Lipid Peroxidation And Vivax Malaria

Adil Raza¹, Sumit Kumar Varshney¹, Hiba Sami², Mohammad Jalal¹, Haris M Khan¹, Vaibhav Gupta¹, M Ashraf Malik³, M. Shahid¹, Abbas A Mehdi⁴

¹(Department Of Microbiology, JNMC, AMU, Aligarh)
²(Department Of Microbiology, SRMSIMS, Bareilly, UP, INDIA)
³ (Department Of Paediatrics, JNMC, AMU, Aligarh)
⁴(Department Of Biochemistry, KGMU, Lucknow, UP, India)

Abstract
Objectives: Plasmodium vivax & Plasmodium falciparum are the important species causing malaria in Indian subcontinent. P. vivax usually causes uncomplicated malaria. Whatever be the cause of malaria weather P. falciparum or P. vivax, there is variable degree of oxidative stress. This oxidative stress leads to peroxidation of membrane lipids. Lipid peroxidation is measured in terms of metabolite malondialdehyde (MDA). This study was designed to estimate lipid peroxidation in vivax malaria only.

Methods: The study population was comprised of 200 patients (age range 18-25 yrs) and 50 age and sex matched, population-based healthy volunteers were included as controls. MDA in the serum of patients and that of healthy controls were estimated.

Calculation of parasitemia:
The parasite count (parasites/μL) was done by counting 200 white blood cells and the number expressed on the basis of 8,000 WBC/μL.

Results: Mean MDA level in the healthy controls was 1.15±0.20 n mole/ml (N=50), and the mean MDA level in the vivax cases was 2.52±1.13 n mole/ml (N=200). Which was significantly higher as compared to the controls (P<0.05). Degree of parasitemia was also calculated. The degree of parasitemia has positive correlation with that of serum MDA level. The coefficient of correlation between serum MDA level and parasitemia was +0.95, which shows strong positive correlation.

Conclusions: There is significant lipid peroxidation in the cases infected with P. vivax. Lipid peroxidation increases as the parasitemia increases i.e. lipid peroxidation has strong positive correlation with that of parasitemia.

Keywords: Plasmodium vivax, MDA, Parasitemia.

I. Introduction

Malaria is caused by different species of a Plasmodium. Plasmodium vivax and Plasmodium falciparum are the important species causing malaria in Indian subcontinent. P. vivax usually causes uncomplicated malaria. It is one of important cause of morbidity and mortality. In 2000, it was estimated that there were 262 million cases of malaria causing 839000 casualties. Now, due to health awareness and facilities provided by the respective government to the public the cases decline to 214 million and deaths to 438000 i.e. 18% decline in number of cases and 48% in deaths and still, 3.2 billion people are at risk of malaria, of whom 1.2 billion are at high risk. In high-risk areas, more than one malaria case occurs per 1000 population[1]. The burden of malaria in Asia is under appreciated, despite recent evidence suggesting that the continent contributes almost 40% of the world’s malaria [2]. In sub-Saharan Africa the overwhelming majority of malaria-associated morbidity and mortality occurs with P. falciparum infections. In India, during 2015, there were 1.13 million cases and 287 deaths due to malaria. [3]

In Asia, P. vivax often accounts for 50% of the malaria prevalence, and yet the morbidity associated with this infection and its spectrum of disease is largely ignored. Several studies have reported evidence indicating physicochemical changes in the membrane, induced by oxidative stress, being responsible for the membrane lipid peroxidation and haemolysis seen in malaria. Whatever be the cause of malaria weather P. falciparum or P. vivax, there is some degree of oxidative stress. Oxidative stress is aggravated by a simultaneous reduced effectiveness of the antioxidant defence system. Since the parasite utilizes erythrocyte proteins for its metabolic requirements, the concentrations of enzymatic antioxidants are decreased with parasite maturaiton [4,5,6]. This oxidative stress leads to peroxidation of membrane lipids. Invasion of human erythrocytes by malaria parasite brings about metabolic changes in the host cell. The host cells may then become more vulnerable to damage due to toxic metabolites derived from both the host and parasites. Reactive oxygen species generated in host-parasite interactions causes the lysis of erythrocytes and alteration in antioxidants. Lipid...
Lipid Peroxidation And Vivax Malaria

DOI: 10.9790/0853-1601106658

peroxidation is measured in terms of metabolite malondialdehyde (MDA). This study was designed to estimate lipid peroxidation in vivax malaria only. Therefore, the present study was designed to analyze the serum MDA concentration in *P. vivax* infected patients and to analyze the relation of serum MDA concentration with *P. vivax* parasitemia.

II. Materials And Methods:

Study population
This study was conducted in confirmed patients of *Plasmodium vivax* infection with clinically proven cases of malaria, who attended out-patient clinics or those admitted to the wards of Jawaharlal Medical College and Hospital, Aligarh Muslim University, Aligarh. The study population was comprised of 200 patients (age range, 18-25 years) in study cohort and fifty age and sex matched, population-based healthy volunteers were also included as controls.

Serum samples
This study was approved by the institutional ethical committee of the Jawaharlal Nehru Medical College and Hospital, AMU, Aligarh UP, India. Blood specimens were obtained from the patients and healthy volunteers after taking informed consent. Venous blood was collected aseptically from the patients and was kept in a dark environment before centrifugation. Serum was obtained by centrifugation at 1,5000 X g for 5 minutes at room temperature, and aliquots were prepared and immediately stored at -70⁰ C until processed further. Estimation of lipid peroxidation was done in terms of malondialdehyde (MDA) by the method described by Ohkhwa et al 1979 [7] spectrophotometrically. Thick and thin Giemsa-stained blood films were screened for the presence of Plasmodium species. The parasite count (parasites/μL) was done by counting 200 white blood cells and the number expressed on the basis of 8,000 WBC /μL [8]

Statistical analysis:
Statistical analysis was done using SPSS, version 17, Statistics software. Unpaired Student’s t was applied for the comparison of serum MDA. Descriptive statistics including mean and SDs were calculated for each continuous variable. Pearson correlation analyses were performed to determine the degree and direction of association between two variables (parasitemia and serum MDA concentration). The P < 0.05 was considered as significant.

III. Results
A significant increase in MDA levels in *P. vivax* malaria group was observed compared to the control group. The mean SD of the fifty age and sex matched healthy controls was 1.14 n mole/ml, and the mean MDA level in the cases was 2.52±1.13 n mole/ml (N=200) (Table 1), which was significantly higher as compared to the controls (P<0.05). Degree of parasitemia was also calculated which shows as degree of parasitemia increases levels of MDA also increases (Table 2). As shown in the figure 1, the degree of parasitemia has positive correlation with that of serum MDA level. The Pearson’s coefficient of correlation between serum MDA level and parasitemia was +0.95, which shows strong positive correlation.

<table>
<thead>
<tr>
<th>Table 1: MDA Levels In Cases And Controls</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
</tr>
<tr>
<td>Controls</td>
</tr>
<tr>
<td>Cases</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Table 2: DEGREE OF PARASITEMIA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Range of Parasitemia (Per μL)</td>
</tr>
<tr>
<td>Number Of Patients</td>
</tr>
<tr>
<td>1400-1800</td>
</tr>
<tr>
<td>1801-2200</td>
</tr>
<tr>
<td>2201-2600</td>
</tr>
<tr>
<td>2601-3000</td>
</tr>
<tr>
<td>3001-3400</td>
</tr>
<tr>
<td>3401-3800</td>
</tr>
<tr>
<td>3801-4200</td>
</tr>
<tr>
<td>4201-4600</td>
</tr>
<tr>
<td>N=200</td>
</tr>
</tbody>
</table>

Figure:1 Correlation of MDA levels with parasitemia
IV. Discussion

The present study shows significant increase in lipid peroxides in malaria patients as compared to control subjects. The level of lipid peroxidation was measured in terms of MDA. The degree of lipid peroxidation was in accordance to the degree of parasitemia i.e. higher the parasitemia more production of reactive oxygen species. These reactive oxygen intermediates triggered more lipid peroxidation. The strategies adopted by the plasmodium to survive in the erythrocytes includes avoidance of host immune system, access to host nutrients [9], transport of macromolecules and ions across the erythrocytes, haemoglobin digestion, heme detoxification, immune evasion tactics[10] and multiple drug resistance. Reactive oxygen intermediates can also denature DNA, inactivate enzyme system, modify the cellular antioxidant defence system. Reports are there that reactive oxygen intermediates can also activate procarcinogens [11]. The malarial parasite itself generates large quantities of reactive oxygen intermediates and also through its interaction with phagocytic cell system [12]. Reactive oxygen intermediates are thought to be involved in both the illness and parasite destruction during malaria. The enhanced oxidative stress to erythrocytes in malaria reportedly comes endogenously from the malaria parasites. ROS are generated during the consumption of haemoglobin by the malaria parasite [13]. Increased production of H2O2 and O2 [14] and a decrease in antioxidant enzymes have been observed in parasitized erythrocytes [4,5,6,9]. A combination of reduced antioxidant enzyme defence and increased production of ROS leads to augmented oxidative stress as evidenced by high levels of erythrocyte lipid peroxidation products in malaria [4,15] and also in cultures of P. falciparum in vitro [6]. In our study there is gradual increase in serum MDA level as the parasitemia increases. Recently D’souza et al 2009, Prasanchandra 2006, et al also found the increment in the serum MDA level [16,17], O B Odonje 2011, Camila Fabri et al 2013 has reported increase in serum MDA level in vivax and falciparum malaria[18,19]. These above mentioned studies were also in accordance to our study. Severe malaria infection causes activation of neutrophils and monocytes resulting increase in cytokine level and endothelial damage. The increased vulnerability of erythrocytes to damage and decreased antioxidant system emphasizes the need for early treatment of P.vivax malaria patients to minimize the red cell destruction and resulting anemia. Because it is evident that serum levels of above mentioned vitamins are also found to be decreased in the malaria cases [17,20,21]. To minimize the effect of ROS on host cells due to malarial parasite and antimalarials used in the treatment of malaria clinician should prescribe some antioxidant substance such as Vit C, Vit A. This may improve the probable outcome of the disease.

V. Conclusions

There is significant lipid peroxidation in the cases infected with P. vivax. Lipid peroxidation increases as the parasitemia increases i.e. lipid peroxidation has strong positive correlation with that of parasitemia.

Conflict of interest statement

We declare that we have no conflict of interest.

References

Lipid Peroxidation And Vivax Malaria


DOI: 10.9790/0853-1601106568 www.iosrjournals.org 68 | Page