IJHG 1(1): (2001)
PRINT: ISSN 0972-3757 ONLINE: 2456-6360
DOI: 10.31901/24566330.2001/01.03.12

Heritability of Some Important Parameters of the Antioxidant Defense System Like Glucose-6-Phosphate Dehydrogenase, Catalase, Glutathione Peroxidase and Lipid Peroxidation in Red Blood Cells by Twin Study

Shila Chakraborty and A. B. Das Chaudhuri

Human Genetics Laboratory, Department of Anthropology, University of Calcutta, 35, Ballygange Circular Road, Calcutta 700 019, West Bengal, India

KEY WORDS Heritability; red blood cell; enzymes; twin study.

ABSTRACT Data on the important parameters of antioxidant defense system like glucose-6-phosphate dehydrogenase (G6PD), catalase, glutathione peroxidase (GSH-PX) and lipid peroxidation (lpx) from 7 pairs of monozygotic (MZ) and 7 pairs of dizygotic (DZ) like-sexed male twin pairs were subjected to variance analysis to examine genetic significance of those biochemical traits. The results of variance analysis and F test show the involvement of significant component of genetic variability in case of G6PD and catalase. The estimates of heritability based on variances of MZ and DZ twins are 0.93 (G6PD), 0.72 (catalase), 0.60 (GSH-PX) and 0.50 (lipid peroxidation).

INTRODUCTION

The examination of role of heredity and environment and their interactions in the variation of biochemical traits specially enzymes involved in physiological and metabolic activities are comparatively less studied than morphometric traits in man.

Considerable evidence indicates that the maintenance of protein redox status is of fundamental importance for cell function. Protein oxidation is related to the components of antioxidant defense system (Altomare et al. 1997). It has been established that environmental toxicant induced intracellular oxidative stress countered by changes in the components of antioxidant defense system (Mukhopadhyay et al. 1988; Kumagai et al. 1997). Glucose-6-Phosphate Dehydrogenase (G6PD), catalase, glutathione peroxidase (GSH-PX) and lipid peroxidation (lpx) are a few of the enzymes associated with the maintenance of glutathione status (Chidambaram and Baradarajan 1996). The levels of these enzymes are often used both in in vivo and in vitro studies on the effects of antioxidants (Mukhopadhyay et al. 1988; Look et al. 1997; Bagchi et al. 1997).

Given above, to assess the influence of genetic and environmental components on some important parameters of the antioxidant defense systems, we studied the levels of G6PD, catalase, GSH-PX and lipid peroxidation in the red blood cells using twin method of study.

MATERIALS AND METHODS

Seven pairs of monozygotic (MZ) male and 7 pairs of dizygotic (DZ) like-sexed male twin pairs were obtained from whom the required blood samples were collected. They denied taking any medication for two weeks prior to blood collection. The zygosity of twins, from whom the blood samples were collected, was determined with special emphasis mainly on serogenetic tests like ABO, Rh, MN, haptoglobin and PTC along with similarity method of diagnosis (Siemens 1924).

G6PD was estimated according to the method described by McKena et al. (1983). The assay was carried out monitoring the change in absorbance of NADP (Sigma). Catalase was estimated according to Beers and Sizer (1952) by measuring the decrease in the absorbance of H_2O_2 at 240 nm. Glutathione peroxidase was determined according to Levander et al. (1988) by measuring the decrease in absorbance of NADPH (Sigma) at 340 nm. Lipid peroxidation was estimated by measuring malondialdehyde formation as described by Takayama (1980).

RESULTS AND DISCUSSION

The intrapair difference in percentage levels

of glucose-6-phosphate dehydrogenase (G6PD), catalase, glutathione peroxidase (GSH-PX) and lipid peroxidation (lpx) in MZ and DZ twins computed on the basis of enzyme levels given in table 1 were utilized to construct figure 1. It will be seen that MZ twins show less difference with respect to all enzymes compared to those DZ twins. The mean intrapair difference in the

Table 1: Intrapair difference in percentage levels of enzymes in monozygotic and dizygotic male twin pairs under study

Pair Number	*Catalase (values in ìmoles of H_2O_2 decomposed / min / mg protein)*		*Glutathione Peroxidase (values in ìmoles NADPH oxidised / min / mg protein)*		*Glucose-6-Phosphate Dehydrogenase (values in ìmoles NADPH formed / min / mg protein)*		*Lipid Peroxidation (values in ìmoles of MDA formed / mg protein)*	
	MZ	*DZ*	*MZ*	*DZ*	*MZ*	*DZ*	*MZ*	*DZ*
1	1.42	13.24	11.76	14.03	11.30	27.27	6.28	6.13
2	3.19	11.35	5.45	2.12	2.94	15.38	6.45	10.42
3	7.65	4.98	1.27	13.80	7.56	0	8.69	10.42
4	2.47	4.25	3.52	2.70	4.85	21.95	3.66	5.39
5	1.0	7.53	3.10	6.85	5.00	14.28	4.12	17.51
6	2.76	4.59	2.04	1.12	5.17	14.28	7.97	5.88
7	1.09	1.42	2.16	9.80	5.88	10.00	2.53	1.43
Mean	2.79	6.76	4.18	7.20	6.1	14.74	5.46	7.56

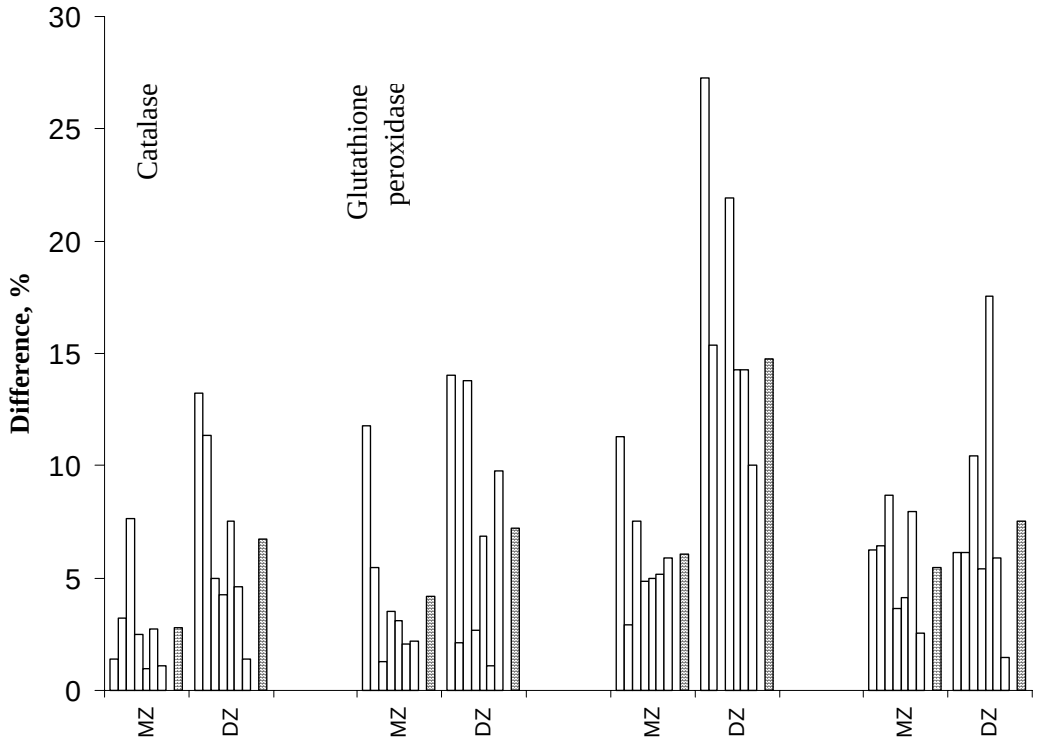

Fig. 1. Intrapir difference in percentage of the enzymes under study

Mean

Table 2: Mean variances of G6PD, catalase, glutathione peroxidase and lipid peroxidation in red blood cells

Types of antioxidants and zygosity	*No. of twin pairs*	*Variance*	*F-ratio*
G6PD			
MZ	7	2.71×10^{-5}	13.50*
DZ	7	3.66×10^{-4}	
Catalase			
MZ	7	1.27	3.63**
DZ	7	4.61	
Glutathione peroxidase			
MZ	7	1.24×10^{-5}	2.52***
DZ	7	3.13×10^{-5}	
Lipid peroxidation			
MZ	7	3.34	2.01***
DZ	7	6.71	

* $P < 0.010$
** $P < 0.100$
*** $P < 0.250$

percentage levels of G6PD, catalase, GSH-PX and lpx are always less in MZ twins than their counterparts in DZ twins. This simply indicates the genetic basis of the enzymes concerned. The mean intrapair variances in G6PD, catalase, GSH-PX and lpx for MZ and DZ twin pairs along with MZ/DZ variances are shown in table 2.

The F-ratios in case of G6PD and catalase are found to be significant at 1% and 10% levels. This is suggestive of significant genetic component of variability in those two enzymes. With some limitations heritability or h^2 statistic (Cavalli-Sforza and Bodmer 1971) has been calculated using formula $h^2 = (V_{DZ} - V_{MZ}) / V_{DZ}$. Here h^2 is the proportion of variability in DZ twins attributable to genetic variation and V_{DZ} and V_{MZ} are intrapair variances of DZ and MZ twin pair respectively. For G6PD, heritability has been found to be 0.93. The estimates of heritability based on variances of MZ and DZ twins were observed for catalase, GSH-PX and lpx to be 0.72, 0.60 and 0.50 respectively. Heritability is found to be least in case of lpx. It is well known that in the vast majority of situations involving lpx proceed through a free radical mediated chain reaction initiated by the abstraction of a hydrogen atom from the unsaturated lipid by a reactive free radical followed by a complex series of propagative reaction. So lpx is the end product estimated whenever unsaturated lipid material undergoes reaction with molecular oxygen to yield lipid hydroperoxides. So it is expected that influence of environmental factors will play a very important and major role in lpx. Our results are in agreement with this idea.

ACKNOWLEDGEMENTS

Financial assistance in a project awarded (Sanction No. 9/28/(379)/93 EMR-1 dated 21.12.93) by the Council of Scientific & Industrial Research (CSIR), Government of India to Dr (Mrs) Shila Chakraborty is gratefully acknowledged. The authors are thankful to the members of twin pairs who participated in the study. The authors acknowledge with thanks the help and cooperation extended by Dr. I. Dasgupta, Ramakrishna Mission seva Prathisthan Hospital, Calcutta while preparing the manuscript.

REFERENCES

Altomare E, Grattagliano I, Vendemaile G, Micelli-Ferrari T, Signorile A, Cardia L 1997. Oxidative protein damage in humans diabetic eye: evidence of a retinal participation. *Eur J Clin Invest*, **27**: 141-147.

Bagchi D, Vuchetich PJ, Bagchi M, Harsoun EA, Tran MX, Tang L, Stohs SJ 1997. Induction of oxidative stress by chronic administration of sodium dichromate (chromium VI) and cadmium chloride (cadmium II) to rats. *Free Rad Biol Med*, **22**: 471-478.

Beers RF Jr, Sizer IW 1952. A spectrophotometric method for measuring the breakdown of hydrogen peroxide by catalase. *J Biol Chem*, **195**: 133-140.

Cavalli-Sforza LL and Bodmer WF 1971. *The Genetics of Human Populations.* San Francisco: WA Freeman and Company.

Chidambaram N, Baradarajan A 1996. Influence of selenium on glutathione and some associated enzymes in rats with mammary tumor induced by 7, 12-dimethylbenz(a) anthracene. *Mol Cell Biochem*, **156**: 101-107.

Coletto GMDD, Krieger H, Magalhaes Jr 1983. Genetic and environmental determinants of 17 serum biochemical traits in Brazilian twins. *Acta Genet Med Gemellol*, **32**: 23-29.

Kumagai Y, Arimoto T, Shinyashiul M, Shimojo N, Nakai Y, Yoshikawa T, Sagai M 1997. Generation of reactive oxygen species during interaction of diesel exhaust particle components with NADPH-cytochrome P450 reductase and involvement of the bioactivation in the DNA damage. *Free Rad Biol Med*, **22**: 479-487.

Levander OE, Deloach DP, Morris VC, Moses PB 1983. Platelet glutathione peroxidase activity as an index of selenium status in rats. *J Nutr*, **113**: 53-63.

Look MP, Rocustroh JK, Rao GS, Kreuzerk A, Barten S, Lemoch H, Sudhop T, Hoch J, Stocuinger K, Spengler U, Sanerbruch T 1997. Serum selenium, plasma glutathione (GSH) and erythrocyte glutathione peroxidase

(GSH-PX)-levels in asymptomatic versus symptomatic human immunodeficiency virus 1 (HIV-1) -infection. *Eur J Clin Nutr,* **51(4)**: 266-272.

McKena R, Ahmad T, Tsao C, Frischen H 1983. Glutathione reductase deficiency and platelet dysfunction induced by 1, 3 bis (2-chloroethyl) 1 - nitrosourea. *J Lab Clin Med,* **102**: 102-115.

Mukhopadhyay S, Mukhopadhyay S, Addya S, Bhattacharya DK, Chaterjee GC 1988. Effects of cadmium treatment in vitro on the antioxidant protection mechanism and activation of human blood platelets. *Thromb Res,* **50**: 419-427.

Siemens HW 1924. *Die Zwillingspathologie*. Berlin: Springer.

Takayama H, Okama M, Uchino H 1980. Estimation of lipoxygenase and cycloxygenase pathways in human platelets. Use of TBA reaction. *Thromb Haemostas,* **44**: 111-114.