Biochemical Profiles of Children with Severe *Plasmodium falciparum* Malaria in Central Sudan: a case-control study

Hani Y. Zaki, 1, Badreldin E. Abdalla 1, Hayder E. Babkier 2

1 Department of Biochemistry and Nutrition, Faculty of Medicine, University of Gezira.
2 Department of Pediatrics, Faculty of Medicine, University of Gezira.

**Correspondence:** Hani Y. Zaki, Department of Biochemistry and Nutrition, Faculty of Medicine, University of Gezira, Sudan.
Tel: +249 511 854279
Fax: +249 511 843415
email: hazaki29@hotmail.com

---

**ABSTRACT**

**Background** Malaria is the leading cause of morbidity and mortality in Sudan. The objective of this study was to investigate the effect of severe *falciparum* malaria on some biochemical parameters in Sudanese children. **Methods** Hyperparasitemia was considered as an important criterion for selection of children with severe malaria. Urea, creatinine, sodium, potassium, glucose, uric acid, total protein, albumin, alkaline phosphatase, aspartate aminotransferase, alanine aminotransferase, iron, total iron binding capacity, and ferritin were measured in patients (n=127) and control subjects (n=53). **Results** Children with severe malaria had significantly increased levels of urea, creatinine, Na⁺ and K⁺, and a significantly increased level of uric acid. In addition, the levels of total protein, albumin, ALP, AST, iron and ferritin were significantly decreased when compared with the control group. **Conclusion** The investigated biochemical parameters showed certain changes of a sub-clinical pattern. Consideration of these changes in children with severe *falciparum* malaria might effectively reduce morbidity and mortality.

**Key words** Malaria, children, Sudan.

---

**INTRODUCTION**

Malaria is the leading cause of morbidity and mortality in Sudan. In 2008, there were 3,073,966 reported malaria cases and 1,125 deaths. *Plasmodium falciparum* is responsible for 90% of all infections being the species associated with most severe cases, especially young children and pregnant women. Clinical presentation varies from asymptomatic to multi-organ manifestation and death, depending on host factors (e.g., immunity, age), and parasite factors. The frequent presentations of severe *falciparum* malaria include cerebral malaria, metabolic malaria (hyperlactaemia, acidosis or respiratory distress) and severe anaemia.

Malaria can affect single or multiple organs with different levels of severity which can be determined as neurologic and renal dysfunction, haematologic, cardiovascular, and respiratory dysfunction, as well as hepatic and metabolic dysfunction.
The objective of this study was to investigate the effects of severe *falciparum* malaria in children on some biochemical parameters that could provide a credential clues in understanding malaria pathogenesis, diagnosis and management.

**MATERIALS AND METHODS**

One hundred twenty seven children (66 males and 61 females) with severe *falciparum* malaria were recruited from Wad-Medani Pediatric Teaching Hospital-Gezira State-Central of Sudan. Upon admission, clinical and biological data were registered in the study application form documenting the severity of malaria according to the WHO guidelines. The main foremost inclusion criterion was the level of parasitaemia, children with 10,000–100,000 asexual forms per μl were enrolled. Giemsa stained, peripheral blood smear was used for detection and diagnosis of *falciparum* malaria. Children were treated with parenteral quinine dihydrochloride with a loading dose of 20 mg/kg, followed by 10 mg/kg at 8 hour intervals. Oral quinine was administered when they became able to take it orally. A control group (with negative blood film for malaria) was made up of 53 children (32 males and 21 females) for comparative purposes. Those who satisfied the selection criteria including residence in the Wad-Medani and whose parents voluntarily gave informed consent were enrolled.

Prior to the start of treatment serum was separated from 2ml of blood and stored at -20°C. Estimation of urea, creatinine, glucose, uric acid, total protein, albumin, alkaline phosphatase (ALP), aspartate aminotransferase (AST), alanine aminotransferase (ALT), serum iron, total iron binding capacity (TIBC), and ferritin by Roche Cobas chemistry analyzer. The sodium (Na+) and potassium (K+) were measured by electerolytes analyzer. Laboratory analyses were run at the same time by the same staff using the same equipment and methods. Data were entered in GraphPad Prism 4 software for descriptive and comparative statistical analysis test. T-test was used, and *P*<0.05 was considered statistically significant.

**RESULTS**

The mean age of the patients was 59.41±42.82 months, 57.14% of them were under 5 years. Considering father education as an index of socioeconomic status (Figure1), almost 80% of patients’ father level of education doesn’t exceed the primary level (illiterate, khalwa and primary).
Out of the total of 127 children screened, 96.9% (123/127) had a high grade fever, 18.9% (24/127) had convulsions, and 7.9% (10/127) had a hemoglobin level less than 6g/dl. Figure 2 shows the clinical manifestations of the severe malaria in children at admission.

![Diagram showing clinical manifestations of severe malaria in children group.](image)

**Figure 2 Clinical manifestations of severe malaria in patients group.**

The urea, creatinine, Na\(^+\) and K\(^+\) levels were found significantly increased in patients than that of controls \((p<0.0001, <0.0001, <0.0090\) and \(<0.0040\) respectively) (Table 1). Although there was slight decrease in the blood glucose level of the patients’ group \((58.39\pm27.07\text{mg/dL})\) than that of the controls \((61.77\pm26.17\text{dL})\) but the difference was not significant (Table 2). Significant increased level of uric acid was found in malaria patients \((p=0.0014)\), while the total serum protein and albumin in malaria-infected patients were found to be significantly decreased when compared with control group \((p=0.0024, <0.0001\) respectively) (Table 2). Levels of ALP and

© 2013 Al Neelain Medical Journal  vol.3No. 8  ISSN 1858-627
AST showed significant decrease in malaria patients ($p<0.0001$ for both enzymes), while no significant changes were observed in ALT level (Table 2). The mean levels of serum iron, TIBC, and serum ferritin were significantly different between patients and controls, the decreased serum iron and ferritin were significantly noted in patients ($p<0.0001$ for both parameters), consistently the TIBC was significantly increased in patients ($p<0.0001$) (Table 3).

**Table 1** Levels (mean±SD) of urea, creatinine, Na$^+$ and K$^+$ in patients with severe malaria and controls

<table>
<thead>
<tr>
<th>Variable</th>
<th>Patients (n=127)</th>
<th>Controls (n=53)</th>
<th>$P$ value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urea (mg/dl)</td>
<td>31.88±13.57</td>
<td>17.94±4.33</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Creatinine (mg/dl)</td>
<td>0.91±0.49</td>
<td>0.55±0.09</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Na$^+$ (mmol/l)</td>
<td>138.2±6.69</td>
<td>135.5±5.25</td>
<td>0.0090</td>
</tr>
<tr>
<td>K$^+$ (mmol/l)</td>
<td>4.07±0.60</td>
<td>3.8±0.46</td>
<td>0.0040</td>
</tr>
</tbody>
</table>

**Table 2** Concentrations (mean±SD) of glucose, uric acid, total protein, albumin, ALP, AST and ALT in malaria patients and controls

<table>
<thead>
<tr>
<th>Variable</th>
<th>Patients (n=127)</th>
<th>Controls (n=53)</th>
<th>$P$ value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glucose (mg/dl)</td>
<td>58.39±27.07</td>
<td>61.77±26.17</td>
<td>0.4780</td>
</tr>
<tr>
<td>Uric acid (mg/dl)</td>
<td>3.39±1.62</td>
<td>2.62±0.79</td>
<td>0.0014</td>
</tr>
<tr>
<td>Total protein (g/dl)</td>
<td>6.57±0.95</td>
<td>7.02±0.63</td>
<td>0.0024</td>
</tr>
<tr>
<td>Albumin (g/dl)</td>
<td>3.68±0.57</td>
<td>4.53±0.51</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>ALP (U/l)</td>
<td>211.3±74.23</td>
<td>530±147.7</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>AST (U/l)</td>
<td>18.83±14.1</td>
<td>29.17±8.71</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>ALT (U/l)</td>
<td>10.4±7.48</td>
<td>10.91±5.52</td>
<td>0.6580</td>
</tr>
</tbody>
</table>

**Table 3** Iron profiles in severe malaria children and controls

<table>
<thead>
<tr>
<th>Variable</th>
<th>Patients (n=127)</th>
<th>Controls (n=53)</th>
<th>$P$ value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Iron (μg/dl)</td>
<td>87.38±58.32</td>
<td>135.1±38.67</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>TIBC (μg/dl)</td>
<td>368.4±118.7</td>
<td>260.5±65.2</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Ferritin (μg/l)</td>
<td>75.1±50.46</td>
<td>109±27.2</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

**DISCUSSION**
Almost all severe forms and deaths from malaria are caused by *P. falciparum*. The evaluation of children with severe malaria needs comprehensive investigations for early treatment and the treatment response monitoring. In this study the assessment of renal function in children with severe *falciparum* malaria showed slight renal impairment; creatinine and urea were found significantly elevated in patients than controls. Since our study patients were hyperparisteamic, it is possible that parasite sequestration in the renal vascular (microvascular) bed might account for these changes, leading to ischemia or abnormal cytokines expression. With increasing renal impairment as it appears to be a fall in the renal excretion of catabolites (e.g. conjugated bilirubin, myoglobin) that may further worsen the renal impairment. Assessment of renal function in acute malaria in Indian children has shown that endogenous creatinine clearance was reduced during the acute illness and returned to normal after recovery. In a study performed in Ghana, plasma concentrations of cystatin C were found to be elevated in 17% of the children, indicating subclinical impairment of renal function.

Fluid and electrolytes changes in malaria are of clinical and physiologic interest. In this study sodium and potassium concentrations were significantly elevated in malaria children when compared with the controls. The intraerythrocytic amplification of the *Plasmodium falciparum* induces new pathways of solute permeability in the host cell's membrane, that might be deleterious to erythrocytic membrane cation transport and increases the rate of hemolysis. During severe parasitemia, hyperkalemia results from intravascular hemolysis or rhabdomyolysis, and occasionally from decreased activity of Na+, K+-ATPase. According to literature, hyperkalemia associated with *falciparum* malaria was seen in Kenyan children, 9 children (16%), of whom 7 (78%) died, generally soon after admission. On the other hand, some authors showed significant lowering of the Na+ and K+ levels in malaria infection.

Biomarkers of catabolic activity (glucose and uric acid), nutritional status (total protein and albumin), and hepatic dysfunction (ALP, AST and ALT) were assessed in malaria children in comparison with healthy children. The malaria children showed insignificant decrease in the glucose concentration, while the uric acid was significantly higher in the patients group. Hypoglycemia (blood glucose concentration ≤2.2 mmol/L) is a complication of many different pediatric illnesses and is usually associated with a poor outcome. Malaria infection has been reported to be associated with pleiotropic changes in glucose metabolism, such as decrease of glycogenolysis, decrease of glucose uptake, and increase in insulin resistance that might essentially been mediated by TNF. Accordingly we stratified the study patients to the cut off levels of blood glucose ≤40mg/dl as hypoglycaemic (42/127) and ≥125mg/dl as hypoglycaemic (14/127). Hence, these complications (hypoglycaemia or hyperglycaemia) associated with the change in blood concentration must be evaluated in all children with severe *falciparum* malaria.

Uric acid is a major contributor of the inflammatory response triggered by *P. falciparum* in human peripheral blood mononuclear cells. Recently, uric acid derived from hypoxanthine accumulated by the parasite was described as a source of inflammation in *Plasmodium yoelii*, a mouse malaria model, which triggers the secretion of TNF. Renal dysfunction that is
observed during severe malaria infections probably contributes to the increased uric acid levels observed in plasma. It was shown that elevated levels of xanthine oxidase and liver enzymes are biochemical features of *Plasmodium falciparum* parasitaemia in Nigerian children. Therefore, uric acid is considered a "danger signal" released by dying cells; with plasmodium the immune system has been alerted. Identifying the mechanisms used by the parasite to induce the host inflammatory response is essential to develop urgently needed therapies against this disease.

In this study, a significant decrease in the levels of serum total protein and albumin was observed in children with malaria compared with the control group. In areas of poor socio-economic condition, with unavoidable nutritional deficiencies, the parasitization of the vulnerable children is of alarming rate. Proteins have multiple functions which include immune defense and relationship of protein to diseases can be well explained by antigen and antibodies since both are examples of proteins and as well related to immune defense. Moreover, the effect of malaria infection on the nutritional status of these children, from two rural communities of Ebonyi State, Nigeria was investigated, the total protein and albumin levels showed negative correlation with the degree of parasitemia. The invasion of hepatocytes by malaria parasite can cause organ congestion, sinusoidal blockage, cellular inflammation, cholestasis, nuclear vacuolation, and liver cell necrosis. In this study, mean levels of AST and ALP were significantly decreased in children with malaria than in controls. The serum transaminases of adult patients in Karachi with jaundice and smear positive *falciparum* infection were not significantly raised in comparison with rising serum bilirubin. Our findings appear different from the data obtained in the study conducted in The Infectious Diseases Unit and Medical Wards at Rashid Hospital, Dubai, United Arab Emirates indicating hepatic dysfunction in acute *falciparum* malaria ranged from mild elevation of liver enzymes to acute hepatitis. Decreased levels of liver enzymes in the study patients could be explicated in the following argument: malarial infection induces hepatic apoptosis through augmentation of oxidative stress; with fewer liver cells remaining to produce and release enzymes, aminotransferases levels drop to normal or below normal. Furthermore, investigation of novel genes dysregulation in response to parasite and its antigens demonstrated decrease in transcriptional expression.

This study showed a significant reduction in serum iron and ferritin levels in malaria children compared with the controls. The intense inflammation of severe malaria increases cytokine concentrations, such as interleukin-6, IFN-gamma, and TNF which can stimulate hepatic hepcidin production. Hepcidin is an acute-phase protein with specific antimicrobial and iron regulatory properties. Increased hepcidin production may contribute by directly inhibiting erythroid progenitor proliferation and survival. Several additional mechanisms in severe malaria may lower the haemoglobin and affect the iron profiles. The deranging blood coagulation, thrombocytopenia, is one of the most persisting features of acute malarial infection. In Tanzanian children, micronutrient supplementation improves childhood anaemia in malaria holoendemic areas and this effect is synergistically enhanced by temporary clearance of parasitaemia. Bone-marrow suppression appears to have an insignificant role but pre-
existent iron deficiency aggravates the severity of the anaemia. Moreover, it was reported that iron combined with effective antimalarial therapy promotes haematological recovery in African children after acute *falciparum* malaria.

In conclusion, our study sounds alarm over severe malaria and its complications in Gezira State, Central Sudan. Earlier considerations for the malaria-associated complications, even the sub-clinical complications, are very crucial in children and might effectively reduce morbidity and mortality.

**Acknowledgements** We thank the staff of Wad Medani Pediatric Teaching Hospital for their professional support. We are also grateful to the children and their families who participated in the study.

**REFERENCES**

and implication of mitochondrial pathway, FASEB J 2006, 20:1224-1226