

The correlation between plasma homocysteine and malondialdehyde levels in preeclampsia

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Submitted: November 20, 2002

Accepted: December 5, 2002

Key words: malondialdehyde: homocysteine: preeclampsia

Neuroendocrinol Lett 2003; 24(6):445-448 NEL240603A10 Copyright © Neuroendocrinology Letters www.nel.edu

Abstract

OBJECTIVE: To investigate the possible correlation between plasma malondialdehyde and homocysteine levels of preeclamptic patients.

DESIGN: Venous blood samples of 20 preeclamptics and 20 healthy pregnant controls were collected. Plasma malondialdehyde and homocysteine concentrations were measured and the correlation between them was investigated. Mann Whitney U test and Spearman correlation analysis were used for statistical analysis.

SETTING: University of Firat, Medical School.

RESULTS: Plasma malondialdehyde and homocysteine concentrations were higher in preeclamptic patients ($p < 0.05$) and a positive correlation between these parameters was found ($r: 0.77, p < 0.01, n: 20$).

CONCLUSION: Our results may put forward some new strategies in the research of etiopathogenesis and treatment of preeclampsia.

Abbreviations & Units

MDA: Malondialdehyde
EDTA: Ethylenediaminetetraacetate
ELISA: Enzyme-linked immunosorbent assay
NO: Nitric oxide
 μmol : Micromol
nmol: Nanomol
g: Gram
mg: milligram
U/L: Unit/Liter

Introduction

Preeclampsia is a pregnancy-specific disorder characterized by vasospasm and endothelial dysfunction, and complicates 7-10 % of all gestations with serious fetomaternal morbidity and mortality. Aetiology of preeclampsia is still obscured but one of the most favored hypotheses is the endothelial dysfunction secondary to the peroxidation of membrane lipids [1]. Decreased antioxidant activity and increased lipid peroxides was shown clearly in preeclamptics [2].

Homocysteine is an intermediate amino acid in the methionine metabolism, which does not take place in the structure of proteins. It is eliminated from the body via conversion into 1- cystathione by a reaction catalyzed by vitamin B-6, and 2- methionine catalyzed by vitamin B-12 and folic acid. Homocysteine is found in low concentrations in all tissues under normal conditions where as accumulates in tissues and plasma if those catalytic vitamins are depleted. Hyperhomocysteinemia is an independent risk factor for cardiovascular diseases and common obstetric problems [3]. Preeclamptic patients also tend to have higher plasma homocysteine levels [4,5].

Normal cellular oxidative metabolism in various tissues free oxygen radicals are released which are potentially harmful molecules causing degeneration of certain structural molecules of the cell, namely various proteins, lipids and nucleic acids. By this way subcellular morphologic and functional changes take place if those radicals are not detoxified by endogen antioxidants [6]. Results of some animal studies demonstrated that high plasma homocysteine levels correlates with decreased overall antioxidant activity and increased lipid peroxides [7].

Plasma homocysteine and lipid peroxide concentrations are frequently found to be elevated in preeclampsia but a causal relationship between these two has not been clearly demonstrated yet.

In this study, a possible correlation between plasma total homocysteine and a lipid peroxidation end product malondialdehyde (MDA) levels was investigated.

Material and methods

This study was conducted in Obstetrics Department of Firat University Medical Faculty Hospital; Elazig-Turkey. Twenty severe preeclamptic patients (Criteriae of severe preeclampsia: systolic blood pressure of 160 mmHg or more and diastolic blood pressure of 110 mmHg or more on measurements at least six hours apart, persistent proteinuria of at least 2+ by dipstick or 24 urinary excretion of 2 g or more, persistent headache, oliguria, nausea-vomiting, epigastric pain, pulmonary edema, thrombocytopenia) and twenty gestational age matched healthy pregnant controls were enrolled into the study. Preeclamptic patients with HELLP syndrome (Hemolysis, elevated liver enzymes, lowered platelet count) were excluded. Sociodemographic features of the subjects were given in table I.

Following an at least eight hours fasting period, and one hour rest, between 08:00–10:00 a.m., blood samples were collected into the tubes containing EDTA from antecubital veins of the subjects in supine position before any medication given. Samples were centrifuged at 3000 rpm. and plasma fractions were stored at -70 degree celcius until examined. Plasma total homocysteine levels were measured by ELX₈₀₀ ELISA machine at 450 nm wavelength using Axis (Norway) kits and results were recorded as $\mu\text{mol/L}$. MDA levels were measured spectrophotometrically (Schimadzu UV-1201 spechtrophotometer) at 532 nanometer wavelength using thiobarbituric acid [8] and recorded as nmol/ml. Statistical analysis was performed using Mann Whitney U test and Spearman Correlation Analysis. All results are given as mean \pm Standard Deviation (Mean \pm SD). $P < 0.05$ was considered to be significant.

Results

Regardless of the preeclampsia, non of the subjects had any chronic disease or pregnancy complication. No significant difference was found between clinical features of the preeclamptics and the normotensive pregnant controls other than hypertension, proteinuria, and pretibial edema ($p > 0.05$), [Table I].

Plasma homocysteine and MDA and other laboratory measurements of normotensive pregnant and preeclamptics were given in table II. Plasma homocysteine concentration of preeclamptics were higher than those of controls ($p < 0.01$). Likewise, plasma MDA levels were also higher in preeclamptic patients ($p < 0.01$). There was a significant correlation between plasma homocysteine and MDA levels of preeclamptic patients ($r: 0.77$, $p < 0.01$, $n: 20$), [Figure I]. where as no correlation was found between these two of healthy pregnant subjects ($r: 0.28$, $p < 0.05$, $n: 20$).

Table 1: Sociodemographic features of the subjects

	Preeclamptic pregnancy	Normotensive pregnancy	P Value
Age (year)	25 \pm 2	26 \pm 2	>0.05
Gestational age (day)	232 \pm 13	225 \pm 15	>0.05
Pregnancy (number)	4.2 \pm 1.4	4.4 \pm 2.1	>0.05
Parturition (number)	3.3 \pm 1.1	3.4 \pm 1.6	>0.05
Blood Pressure (mmHg)			<0.05*
Systole	164 \pm 14	116 \pm 12	
Diastole	114 \pm 5	74 \pm 6	
Proteinuria (mg/24 hour)	5200 \pm 1350	54 \pm 24	<0.05*
Pretibial Odema (+)	2.7 \pm 0.6	0.6 \pm 0.3	<0.05*

*: Significant

Table II: The plasma homocysteine, malondialdehyde levels and some laboratory parameters of the preeclamptic and normotensive pregnant women (Mean \pm Standard Deviation).

	Preeclamptic pregnancy	Normotensive pregnancy	P Value
Homocysteine ($\mu\text{mol/L}$)	20.4 \pm 3	9.1 \pm 2	<0.05*
Malondialdehyd (nmol/ml)	1.9 \pm 0.2	0.84 \pm 0.2	<0.05*
Hemoglobine (g/dL)	12.3 \pm 0.8	12.6 \pm 0.9	>0.05
Urea (mg/dL)	26 \pm 2.2	23 \pm 3.2	>0.05
Creatine (mg/dL)	0.7 \pm 0.1	0.5 \pm 0.2	>0.05
SGOT (U/L)	19.4 \pm 2.3	18.3 \pm 1.9	>0.05
SGPT (U/L)	14.5 \pm 2.1	16.3 \pm 2.4	>0.05
LDH (U/L)	320 \pm 12	334 \pm 18	>0.05

*: Significant

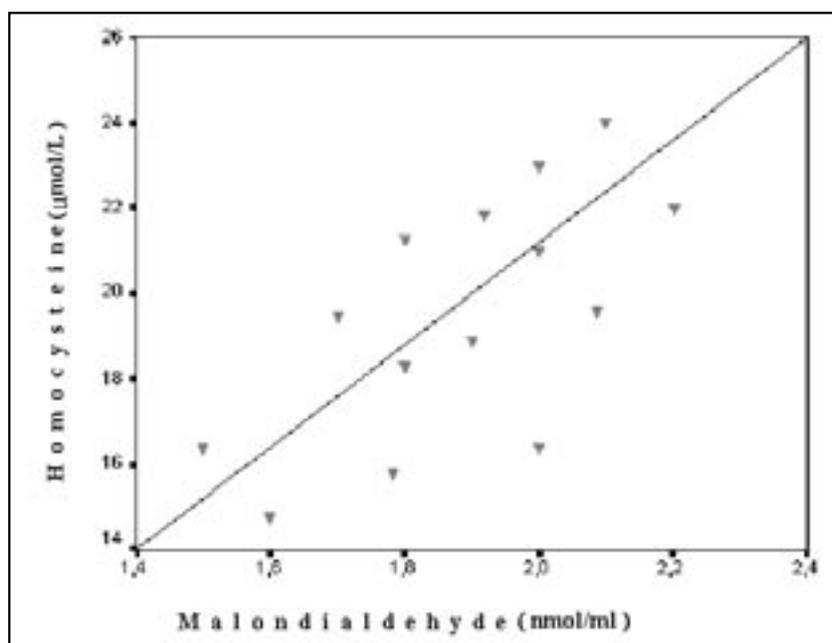


Figure 1: The correlation of homocysteine and malondialdehyde between severe preeclamptic and normotensive pregnancy ($r:0.77$, $p<0.01$, $n:20$).

Discussion

In this study, plasma total homocysteine and MDA concentrations were shown to be increased and positively correlated in preeclamptics. No correlation was observed between MDA and homocysteine levels of healthy pregnant controls.

Serum concentrations of homocysteine decrease during normotensive pregnancy parallel to the physiologic fall of albumin concentration and folic acid supplementation, but increases in preeclampsia like some other pregnancy complications [9–11]. Hyperhomocysteinemia here might be a cause rather than just a marker of adverse pregnancy outcome. In a study conducted on early pregnancy losses, hyperhomocysteinemia was shown to decrease total vessel surface and hence to disrupt placental perfusion [12]. In normal endothelium, nitric oxide (NO) suppresses the smooth muscle proliferation in vessel walls. Decreased NO activity by the effect of homocysteine might contribute to the pathology in those patients.

Hyperhomocysteinemia increases the risk of atherosclerosis through a mechanism involving oxidative damage. When added to the plasma, homocysteine is readily oxidized to form homocystine, homocysteine mixed disulfides and homocysteine thiolactone leading to the formation of oxygen radicals and lipid peroxidation [13].

Endothelial cells detoxify homocysteine by NO and S-nitrosothiol compounds. In addition, hyperhomocysteinemia decreases endothelial NO production probably through increasing synthesis of asymmetric dimethyl arginine which is an endogenous inhibitor of NO synthesis [14, 15]. S-nitrosylation decreases the oxygen radical producing capacity of homocysteine and converts this molecule to a potent vasodilator, S-nitroso-homocysteine. This protective effect of NO

is attenuated in long term leaving endothelium more susceptible to oxygen radicals. Increased concentrations of oxygen radicals also decrease the bioavailability of NO, which amplifies their effect on endothelium. Homocysteine, on the other hand decreases the activity of glutathione peroxidase enzyme, which plays an important role in the detoxification of lipid peroxides. As a result, endothelial dysfunction worsens [13].

Steady-state urinary excretion of nitrites is the best indicator of overall *in vivo* nitric oxide production. In preeclampsia, urinary excretion, but not plasma levels, of these nitric oxide metabolites are decreased [16]. In addition, plasma concentrations of endogenous nitric oxide synthetase inhibitors are also raised in patients with preeclampsia as compared with the levels in patients with normotensive pregnancies or patients with gestational hypertension [17, 18]. Nitric oxide donors cause a significant decrease in uterine artery resistance in patients with an elevated uterine artery resistance index [19]. Thus alterations in endothelial nitric oxide synthetase expression in preeclamptic placentas might also be a result of placental damage and repair processes rather than the primary mediator [1].

Preeclampsia is a pregnancy specific disease characterized by endothelial dysfunction. However pregnancy is not the cause of this condition but is simply a condition of stress that uncover the endothelial malfunction. Blood pressure although decreases postpartum, these patients more readily become chronic hypertensives by ageing [20]. It is well known that plasma homocysteine and MDA concentrations increase in preeclampsia [21, 22]. The obvious correlation observed between total plasma homocysteine and MDA levels in this study. Under the light of literature, and according to the results of this current study, one

can speculate that, high concentrations of homocysteine in preeclampsia, increasing total oxidant and decreasing antioxidant activities might be the mechanism of endothelial injury and hence vasospasm.

It has been speculated that, folic acid supplement could prevent the unwanted effects of homocysteine in preeclampsia [23, 24]. According to the correlation found between the homocysteine and the MDA levels in preeclampsia one can speculate that administration of antioxidants and/or NO donors could be a chance of restoring the endothelial functions in these patients.

In conclusion, the results of this study, in preeclampsia; 1- Vitamin supplementation which decrease homocysteine levels can also decrease the increased oxidant activity 2- antioxidant and/or NO donors can restore the endothelial malfunction. Clinical trials are needed to enlighten this speculation.

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