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Oxidative stress plays an important role in malaria immunity and pathogenesis. Malaria-induced oxidative stress is thought to originate from immuno-defensive reactions of the host cells against the parasite and as a result of parasite metabolism. The parasite feeds on hemoglobin and releases the highly reactive and toxic heme. This can react with molecular oxygen to form hemin and superoxide radical (O$_2^-$), a highly reactive oxygen species. In the parasite’s food vacuole heme is, however, rendered inert and nontoxic through conversion into hemozoin, the malaria pigment. Most of the quinoline antimalarials interfere with the conversion of heme to hemozoin thereby inducing its accumulation inside the food vacuole and eventually killing the parasite.

In severe malaria, parasite toxins may trigger the release of oxygen free radicals and stimulate a variety of pro-inflammatory cytokines, such as tumor necrosis factor-alpha, interleukins, gamma interferon, and nitric oxide. These pro-inflammatory factors are believed to cause much of the clinical complications observed in severe malaria with multiple organ involvement. Several studies have implicated malaria-induced oxidative stress in complications such as reduced macrophage function, reduced erythrocyte deformability, and increased activation of pro-inflammatory cytokines. In children with malaria, both blood plasma and erythrocytic lipid peroxidation are increased, whereas erythrocytic antioxidants such as glutathione (GSH) were shown to be lower in patients than in controls. Polymorphisms resulting into absence or reduced enzyme activity have been identified and linked with pathogenesis in a number of disorders and diseases characterized with increased oxidative stress. In a previous study, we observed that GSTM1-null genotype was associated with severe malaria in Cameroonian children. In this study, we have investigated the genotypic distribution of human GSTM1, GSTT1, and GSTP1 polymorphisms in mild versus severe malaria in Tanzanian children.

The clinical data and DNA samples of this study were collected in the period between July and September 2006, in a drug efficacy study of mild malaria cases in Mnyuzi, Tanga Region, Tanzania. Briefly, children 3–15 years of age with a temperature > 37.5°C or a history of fever within the last 48 hours and with Plasmodium falciparum mono-infection at a density between 500 and 100,000 parasites/μL were eligible for recruitment. Children with a hemoglobin (Hb) concentration below 8 g/dL, as measured by HemoCue (HemoCue AB, Angelholm, Sweden), were excluded. Children with severe malaria were included for the current study in the same study period. Severe anemia (Hb < 5 g/dL) was observed in all children who attended the clinic with severe malaria. Additional signs of severe disease that were examined: hyper parasitaemia (≥ 250,000 parasites/μL), metabolic acidosis manifested by respiratory distress as described by Marsh and others, cerebral malaria presented as coma score ≤ 2 (Blantyre coma scale) or impaired consciousness with Blantyre score < 3 and prostration or extreme weakness (e.g., inability to sit or stand). For severe cases treatment was initiated with quinine, according to Tanzanian National Guidelines and referred to the nearby district hospital in Korogwe in case the study physician considered this appropriate. There was no active follow-up of the outcome of severe malaria cases after the appropriate treatment was installed.

For all mild and severe malaria cases, a malaria blood slide, Hb measurement, and filter paper DNA sample were collected. A short questionnaire was used to obtain information on sex, age, disease presentation, and ethnicity of the patient. The ethical clearance for the collection of the mild malaria material was obtained from the Tanzanian National Institute for Medical Research (NIMR/HQ/R.8a Vol. XIII/446) and clearance for the collection of material of severe malaria cases was obtained from Kilimanjaro Christian Medical Center (KCMC 2006#28). In the informed consent obtained from the parents or guardians of the children, they approved the use of their children’s DNA samples to study the relation between human genetic factors and malaria disease presentation. A total of 107 mild and 50 severe malaria cases were enrolled. Parasite density in the severe malaria group ranged from 4,640–174,000 parasites/μL and was higher than in the mild malaria group (Wilcoxon-rank sum test, P < 0.001). For more group characteristics see Table 1.

The DNA extracted from the dried filter papers was done using Nucleospin Tissue kits (Macherey-Nagel, Düren,
Glutathione S-transferase (GST) polymorphisms can change the enzyme activity, which can lead to reduced detoxification of the host cell or increased availability of host GSH that might be used by the parasite. In both cases the malaria pathology could be accelerated. It is also likely that the impact of GSTs is not direct on erythrocytes but on other cells that are involved in the immune response mechanisms and that severe malarial anemia as an outcome can partly be attributed to such responses. Therefore, further studies including in vitro cellular studies to assess malarial outcomes for specific GST polymorphism genotypes are important.

In conclusion, GSTP1 and possibly GSTM1 may have protective effects against severe falciparum malaria in children. The contribution of specific GST polymorphisms to severe disease may differ between populations or geographic areas. These findings do not undermine the importance of oxidative stress in malaria clearance, but rather provide a broader perspective on the impact of oxidative stress on both the host and parasite cells.

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