ASSOCIATION OF HEPATOMEGALY AND JAUNDICE WITH ACUTE RENAL FAILURE BUT NOT WITH CEREBRAL MALARIA IN SEVERE FALCIPARUM MALARIA IN THAILAND

MATHIEU NACHER, SOMBAT TREEPRASERTSUK, PRATAP SINGHASIVANON, UDOMSAK SILACHAMROON, SUPARP VANNAPHAN, FRÉDÉRICK GAY, SORNCHAI LOOAREESUWAN, AND POLRAT WILAIRATANA

Unité INSERM 511, Immunobiologie Cellulaire et Moléculaire des Infections Parasitaires, Faculté de Médecine Pitié-Salpêtrière, Paris, France; Faculty of Tropical Medicine, Mahidol University, Hospital for Tropical Diseases, Bangkok, Thailand

Abstract. We conducted a case record study comparing liver tests abnormalities in 20 malaria-related acute renal failure cases without cerebral malaria, 52 cerebral malaria cases without other organ impairment, 189 cases of nonsevere malaria associated with a high parasite burden, and 131 cases of mild Plasmodium falciparum malaria. Jaundice and hepatomegaly were significantly associated with renal failure (adjusted odds ratio [AOR], 3.3, 95% confidence interval [CI], 1.3–8.6, P = 0.01; and AOR, 1.7 95% CI, 1.13–2.4, P = 0.01) but not with cerebral malaria (AOR, 1, 95% CI, 0.5–2, P = 0.8; and AOR, 1.08, 95% CI, 0.8–1.8, P = 0.5). Patients with acute renal failure were significantly older and had increased liver abnormalities compared with other groups. Although an increase in the proportion of mature schizonts over ring forms was significantly associated with cerebral malaria, it did not seem to have affected acute renal failure. These results suggested that cytoadherence was not the main determinant for renal failure and that jaundice itself may have potentiated the effects of hypovolemia.

INTRODUCTION

Malaria still kills > 1 million people every year. Numerous specific and nonspecific effectors of the immune system are involved in the host response to control the multiplication of Plasmodium falciparum. Among these, the liver and the reticuloendothelial system in general have a role in phagocytosis, soluble mediator release, and initiation of the humoral response. During mild malaria, it is common to observe nonspecific hepatitis, indicated by a slight hepatomegaly and liver enzyme elevation. Jaundice is common in adults with severe malaria. Notably, it has often been reported in patients with cerebral malaria, acute renal failure, or both. Jaundice in patients with malaria can result from hemolysis of parasitized and nonparasitized red blood cells and liver dysfunction. Few studies have focused on hepatomegaly during severe malaria. Hepatic failure is exceptional, but decreased hepatic blood flow and microsomal activity are common during severe malaria. There are several clinical manifestations of severe malaria, which may differ in pathophysiology. From a pragmatic perspective, these manifestations have in common the following: they are life-threatening, and they are often grouped as a multifaceted entity requiring intensive care. However, in research, this may hamper progress in the elucidation of underlying pathogenesis. Definition of both cases and controls is of paramount importance in the design of epidemiological studies. We therefore split the definition of severe malaria into nonoverlapping subgroups and conducted a cross-sectional case record study to determine whether liver abnormalities had any preferential association with different manifestations of disease severity.

MATERIAL AND METHODS

Study site. The study took place at the Hospital for Tropical Diseases, Bangkok, Thailand. We chose mutually exclusive clinical categories that are common entities in Thailand. We conducted a cross-sectional study on 20 patients with malarial acute renal failure without neurological impairment, 52 patients with cerebral malaria without renal failure, 189 patients with nonsevere malaria despite a high parasite biomass, and 131 patients with mild Plasmodium falciparum malaria. All were admitted between January 1998 and December 1999. The duration of illness was recorded. The liver size was measured in centimeters below the ribcage on the midclavicular line on a supine patient. As others, we considered the measure of a palpable liver as a reflection of hepatomegaly. However, liver span and tenderness were not recorded. All patients underwent clinical examination and had blood smear, blood count, and blood chemistry analyses performed at the time of admission.

Definitions. Cases. Because acute renal failure and cerebral malaria may not have the same pathophysiology, we decided to create a separate group for each of them. Acute renal failure was defined as a serum creatinine level > 3 mg/dL.; cerebral malaria was defined as a Glasgow Coma Scale < 10.

Controls. Moderately severe controls were patients with no cerebral or renal complications despite having a high parasite biomass, which was defined as the presence of asexual parasitemia > 5% or 200,000 parasites/μL, or the presence of circulating schizonts and the absence of any severity-defining criteria.

Mild controls had mild falciparum malaria, defined as the presence of P. falciparum on the blood smear and the absence of any of the above criteria.

Patients with mixed Plasmodium infections were excluded from the study.

Comparison groups. It has been suggested that at a population level, the relative rarity of severe complications of malaria reflects that of the most virulent strains. In this perspective, the best control group to study host factors related to severe complications of malaria would have been patients infected by the same strain who did not develop severe complications of malaria. In the absence of known stable virulence markers, and because there is a positive correlation between parasite biomass and complications, we chose “moderately severe controls” with a high parasite bio-

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mass and then adjusted for symptom duration. Patients thus had parasites of comparable growth kinetics and only differed from the clinical status reflecting presence or absence of cerebral or renal dysfunction. Thus, after frequency matching for parasite biomass and adjusting for the time we assume this reduced the specific effect of the strain, we therefore facilitated the detection of host-related protective or risk factors. A previous study showed that in addition, in patients with cerebral malaria the early trophozoite/schizont ratio,9 were observed in the cerebral malaria group, whereas the highest parasite counts were in the renal failure group (Table 1).

Table 1 shows that the mean age was significantly higher in the acute renal failure group (P = 0.005). The most mature infections, in terms of the stage of parasite development as reflected by the early trophozoite/schizont ratio9 were observed in the cerebral malaria group, whereas the highest parasite counts were in the renal failure group (Table 1). Patients with renal failure and cerebral malaria had significantly lower hemoglobin concentrations than moderately severe malaria (P = 0.0001). When comparing moderately severe malaria with mild malaria, there was also a linear trend (P = 0.009).

Liver tests. Patients with acute renal failure often had abnormal liver tests; 90% of them had jaundice (Tables 1 and 2). There was a significantly higher proportion of con-
jugated bilirubin in renal failure than in cerebral malaria (P = 0.002) and in mild or moderately severe malaria (P < 0.0001). No significant difference between the proportion of conjugated bilirubin in cerebral malaria and moderately severe malaria was observed (P = 0.7). Among patients with acute renal failure, there was also a significantly higher aspartate aminotransferase level (P = 0.005) than in patients with cerebral malaria or in moderately severe controls (Table 1). Among patients with acute renal failure, there was also a significantly higher globulin level than in patients without renal failure (P = 0.005) (Table 1). Among patients with acute renal failure, there was a significantly lower albumin level than in patients with cerebral malaria or in moderately severe controls (P = 0.02) (Table 1). However, there was no difference in total protein level between groups (P = 0.21). Alkaline phosphatase seemed lower in the acute renal failure group, but this difference was not significant (P > 0.5) (Table 1).

**Adjusted odds ratios.** Table 2 shows that jaundice and increased liver size were associated with acute renal failure, but not with cerebral malaria. In the logistic regression models, when comparing cerebral malaria with moderately severe malaria and adjusting for parasitemia, schizont counts, and evolution duration, we observed that splenomegaly was associated with cerebral malaria (adjusted odds ratio [AOR], 1.4, 95% confidence interval [CI], 1.04–1.9, P = 0.02). A longer evolution duration (in days) (AOR, 1.14, 95% CI, 1.05–1.25, P = 0.002) and increased schizont counts (AOR, 1.12, 95% CI, 1.02–1.24, P = 0.02) were associated with cerebral malaria. We then replaced parasitemia and schizont counts with the schizont/trophozoite ratio, a reflection of the percentages of parasites expressing cytoadherence-mediating knobs. After transforming the schizont/trophozoite ratio into a categorical variable with equivalent patient numbers in each group, the increase in the proportion of mature schizonts over young asexual forms was associated with cerebral malaria (AOR, 1.76, 95% CI, 1.15–2.7, P = 0.009). When comparing acute renal failure with moderately severe malaria, neither evolution duration (AOR, 0.97, 95% CI, 0.8–1.2, P = 0.7) nor parasite counts (AOR for parasitemia, 1, 95% CI, 0.9–1, P = 0.6; AOR for schizont counts, 1.04, 95% CI, 0.9–1.2, P = 0.6) were significant. The proportion of mature schizonts or young forms was not associated with significant risk for renal failure (AOR, 1.3, 95% CI, 0.6–2.6, P = 0.4). The most parsimonious model comparing acute renal failure and moderately severe malaria retained jaundice and increased liver size as risk factors and splenomegaly, which was associated with protection from acute renal failure (AOR, 0.38, 95% CI, 0.14–1, P = 0.05). Recoding bilirubin in categorical variables showed that there was a linear trend between the concentration of total bilirubin and acute renal failure (chi-square, 27, P < 0.0001) (Figure 1). Liver enzymes and albumin levels were removed from the models because after adjustment, they did not predict either cerebral malaria or acute renal failure (data not shown).

**Liver size.** Because the palpable portion of the liver is not the most appropriate measure of hepatomegaly, we defined hepatomegaly as when patients had a palpable liver of ≥ 2 cm. By use of this definition, hepatomegaly was significantly associated with acute renal failure (AOR, 5.4, 95% CI, 1.8–16.6, P = 0.003) but not with cerebral malaria (AOR, 1.4, 95% CI, 0.6–4, P = 0.4) when we used moderately severe malaria as the control. There was a positive

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**Table 2.** Crude and adjusted odds ratios for palpable liver size and jaundice using different comparison groups

<table>
<thead>
<tr>
<th>Comparison groups</th>
<th>Exposure</th>
<th>Crude odds ratio (95% CI)</th>
<th>Adjusted odds ratio (95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>CM/moderately severe malaria</td>
<td>Palpable liver</td>
<td>1.8 (0.96–3.3)</td>
<td>1.08 (0.8–1.8)†</td>
<td>0.5</td>
</tr>
<tr>
<td>CM/moderately severe malaria</td>
<td>Jaundice</td>
<td>1.1 (0.6–2.1)</td>
<td>1.0 (0.5–2)†</td>
<td>0.8</td>
</tr>
<tr>
<td>ARF/moderately severe malaria</td>
<td>Palpable liver</td>
<td>5.2 (1.8–15.5)</td>
<td>1.7 (1.13–2.4)†</td>
<td>0.01</td>
</tr>
<tr>
<td>ARF/moderately severe malaria</td>
<td>Jaundice</td>
<td>15 (3.7–∞)</td>
<td>3.3 (1.3–8.6)†</td>
<td>0.01</td>
</tr>
<tr>
<td>ARF/CM</td>
<td>Palpable liver</td>
<td>2.9 (0.9–9.5)</td>
<td>1.35 (0.9–2)†</td>
<td>0.13</td>
</tr>
<tr>
<td>ARF/CM</td>
<td>Jaundice</td>
<td>12.1 (2.8–∞)</td>
<td>13.3 (2.6–64)†</td>
<td>0.002</td>
</tr>
<tr>
<td>CM/mild</td>
<td>Palpable liver</td>
<td>2.5 (1.3–4.8)</td>
<td>1.1 (0.8–1.6)¶</td>
<td>0.5</td>
</tr>
<tr>
<td>ARF/mild malaria</td>
<td>Palpable liver</td>
<td>7.4 (2.4–22.2)</td>
<td>3 (1.24–7.5)¶</td>
<td>0.01</td>
</tr>
<tr>
<td>Moderately severe malaria/mild malaria</td>
<td>Palpable liver</td>
<td>1.4 (0.9–2.2)</td>
<td>1.3 (0.99–1.7)¶</td>
<td>0.055</td>
</tr>
</tbody>
</table>

* AOR = adjusted odds ratio. ARF = malaria with acute renal failure; CI = confidence interval. CM = cerebral malaria.
† Adjusted using a logistic regression model with liver size, jaundice, spleen size, parasitemia, schizont counts, and symptom duration as independent variables. Interaction terms were created but did not show significant interaction.
‡ Adjusted using a logistic regression model with liver size, jaundice, spleen size, direct bilirubin, parasitemia, and symptom duration as independent variables.
¶ Adjusted using a logistic regression model with liver size, direct bilirubin level, 1.4 (95% CI, 1.1–1.7) (P = 0.002).
§ Adjusted using a logistic regression model with liver size, jaundice, spleen size, direct bilirubin, parasitemia, schizont counts, and symptom duration as independent variables.
# AOR for direct bilirubin level, 1.6 (95% CI, 1.16–2.2) (P = 0.004).

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**Figure 1.** Odds of developing malaria-associated acute renal failure for different total bilirubin concentrations, with moderately severe malaria used as the control.
correlation between liver size and spleen size (Spearman’s rho = +0.42, P < 0.0001). Liver size increased with parasitemia and schizont counts (Spearman’s rho, respectively, +0.14, P = 0.003, and +0.29, P < 0.0001). Liver size was not correlated with the ratio of trophozoites/schizonts (Spearman’s rho = 0.02, P = 0.7). There was a positive correlation between liver size and both conjugated (Spearman’s rho = +0.30, P < 0.0001) and unconjugated bilirubin (Spearman’s rho = +0.28, P < 0.0001). Adjusting for parasitemia and schizont counts did not alter these associations (P < 0.0001). After excluding patients with renal failure, there was a positive correlation between liver size and creatinine level (Spearman’s rho = +0.17, P = 0.006). Adjusting for parasitemia and schizonts did not alter this association (P < 0.0001), but after adding bilirubin to the model, the correlation between elevated creatinine level and liver size disappeared (P = 0.22).

**Bilirubin, creatinine, and age.** Both conjugated and unconjugated bilirubin median concentrations were higher among patients with acute renal failure (Table 1, P < 0.0001); this remained significant after adjusting for parasitemia and schizont counts. The conjugated bilirubin/total bilirubin ratio was significantly higher in the acute renal failure group (Table 1). There was a positive correlation between age and conjugated bilirubin (Spearman’s rho = 0.25, P < 0.0001) and between age and unconjugated bilirubin (Spearman’s rho = 0.19, P = 0.0001). After excluding patients with acute renal failure and adjusting for weight and parasite counts (nonparametric regression), we observed that the median creatinine level was not influenced by age (P = 0.66). After excluding patients with acute renal failure and adjusting for weight and parasite counts (nonparametric regression), we observed that the median creatinine concentration was positively correlated with conjugated bilirubin (P = 0.005) but not unconjugated bilirubin (P = 0.14). Among patients with jaundice, those with acute renal failure had significantly higher median concentrations of conjugated bilirubin, 6.5 IU (interquartile range [IQR] = 2.34–13.2) than moderately severe controls with jaundice, 3 IU (IQR = 1.6–6.3) (P = 0.008). After adjusting for parasitemia and schizont counts, this remained significant (P = 0.01).

**Bilirubin in patients with jaundice.** After excluding acute renal failure, the proportion of conjugated bilirubin/total bilirubin was significantly higher in patients with jaundice (ratio = 0.55, IQR = 0.4–0.62) than in those without jaundice (ratio = 0.27, IQR = 0.22–0.36) (P < 0.0001).

Among patients with jaundice, the median unconjugated bilirubin level was slightly higher among patients with renal failure (3.5 mg/dL; IQR = 2.4–7.7) than among moderately severe controls with jaundice (2.8 mg/dL; IQR = 2.1–4.3), but this failed to reach statistical significance (P = 0.06) before and after adjustment for parasitemia and schizont counts (P = 0.26). Similarly, in patients with jaundice, there was no significant difference in the median conjugated/total bilirubin concentrations ratio between patients with acute renal failure (ratio = 0.61; IQR = 0.52–0.67) and moderately severe controls (ratio = 0.55; IQR = 0.39–0.63) (P = 0.08).

**Bilirubin, parasite counts, and trophozoite/schizont ratio.** Levels of conjugated and unconjugated bilirubin were positively correlated with parasitemia (Spearman’s rho = +0.36, P < 0.0001, and Spearman’s rho = +0.33, P < 0.0001, respectively) and schizont counts (Spearman’s rho = +0.43, P < 0.0001, and Spearman’s rho = +0.42, P < 0.0001, respectively).

By using nonparametric regression analysis in data for the patient groups without renal failure, we observed that conjugated bilirubin was positively correlated with early trophozoite counts (P = 0.036) and schizont counts (P = 0.02), but not by the early trophozoite/schizont ratio (P = 0.5). Similarly, after adjusting for the patient’s weight, we observed that the creatinine level was positively correlated with parasite counts (P = 0.037), but not by the early trophozoite/schizont ratio (P = 0.4).

**Predictive values.** We placed patients with cerebral malaria, acute renal failure, and moderately severe malaria into one group: patients with a high parasite biomass. In this group, the sensitivity of having a palpable liver for predicting acute renal failure was 76% (95% CI, 54–90), the specificity was 54% (95% CI, 47–60), the positive predictive value was 14% (95% CI, 8–21), and the negative predictive value was 96% (95% CI, 91–98). In the same group, the sensitivity of jaundice to predict for acute renal failure was 92% (95% CI, 73–99), the specificity was 61% (95% CI, 54–67), the positive predictive value was 19% (95% CI, 12–27), and the negative predictive value was 99% (95% CI, 95–100).

**DISCUSSION**

In Thailand, malaria transmission is low, and all ages may develop severe malaria. In adults, jaundice and acute renal failure are common presentations of severe malaria. Here, when we used homogenous case definitions, we observed that jaundice and hepatomegaly were associated with acute renal failure, but not with cerebral malaria. Among patients with acute renal failure, there seemed to be an increase in liver test abnormalities. A notable difference was the higher percentage of both conjugated and unconjugated bilirubin in patients with renal failure than in patients in other groups. Although renal failure itself may increase conjugated bilirubin, the significant increase of the conjugated bilirubin proportion among patients with jaundice with and without renal failure suggests that patients with jaundice and malaria had cholestasis. The linear relationship between renal failure and total bilirubin and studies on the renal consequences of obstructive jaundice could therefore support a possible causal relationship.

The pathophysiology of renal failure in malaria is still unclear. It is thought that cytoadherence, multifactorial changes in cortical perfusion, cytokine release, and hypovolemia lead to tubular necrosis. The association between renal failure and jaundice is a recurrent finding in studies on severe malaria. Acute renal failure can be found both in patients with and without cerebral malaria. In the case of isolated acute renal failure, considering cytoadherence as the major mechanism would imply that it must be organ selective, because cerebral function is highly sensitive to vascular disturbances. However, in our logistic regression models, schizont counts and their relative proportion to ring forms, which reflect the proportion of sequestrated parasites, were not risk factors when compared with moderately severe con-
trols. This suggested that sequestration was not the most im-
portant determinant of acute renal failure.

On the contrary, schizont counts and their relative pro-
portion to ring forms were risk factors for cerebral malaria, sup-
sporting the hypothesis of cytoadherence-related neuro-
logical impairment. We also observed that creatinine in-
creased with parasite counts, but that the proportion of ma-
ture forms did not seem to influence its concentration. In
other words, a high parasite biomass was a prerequisite for
acute renal failure, but whether most parasites were seques-
tered or not was not a crucial determinant. Severe malaria
results from a combination of host and parasite factors.1 Par-
asite virulence can be defined as its intrinsic ability to cy-
toadhere and to upregulate endothelial cytoadherence recep-
tors and escape splenic destruction; in addition, virulence
activity may have selected cytoadhesive clones of
the patient from renal failure. On the contrary, increased splenic
function, thus reducing hepatic inflammation and protecting the
splenic destruction of parasitized red blood cells
might be important for cerebral malaria.

This is consistent with a pathological study on severe ma-
laria showing that the presence of parasitized red blood cells
in the kidney was much lower in the kidneys than in the
brains of patients with cerebral malaria.17 An extension of
this would be that parasites that have both cytoadhesive and
multiplicative dimensions of virulence would lead to the
association of cerebral malaria and acute renal failure. Host
factors also seem important: we observed here that patients
with acute renal failure were significantly older than the rest.
Age-related differences of liver function may explain this.
Given the increase of both conjugated and unconjugated bil-
irubin with age, a hypothesis would be that the hepatic (and
maybe renal) function of older patients was more fragile
under the stress of infection and hemolysis. In these patients, jaundice may have resulted from both an increase in biliru-
bin production due to massive hemolysis and some degree
of retention. An increased stimulation of liver macrophages
in patients with high parasite burdens may have generated
intra hepatic cholestasis.1 Bilirubin in general is toxic for
tubular cells, but it has been shown that patients with ob-
structive jaundice have a high risk of tubular necrosis when
exposed to hypovolemia and hypoxia.13 Here, patients with
acute renal failure had hypoalbuminemia, which may have
contributed to hypovolemia.

The correlation between hepatomegaly and splenomegaly
suggested that in patients with a high biomass, there is a
global activation of the reticuloendothelial system. Here, we
found that splenomegaly seemed protective against renal
failure but was associated with cerebral malaria. It is pos-
sible that splenic destruction of parasitized red blood cells
reduced intravascular and intrahepatic red blood cell destruc-
tion, thus reducing hepatic inflammation and protecting the
patient from renal failure. On the contrary, increased splenic
activity may have selected cytoadhesive clones of \( P. falciparum \), increasing the risk for cerebral malaria.18 This may
have explained why in patients with cerebral malaria there
was no correlation between liver size and symptom duration.

Reports of renal failure and jaundice during severe ma-
laria originate mainly from Asian countries, probably be-
cause of differences in malaria transmission. However, re-

notably for glucuronidation, could also contribute to these
differences.

Among patients with a smear suggesting high parasite bio-
mass, having jaundice or a palpable liver had low positive
predictive value. Nevertheless, given the strong association
between jaundice and renal failure, a question arises: how
useful would chelating agents be in the prevention of renal
failure among patients with jaundice? Similarly, it has been
shown that careful rehydration in patients with obstructive
jaundice could significantly reduce acute renal failure.19
These simple measures could prevent a complication that is
associated with both high mortality and increased costs.

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Authors’ addresses: Mathieu Nacher, and Frédéric Gay, Unité IN-
SERM 511, Immunobiologie Cellulaire et Moléculaire des Infec-
tions Parasitaires, Faculté de Médecine Pitié-Salpêtrière, 91 Boule-
vard de l’Hôpital, 75634 Paris Cedex 13, France; Sombat Treepras-
ertsuk, Pratap Singhaisvanon, Udomsak Silachanroom, Suparn Van-
naphan, Sorochai Loaoresuwan, and Prolrat Wilaratanarat, Faculty of
Tropical Medicine, Mahidol University. Hospital for Tropical Dis-
Eases, 420/6 Ratchavithi Road, 10400, Bangkok, Thailand.

Reprint requests: Mathieu Nacher, Faculty of Tropical Medicine,
Mahidol University, Hospital for Tropical Diseases, 420/6 Ratchav-
thi Road, 10400 Bangkok, Thailand, Fax 662–245 72–88 (e-mail: m.nacher@lycos.com).

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