EVALUATING ROLE OF OXIDATIVE STRESS IN DETERMINING THE PATHOGENESIS OF FALCIPARUM MALARIA INDUCED ACUTE RENAL FAILURE

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ABSTRACT

Serum malondialdehyde was measured in sixty-one falciparum malaria cases, which include thirty uncomplicated, and thirty-one complicated with acute renal failure. Twenty-six healthy individuals were also studied as controls. Serum malondialdehyde level was found to be significantly elevated in falciparum malaria induced acute renal failure cases when compared with uncomplicated falciparum malaria ($p < 0.001$) and healthy controls ($p < 0.001$). A positive correlation with the raised urea, creatinine and bilirubin levels were significant ($r = 0.62$, $p < 0.025$; $r = 0.65$, $p < 0.05$ and $r = 0.72$, $p < 0.001$ respectively) indicating the severity of complication with rise of lipid peroxides in falciparum malaria induced acute renal failure cases.

KEY WORDS

Acute renal failure, Falciparum malaria, Reactive oxygen species, Malondialdehyde

INTRODUCTION

Acute renal failure (ARF) is a serious complication whose incidence is less than one percent among all cases of falciparum malaria, but occurs in about sixty percent of complicated cases (1, 2). The malaria parasite has been shown to generate abnormally large quantities of reactive oxygen species (ROS) within the infected erythrocytes (3). Extracellularly, ROS is also generated as a result of the hosts immune response (4, 5), which bring about extensive damage to parenchymatous organs and endothelial cells observed often in severe falciparum malaria (6). Studies in experimental models have incriminated free radicals as primary mediators in the pathogenesis of ischemic, toxic and immunologically mediated renal injury (7-9).

The aim of the present work is to identify the role of peroxidation in pathogenesis of falciparum malaria induced ARF.

MATERIALS AND METHODS

Thirty-one adult patients with falciparum malaria induced ARF and thirty uncomplicated falciparum malaria patients admitted to the Nephrology and Medicine wards of SCB Medical College Hospital, Cuttack were included in the study along with twenty-six age and sex matched healthy controls. None of the malaria patients had earlier been infected with the parasite. In the ARF group three patients had multiorgan affection. Confirmation of falciparum

infection was by a positive peripheral smear and immunochromatographic test. ARF was defined using WHO criteria, i.e., urine output of less than 400ml/24hr, failure to improve after rehydration and rise in serum creatinine to more than 3mg/dl. Routine biochemical tests done to assess the glycemic state, renal and hepatic functions were estimated on a Technicon RA-1000 autoanalyser. Peroxidation status in serum was determined by estimating malondialdehyde (MDA) as thiobarbituric acid reactive substances at 532nm prior to dialysis. The average number of dialysis per patient was 2.3. Five patients died resulting in mortality of 16.1 percent. As increased bilirubin in jaundice patients could possibly interfere with MDA estimation, a correction factor of 0.008 was reduced for every unit rise in serum bilirubin beyond 1mg/dl (6). Study was conducted after clearance from ethics committee was taken.

All values were expressed as means + SE. Data were analyzed using ANOVA; Students 't' was used to denote significance of difference and Pearsons coefficient was used to establish the level of any correlation.

RESULTS

Table 1 shows the mean +SE of the various parameters studied in patients and controls. Hemoglobin level is lower in patients than control group and statistically significant. Glycemic status in the three groups is within normal range. Serum bilirubin is higher in falciparum malaria cases than controls and when compared had statistically significant and higher value in complicated malaria cases than controls ($p < 0.001$). Raised urea and creatinine values in complicated malaria cases are significant ($p < 0.001$) than both control and uncomplicated cases. The mean Serum

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Table 1 : Levels (Mean + SE) of the parameters studied in patients and control

Parameters	Healthy (n=26)	F. Malaria without ARF (n=30)	F. Malaria with ARF (n=31)	(ANOVA) $F_{(2,84)}$
Hemoglobin (gm/dl) Fasting Blood	11.8 + 0.33	9.7 + 0.58	9.58 + 0.49	22*
Glucose (mg/dl)	86 + 3.13	79.93 + 3.97	84.06+3.72	2.4
Bilirubin (mg/dl)	0.6 + 0.07	0.99 + 0.19	3.03 + 0.80	25.01*
Urea (mg/dl)	31.34 + 1.17	39.13 + 2.09	154.96+ 21.4	107.17*
Creatinine (mg/dl)	1.22 + 0.13	1.03 + 0.07	6.73 + 1.13	14.57*
Sodium (mEq/L)	140.07 +1.35	140.3 + 2.7	135.25 + 3.29	0.94
Potassium (mEq/L)	4.1 + 0.17	4.12 + 1.47	3.89 + 0.31	0.91
MDA (nmol/ml)	1.46 + 1.21	3.45 + 0.19	5.66 + 0.21	361.22*

* p<0.05 - significant

Fig. 1: Correlation between Serum MDA and Serum Bilirubin in Patients of falciparum malaria with RAF

level of MDA was high in complicated (ARF) when compared to uncomplicated malaria cases ($p<0.001$) and healthy controls and it differs significantly. A good positive correlation of Serum MDA with urea ($r=0.62$, $p<0.025$), creatinine ($r=0.65$, $p<0.05$) and bilirubin ($r=0.72$, $p<0.001$) was obtained (Figure 1).

DISCUSSION

In our study we found a statistically significant rise (3.8 times) in MDA level in falciparum malaria with ARF than healthy controls, and when compared with falciparum malaria without ARF, the increase in MDA

Fig. 2: Correlation between Serum MDA and Creatinine in patients of falciparum malaria with ARF

was by a factor of 1.6. This was in consistency with other reported work where such similar rise in MDA in falciparum malaria has been attributed to be a measure of severity of tissue damage (6, 10-12). The assumption that serum MDA reflects the oxidative damage of tissues particularly in erythrocytes, which are in direct and continuous contact with plasma and in which the peroxide is generated as a result of oxidative stress perhaps, holds good for ARF patients in our study. The parasitised red blood cells express a central role in the underlying hemodynamic derangement in falciparum malaria induced ARF. The

merozoites adhere to the vascular endothelial cells disturbing the microcirculation and development of renal failure(1). Apart from the intraerythrocytic parasite metabolism, where infected red blood cells were shown to produce hydrogen peroxide and hydroxyl radicals twice as much as normal erythrocytes. ROS generation is also by non-specific activation of blood monocytes and tissue macrophages. The stimulated phagocytes result in respiratory burst due to the oxidation of NADPH-by-NADPH oxidase which catalyses one electron reduction of oxygen with formation of superoxide anion radical (19, 23). This results in the most undesirable cycle of imbalance between oxidants and antioxidants. The mesangial cells in the glomerulus are the potential sites to generate reactive species.

Our studies revealed a positive correlation of MDA with raised serum urea and creatinine, reflection of the degree of renal failure, which corroborates with similar work where it has been concluded that the level of severity of disease process is reflected by the serum lipid peroxide level (13). A suggestion that lipid peroxidation could be responsible for mitochondrial disintegration combined with depressed energy metabolism and diminished transport performance of the nephron may hold good in our case (14). A further conclusion in experimentally induced ARF reveals that the extent of lipid peroxidation in the early phase of ARF may be decisive for morphologic and functional impairment and for maintenance of this syndrome (15-18).

Both conjugated and unconjugated bilirubin and bile acid as well have been shown to be involved in the pathogenesis of acute renal failure (20-22). In our study, seventy seven percent of patients in complicated (ARF) malaria had hyperbilirubinemia. There is a positive and significant correlation of MDA with bilirubin. Endotoxin released in jaundice increases the vascular response to catecholamines, increases plasma renin activity further increasing renal ischemia and compromising renal function.

Suggestions that have also come forth are that human malaria infection is associated with an increase in ROS, which depending on the situation may participate in host defence against the parasite or cause extensive tissue damage observed in severe form of malaria (4, 5).

In conclusion our results suggest that raised lipid peroxidation is due to oxidative burden in erythrocytes induced by parasitic infection. ROS is implicated in severe tissue damage observed in falciparum malaria and the resulting complications. In our study there is a positive correlation of severity with the lipid peroxide parameter, which is a reflection of the oxidative stress.

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