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Antioxidant Enzymes in Erythrocyte and Placenta of Pre-Eclamptic Toxemia

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Summary

Antioxidant enzymes superoxide dismutase (SOD), catalase and Glutathine peroxidase (GPX) were studied in haemolysate and placenta of 125 cases of normal pregnancies and 100 cases of pre-eclamptic toxemic (PET) pregnancies. SOD catalase and GPx were significantly lowered in the red blood cells in PET. However, in placenta the enzyme activities were elevated.

Introduction

Pre-eclamptic toxemia (PET) is one of the major causes of high prenatal mortality and morbidity (Chesley, 1984). Pre-eclampsia remains a major health problem (Shapiro et al 1968), particularly among underprivileged people (Brewer, 1966). The cause of the typical disturbance, the triad of hypertension,oedema and proteinuria, occurring after the twentieth week of gestation in primigravid women remains obscure (Sims, 1970).

A number of enzymes and compounds present in the cell, function to protect cellular components from oxidative damage. Antioxidant enzyme studies in erythrocytes assume special significance. Activated forms of oxygen such as superoxide and hydrogen peroxide are considered to play an important role in the oxidative degradation of haemoglobin (Carrel et al., 1975; Itano et al., 1977., Imanishi et al, 1981). Red cells were protected against the oxidative stresses by antioxidant enzymes such as SOD, catalase and GPx. Red cell antioxidant levels might reflect the metabolic or antioxidant/pro-oxidant balance in the interior organs and tissue cells.

Materials and Methods

The investigation was carried out in 225 pregnant women; 125 had normal course of pregnancy and the rest had symptoms of PET. Blood and placentae were collected from the Government Hospital for women and children, Egmore, Madras 600 008, and Government Kasturba Gandhi hospital for women and children, Triplicane, Madras 600 005.

The pregnant women were in the age group of 19-30 years. 59% of them were primigravidae women with symptoms of any other obstetric disease and diabetes mellitus were excluded from this study.

Erythrocytes separated from plasma (obtained with EDTA) were analysed for the antioxidant enzymes SOD by the method of Misra and Fridovich (1972),

catalase by the method of Sinha (1972); and GPx by modified method of Rotruck et al., (1973).

Placenta removed during delivery were kept in ice and used within the next 2 hours. The placental tissue close to the umbilical cord was taken and a 10% homogenate was prepared and used for the assay of antioxidant enzymes.

The values obtained for the subjects with normal pregnancies and PET were expressed as mean ±(SD) standard deviation. Statistically significant differences between the 2 groups were arrived at using student 't' test and 'p' values.

Frequency distribution of antioxidant levels in the normal pregnancies and PET patients were plotted.

Results

There was no maternal age difference in PET and normal pregnant women. Placental weight was lowered significantly in PET when compared to normal pregnancies

Table I shows the antioxidant enzyme in the erythrocyte of pre eclamptics and normal pregnancies. The activities of SOD, catalase and GPx in haemolysate were significantly reduced ($p<0.001$) in PET when compared to the normal pregnancies.

It can be seen from fig 1 that the frequency distributions of erythrocyte SOD and catalase show an overlapping between the PET and normal pregnancies.

Fig 2 shows the frequency distribution of placental SOD, catalase and GPx in PET and normal pregnancies. SOD showed marked differentiation and in the case of catalase and GPx lesser degree of overlapping were observed between PET and normal pregnancies.

Discussion

Cellular antioxidants fight pro oxidant stress which appear in various metabolic processes essential for life. During respiration, molecular oxygen was reduced to form water. During these processes, electrons were added sequentially to oxygen and some of the intermediate ones toxic to cell life. The toxic intermediates of respiration were superoxide, peroxide and other free radicals formed in the metabolic processes. Exposure of the oxidants on to cell components was observed in terms of lipid peroxide formed.

These pro-oxidants and free radicals were eliminated by a series of enzyme systems, SOD, CAT and GPx and antioxidant scavengers, Vitamin A, Carotene, Vitamins C, E, selenium and glutathione. Deficiency of one or more of these antioxidants was observed in a variety of inflammatory conditions (Birnboim and Kanabus 1985, Flohe et al., 1985 and several others).

All inflammatory cells except lymphocytes produce superoxide radicals when activated (Babior et al., 1973) and extracellular administration of Cu, Zn SOD has been shown to protect against inflammatory damages (Michelson and Puget, 1979).

In PET, RBC's showed significantly lower levels of SOD, CAT and GPx, while in the placenta, SOD, CAT and GPx were elevated (Table I & II). The marked increase in lipid peroxide with concominant decrease in SOD, CAT activities were observed in blood samples of PIH as compared to normal pregnancies (Pandey et al., 1996). The enzymes SOD, CAT and GPx were of endogenous origin and their synthesis might be increased in the placenta as a compensatory mechanism to protect the foetus from free radical induced damage. Alternatively, their degradation might be delayed. It would be of interest to study the induction of these enzymes with respect to the nature of the inducer and the synchrony of

Table – I
Antioxidant enzyme activities in haemolysate of PET

Enzymes	Normal pregnancies (125)	Pre-eclamptic toxemia (100)
SOD units / mg Hb	76±11	51±7.5***
Catalase umoles H_2O_2 consumed / mt / mg Hb	1.7±0.7	1.2±0.5***
GPx µg GSH / mt / g / Hb	7.16±0.61	3.68±0.41

- Values are mean +/- standard deviation
Figures in parenthesis indicate number of cases
Statistically significant difference from normal pregnancies ***P<0.001.

FIG 1

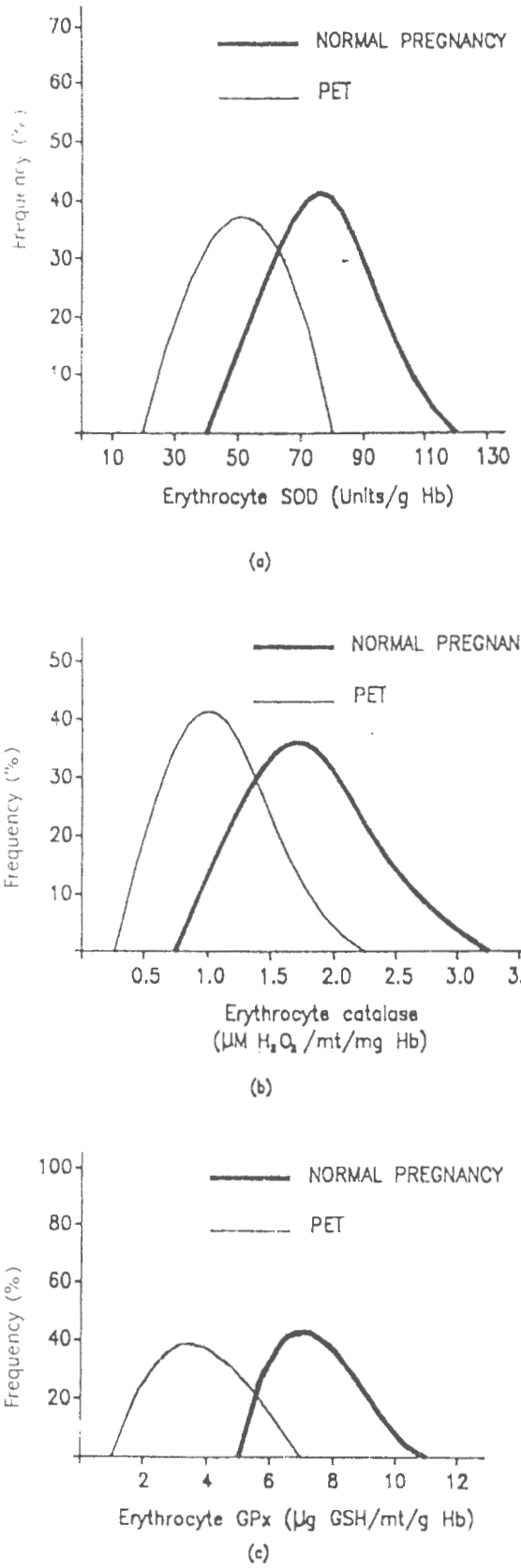
NORMAL PREGNANCY
PET
Frequency (%)
Erythrocyte SOD (Units/g Hb)
(a)
Frequency (%)
Erythrocyte catalase
(μM H_2O_2 /mt/mg Hb)
(b)
Frequency (%)
Erythrocyte GPx (μg GSH/mt/g Hb)
(c)

FIG 2

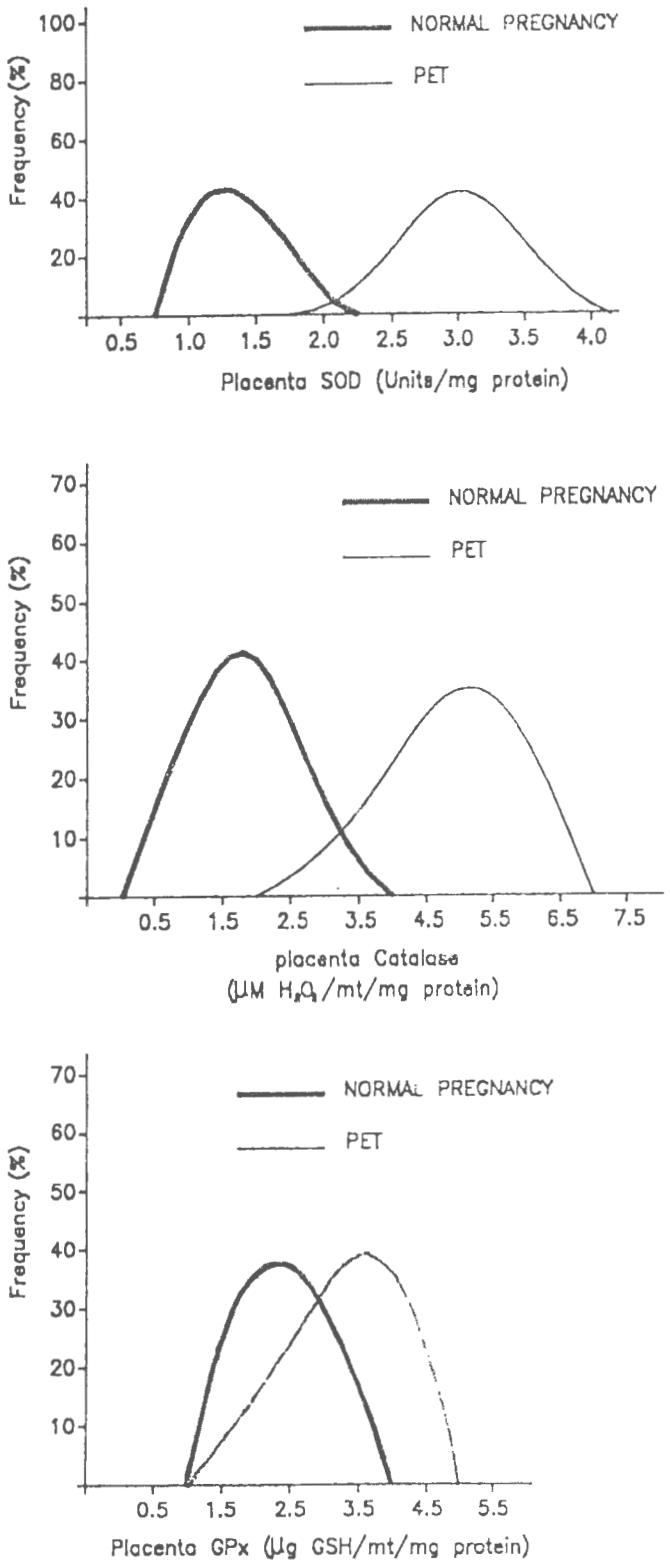
NORMAL PREGNANCY
PET
Frequency (%)
Placenta SOD (Units/mg protein)
placenta Catalase
(μM H_2O_2 /mt/mg protein)
Placenta GPx (μg GSH/mt/mg protein)

Table II
Activities of placental antioxidant enzymes in PET

Enzymes	Normal pregnant (125)	PET (100)
Superoxide dismutase units/mg protein	1.4±0.1	3.1+0.4***
Catalase		
μMH2O2 utilized mt/mg protein	4.02±2.1	5.4+2.0***
GPx		
μg GSH mt mg protein	2.3±0.4	3.4+0.7***

- Values are mean +/-standard deviation
- Figures in parenthesis indicate number of cases
- Statistically significant variations from normal pregnancies *** P<0.001

any in their synthesis under induction. The possibility of the structural genes for the three enzymes occupying the same operon also cannot be ruled out.

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