Hemoglobin E and Glucose-6-Phosphate Dehydrogenase Deficiency and *Plasmodium falciparum* Malaria in the Chittagong Hill Districts of Bangladesh

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Abstract. Hemoglobin E is largely confined to south and southeast Asia. The association between hemoglobin E (HbE) and malaria is less clear than that of hemoglobin S and C. As part of a malaria study in the Chittagong Hill Districts of Bangladesh, an initial random sample of 202 individuals showed that 39% and 49% of Marma and Khyang ethnic groups, respectively, were positive for either heterozygous or homozygous hemoglobin E. In this group, 6.4% were also found to be severely deficient and 35% mildly deficient for glucose-6-phosphate dehydrogenase (G6PD). In a separate *Plasmodium falciparum* malaria case–uninfected control study, the odds of having homozygous hemoglobin E (HbEE) compared with normal hemoglobin (HbAA) were higher among malaria cases detected by passive surveillance than age and location matched uninfected controls (odds ratio [OR] = 5.0, 95% confidence interval [CI] = 1.07–46.93). The odds of heterozygous hemoglobin E (HbAE) compared with HbAA were similar between malaria cases and uninfected controls (OR = 0.71, 95% CI = 0.42–1.19). No association by hemoglobin type was found in the initial parasite density or the proportion parasite negative after 2 days of artemether/lumefantrine treatment. HbEE, but not HbAE status was associated with increased passive case detection of malaria.

INTRODUCTION

There is strong epidemiologic and biomolecular evidence supporting the protective effects of hemoglobin S and C on severe malaria, but their protective effects on uncomplicated and asymptomatic malaria are more contested.^{1–5} In southeast Asia hemoglobin E (HbE; $\beta^{26\text{Glu}\rightarrow\text{Lys}}$) predominates, with reported prevalence of 30–45% (as high as 74% documented among an ethnic minority group in northeastern Thailand).^{6,7} Those with homozygous hemoglobin E (HbEE) have mild disease with anemia, splenomegaly, and hemolysis, while the heterozygous state is generally asymptomatic.⁸ A study suggested a selective population advantage of high HbE allele frequencies,⁹ consistent with higher rates of hemoglobinopathies in malaria-infected areas, a theory proposed by Haldane over 60 years ago.¹⁰

However, there is a general paucity of evidence on the role of HbE in malaria pathogenicity. One study showed the rate of development of severe, acute malaria was only 2.4% for heterozygous hemoglobin E (HbAE) patients compared with 18% for patients without HbE.¹¹ In Myanmar, patients with HbAE and normal hemoglobin (HbAA) showed similar parasitemia rates and clinical severity of malaria. However, the number of HbEE patients was small and none had severe disease.¹² Another study with acute *falciparum* malaria patients being treated with artemisinin derivatives showed that patients with HbAE had significantly faster parasite clearance compared with controls with no hemoglobinopathies, a difference not demonstrated with the use of other antimalarial drugs.¹³ The authors hypothesize that this result might be related to potentiating effects of the oxidative reactions of artemisinins. One study in Thailand showed that there was

no significant difference in allele frequency of HbE between patients with mild malaria and those with cerebral malaria.⁷ This study only had three HbEE patients, none of which developed cerebral malaria.

During a 3-year malaria epidemiologic project in the Chittagong Hill Districts of Bangladesh, an area hypoendemic for *P. falciparum* and *P. vivax*, we first investigated the prevalence of HbE and glucose-6-phosphate dehydrogenase (G6PD) deficiency among the two most populous ethnic minority groups. A separate subsequent malaria case–uninfected control study assessed the association of HbE in malaria cases detected principally by passive means compared with age and location matched uninfected controls, to address the contribution of HbE to malaria hot spots. Additional questions involved the relationship of HbE and initial parasite density as well as the clearance of parasites 2 days after treatment initiation.

METHODS

The study area consisted of two unions in the Chittagong Hill Districts of Bangladesh, an area now known as hypoendemic for *P. falciparum* and *P. vivax*, with more than a dozen resident ethnic groups. As part of an epidemiology cohort study, we monitored populations year round in all age groups, performing demographic surveys, active and passive surveillance, entomological sampling and mapping. The unions consisted of about 24,000 people, divided into 24 geographic clusters of about 2.5 km² and similar population sizes of about 1,000 persons.^{14,15} Informed consent was obtained from all adult participants and guardians of child participants. This study has been approved by the Johns Hopkins Bloomberg School of Public Health and International Center For Diarrheal Disease Research, Bangladesh (icddr,b) Institutional Review Board (IRB) committees.

There were three aspects to the analysis in this study: 1) determining the cross-sectional prevalences of HbE and

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G6PD deficiency in two of the more populous local ethnic groups in which blood sampling took place in the dry season (December 2010 and April–May 2011), 2) examining the associations between HbE and *P. falciparum* malaria cases (dates of malaria range from May 29, 2010 to February 16, 2012) compared with uninfected controls with HbE sampling performed from May 2012 to August 2013 (3 months to several years after malaria episode, with all but a few over a year later), and 3) assessing the association of HbE on both initial parasite density and absence of parasites 2 days after initiation of treatment of *P. falciparum*-infected cases, as a proxy for clearance.

Estimation of prevalences of HbE and G6PD deficiency. A total of 112 Marma and 90 Khyang individuals, aged 19–31 years, already part of the larger epidemiologic study, were randomly selected using a random number generator with blood analysis done in December 2010 and March–April 2011. In the Marma tribe, there were 73 females and 39 males. In the Khyang tribe, there were 50 females and 40 males. Five women were pregnant at the time of blood draw based on documented pregnancy and birth records from our larger cohort study.

The blood samples were analyzed for malaria by rapid diagnostic test (RDT, FalciVax, Zephyr Biomedical Systems, Verna, Goa, India) and microscopy. Analysis of hemoglobin amounts and types was done using an automated hemoglobin testing system (VARIANT™ II Hemoglobin Testing System, Biorad Incorporated, Hercules, CA). Pregnant woman were excluded from the hemoglobin analysis. Laboratory analysis for G6PD deficiency was performed on the Hitachi 902 instrument (Roche Diagnostics, Basel, Switzerland) using Randox reagents (Randox Laboratories, Crumlin, United Kingdom). The G6PD level was categorized by the World Health Organization (WHO) classification using a population-defined mean of 11.8 U/g Hb, from Cambodia,¹⁶ since we lack a local population-defined mean for Bangladesh. Using 11.8 U/g Hb, we established five classes: Class I: very severely deficient with < 1% residual activity (≤ 0.12 U/g Hb); Class II: severely deficient, > 1–10% residual activity (> 0.12 – 1.2 U/g Hb); Class III: mildly deficient, > 10–60% residual activity (> 1.2 – 7.1 U/g Hb); Class IV: normal activity, > 60–150% residual activity (> 7.1 – 17.7 U/g Hb); and Class V: increased activity, > 150% residual activity (> 17.7 U/g Hb).

Examining the associations between HbE and *P. falciparum* malaria. A second and separate aspect of the study was a nested *P. falciparum* malaria case–uninfected control analysis for HbE status and RDT- or microscopy-positive *P. falciparum* malaria among the most populous Marma ethnic group. Malaria was detected by both passive and active means from our cohort study data. The malaria case definition required a positive RDT or microscopy and was divided between 1) passive case detection when individuals had sought treatment of fever, history of fever, or other symptoms, and were found to have a positive RDT and/or microscopy and 2) active case detection in which individuals tested positive after being randomly selected for RDT/microscopy testing within a pre-determined sampling structure according to cluster and age, as used for the larger cohort study.^{14,15} From the larger epidemiologic cohort study, 170 passive malaria cases were randomly selected from 472 using a random number generator.¹⁴ All cases detected by active surveillance meeting this criteria from our cohort study were also included in this analysis. Uninfected controls were matched by age (6 months–4 years; 5 years–14 years; 15+ years), cluster (by 24 geographic areas),

and ethnic group (all Marma). Uninfected controls were chosen initially on the basis of never having had documented positive malaria by RDT/microscopy during the cohort study that began in October 2009. They also had to have been selected for random active surveillance and to have specifically tested negative for malaria within 12 weeks of the time the case tested positive, with preference to those tested closer in time to the case. Participants with HbEE and HbAE were compared with those with HbAA, including those with low A2 (excluding those with β -thalassemia trait and the two cases of *P. vivax* malaria). The two cases that had malaria twice were only included one time for this analysis. Analysis was done using McNemar's test on matched pairs with a sensitivity analysis using conditional logistic regression, including those without matched pairs.

Using McNemar's test for the passive case–control analysis, 142 matched pairs were available after eliminating those with β -thalassemia. For the 142 eligible pairs, when comparing any HbE versus normal, if we assume a 50% probability of exposure, a power of 80%, a probability of an exposure discordant pair of 50%, and an alpha of 5%, we would at a minimum be able to detect an odds ratio (OR) of 2. When comparing HbEE versus normal, the minimum OR detectable (after reducing sample size by 36%) is 2.8. For the active sample (21 matched pairs), when similar comparisons are made using the same assumptions, we do not have the power to detect anything but the most extreme relationships. For any HbE versus normal, there is a minimum detectable OR of 7.8 and for HbEE versus normal, the minimum detectable OR would be 12.5 or greater.

Assessment of initial parasite density and absence of parasites 2 days after initiation of treatment. Some of the malaria cases had only an initial RDT available, rather than a blood film for quantification. The geometric means and 95% confidence intervals (CIs) were calculated on the log-normalized parasite density using Student *t* tests. The means were compared through analysis of variance (ANOVA) tests. We also conducted a case–control analysis comparing the HbE status of subjects from any ethnic group who had persistent parasitemia 2 days after the initiation of treatment with artemether/lumefantrine ($N = 48$), with those *P. falciparum* malaria patients with absent parasites 2 days later ($N = 288$). The controls were a convenience sample selected from those *P. falciparum* malaria patients who were not positive on Day 2 in our active and passive studies described above.

All analyses were conducted using R Statistical software (Vienna, Austria).¹⁷

RESULTS

Population hemoglobin analysis. Initially, blood samples were taken for hemoglobin analysis on 202 study participants, 19–31 years of age, from the two most populous ethnic groups, in the dry season (December 2010 and late April to early May 2011). None of the participants tested positive for malaria by RDT or microscopy at this time. In total, 36% were heterozygote for HbE and 8% were homozygous, as shown in Table 1. The Khyang ethnic group had 11% ($N = 10$) homozygous for HbE and 38% ($N = 34$) heterozygous, while the Marma ethnic group had 5% ($N = 6$) homozygous and 34% ($N = 38$) heterozygous. These differences were not significant by Fisher's exact test (two-sided *P* value of 0.12). Hemoglobin Hope, a beta chain Gly136(H14)Asp mutation, was

TABLE 1

Hemoglobin type and quantity analysis for random sampling of the Marma and Khyang ethnic groups excluding six pregnant women ($N = 196$)

Frequency distribution	Number	Percentage	Hemoglobin mean (g/dL)	Hemoglobin quartiles 25%; 75%	Hemoglobin minimum; maximum
HbEE	16	8.2	12.2	10.9; 13.1	9.8; 14.8
HbAE	69	35.2	12.5	11.4; 13.7	7.1; 16.0
Normal study	98	50.0	12.9	11.7; 14.1	6.9; 18.1
Hb Hope	3	1.5	12.5	11.4; 14.3	8.9; 14.6
Low Hb A2 level	10	5.1	11.45	10.8; 12.8	7.9; 13.2
Total	196	100.0	12.6	11.5; 13.8	6.9; 18.1

HbAE = heterozygous hemoglobin E; HbEE = homozygous hemoglobin E; Hb Hope = a beta chain Gly136(H14)Asp mutation.

found in 1.5% of those sampled ($N = 3$). Low hemoglobin A2 was found in 5% ($N = 10$). Hemoglobin A2, a normal variant consisting of two alpha and two delta chains, may be elevated in people with β -thalassemias and has a lowered value in people with iron-deficient anemia or α -thalassemias, persistence of fetal hemoglobin, or hemoglobin H (thalassemia intermedia).¹⁸

The overall mean hemoglobin among the 196 (6 removed because of pregnancy at the time of blood draw) malaria-negative Marma and Khyang people (aged 19–31 years) tested in the initial prevalence survey was 12.6 g/dL (13.6 g/dL male, 12.0 g/dL female). We did not find a significant difference in hemoglobin levels by race or by HbE type in this population when controlling for age, sex, and pregnancy status (P value = 0.43).

G6PD deficiency. Of the 202 participants tested for G6PD deficiency, the mean was 7.42 U/g Hb, with 58.9% having normal G6PD activity, 34.7% having mild deficiency, and 6.4% having severe deficiency (Figure 1). Unlike with hemoglobin levels, where no significant difference was found by ethnic group, we found that there was a significantly higher proportion of cases that were G6PD deficient among the

Marma, with a mean 6.68 U/g Hb, compared with that of Khyang who had a mean G6PD activity of 8.35 U/g Hb, a difference of 1.68 (95% CI = -2.57 to -0.78), and a P value of 0.0003 using Welch two-sample t test. Thus, half the Marma population had at least a mild deficiency compared with 30% of the Khyang population.

Case-control analysis of HbE and malaria. From the larger epidemiologic cohort, 170 passive malaria cases were randomly selected from a total of 472 cases appearing over the 2-year period. A total of 146 uninfected controls matched by age, cluster, and ethnic group were identified, located, and tested for hemoglobin amount and type. Of these 146 matched pairs, three were eliminated with β -thalassemia trait, one was eliminated as the case had *P. vivax* malaria, leaving 142 *P. falciparum* cases and matched uninfected controls (Table 2). We found significantly higher odds of malaria among patients with HbEE compared with those with HbAA (including those with low A2), based on our sample of age, location and ethnic group matched pairs using McNemar's exact test (OR = 5.0, 95% CI = 1.07–46.93; P value = 0.039). If the matched design is ignored, the unpaired analysis has similar prevalence of HbEE among cases and controls (10 and 12 out of the 142, respectively), while the paired analysis reached significance in the HbEE matched comparison of 10 to 2 normal Hb. Although not statistically significant, we found the opposite trend when comparing those with HbAE to HbAA (OR = 0.71, 95% CI = 0.42–1.19; P value = 0.22). A sensitivity analysis using conditional logistic regression, including the cases or controls that were collected without a match for both the active and passive analysis, resulted in similar ORs and no change in the conclusions of the study.

To examine malaria cases identified from the active surveillance, 24 randomly selected subjects who tested positive for

FIGURE 1. Frequency of glucose-6-phosphate dehydrogenase (G6PD) deficiency and World Health Organization (WHO) classification categories. The frequency of 202 G6PD enzyme levels from the random sampling of Marma and Khyang tribal members. Class I: very severely deficient with < 1% residual activity (≤ 0.12 U/g Hb); Class II: severely deficient, > 1–10% residual activity (> 0.12 – 1.2 U/g Hb); Class III: mildly deficient, > 10–60% residual activity (> 1.2 – 7.1 U/g Hb); Class IV: normal activity, 60–150% residual activity (7.1 – 17.7 U/g Hb); Class V: increased activity, > 150% residual activity (> 17.7 U/g Hb).

TABLE 2

Hemoglobin type and malaria in matched pairs for passive *Plasmodium falciparum* malaria case—uninfected control analysis

Uninfected controls	Cases			Total
	HbEE	HbAE	Normal	
HbEE	1	7	2*†	10
HbAE	1	26	38†‡	65
Normal	10*†	27†‡	30	67
Total	12	60	70	142

HbAE = heterozygous hemoglobin E; HbEE = homozygous hemoglobin E.

McNemar's test results:

*For HbEE vs. HbAA analysis: Odds ratio (OR) = 5.0, 95% confidence interval (CI) = 1.07–46.93; McNemar's exact two-tailed P value = 0.039.†For any HbE vs. HbAA analysis: OR = 0.93, 95% CI = 0.58–1.48; McNemar's exact two-tailed P value = 0.82.‡For HbAE vs. HbAA analysis: OR = 0.71, 95% CI = 0.42–1.19; McNemar's exact two-tailed P value = 0.22.

Conditional logistic regression results:

HbEE vs. HbAA: OR = 5.0, 95% CI = 1.1–22.8; P value = 0.038.Any HbE vs. HbAA: OR = 0.92, 95% CI = 0.59–1.45; P value = 0.73.HbAE vs. HbAA: OR = 0.71, 95% CI = 0.43–1.16; P value = 0.18.

TABLE 3
Hemoglobin type and initial parasite density

	HbEE	HbAE	β -Thalassemia trait	HbAA
Active cases ($N = 23$)*				
Absent parasitemia N (%)	—	2 (18)	0 (0)	1 (9)
Positive parasitemia N (%)	—	9 (82)	1 (100)	10 (91)
Range (parasites/ μ L)†	—	200–17,320	—	200–11,200
Geometric mean parasite density (parasites/ μ L)‡§ (95% CI)	—	1,694 (561–5,113)	1,000	1,084 (391–3,009)
Passive cases ($N = 147$)§				
Absent parasitemia N (%)	1 (8)	9 (14)	0 (0)	15 (22)
Positive parasitemia N (%)	12 (92)	54 (86)	3 (100)	53 (78)
Range (parasites/ μ L)†	480–31,400	240–112,000	3,280–14,240	480–60,000
Geometric mean parasite density (parasites/ μ L)‡ (95% CI)	2,232 (845–5,900)	4,175 (2,815–6,193)	6,175 (947–40,271)	4,912 (3,425–7,044)

CI = confidence interval; HbAA = normal hemoglobin; HbAE = heterozygous hemoglobin E; HbEE = homozygous hemoglobin E.

*Blood samples not collected for one of 24 active cases (1 HbEE).

†Range and geometric mean parasite density are calculated including all positive values.

‡ANOVA comparison of geomeans $F = 0.25$, P value = 0.78.

§Blood films not available for 22 of 169 passive cases (1 β -thalassemia trait, 8 HbAE, 1 HbEE, 12 HbAA).

||ANOVA comparison of geomeans $F = 1.13$, P value = 0.34.

malaria during the active surveillance were compared with age, cluster, and ethnic group matched uninfected controls. Of the 24 infected subjects, 23 had participating matched pairs. Of these, one case and one control had β -thalassemia trait, leaving 21 matched pairs for analysis. With this small sample size, we did not see any significant difference in proportion having HbEE and HbAE among matched malaria cases and controls. The conditional logistic regression analysis did not change this finding.

Next we examined if the initial parasite density of malaria cases might be different by hemoglobin status. Of the 24 cases from active surveillance (some were RDT only), we found an initial parasite density on 20, with a geometric mean of 1,320 (95% CI = 694–2,511). Of the 170 passive *P. falciparum* malaria cases, 122 had available parasite density, with a geometric mean of 4,254 (95% CI = 332–5,455). The mean parasite density did not significantly differ by hemoglobin type in either the active ($F = 0.25$, P value = 0.78) or passive studies ($F = 1.13$, P value = 0.34) (Table 3).

The last part of the analysis related to an examination of the hemoglobin status of those patients who had positive *P. falciparum* parasitemia (possible delayed clearance) 2 days after initiating treatment with artemether/lumefantrine ($N = 47$). This group of 47 patients, parasite positive on day 2, was made up of seven malaria case found positive through our active surveillance and 40 patients with malaria detected by the passive surveillance, representing a range of ethnicities including 31 Marma, four Chakma, one Tripura, two Khyang, seven Bengali, and two from other ethnic groups. The matched infected controls in this analysis were all the *P. falciparum* cases for which we had conducted hemoglobin type analysis and had cleared parasites by day 2 (20 active and 165 passive). We also compared the Marma cases that had persistent parasitemia to controls, as all controls were Marma. Although the

point estimates of the ORs were below 1 (suggestive of a possible faster clearance with HbE), we did not see a significant difference of absence of parasites by day 2 stratified by HbE status (Table 4). This result did not change if we separated active and passive malaria cases.

DISCUSSION

In southeast Asia, an area that was hyperendemic for malaria, HbE has high prevalences.

In the Chittagong Hill Districts, malaria has persisted in this forested, hilly environment with seasonal heavy rains. In this study, a little under half of the randomly sampled Marma and Khyang populations had some variant of HbE. Among mild clinical malaria cases collected through passive sampling, there is some evidence of an increased proportion of homozygous HbEE compared with HbAA among those with malaria compared with matched uninfected controls. There was no significant difference in HbAE levels compared with normal levels among cases and controls. With our asymptomatic malaria case-control study collected through active sampling, we had too small sample size to define the relationship between malaria and HbE.

Differences have been noted in hospitalized, severe malaria cases in regard to HbE status with some evidence of a protective effect of HbEE¹² and HbAE¹¹ from severe complications of malaria in a hospital setting. In this study, we did not have enough severe malaria cases to analyze the association between severe disease and HbE status, but rather we compared those without infection to those who presented with mild clinical infection in a community setting. It is possible that the protective effects may only apply to reducing severe symptoms, a result found by Hutagalung and others,¹¹ rather than preventing infection or onset of

TABLE 4
Hemoglobin type in parasite clearance analysis

	HbEE N (%)	HbAE N (%)	Any HbE N (%)	β -Thalassemia trait N (%)	HbAA N (%)	N
All day 2 positive cases	3* (6)	16 (34)	19‡ (40)	3 (6)	25 (53)*‡	47
Day 2 positive Marma cases	2† (6)	11 (35)	13§ (42)	1 (3)	17 (55)†§	31
Day 2 negative Marma cases	13*† (7)	80 (43)	93‡§ (50)	5 (3)	87 (47)*†‡§	185

HbAA = normal hemoglobin; HbAE = heterozygous hemoglobin E; HbE = hemoglobin E; HbEE = homozygous hemoglobin E.

*For HbEE vs. HbAA analysis using all day 2 + cases: OR = 0.80, 95% CI = 0.14–3.26; Fisher's exact two-tailed P value = 1.

†For HbEE vs. HbAA analysis using only Marma day 2 + cases: OR = 0.79, 95% CI = 0.08–4.01; Fisher's exact two-tailed P value = 1.

‡For any HbE vs. HbAA analysis: OR = 0.71, 95% CI = 0.34–1.45; Fisher's exact two-tailed P value = 0.40.

§For any HbE vs. HbAA analysis using only Marma day 2 + cases: OR = 0.72, 95% CI = 0.30–1.67; Fisher's exact two-tailed P value = 0.44.

mild symptoms of infection. As we lacked sufficient power to compare asymptomatic cases found through active surveillance, it is not possible to parse out this difference with this analysis. However, as Billo and others¹⁹ have suggested, it is possible that the cross-sectional analyses used were not robust enough to identify these differences and that a longitudinal approach was needed. The small sample size and lack of significance when comparing the marginal frequencies of hemoglobin status by malaria suggest that further research on this question is needed to fully understand the relationship of HbE and malaria. With increased power, factors such as age and geographic clusters could be further explored. Furthermore, a study including populations with asymptomatic/submicroscopic infections, clinical mild malaria infections, as well as severe malaria may help to parse out whether the associations we found relate to HbEE or HbAE being associated with the initial malaria infection, becoming clinically symptomatic, and/or having severe symptoms and complications.

We did not find any evidence that hemoglobin type is associated with either geometric mean parasite density on the day of diagnosis or clearance of parasitemia by day 2 after beginning treatment. The day 2 estimates were approximate, as the day 2 sample was collected at any time during day 2 and could have ranged by a number of hours, possibly impacting our results. Bangladesh, despite repeated measurements, has yet to show delayed clearance with artemisinin treatment as seen in Cambodia.^{20–23}

With respect to G6PD deficiency, we found 6% with severe deficiency in this population. This suggests one should screen for deficiency with a point-of-care test in this population before use of a 14-day course of primaquine used to eliminate the liver stage in *P. vivax* infections or long half-life tafenoquine.^{24,25} Single dose primaquine at 0.25 mg base/kg is well tolerated even in those severely deficient for G6PD, and is recommended by the WHO in addition to artemisinin combination therapy, for all but pregnant woman and infants under 1 year as a gametocytocidal drug for those with *P. falciparum* infections as part of an elimination strategy.²⁶ The high levels of G6PD deficiency in this area should thus not be a reason to stop the implementation of these recommendations.

Limitations of this study include inability to locate all of the controls that had been selected for our cases. The population that did not have controls was slightly (although not statistically significantly) younger. As age and location may be related to our outcome, this was a limiting factor of our analysis. The analysis, particularly for the malaria cases detected through active surveillance, was limited by sample size. For the elimination studies, the number of hours defined as '2 days' varied depending on when during the day the patients were originally tested/treated and when field workers were able to return to their villages. Thus the timing is not as controlled nor measured as often as some hospital-based parasite elimination studies. With respect to G6PD measurements, reticulocyte count was not taken at the time of blood draw. As patients were not ill at the time of blood draw, this is unlikely to substantially impact results. However, we would underestimate true rates of deficiency in the rare cases of high reticulocytosis elevating levels of G6PD.

In summary, we found high levels of G6PD deficiency, HbE, and anemia in this population. Uncomplicated malaria was more common among patients with homozygous HbE compared with controls, while no difference was found in the

prevalence of malaria in patients with HbAE compared with normal patients. Focal populations of certain ethnic groups with remnant high prevalence of HbEE may provide an increased risk of uncomplicated malaria secondary to either longer duration of infection or increased risk of infection, and thus malaria control in these populations is necessary for elimination programs to be successful.

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