein (CRP), and Ca-125 values in patients with endometrioma.

MATERIAL and METHODS

Study design

This prospective study was carried out at the Infertility Department of the Etlik Zubeyde Hanim Women's Health Training and Research Hospital between April 2016 and December 2016. A total of 80 patients recruited to the study underwent laparoscopy of which 40 had removal of endometrioma and a confirmed histological diagnosis of the disease. Forty consecutive patients with a voluntary laparoscopic tubal ligation and no macroscopically observed endometriotic foci in the abdominal cavity during surgery were taken as the control group. The study was approved by the University of Health Sciences Turkey Etlik Zubeyde Hanim Women's Health Training and Research Hospital Ethics Committee (Date:08.04.2016, Decide No:2016/2). Informed consent was obtained from all the study participants.

Patient selection

Medical and gynecological histories of the study subjects were taken, followed by clinical and gynecological examinations. Sociodemographic and medical characteristics of the patients (age, parity, medical history, menstrual cycle pattern, presence of dysmenorrhea, dyspareunia, dysuria, dyschezia) were recorded. All patients were operated on laparoscopically by the same surgical team. Inclusion criteria were the presence of symptomatic endometrioma with a diameter >5 cm or symptomatic ruptured endometrioma with a diameter less than 5 cm. Patients with a systemic disease received hormonal treatment such as oral contraceptives, gonadotropin-releasing hormone agonist/antagonist, antidepressants, oral anticoagulants, non-steroidal anti-inflammatory drugs, and corticosteroids within the preceding 3 months were excluded from the study. Other exclusion criteria were a history of a disease that may affect inflammatory markers (gastrointestinal infection, upper respiratory-lower respiratory tract infections, genito-urinary system diseases). The inclusion criteria for the study group was having histopathologically confirmed endometrioma. Patients having a concomitant ovarian pathology accompanying endometrioma diagnosed during the operation or having a histopathology report that was not compatible with endometrioma were also excluded from the study. The stage of the cases with endometrioma was evaluated and recorded during laparoscopy by using the "revised American Society for Reproductive Medicine (r-ASRM) classification" (30).

Laboratory analysis

Venous blood samples were collected from the study and the control group into tubes containing ethylenediaminetetraacetic acid after overnight fasting before the operations, and the samples were processed within one hour of collection. The plasma samples were centrifuged at 1,500 g for 10 minutes before being stored at –80 ° C. Ca 125 IU/ml (Roche HITACHI COBAS 8000 Switzerland), sedimentation mm (Rapida Medical Limited Company ESR-40 Turkey), and C-Reactive Protein (CRP) mg/L (Beckman Coulter AU680 USA) levels were assessed with an automated chemiluminescence system from the blood samples taken routinely from the patients in the preoperative period.

Thiol/Disulfide homeostasis parameter measurements

Thiol/disulfide homeostasis tests (plasma native thiol, total thiol, and disulfide levels) were measured by a new and fully automated spectrophotometric method described by Erel and Neselioglu21. Disulfide bonds were first reduced to form free functional thiol groups with sodium borohydride (NaBH 4). The unused reductant (NaBH 4) was consumed and removed with formaldehyde to prevent the reduction of DTNB (5,5 ′ -dithiobis-(2-nitrobenzoic acid)), and all of the thiol groups, including the reduced and native thiol groups, were determined after the reaction with DTNB. One-half of the difference between total thiols and natural thiols provides the dynamic disulfide amount. Then the native and total thiols (TT), disulfide (SS) amounts, disulfide/total thiol (SS/TT), disulfide/native thiol (SS/NT), and native thiol/total thiol (NT/TT) ratios were calculated.

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Statistical analysis: Sample size calculation was based on G*Power Version 3.1.9.4 (Franz Faul, Universitat Kiel, Germany) with 0.05 alpha error end 95% power and thus 40 patients were recruited to each group in order to provide an effect strength of 0.708. Statistical analyses were performed using IBM SPSS 26.0 software (IBM Corp. Released 2019. IBM SPSS Statistics for Windows, Version 26.0. Armonk, NY: IBM Corp). The distributions of data were determined using the Kolmogorov–Smirnov test. Variables were expressed as the mean ± SD or median (minimum-maximum) and percentages. Parametric data were compared using the independent samples t-test, Pearsonc's correlations were performed to determine correlations between thiol/disulfide levels and Ca 125, CRP, sedimentation levels, and endometrioma stage. A p-value of less than 0.05 was accepted as statistically significant for all tests.

RESULTS

The distribution of sociodemographic characteristics of the patients in the study and the control group is given in Table-1. The mean age was similar in both groups; 33.1 ± 7.8 years in the study group and 35.1 ± 6.8 in the control group (p>0.05) (**Table1**). Overall three patients had dysmenorrhea and 4 patients had dyspareunia in the control group while 38 patients had either dysmenorrhea or dyspareunia. The symptoms of dysmenorrhea and dyspareunia were found to be statistically significantly higher in the endometrioma group (p<0.05), and none of the patients complained of dysuria and dyschezia in both groups.

Ca 125 values were found to be statistically significantly higher in the endometrioma group when compared with the control group (p=0.023) (**Table1**).

When total thiol (TT), native thiol (NT), disülphide (SS), disulfide/total thiol (SS/TT), disulfide/native thiol (SS/NT), native thiol/total thiol (NT/TT) values were measured for dynamic thiol hemostasis, two parameters; TT, and NT were statistically significantly lower in the endometrioma group (**Fig.1-2**), while SS/TT, SS/NT were found to be higher than the control group (**Table 2**). Although SS was found to be increased in the endometrioma group the difference was not statistically significant. (**Table 2**).

All the patients in the study group had stage 3-4 endometriosis. Out of 40 patients, 42,5% (n=17) had stage 3 endometriosis while the remaining 57,5% (n=23) were reported to have stage 4 endometrioma. Out of 40 patients, 27,5% (n=11) of the patients had bilateral endometriomas. In addition, when the cyst sizes were evaluated, a cyst diameter of 5 to 10 cm was observed in 70% (n=28) patients, and a cyst diameter >10 cm was detected in 22.5% (n=9) patients.

A positive correlation was found between bilaterality, Ca-125 levels, endometrioma diameter, and the r-ASRM score (p<0,05). Among the thiol parameters, a positive correlation was found between native thiol and the r-ASRM score, showing a higher native thiol level with increasing r-ASRM scores (p=0.041) (**Table 3**).

Table 1: Demographic and clinical characteristics of the endometrioma and the control group

Characteristics	Endometrioma group (n= 40) Mean ± Std	Control group (n= 40) Mean ± Std	p value
Age (years)	33,1 ± 7,8	35,1 ± 6,8	0,215
Dysmenorrhea (N,%)	25 (62,5%)	3 (7,5%)	<0,001*
Dyspareunia (N,%)	13 (32,5%)	4 (10%)	0,014*
C-Reactive Protein (CRP) mg/l	0,97±2,29	0,28±0,64	0,069
Sedimentation mm	18,1 ±13,1	11,9 ± 6,7	0,589
Ca-125 IU/ml	115,2 ± 277,6	13,3 ± 5,7	0,023*

Independent T Test applied *p<0,05 is statistically significant

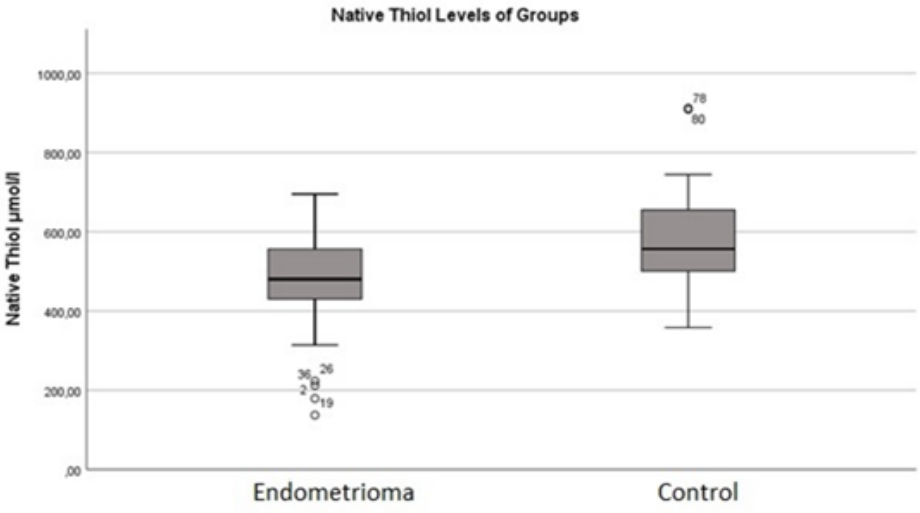

Figure 1. Native Thiol Levels of the Endometrioma and Control Group

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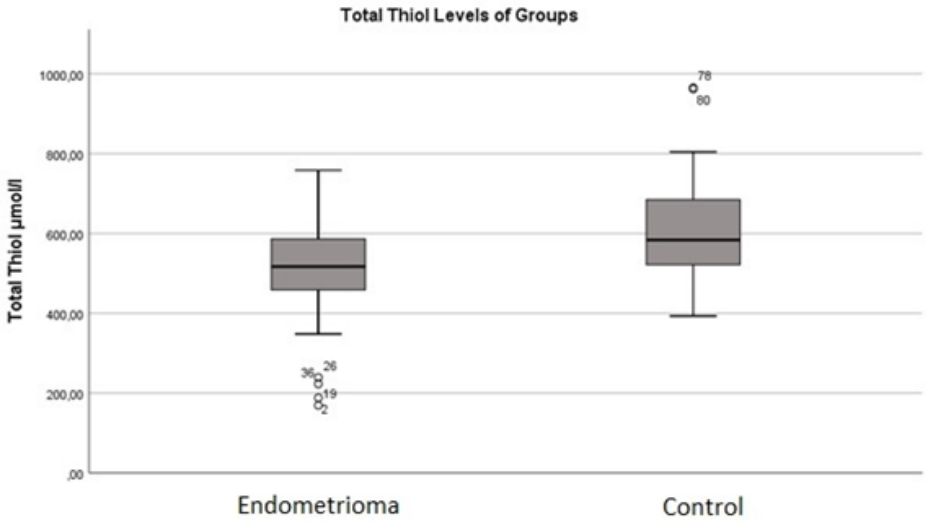

Figure 2. Total Thiol Levels of Endometrioma and Control Groups

Table 2: Comparison of oxidative stress parameters between the endometrioma and the control group

Thiol Parameters	Endometrioma Group (N:40) Mean±Std	Control Group (N:40) Mean±Std	p value
Total Thiol (TT) µmol/l	505,17 ± 133,88	603,0±134,22	0,02*
Native Thiol (NT) µmol/l	469,30±126,52	571,72±125,32	0,00*
Disulfide (SS) µmol/l	17,42±11,17	15,32±8,19	0,341
Disulfide / Native Thiol*100	3,12±2,02	2,05±1,21	0,005*
Disulfide / Total Thiol*100	3,50±2,52	2,22±1,36	0,006*
Native Thiol / Total Thiol*100	92,37±4,16	94,40±2,40	0,007*

Independent T Test applied *P<0,05 is statistically significant

Table 3: Cross-correlation of plasma Thiol-Disulfide levels and bilaterality, diameter, and the r-ASRM score of the endometrioma cases

		Bilaterality	Diameter	Postop Stage	Sed.	C-RP	Ca 125
Bilaterality	Corr.	1,000	,583**	,416**	0,062	-0,021	,279*
	Sig.		**0,000**	**0,008**	0,588	0,856	**0,012**
	N	80	40	40	80	80	80
Diameter	Corr.	,583**	1,000	,431**	0,183	0,014	0,274
	Sig.	**0,000**		**0,006**	0,259	0,932	0,087
	N	40	40	40	40	40	40
Postop. Stage	Corr.	,416**	,431**	1,000	-0,147	0,002	0,147
	Sig.	**0,008**	**0,006**		0,365	0,989	0,366
	N	40	40	40	40	40	40

		Total Thiol	Native Thiol	Disulfide	Disulfide /TT	Disulfide /NT	NT /TT
Bilaterality	Corr.	-0,133	-0,155	0,064	0,186	0,148	-0,172
	Sig.	0,240	0,170	0,574	0,099	0,190	0,126
	N	80	80	80	80	80	80
Diameter	Corr.	0,154	0,114	0,167	0,163	0,132	-0,125
	Sig.	0,342	0,484	0,304	0,315	0,416	0,444
	N	40	40	40	40	40	40
Postop. Stage	Corr.	0,252	,324*	-0,053	-0,122	-0,137	0,152
	Sig.	0,117	**0,041**	0,747	0,454	0,400	0,348
	N	40	40	40	40	40	40

* Correlation is significant at the 0.05 level (2-tailed). Ca-125:Cancer Antigen 125, C-RP: c-Reactive Protein, r-ASRM Score: Revised American Society for Reproductive Medicine Classification Score, Sed: Sedimentation,

DISCUSSION

This is a preliminary study evaluating the relationship of disulfide, native thiol, and total thiol levels with endometrioma. Previous methods used to measure thiol and disulfide levels failed to reflect plasma thiol/disulfide homeostasis. In this study, a novel technique (29) was used to cumulatively measure the thiol/disulfide balance by measuring both thiol and disulfide levels. Cumulative measurement of thiol/disulfide balance, and both thiol and disulfide levels are performed with this new method as it has the advantage of being user-friendly and cost-effective.

Türkyılmaz et al. (31) evaluated the patients with endometrioma, serum native thiol and total thiol levels in the study group were significantly lower than the control group, while serum catalase levels were significantly higher when compared with the control group. Thiols act as oxygen scavengers during oxidative stress and provide redox balance. Plasma values of thiols are good indicators of tissue redox potentials (32). In the presented study group with histologically proven endometrioma, increased disulfide, disulfide/native thiol, and disulfide/total thiol levels, and decreased native thiol, total thiol levels were detected.

Endometriosis is a multifactorial degenerative disease that creates a chronic inflammatory injury. Thus, glandular fibrosis occurs with the accumulation of extracellular matrix (ECM) components on tissues (33,34). Although the exact pathophysiology of endometriosis is not clear, the immune system changes and chronic inflammation are thought to be very important in the progression of the disease. In a systematic review published in 2012, a total of 36 oxidative stress biomarkers were evaluated in patients with endometriosis. The authors reported that 23 of the 36 markers were significantly higher in patients with endometriosis in comparison to the control group. These findings emphasize the importance of oxidative stress in the pathogenesis and progression of endometriosis (11). We also observed an increase in Sedimentation, C-Reactive Protein, and Ca-125 levels in the endometrioma group in our study.

Various non-hormonal treatment modalities that demonstrate antioxidant, anti-inflammatory, anti-tumor, anti-angiogenic, and anti-metastatic properties, such as curcumin, Vitamin C have been used to regress the endometriotic lesions and reduce endometriosis-related symptoms (35, 36). Guney et al. (37) reported the anti-oxidant, anti-inflammatory, and immunomodulatory effect of melatonin in a rat model after showing regression of the endometriotic implants with melatonin. In rats treated with melatonin, the level of malondialdehyde (MDA) in the endometriotic implants decreased statistically significantly and activities of antioxidant enzymes superoxide dismutase (SOD) and catalase (CAT) were significantly increased thus the endometriotic foci regressed. Antioxidant treatments in pre-clinic studies and regression of the endometriotic foci in animal studies direct the researchers to investigate new treatment modalities.

A positive correlation was found between cyst diameter and serum Ca-125 level and r-ASRM scores in patients with endometrioma. In addition, a positive correlation was found between native thiol and the r-ASRM scores. Recently, the relationship between the severity of endometriosis defined by using the r-ASRM score and oxidative stress was investigated by Amreen et al. The researchers reported the lowest median superoxide dismutase activity and glutathione peroxidase and highest lipid peroxide median activity in patients with the severe endometriosis cases who defined by r-ASRM score (38). This new thiol-disulfide method can be used by the clinician before the operation to predict the severity of the disease.

The strength of this study is the utilization of the novel thiol/disulfide homeostasis method in understanding the relationship between the severity of endometrioma and oxidative stress. The limitation of this study is the restricted number of patients recruited. Despite this limitation, the presented research is a pioneering study that evaluated a novel technique in cases with endometrioma.

CONCLUSION

Although the pathophysiology of endometrioma is unknown, it is recognized as a multifactorial disease that is exacerbated by oxidative stress. The significant decrease in serum total thiol, native-thiol levels and increased disulfide levels observed in patients with endometrioma supports the hypothesis that oxidative stress plays a role in endometriosis. These non-invasive serum markers can assist to the clinicians during the treatment, pre-operation, and follow-up of the disease. This preliminary study demonstrates novel thiol/disulfide homeostasis in patients with endometrioma. However, there is a need for more studies using this new method with a larger number of cases recruited.

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Ethical approval: All procedures performed in studies involving human participants were in accordance with the institutional and/or national research committee's ethical standards and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards.

REFERENCES

1. Giudice LC, Kao LC. Endometriosis. Lancet [Internet]. 2004 Nov 13 [cited 2022 Mar 17];364(9447):1789–99. Available from: https://pubmed.ncbi.nlm.nih.gov/15541453/

2. Vercellini P, Viganò P, Somigliana E, Fedele L. Endometriosis: Pathogenesis and treatment. Nature Reviews Endocrinology [Internet]. 2014 [cited 2021 Jun 19];10(5):261–75. Available from: https://pubmed.ncbi.nlm.nih.gov/24366116/

3. Balasch J, Creus M, Fábregues F, Carmona F, Ordi J, Martinez-Román S, et al. Visible and non-visible endometriosis at laparoscopy in fertile and infertile women and in patients with chronic pelvic pain: a prospective study. Hum Reprod [Internet]. 1996 [cited 2022 Mar 17];11(2):387–91. Available from: https://pubmed.ncbi.nlm.nih.gov/8671229/

4. Meuleman C, Vandenabeele B, Fieuws S, Spiessens C, Timmerman D, D'Hooghe T. High prevalence of endometriosis in infertile women with normal ovulation and normospermic partners. Fertil Steril [Internet]. 2009 Jul [cited 2022 Mar 17];92(1):68–74. Available from: https://pubmed.ncbi.nlm.nih.gov/18684448/

5. S Jenkins, D L Olive, A F Haney. Endometriosis: pathogenetic implications of the anatomic distribution - PubMed [Internet]. Obstet Gynecol. 1986 [cited 2022 Mar 17]. p. 335–8. Available from: https://pubmed.ncbi.nlm.nih.gov/3945444/

6. Redwine DB. Ovarian endometriosis: a marker for more extensive pelvic and intestinal disease. Fertil Steril [Internet]. 1999 Aug [cited 2022 Mar 17];72(2):310–5. Available from: https://pubmed.ncbi.nlm.nih.gov/10439002/

7. Jenkins S, Olive DL, Haney AF. Endometriosis: Pathogenetic implications of the anatomic distribution. Obstetrics and Gynecology [Internet]. 1986 [cited 2022 Mar 17];67(3):335–8. Available from: https://pubmed.ncbi.nlm.nih.gov/3945444/

8. Gupta S, Harlev A, Agarwal A. Endometriosis: a comprehensive update. 2015 [cited 2022 Mar 17]; Available from: https://link.springer.com/content/pdf/10.1007/978-3-319-18308-4.pdf

9. Turgut A, Özler A, Görük NY, Tunç SY, Evliyaoğlu O, Gül T. Copper, ceruloplasmin and oxidative stress in patients with advanced-stage endometriosis. Eur Rev Med Pharmacol Sci [Internet]. 2013 [cited 2022 Mar 17];17(11):1472–8. Available from: https://pubmed.ncbi.nlm.nih.gov/23771536/

10. Matsuzaki S, Schubert B. Oxidative stress status in normal ovarian cortex surrounding ovarian endometriosis. Fertility and Sterility [Internet]. 2010 May 1 [cited 2021 Jun 19];93(7):2431–2. Available from: https://pubmed.ncbi.nlm.nih.gov/19819438/

11. Carvalho LFP, Samadder AN, Agarwal A, Fernandes LFC, Abrão MS. Oxidative stress biomarkers in patients with endometriosis: Systematic review. Archives of Gynecology and Obstetrics [Internet]. 2012 Oct 1 [cited 2021 Jun 19];286(4):1033–40. Available from: https://pubmed.ncbi.nlm.nih.gov/22791380/

12. Augoulea A, Mastorakos G, Lambrinoudaki I, Christodoulakos G, Creatsas G. The role of the oxidative-stress in the endometriosis-related infertility [Internet]. Vol. 25, Gynecological Endocrinology. Gynecol Endocrinol; 2009 [cited 2021 Jun 19]. p. 75–81. Available from: https://pubmed.ncbi.nlm.nih.gov/19253102/

13. da Broi MG, Jordão-Jr AA, Ferriani RA, Navarro PA. Oocyte oxidative DNA damage may be involved in minimal/mild endometriosis-related infertility. Molecular Reproduction and Development [Internet]. 2018 Feb 1 [cited 2021 Jun 19];85(2):128–36. Available from: https://pubmed.ncbi.nlm.nih.gov/29247565/

14. da Broi MG, de Albuquerque FO, de Andrade AZ, Cardoso RL, Jordão Junior AA, Navarro PA. Increased concentration of 8-hydroxy-2′-deoxyguanosine in follicular fluid of infertile women with endometriosis. Cell and Tissue Research [Internet]. 2016 Oct 1 [cited 2021 Jun 19];366(1):231–42. Available from: https://pubmed.ncbi.nlm.nih.gov/27250533/

15. Garrido N, Pellicer A, Remohí J, Simón C. Uterine and ovarian function in endometriosis [Internet]. Vol. 21, Seminars in Reproductive Medicine. Semin Reprod Med; 2003 [cited 2021 Jun 19]. p. 183–91. Available from: https://pubmed.ncbi.nlm.nih.gov/12917788/

16. Biswas S, Chida AS, Rahman I. Redox modifications of protein-thiols: Emerging roles in cell signaling [Internet]. Vol. 71, Biochemical Pharmacology. Elsevier Inc.; 2006 [cited 2021 Jun 19]. p. 551–64. Available from: https://pubmed.ncbi.nlm.nih.gov/16337153/

17. Jones DP, Liang Y. Measuring the poise of thiol/disulfide couples in vivo [Internet]. Vol. 47, Free Radical Biology and Medicine. Free Radic Biol Med; 2009 [cited 2021 Jun 19]. p. 1329–38. Available from: https://pubmed.ncbi.nlm.nih.gov/19715755/

18. Uysal P, Avcil S, Neşelioğlu S, Biçer C, Çatal F. Association of oxidative stress and dynamic thiol-disulphide homeostasis with atopic dermatitis severity and chronicity in children: a prospective study. Clinical and Experimental Dermatology [Internet]. 2018 Mar 1 [cited 2021 Jun 19];43(2):124–30. Available from: https://pubmed.ncbi.nlm.nih.gov/29164676/

19. Matteucci E, Giampietro O. Thiol signalling network with an eye to diabetes. Molecules [Internet]. 2010 Dec [cited 2021 Jun 19];15(12):8890–903. Available from: https://pubmed.ncbi.nlm.nih.gov/21135801/

20. Go YM, Jones DP. Cysteine/cystine redox signaling in cardiovascular disease [Internet]. Vol. 50, Free Radical Biology and Medicine. Free Radic Biol Med; 2011 [cited 2021 Jun 19]. p. 495–509. Available from: https://pubmed.ncbi.nlm.nih.gov/21130865/

21. Tetik S, Ahmad S, Alturfan AA, Fresko I, Disbudak M, Sahin Y, et al. Determination of oxidant stress in plasma of rheumatoid arthritis and primary osteoarthritis patients. Indian Journal of Biochemistry and Biophysics. 2010 Dec;47(6):353–8.

22. Rodrigues SD, Batista GB, Ingberman M, Pecoits-Filho R, Nakao LS. Plasma cysteine/cystine reduction potential correlates with plasma creatinine levels in chronic kidney disease. Blood Purification [Internet]. 2013 Mar [cited 2021 Jun 19];34(3–4):231–7. Available from: https://pubmed.ncbi.nlm.nih.gov/23147870/

23. Korkmaz V, Kurdoglu Z, Alisik M, Turgut E, Sezgın OO, Korkmaz H, et al. Thiol/disulfide homeostasis in postmenopausal osteoporosis. Journal of Endocrinological Investigation [Internet]. 2017 Apr 1 [cited 2021 Jun 19];40(4):431–5. Available from: https://pubmed.ncbi.nlm.nih.gov/27858341/

24. Chen W, Zhao Y, Seefeldt T, Guan X. Determination of thiols and disulfides via HPLC quantification of 5-thio-2-nitrobenzoic acid. J Pharm Biomed Anal [Internet]. 2008 Dec 15 [cited 2022 Jul 2];48(5):1375–80. Available from: https://pubmed.ncbi.nlm.nih.gov/18926658/

25. Głowacki R, Bald E. Fully automated method for simultaneous determination of total cysteine, cysteinylglycine, glutathione and homocysteine in plasma by HPLC with UV absorbance detection. J Chromatogr B Analyt Technol Biomed Life Sci [Internet]. 2009 Oct 15 [cited 2022 Jul 2];877(28):3400–4. Available from: https://pubmed.ncbi.nlm.nih.gov/19559659/

26. Carru C, Deiana L, Sotgia S, Pes GM, Zinellu A. Plasma thiols redox status by laser-induced fluorescence capillary electrophoresis. Electrophoresis [Internet]. 2004 Mar [cited 2022 Jul 2];25(6):882–9. Available from: https://pubmed.ncbi.nlm.nih.gov/15004850/

27. Mourad T, Min KL, Steghens JP. Measurement of oxidized glutathione by enzymatic recycling coupled to bioluminescent detection. Anal Biochem [Internet]. 2000 Aug 1 [cited 2022 Jul 2];283(2):146–52. Available from: https://pubmed.ncbi.nlm.nih.gov/10906234/

28. Winther JR, Thorpe C. Quantification of thiols and disulfides. Biochim Biophys Acta [Internet]. 2014 Feb [cited 2022 Jul 2];1840(2):838–46. Available from: https://pubmed.ncbi.nlm.nih.gov/23567800/

29. Erel O, Neselioglu S. A novel and automated assay for thiol/disulphide homeostasis. Clinical Biochemistry [Internet]. 2014 Dec 1 [cited 2021 Jun 19];47(18):326–32. Available from: https://pubmed.ncbi.nlm.nih.gov/25304913/

30. Brosens IA, Cornillie F, Koninckx P, Vásquez G. Evolution of the Revised American Fertility Society Classification of Endometriosis. [Internet]. Vol. 44, Fertility and sterility. Fertil Steril; 1985 [cited 2021 Jun 19]. p. 714–6. Available from: https://pubmed.ncbi.nlm.nih.gov/4054355/

31. Turkyilmaz E, Yildirim M, Cendek BD, Baran P, Alisik M, Dalgaci F, et al. Evaluation of oxidative stress markers and intra-extracellular antioxidant activities in patients with endometriosis. European Journal of Obstetrics and Gynecology and Reproductive Biology [Internet]. 2016 Apr 1 [cited 2021 Jun 19];199:164–8. Available from: https://pubmed.ncbi.nlm.nih.gov/26930044/

32. Moriarty-Craige SE, Jones DP. Extracellular thiols and thiol/disulfide redox in metabolism [Internet]. Vol. 24, Annual Review of Nutrition. Annu Rev Nutr; 2004 [cited 2021 Jun 19]. p. 481–509. Available from: https://pubmed.ncbi.nlm.nih.gov/15189129/

33. Hoffmann C, Ellenberger C, Mattos RC, Aupperle H, Dhein S, Stief B, et al. The equine endometrosis: New insights into the pathogenesis. Animal Reproduction Science [Internet]. 2009 Apr [cited 2021 Jun 19];111(2–4):261–78. Available from: https://pubmed.ncbi.nlm.nih.gov/18468817/

34. Aresu L, Benali S, Giannuzzi D, Mantovani R, Castagnaro M, Falomo ME. The role of inflammation and matrix metalloproteinases in equine endometriosis. Journal of Veterinary Science [Internet]. 2012 Jun [cited 2021 Jun 19];13(2):171–7. Available from: https://pubmed.ncbi.nlm.nih.gov/22705739/

35. Arablou T, Kolahdouz-Mohammadi R. Curcumin and endometriosis: Review on potential roles and molecular mechanisms [Internet]. Vol. 97, Biomedicine and Pharmacotherapy. Elsevier Masson SAS; 2018 [cited 2021 Jun 19]. p. 91–7. Available from: https://pubmed.ncbi.nlm.nih.gov/29080464/

36. Erten OU, Ensari TA, Dilbaz B, Cakiroglu H, Altinbas SK, Çaydere M, et al. Vitamin C is effective for the prevention and regression of endometriotic implants in an experimentally induced rat model of endometriosis. Taiwanese Journal of Obstetrics and Gynecology [Internet]. 2016 Apr 1 [cited 2021 Jun 19];55(2):251–7. Available from: https://pubmed.ncbi.nlm.nih.gov/27125410/

37. Güney M, Oral B, Karahan N, Mungan T. Regression of endometrial explants in a rat model of endometriosis treated with melatonin. Fertility and Sterility [Internet]. 2008 Apr [cited 2021 Jun 19];89(4):934–42. Available from: https://pubmed.ncbi.nlm.nih.gov/17582405/

38. Amreen S, Kumar P, Gupta P, Rao P. Evaluation of Oxidative Stress and Severity of Endometriosis. J Hum Reprod Sci [Internet]. 2019 Jan 1 [cited 2022 Mar 17];12(1):40–6. Available from: https://pubmed.ncbi.nlm.nih.gov/31007466/