Correlation between oxidative stress and level of gamma glutamyl transpeptidase in pre and post-menopausal women

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Abstract

Introduction: Menopause is the permanent end of menstruation and fertility. It is a natural process and not a disease. Menopause can lead to a decrease in estrogen production. Estrogen, an immune-modulating hormone is associated with proper functioning of the immune system. Oxidative stress is defined as an imbalance between production of free radicals and reactive metabolites. Normally, antioxidants neutralize ROS and thus help to prevent over exposure from oxidative stress. The association of this increase in serum GGT with enhanced oxidative stress and reduced antioxidant defense system in the post-menopausal women may lead to the speculation that GGT could be considered an index or a marker of oxidative stress.
Objective: To measure the Oxidative Stress and estrogen (Estradiol) in pre- and post-menopausal woman and correlate it with the level of enzymatic markers Gamma glutamyl transferase (GGT) in serum with the amount of oxidative stress in pre- and post-menopausal women.
Material and Method: The study was carried out in the Department of Biochemistry, NIMS Medical College and Hospital, Shobha Nagar, Jaipur, Rajasthan. A total of 100 patients attending Obs-Gynaecology OPD were taken, out of which 50 were Post-menopausal women and 50 were Pre-menopausal women.
Results: The level of oxidative stress was high in post-menopausal female as compared to pre-menopausal female. The level of Gamma glutamyl transferase (GGT) was higher in postmenopausal women as compared to premenopausal women. The level of hormone estradiol in postmenopausal women is lower than the premenopausal women indicating that oxidative stress decreases the level of antioxidant property of estrogen hormone (Estradiol). The study highlights the role of Gamma GT as an indicator of oxidative stress.

Keywords: Menopause, estrogen, oxidative stress, free radicals, estradiol

Introduction

Ovaries are the organs of female reproductive system, which secrete female hormones estrogen and progesterone. Estrogen and progesterone are responsible for menstruation (monthly periods). When the secretions of these hormones are gradually reduced by ovaries, the process of termination of menstrual cycles start. It is a natural process and not a disease.[1] Menopause literally means the "end of monthly cycles" (the end of monthly periods or menstruation), from the Greek word pausis (cessation) and the root men-(month). Menopause is an event that typically occurs in women in midlife, during their late 40s or early 50s, and it signals the end of the fertile phase of a woman's life. [2] Estrogen (E2) is a C-18 steroid hormone with a phenolic ring. It is the most important natural estrogen present in male and female, whereas estrone (E1) is a less potent estrogen than estradiol and is the major circulating estrogen in the postmenopausal women. The main source of estrogen in the premenopausal women is the dominant follicle and its subsequent development into the corpus luteum after ovulation. [3] Menopause can lead to a decrease in estrogen production. Estrogen, an immune-modulating hormone is associated with proper functioning of the immune system. Because estrogen production decreases following menopause, the immune system of post-menopausal women may be compromised. The immune system encompasses an array of defenses that help to guard against the development of several diseases, some of them age- related. An increase in oxidative stress and a decrease in estrogen places postmenopausal women at increased risk for several diseases. [4] Oxidative stress is defined as an imbalance between production of free radicals and reactive metabolites. Oxidative stress may be involved in many different types of diseases such as coronary artery disease, stroke, arthritis and cancer. [5] The damage to the cell because of decreased estrogen production combined with DNA damage from reactive oxygen species (ROS) alters the mitochondrial physiology that may contribute to a greater cellular stress response, cell growth arrest, and subsequent apoptosis. [6] Specifically, in the ovary, estrogen is produced from the conversion of androgens via the enzyme aromatase. Moreover, estrogen is synthesized in three forms: Estradiol, estriol, and estrone. Specifically, 17β-estradiol is the most common and potent form of estrogen predominating during the premenopausal and perimenopausal periods, whereas estrone, the much weaker form, is prevalent during the postmenopausal phase. The latter form of estrogen is normally produced from the conversion of androstenedione in adipose tissue and the liver. Estrogen is synthesized in smaller amounts by other tissues, such as adrenal glands, fat cells, breast tissue, and hepatocytes. [7] Gamma-glutamyl transferase (GGT) is an enzyme involved in the transfer of the γ-glutamyl residue from γ-glutamyl peptides to amino acids, H_2O and other small peptides. In most biological systems, glutathione serves as the γ-glutamyl donor. [19] On the other hand, GGT is also involved in the synthesis of glutathione. [8] The intracellular glutathione (GSH) level depends upon the equilibrium between processes during which it is consumed and its biosynthesis is limited by cysteine availability. [9] The availability of cysteine, necessary for the biosynthesis of cellular glutathione, the most important cell antioxidant, depends upon gamma glutamyl

transpeptidase (GGT) activity; hence this enzyme may play an important role in the anti-oxidative defense system of the cell. [10] Plasma concentrations of estrogens fall and those of follicle stimulating hormone (FSH), and, to a lesser extent, luteinizing hormone (LH) increases after removal of the negative feedback to the pituitary. [11] The association of this increase in serum GGT with enhanced oxidative stress and reduced antioxidant defense system in the post- menopausal women may lead to the speculation that GGT could be considered an index or a marker of oxidative stress. [12]
The aim of the present study was to measure the Oxidative Stress and estrogen (Estradiol) in pre-and post-menopausal woman and correlate it with the level of enzymatic markers Gamma glutamyl transferase (GGT) in serum with the amount of oxidative stress in pre-and post-menopausal women.

Aims & Objectives

The overall aims of the study were:

Measure the Oxidative Stress and estrogen (Estradiol) in pre- and post-menopausal woman and correlate it with the level of enzymatic markers Gamma glutamyl transferase (GGT) in serum with the amount of oxidative stress in pre- and post-menopausal women.

Material & Methods

The study was carried out in the Department of Biochemistry, NIMS Medical College and Hospital, Shobha Nagar, Jaipur, Rajasthan.

Study population and selection criteria

A total of 100 patients attending Obs-Gynaecology OPD were taken, out of which 50 were post-menopausal women and 50 were pre-menopausal women.

Group I: Premenopausal women (n = 50).

The normally menstruating, and women with some sort of menstrual disorders e.g., irregular menses, menorrhagia, etc. were included in this group; but women experiencing amenorrhea were excluded. These selected subjects were considered as control group and their age range was 25-40 years.

Group II: Postmenopausal women (n = 50).

These women who had at least one year of amenorrhea and were not receiving hormone replacement therapy. These selected subjects were considered as study group and had an age range of 41-70 years.

Exclusion criteria

The subjects suffering from hypertension, cardiovascular diseases, diabetes, venereal diseases.
The subject showing any pathology (including carcinoma).
Women taking oral contraceptives, antioxidants, or any other drug.
Women of age <25.
Based on the history, the patients with any concurrent sickness.

Pregnant or lactating females.

Sample collection procedures and instruments
Using aseptic techniques, collected venous blood which divided into:
2.0 ml of fasting sample (8-12 hrs.) was collected in a plain tube for estimating estradiol hormone and Gamma GT.
3.0 ml of whole blood was collected in EDTA vial for the estimation of Ferric Reducing Ability of Plasma (FRAP).

Biochemical parameters: All subjects were screened for their general and medical history, especially menstrual. Clinical examination was carried out with the aid and advice of a competent gynecologist. We evaluated the level of estradiol hormone, gamma glutamyl transferase and oxidative stress by FRAP method in the serum/plasma the subjects to find out the correlation, between pre menopause and post menopause women.

Quality control: Internal and external quality control of convenient samples was done as per standard operative procedures (SOPs) of the Department of Biochemistry.

Data analysis and interpretation: The collected data was entered in Excel spreadsheet. Mean and standard deviation was calculated for quantitative data.

Observation

Table 1: Comparison of Ferric reducing ability of plasma and gamma glutamyl transpeptidase of control group & study group

Parameter	Control group (n=50)	Study group (n=50)
Ferric Reducing Ability of Plasma (FRAP). (μ moles of $FeSO_4$ equivalent/L of plasma)	916.08 ± 131.08	483.12 ± 53.64 *
Gamma G T (U/L)	29.86 ± 7.73	59.87 ± 6.35 *

*= $p < 0.001$ (statistically significant)

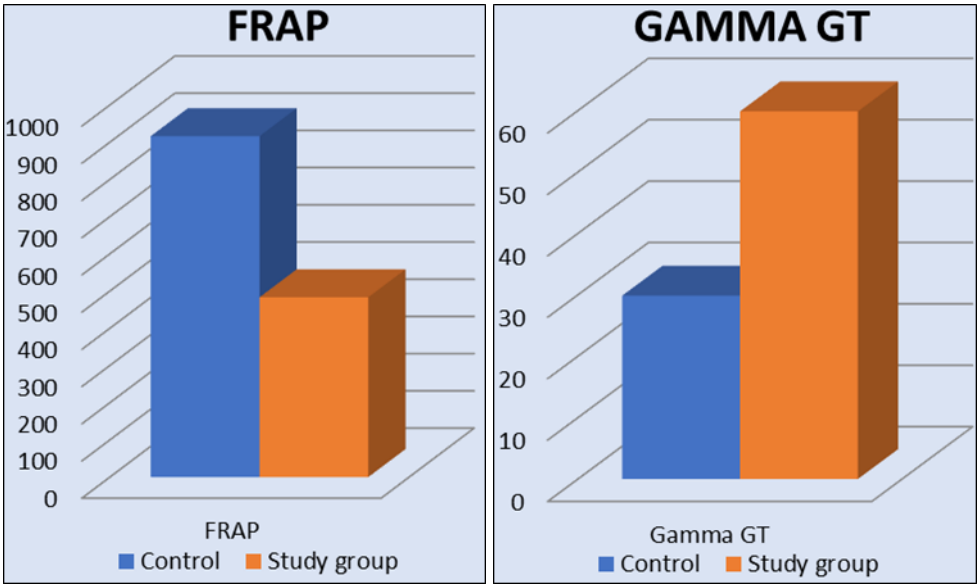

Fig 1: Comparison of Ferric reducing ability of plasma and gamma glutamyl transpeptidase of control group & study group

In these study the value of FRAP and Gamma GT were compared between control and study group. The level of FRAP was significantly decreased in study group compared to control ($p< 0.001$). The value of Gamma GT was significantly increased in study group compared to control ($p<0. 001$).

Table 2: Comparison of FRAP and E2 in Control and Study group

Parameter	Control group (n=50)	Study group (n=50)
Ferric Reducing Ability of Plasma (FRAP). (µ moles of $FeSO_4$ equivalent/L of plasma)	916.08 ± 131.08	483.12 ± 53.64 *
Estradiol (pg/ml)	152.95 ± 60.18	18.32 ± 5.27 *

*= $p< 0.001$(statistically significant)

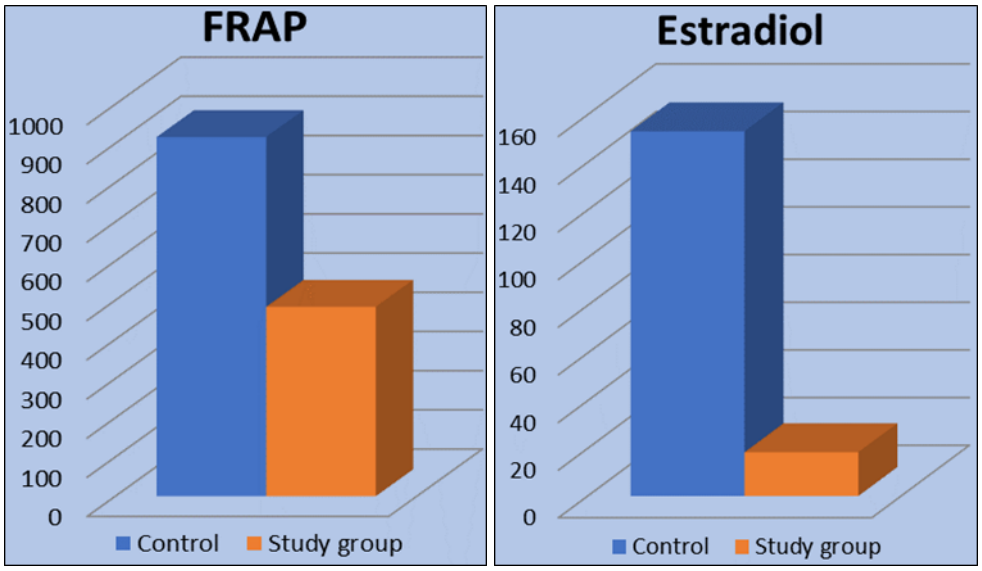

Fig 2: Comparison of FRAP and estradiol in Control and Study group

In these studies, value of FRAP and E2 were compared between control and study group. The levels of FRAP and E2 were found to be significantly decreased in study group as compared to control ($p< 0.001$).

Table 3: Comparison of activity of Gamma GT and Estradiol in Control and Study group

Parameter	Control group (n=50)	Study group (n=50)
Gamma GT (U/L)	29.86 ± 7.73	59.87 ± 6.35 *
Estradiol (Pg/ml)	152.95 ± 60.18	18.32 5.27 *

*= $p< 0.001$ (statistically significant)

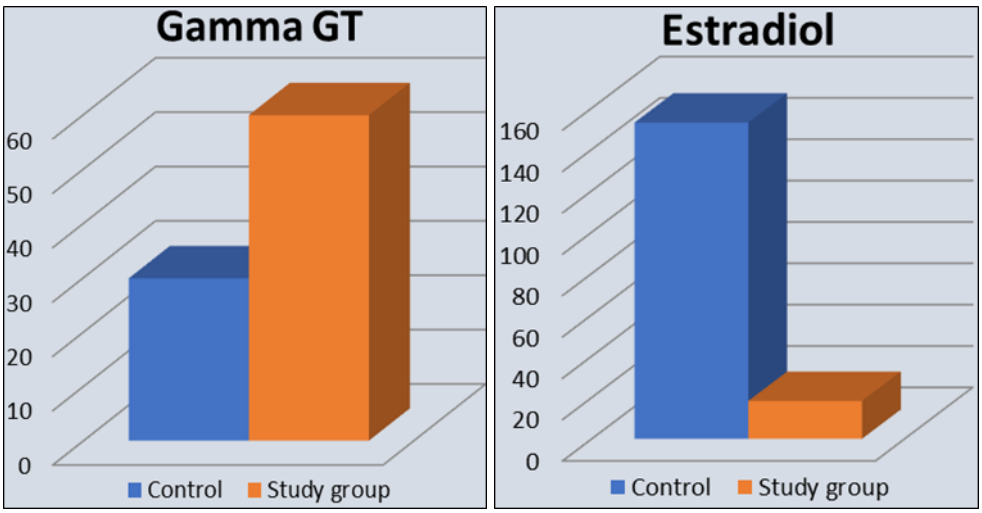

Fig 3: Comparison of activity of Gamma GT and E2 in Control and Study group

In these studies, value of Gamma GT and Estradiol were compared with control and study group. The levels of Gamma GT were found to be significantly increased as study group as compared to control ($p<0.001$). The value of Estradiol in study group was decreased as compared to control group ($p<0.001$).

Discussion

In the study, the pattern and correlation of oxidative stress (as measure of ferric reducing ability of plasma, FRAP), Estradiol hormone, and gamma GT has been examined in healthy pre- and post-menopausal women. The present study included total 100 subjects. All subjects were divided into two groups. Control group consisted of 50 normal healthy pre-menopausal individuals while the study group consisted of 50 post-menopausal women. The results of the study showed a significant variation in the oxidative stress and Gamma GT among the study group and controls. The Gamma GT was significantly increased in study group compared to control group because of reduce antioxidant ability. The level of these Gamma GT also varied with the level of oxidative stress represented by ferric reducing ability of plasma (FRAP). Menopausal phase in a woman's life is an important physiological phenomenon, which is associated with cessation of menstrual cycle due to loss of ovarian function. The

deficiency of estrogen in postmenopausal women develops oxidative stress due to release of free radical or reactive oxygen species (ROS) and becomes the cause of various pathologies.

Various studies conducted by different authors, exhibit a fair measure of ambiguity regarding the effect of menopause on oxidative stress levels. We reported that post-menopausal women had significantly lower concentrations of estrogen and increased concentration of Gamma GT in comparison to pre-menopausal women. We also observed that FRAP levels were decreased in post-menopausal women as compared with normally healthy premenopausal women; however, the variations were statistically significant.

Conclusion

The level of oxidative stress as measured by the ferric reducing ability of plasma was high in post-menopausal female as compared to pre-menopausal female. The ferric reducing ability of plasma (FRAP) was low in post-menopausal women and high in pre-menopausal women.

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