Lipid peroxidation and antioxidant activity in complicated pregnancies

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Summary

Aims and Objectives: Preeclampsia and eclampsia are pregnancy complications with serious consequences for mother and infant. Uncontrolled lipid peroxidation may play an important role in the pathophysiology of preeclampsia and eclampsia by causing vascular endothelial cell dysfunction. Antioxidants serve to control lipid peroxidation. We attempted to ascertain whether antioxidant protective mechanisms are diminished in women with preeclampsia and eclampsia. *Materials and Methods:* Lipid peroxidation and antioxidant markers were assayed in 25 healthy non-pregnant women as a control group, 25 third trimester normal pregnant women, 25 preeclamptic and 25 eclamptic patients of the same trimester by standard spectrophotometer methods. *Results:* In preeclampsia and eclampsia malondialdehyde, a product of lipid peroxidation, was significantly increased while enzymatic antioxidants like superoxide dismutase, glutathione peroxidase, glutathione reductase and catalase were significantly reduced as compared to normal pregnant women and non-pregnant controls. *Conclusion:* Lipid peroxidation is an important factor in the pathogenesis of preeclampsia and eclampsia. The decrease in antioxidants is probably due to a compensatory nature responding to the increased lipid peroxide load in preeclamptic and eclamptic patients and may indicate the severity of the disease.

Key words: Lipid peroxidation; Malondialdehyde; Preeclampsia; Eclampsia.

Introduction

Preeclampsia and eclampsia are leading causes of feto-maternal morbidity and mortality, accounting for more than 40% of iatrogenic premature deliveries. In developing countries they account for 10-15% of maternal deaths [1, 2]. Increased risk of death for women at 20-32 weeks of gestation is higher than for those at 36-40 weeks. Worldwide, preeclampsia and eclampsia contribute to the death of a pregnant woman every three minutes (200,000 maternal deaths worldwide per year) and have a significant implication for the ongoing health of both mother and infant [3]. The age-specific mortality ratios for preeclampsia and eclampsia reflect the trends observed in other studies - slight risks for younger women (under the age of 20 years) and markedly increased risks for older women [4]. Women who develop preeclampsia and eclampsia during pregnancy are at an increased risk of abruptio placentae, acute renal failure, cerebrovascular and cardiovascular complications, and maternal death [5]. Vascular endothelial cell dysfunction may be caused by uncontrolled lipid peroxidation [6]. Lipid peroxidation is an oxidative process that normally occurs at low levels in all cells and tissues. Under normal conditions, a variety of antioxidant mechanisms serve to control this peroxidative process [7]. However, a diminution of normal antioxidant function will allow increased peroxidative activity to occur at the expense of oxygen and polyunsaturated fatty acids. In disease states such as toxemia in pregnancy, an imbalance between lipid peroxidation and antioxidant mechanisms could impair normal endothelial function. Sera lipid peroxidation products are increased in pregnant women and this increase is further augmented in toxemic patients with decreased antioxidant levels [8].

In this study, we investigated whether the normal balance between lipid peroxidation and antioxidant activity observed during normal pregnancies is impaired in preeclamptic and eclamptic pregnancies. To test this hypothesis, we measured antioxidant activity levels relative to lipid peroxide levels in normal pregnant, preeclamptic, eclamptic and non-pregnant healthy controls and compared results in each group.

Materials and Methods

The present study was carried out jointly by the Department of Biochemistry and Obstetrics and Gynecology from July 2000 to June 2004. The study was approved by the ethical committee of J.N. Medical College and District Civil hospital of Belgaum. Informed written consent was given by all subjects. The study included 100 cases: 25 normal healthy controls, 25 normal healthy pregnant women in the third trimester, 25 third trimester preeclamptic patients and 25 eclamptic patients in the same trimester. The subjects selected for the present study were attending and/or admitted to the District Civil Hospital. Age ranged from 20-40 years. The diagnosis of preeclampsia was based on the definition of ACOG [8]: 1) Systolic blood pressure greater than 140 mm Hg or a rise of at least 30 mm Hg or 2) diastolic blood pressure greater than 90 mm Hg or a rise of at least 15 mm Hg (occurring on two occasions at least 6 hours apart), and 3) proteinuria of 300 mg or greater in a 24-hour urine collection or protein concentration of 1 g/l (on two occasions at least 6 hours apart). Eclampsia was defined as the occurrence of seizures in women with preeclampsia. Women with normal pregnancies were normotensive throughout the gestation and had no proteinuria. Preeclamptic and eclamptic

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Table 1. — *Malondialdehyde (MDA) enzymatic antioxidants (superoxide dismutase (SOD), glutathione peroxidase (GSH-Px), glutathione reductase (GSH-Rx) and catalase) levels in non-pregnant women, 3rd trimester normal pregnant women and 3rd trimester toxemic (preeclamptic and eclamptic) patients.*

	MDA n mol/ml	SOD IU g/Hb	GSH-Px IU g/Hb	GSH-Rx IU g/Hb	Catalase IU g/Hb
Non-pregnant women (n = 25)	1.19 ± 0.09	683.90 ± 155.25	31.08 ± 4.45	10.52 ± 4.67	8.13 ± 2.21
3rd trimester normal pregnant women (n = 25)	1.79 ± 0.14	542.64 ± 142.86	23.45 ± 4.79	7.78 ± 3.40	6.20 ± 1.69
3rd trimester preeclamptic patients (n = 25)	2.93 ± 0.54	452.07 ± 106.05	18.58 ± 4.46	6.86 ± 2.33	5.07 ± 1.31
3rd trimester eclamptic patients (n = 25)	4.80 ± 0.61	397.82 ± 108.99	14.55 ± 3.67	5.47 ± 2.41	3.99 ± 1.22
F-value	347.85	25.22	63.75	9.95	27.45
p value	0.0005	0.0005	0.0005	0.0005	0.0005

patients did not receive any antihypertensive medication until the study samples were taken. Blood pressure and proteinuria levels were determined at the time of sampling.

The subjects were of low socio-economic status based on income. Women were excluded if they were obese, had diabetes mellitus under medication or untreated diabetes, suffered from alcoholism, severe anemia (< 6.0 g% of Hb) or any other systemic disorders.

Blood samples (5 ml) were drawn by venipuncture and collected in heparinized tubes. Malondialdehyde, a product of lipid peroxide detectable in blood, was used as an indicator of lipid peroxidation. Malondialdehyde concentrations were determined by using thiobarbituric acid [10]. Hemolysate was prepared to determine antioxidant acivities like superoxide dismutase [10], glutathione peroxidase, glutathione reductase and catalase [11], and hemoglobin [11-13].

Statistical data were expressed as mean ± SD and statistical significance was determined by ANOVA and the Bonferroni multiple comparison test.

Results

The characteristics of the non-pregnant and pregnant women and preeclamptic and eclamptic patients are shown in Table 1. Statistically significant increased levels of circulating malondialdehyde was observed in the third trimester normal pregnant women, preeclamptic and eclamptic patients as compared to non-pregnant controls. Preeclamptic and eclamptic patients had further increases when compared to normal pregnant women. Activation of PGH synthase can generate oxygen radicals that could act on lipids to generate increased lipid peroxides in normal pregnant women, which would be aggravated during pregnancy-induced hypertension [14].

Antioxidants oppose the toxic actions of lipid peroxides and oxygen free radicals by limiting the amount of lipid peroxides that are formed. Enzymatic antioxidants like superoxide dismutase, glutathione perxidase, glutathione reductase and catalase significantly differed in each group by analytical variance.

Discussion

In the present study oxidative stress was evaluated in normal pregnant patients, and preeclamptic and eclamptic patients by analyzing pro-oxidant and enzymatic antioxidants. Lipid peroxidation was considered as a marker for pro-oxidant, whereas superoxide dismutase, glutathione peroxidase, glutathione reductase and catalase were considered as enzymatic antioxidants. Free radicals are difficult to measure directly due to their unstable and transient nature. Their tendency to cause lipid peroxidation has been used as an indirect measurement [15].

Markers of lipid peroxidation (MDA) are increased during the progression of normal pregnancies and further aggravated in pregnancy-induced hypertension patients [15].

Consistent with previous reports [16] we also noted the significant increase in MDA levels in the third trimester of normal pregnancies compared to nonpregnant women, and further increases were observed in preeclamptic and eclamptic patients when compared to normal pregnant women as well as non-pregnant controls. Our findings clearly indicate that lipid peroxidation may be an important factor in the pathogenesis of preeclampsia and eclampsia.

The protective antioxidant mechanisms are complex and multifactorial. The susceptibility of cells to oxidative stress is a function of the overall balance between the degree of oxidative stress and antioxidant defense capability. Previous studies have demonstrated that superoxide dismutase activity was reduced in the gestation period of normal pregnancies and was lowest in pregnancy-induced hypertension with proteinuria. This could be due to reduced enzyme activity and production of enzyme inactivation by lipid peroxides [17, 18]. However in this study, significant decreased activity of superoxide dismutase and catalase was found in normal pregnant women compared to non-pregnant controls. Further decreases were observed in preeclamptic and eclamptic patients compared to normal pregnant and non-pregnant women.

A study by Pathak *et al.* [19] demonstrated a progressive fall in the superoxide dismutase and glutathione peroxidase activity in normal pregnancies. Decreased activity of glutathione peroxidase and significantly increased levels of malondialdehyde were observed in women with preeclampasia compared to women with normal pregnancies. Glutathione peroxidase is one of the primary antioxidants present in tissues and it inactivates lipid peroxides thereby limiting their levels. However in this study significant decreased activity was seen in toxemic patients when compared to normal pregnant and non-pregnant controls.

Increased levels of lipid peroxidation and decreased

activity of antioxidants in women with preeclampsia and eclampsia, as compared to normal and non-pregnant women, indicates that imbalance in oxidant and antioxidant systems may be impaired. This impairment could result in vascular endothelial dysfunction. The primary reaction sites of lipid peroxidation involve membrane-associated molecules. Polyunsaturated fatty acids and cholesterol alters the structural and functional integrity of biological membranes [20], and can affect normal vascular endothelial cell activity. Rodgers *et al.* [21] reported that preeclamptic sera contain cytotoxic factors that damage endothelial cells; their study results correlate with our data.

It is evident from our study that oxidative stress in preeclampsia and eclampsia leads to decreased activity of antioxidants. Supplementation of natural antioxidants like vitamin E may be of some benefit in the prevention of impending complications like preeclampsia and eclampsia.

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