Some Oxidative Stress Markers in Pregnant Anemic Woman

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Abstract

Anemia is common disease during pregnancy which is known to promote oxidative stress due to insufficient tissue oxygen supply leading to increased free radical generation and very low level of circulating red blood cells and mobile free radical scavengers which protect the tissues from reactive oxygen species (ROS) mediated damage. In aerobic organisms, free radicals or reactive Oxygen Species (ROS) are produced by normal cellular metabolism or by endogenous sources. These species can cause damage to all biomolecules such as poly-unsaturated fatty acids, DNA and proteins. Malondialdehyde (MDA) is a product of lipid peroxidation of polyunsaturated fatty acids. We have determined the serum MDA, serum uric acid and erythrocyte SOD in pregnant anemic women and pregnant healthy woman (control). We found significantly increased MDA and uric acid significantly decreased erythrocyte SOD in pregnant anemic women as compared to control. This may indicate increased oxidative stress in pregnant anemic woman.

Keywords: Anemia, Uric acid, Superoxide Dismutase.

Introduction

Anemia is common disease during pregnancy which is known to promote oxidative stress due to insufficient tissue oxygen supply leading to increased free radical generation and very low level of circulating red blood cells and mobile free radical scavengers which protect the tissues from reactive oxygen species (ROS) mediated damage (1).

In aerobic organisms, free radicals or reactive Oxygen Species (ROS) are produced by normal cellular metabolism or by endogenous sources. These species can cause damage to all biomolecules such as poly-unsaturated fatty acids, DNA and proteins. Lipid peroxidation and DNA damage by these ROS may lead to mutagenesis, carcinogenesis and finally cell death, if the antioxidant system is impaired (2,3). Malondialdehyde (MDA) is a product of lipid peroxidation of polyunsaturated fatty acids. This has been found to be increased in conditions of oxidative stress (1).

Superoxide dismutase (SOD) is an antioxidant enzyme which converts superoxide radicals into H₂O₂. Two iso-enzymes of SOD are described; one is mitochondrial manganese dependent other
is cytoplasmic copper-zinc dependent enzyme (4). Uric acid is metabolic breakdown product of purine nucleotide. It is believed that antioxidant role of ascorbic acid in primates is replaced by uric acid (5). In this study we estimated serum malondialdehyde (MDA) concentration as lipid peroxidation product, serum uric acid level and erythrocytes SOD activity in pregnant anemic woman and compared with normal healthy pregnant woman.

Materials and Methods
The present study was carried out in the Department of Biochemistry, Government Medical College and Hospital, Miraj and P.V.P. Government Hospital, Sangli (Maharashtra). Study protocol was approved by ethical committee of Government Medical College, Miraj (Maharashtra, India).

Sample size: The study included total 90 subjects. Control: Consists of 30 normal healthy pregnant woman selected from the OPD of Government Medical College and Hospital. Patients: This includes 60 patients with anemic pregnant women hospitalized or attaining Gynecology Department at Government Medical College and Hospital.

Inclusion criteria: The females with age 18 to 40 years without complications were selected for the study.

Exclusion Criteria: The subjects having history of other disorders which may affect antioxidant status such as heart diseases, diabetes mellitus, cancer etc were excluded from study.

Blood Collection: Informed consent was obtained from the subjects. 3 ml blood was drawn from an anticubital vein through disposable syringe and needle with all aseptic precautions. 2 ml blood was collected in plain bulb; clear serum was separated and used for the estimation of MDA and uric acid. Serum MDA level was estimated by thiobarbituric acid reaction described by Kai Satoh (6) and the levels were expressed in nmol/ml. Serum uric acid concentration was estimated by uricase end point method (7) and concentrations were expressed in mg/dl. 1ml blood was collected in bulb having anticoagulant (heparin). Blood samples from heparin bulb were centrifuged and plasma was removed. Erythrocytes were washed with normal saline for three times and used for estimation of Superoxide dismutase. SOD activity was estimated by method described by Marklund and Marklund (8) and values were expressed in U/gm. The data were statistically evaluated by using student ‘t’.

Results
We found significantly increased (p<0.001) serum MDA and serum uric acid and significantly decreased erythrocyte SOD activity (p<0.001) in pregnant anemic woman as compared to control and are given in table no. 1.

Table No. 2 shows correlation of MDA with serum uric acid and erythrocyte SOD in patients and control. We found highly significant negative correlation of MDA with SOD and uric acid in patients. Whereas in control; we found highly significant negative correlation of MDA with SOD and highly significant positive correlation with uric acid.

Table No. 1: Some oxidative stress markers in patients and control

| Parameters | Patients (n=60) | Control (n=30) |
|------------|----------------|----------------|
|            | 1st trimester | 2nd trimester | 3rd trimester |                  |
| MDA (nmol/ml) | 5.06 ± 0.24* | 5.42 ± 0.596* | 5.98 ± 0.51* | 3.28 ± 0.67 |
| SOD (U/gm)     | 9.42 ± 0.59*  | 9.00 ± 1.11*  | 8.06 ± 1.08* | 12.82 ± 1.11 |
| Uric acid (mg/dl) | 4.64 ± 1.05* | 4.08 ± 0.58* | 5.52 ±1.04* | 3.27 ± 0.22 |

* Highly significant (p< 0.001)
Table No. 2: Correlation of MDA with erythrocyte SOD and serum uric acid in patients and Control

| MDA (nmol/ml) | SOD     | Uric acid | SOD     | Uric acid |
|--------------|---------|-----------|---------|-----------|
|              | Patients| Control   | Patients| Control   |
|              | -0.675**| -0.718**  | -0.893**| 0.735**   |
|              | P<0.001 | P<0.001   | P<0.001 | P<0.001   |

Discussion
We observed significantly increased levels of serum MDA and serum uric acid in patient as compared to control. Increase was gradual with the progression of pregnancy from 1st to 3rd trimester. This may be due to over production of reactive oxygen species (ROS) or deficiency of antioxidant defense system.

The enzymes involved is oxidative metabolism requires iron \(^{(1,9)}\). Ferrous iron used for oral iron therapy in pregnancy may acts as a potent prooxidant. Many studies have suggested that iron deficient women were further susceptible to this iron therapy induced oxidative stress. This may be due to increased production of ROS because of increased oxygen demand during pregnancy reduction in the activities of antioxidant enzymes such as superoxide dismutase and glutathione peroxidise \(^{(1)}\). Similarly previous studies showed increased lipid peroxidation product MDA in pregnant anemic woman \(^{(1,9,10,11)}\).

We observed significantly decreased levels of erythrocyte SOD in patient as compared to control. Decrease was gradual with the progression of pregnancy from 1st to 3rd trimester. This may be due to insufficient nutrition and oxidative stress under hypoxic condition. It is also may be due to inhibition of SOD by reactive oxygen species \(^{(11)}\). Deficiency of the enzyme SOD results in increase in superoxide radical that may lead to erythrocyte damage \(^{(4)}\).

Some previous studies found decreased SOD activity in pregnant anemic woman as compared to healthy pregnant woman \(^{(11,12)}\). We found highly significant negative correlation of MDA with SOD and uric acid in patients. SOD may be utilized to cope up oxidative stress. This may indicate increased oxidative in pregnant anemic woman.

We concluded that pregnant anemic woman may associate with generation of ROS which implies more risk for pregnant women as well as for fetus.

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