



## Lipid peroxidation and nonenzymatic antioxidants in normal pregnancy

SB Patil<sup>1</sup>, MV Kodliwadmath<sup>1</sup>, Sheela M Kodliwadmath<sup>2</sup>

Departments of Biochemistry<sup>1</sup> and of Obstetrics and gynecology<sup>2</sup> J.N. Medical College, Nehru Nagar, Belgaum-590010.

**OBJECTIVE(S)** : To evaluate the lipid peroxidation and nonenzymatic antioxidant status in normal pregnancy.

**METHOD(S)** : The study comprised 25 healthy nonpregnant women as controls and 75 age matched women with normal pregnancy (25 in each trimester) as subjects. Estimation of lipid peroxidation was done by thiobarbituric reactive substance (TBARS) and nonenzymatic antioxidants were estimated by standard methods.

**RESULTS** : Statistically significant increase in levels of lipid peroxidation and marked decrease in levels of nonenzymatic antioxidants were observed in pregnant women as compared to levels in nonpregnant controls.

**CONCLUSION(S)** : Pregnant women are susceptible to oxidative damage.

**Key words:** lipid peroxidation, malondialdehyde, thiobarbituric acid, superoxides

### Introduction

Pregnancy is a stressful condition in which many physiological and metabolic functions are altered to a considerable extent<sup>1</sup>. In recent years, the role of decreasing antioxidants and increasing superoxides is gaining importance as these are a threat to normal pregnancy. Certain biochemical indices are useful in assessing the progress of pregnancy. The generation of free radicals is a normal physiological process and free radicals act on lipids to cause lipid peroxidation<sup>2</sup>. The cells have evolved a number of counteracting antioxidant defences. These antioxidant defence mechanisms can be categorized under the heads of free radical scavenging and chain breaking antioxidants. Reduced glutathione, alpha tocopherol, ascorbic acid and retinol are nonenzymatic chain breaking antioxidants which limit the cellular concentration of free radicals and prevent excessive

oxidative damage<sup>3</sup>. The present study was undertaken to assess the role of antioxidants, lipid peroxidation and superoxide generation in normal pregnancy.

### Methods

The study comprised 25 healthy nonpregnant women as controls and 75 age matched normal pregnant women (25 in each trimester) as subjects. The subjects and the controls belonged to low socioeconomic status, were in the age range of 20-40 years, and were attending for antenatal check up at our District Civil Hospital, Belgaum.

*Inclusion criteria were –*

1. Healthy nonpregnant women.
2. Women with normal pregnancy.

*Exclusion criteria were –*

1. Obesity.
2. Diabetes mellitus.
3. Alcoholism.
4. Severe anemia (Hb < 6.0 g/dL).
5. Any other systemic disorder.

*Paper received on 12/09/2005 ; accepted on 17/03/2006*

Correspondence :

Dr. S. B. Patil

Department of Biochemistry

Jawaharlal Nehru Medical College

Nehru Nagar

Belgaum - 590010.

Tel. 0831 24773777 Ex. 2067

### Collection and storage of blood samples

Ten mL of blood was drawn by venipuncture and collected in a heparinized tube (10 units of heparin per mL of blood). The following parameters were analyzed within 10-15 minutes of collecting blood sample –

#### A. Whole blood

1. Malondialdehyde (MDA) (Thiobarbituric acid reactive substance)
2. Reduced glutathione (Beutler, 1976)

#### B. Plasma

1. Vitamin-E (alpha tocopherol) (Quaife et al, 1949; Baker and Frank, 1968)
2. Vitamin-C (Ascorbic Acid) (Evelyn and Malloy, 1938.)
3. Vitamin-A (Retinol) (Bessey et al. 1946; Karmarkar and Raja Gopal 1952)

Unpaired t test was used for statistical analysis.

**Table 1. Malondialdehyde (MDA) and nonenzymatic antioxidants (reduced glutathione, vitamin E, vitamin C and vitamin A) levels in controls and subjects.**

	MDA (n mol/mL)	P	Reduced glutathione (mg %)	P	Vitamin E (mg %)	P	Vitamin C (mg %)	P	Vitamin A (mg%)	P
Nonpregnant (n=25)	1.19±0.09		59.97±1.30		1.42±0.19		1.11±0.24		28.62±3.45	
Normal pregnant (n=75)										
1 <sup>st</sup> Trimester (n=25)	1.42±0.13	<0.001	57.23±7.72	0.87	0.97±0.13	<0.001	1.06±0.30	0.518	26.24±3.52	0.190
2 <sup>nd</sup> Trimester (n=25)	1.64±0.12	<0.001	54.46±7.72	<0.001	0.91±0.12	<0.001	1.03±0.30	0.303	24.25±5.19	<0.001
3 <sup>rd</sup> Trimester (n=25)	1.79±0.14	<0.001	50.77±6.91	<0.000	0.87±0.15	<0.001	1.03±0.30	0.543	28.24±5.19	0.859

Results are expressed as mean ± SD and P values are compared with nonpregnant women by using unpaired t test

## Results

There was a consistent significant increase in lipid peroxidation (MDA) in all the trimesters as compared to nonpregnant controls ( $P < 0.001$ ). A significant decrease of nonenzymatic antioxidants viz. reduced glutathione (after 1<sup>st</sup> trimester), vitamin-E and retinol (only in 2<sup>nd</sup> trimester) was observed throughout the gestation phase (Table 1). Vitamin C levels decreased but not significantly.

## Discussion

Free radicals by their unstable and transient nature are difficult to measure directly. Their tendency to cause lipid peroxidation has been used as an indirect measure. Markers of lipid peroxidation (MDA) have been increased during the progression of normal pregnancy<sup>4</sup>. Vitamin E and vitamin A are most important chain breaking antioxidants and they protect polyunsaturated fatty acids from peroxidative damage by donating hydrogen to the lipid peroxy radical. Because of the lipophilic property of the tocopherol molecule vitamin E is the major free radical chain terminator in the lipophilic environment. Vitamin C as a reducing and antioxidant agent, directly reacts with superoxides, hydroxyl radicals, and various lipid hydroperoxides. In addition it can also restore

the antioxidant properties of oxidised vitamin E.

Ishihara<sup>5</sup> studied lipid peroxide levels in nonpregnant and normal pregnant women in the three trimesters. Remarkably increased levels of lipoperoxides were observed by him in the 2<sup>nd</sup> and 3<sup>rd</sup> trimesters of pregnancy as compared to levels in nonpregnant women. Similar observation was made by Kodliwadmath et al<sup>6</sup>. In the present study, we found significant increase in lipid peroxides in all the three trimesters.

Yu<sup>7</sup> reported that reduced glutathione is an effective reductant and plays an important role in a variety of detoxification processes. The enzyme glutathione reductase plays a pivotal role in replenishing and maintaining optimum concentrations of reduced glutathione in biological systems.

Jendryczko and Drozd<sup>8</sup> and Kharb et al<sup>9</sup> reported decreased levels of alpha tocopherol and retinol in healthy pregnant women as compared to levels in controls. It was suggested that measurement of alpha tocopherol and retinol concentrations might be useful as a predictor of the likely development of preeclampsia. Kumar and Das<sup>10</sup> found decreasing trend in the levels of vitamin C throughout the gestational

phase but the decrease was not significant compared to the levels in controls. The present study reveals, decreased levels of glutathione, vitamin E, vitamin C and vitamin A. This may be due to increased lipid peroxidation

### **Conclusion**

Our study shows a significant increase in the lipid peroxidation as pregnancy advances, which is associated with decreased antioxidant levels. Supplementation of natural antioxidants like alpha tocopherol, ascorbic acid and beta carotene may be beneficial in preventing complications like pregnancy induced hypertension.

### **Reference**

1. Scott W. Lipid peroxidation in pregnancy. *Hypertension in Pregnancy*. 1994;13:1-32.
2. Cheesman KH, Slater TF. An introduction to free radical biochemistry. *Br Med Bull* 1993;49:481-93.
3. Corrocher R, Casril M, Belliosala G et al. Severe impairment of antioxidant system in human hepatoma. *Cancer* 1986;58:1658-62.
4. Wickens D. Free radical oxidation (peroxidation) products in plasma in normal and abnormal pregnancy. *Ann Clin Biochem* 1981;18:158-62.
5. Ishihara M. Studies on lipoperoxide of normal pregnant women and of patients with toxemia of pregnancy. *Clin Chim Acta* 1978;84:1-9.
6. Kodliwadmath SM, Kodliwadmath MV, Sadashivadu B. Serum malondialdehyde and ceruloplasmin levels in toxemia of pregnancy. *J Obstet Gynecol India* 1989;39:648-51.
7. Yu BP. Cellular defenses against damage from reactive oxygen species. *Phy Rev* 1994;74:139-62.
8. Jendryczko A, Drozd M. Plasma retinol, betacarotene and vitamin E levels in relation to the future risk of pre-eclampsia. *Zentralbl Gynecol* 1989;111:1121-3.
9. Kharb S, Gulati N, Ghalaut VS et al. Vitamin E concentration in normal pregnant women. *J Obstet Gynecol India* 2000;50(1): 48-9.
10. Kumar CA, Das UN. Oxidant stress in pre-eclampsia and essential hypertension. *J Assoc Physicians India*, 2002;50:1372-5.