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HEPATIC SUPEROXIDE DISMUTASE ACTIVITY IN THE MOUSE INFECTED WITH PLASMODIUM BERGHEI

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Abstract. The effects of malarial infection, induced by infecting the mouse with *Plasmodium berghei* (30—40 % parasitemia), on hepatic superoxide dismutases (SODs) were studied. Total SOD and Cu—Zn SOD activities were found to be significantly reduced. Mn SOD activity, however, was not found to be altered.

Malarial infection in experimental animals is associated with various biochemical changes. Notable among these is the altered hepatic lipid profile in the rodents infected with *P. berghei* (Rao et al. 1967, 1969). The altered lipid profile is considered to be mainly due to increased lipid peroxidation in the infected animals (Sharma et al. 1979). Superoxide radicals are believed to enhance lipid peroxidation in biological systems (Chance et al. 1979). The increased levels of superoxide radicals in the liver are implicated to be main cause of observed elevation in lipid peroxidation (Sharma et al. 1979). This implication is based on the decreased activity of hepatic SODs, enzymes primarily responsible for scavenging the superoxide radicals, observed in the rodents infected with *P. berghei* (Sharma et al. 1979, Kulkarni et al. 1981b). There are two distinct forms of SODs present in the eukaryotic cells (Fridovich 1981). Cupro-zinc superoxide dismutase (Cu—Zn SOD) is present in the cytosol and in the intramembranous space of mitochondria and manganese superoxide dismutase (Mn SOD) is present in the matrix of the mitochondria (McCord and Fridovich 1969, Keele et al. 1970, Weisiger and Fridovich 1973, Fridovich 1981). These two forms of SODs are differentially affected in various experimental conditions (Kimball et al. 1976, Stevens and Autor 1977, Shatzman and Kosman 1978). We have earlier reported decrease in hepatic Cu—Zn SOD activity in the rats infected with *P. berghei* (Kulkarni et al. 1981b). We now report the effects of *P. berghei* infection on both forms of SOD in mouse liver.

MATERIAL AND METHODS

Albino mice (Haffkine strain) weighing 20 g on the average were used for the study. The strain of *Plasmodium berghei* was generously supplied by the National Institute of Communicable Diseases New Delhi and was maintained in mice by serial blood passage. For determining the percentage of parasitemia, thin smears of blood taken from the tail of the infected mouse were stained with 5 % Giemsa in the conventional way and the percentage of the infected red blood cells was determined. The inoculum of the infected blood was prepared in citrated saline and each mouse was injected intraperitoneally with an inoculum of one million parasitized RBCs. When 30—40 % of parasitemia had been established, mice were killed by decapitation. Control mice were intraperitoneally injected with normal saline. Livers were removed quickly, rinsed with ice cold saline and homogenized in 0.02 M phosphate buffer, pH 7.0. An aliquot of the homogenate was used for protein estimation (Lowry et al. 1951). The rest of the homogenate was treated with Triton—X 100 (0.1 % w/v) for 30 min and centrifuged at 14 000 g for 60 min. A part of the supernatant was used for estimation of total SOD activity. The remaining supernatant was mixed with equal volume of chilled chloroform: ethanol (3 : 5) and was subjected to a gentle shaking for 60 min and then centrifuged as mentioned

above. The adequous supernatant was used for estimation of Cu—Zn SOD activity (Weisiger and Fridovich 1973). Mn SOD activity was deduced by subtracting Cu—Zn SOD activity from total SOD activity. The activity of both forms of SOD was determined by the method of Marklund and Marklund (1974). One unit of SOD activity was defined as the volume of supernatant required to inhibit autooxidation of pyrogallol by 50 % under experimental conditions reported earlier (Kulkarni et al. 1981b). The results are expressed as mean ± s.e.m. and the statistical difference in the two groups was assessed by the Student's "t" test.

RESULTS AND DISCUSSION

The results of this study are summarized in Table 1. The reduction observed in hepatic protein levels in the mouse infected with *P. berghei* is in agreement with other reports (Sharma et al. 1979, Kulkarni et al. 1981b). In our studies, hepatic total SOD activity was reduced by 15 % in the infected mice. About 15 % reduction of the hepatic total SOD activity has been reported in the rats infected with *P. berghei* (Sharma et al. 1979). Experiments presented in this paper suggest that only the Cu—Zn SOD activity decreases (by 17 %) while the Mn SOD activity remains unaltered in the liver of infected mice.

Table 1. Hepatic superoxide dismutase activity in the mice infected with *Plasmodium berghei*

Group	Liver protein (mg/gm)	Superoxide dismutase (units/mg liver protein)		
		Total SOD	Cu—Zn SOD	Mn-SOD
Control (5)[1]	175.94 ± 3.74	3.81 ± 0.15	2.21 ± 0.13	1.59 ± 0.10
Infected (5)	157.29**[2] ± 3.31	3.22* ± 0.19	1.84* ± 0.10	1.54 ± 0.06

[1] = Figures in parenthesis indicate number of mice in each group
[2] = Values significantly different from the controls, * $p < 0.05$; ** $p < 0.01$.

Of the two forms of SODs, the CU—Zn SOD has been shown to be inhibited in rat pancreas by diabetogenic drugs (Crouch et al. 1981). On the other hand, hyperoxea is known to induce primarily pulmonary Mn SOD (Stevens and Autor 1977). We have found that the hepatic Cu—Zn SOD (in Tsuchibashi extract) is reduced in monkeys infected with hepatitis B virus (Kulkarni et al. 1981a). Moreover, the ability of one form of SOD to substitute for the other is also reported (Shatzman and Kosman 1978, Fridovich 1981). *P. berghei* infection in mice thus seems to have an effect on the two forms of SODs of which Cu—Zn SOD is significantly reduced in the liver. These results may contribute to understaining of the phenomenon of elevated lipid peroxidation (Sharma et al. 1979) reported in the malarial infection in rodents.

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АКТИВНОСТЬ ПЕЧЕНОЧНОЙ СУПЕРОКСИД ДИСМУТАЗЫ У МЫШИ, ЗАРАЖЕННОЙ *PLASMODIUM BERGHEI*

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Резюме. Изучали действие заражения малярией, вызванного *Plasmodium berghei* (30—40 % паразитемия) на печеночные супероксид дисмутазы (СОД). Активности общей СОД и медно-цинковой СОД очень понижались, тогда как активность марганцевой СОД не изменилась.

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