α^+-Thalassemia Protects against Anemia Associated with Asymptomatic Malaria: Evidence from Community-Based Surveys in Tanzania and Kenya

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Background. In hospital-based studies, α^+-thalassemia has been found to protect against severe, life-threatening falciparum malaria. α^+-Thalassemia does not seem to prevent infection or high parasite densities but rather limits progression to severe disease—in particular, severe malarial anemia. We assessed to what extent α^+-thalassemia influences the association between mild, asymptomatic *Plasmodium falciparum* infection and hemoglobin concentration.

Methods. The study was based on 2 community-based surveys conducted among afebrile children (0.5–8 years old; $n = 801$) in Kenya and Tanzania.

Results. Among children without inflammation (whole-blood C-reactive protein concentration $\leq$10 mg/L), *P. falciparum* infection was associated with only small reductions in hemoglobin concentration, and effects were similar across α-globin genotypes. By contrast, the reduction in hemoglobin concentration associated with *P. falciparum* infection accompanied by inflammation was larger and strongly depended on genotype (normal, −21.8 g/L; heterozygous, −16.7 g/L; and homozygous, −4.6 g/L). Relative to children with a normal genotype, this difference in effect was 5.1 g/L (95% confidence interval [CI], −1.0 to 11.1 g/L) for heterozygotes and 17.2 g/L (95% CI, 8.3 to 26.2 g/L) for homozygotes (estimates are adjusted for study site, age, height-for-age *z* score, and iron deficiency).

Conclusions. α^+-Thalassemia limits the decline in hemoglobin concentration that is associated with afebrile infections, particularly those that are accompanied by inflammation.

α^+-Thalassemia is highly prevalent in sub-Saharan Africa, Asia, and Melanesia [1]. Normal individuals have duplicate α-genes on each chromosome 16. By contrast, those with heterozygous α^+-thalassemia have a loss of 1 α-gene, resulting in 3 functional α-genes ($-\alpha/\alpha\alpha$), whereas homozygotes have only 2 functional α-genes ($-\alpha/-\alpha$). Heterozygosis is characterized by slight hematological changes, whereas homozygotes generally have mild microcytic anemia [1–3].

Case-control studies have consistently shown that α^+-thalassemia protects against severe, life-threatening malaria [4–7]. This protective effect is most pronounced in homozygotes and acts primarily against severe malarial anemia [4–6, 8]. No study has found evidence of a reduced incidence of uncomplicated malaria episodes in α^+-thalassemic children [8–10]. Similarly, none of the cited studies found an association between α^+-thalassemia and parasite prevalence or density. Although the mechanisms of protection are largely unknown, these findings suggest that α^+-thalassemia prevents disease progression through mechanisms other than limiting parasite replication and that the protection conferred by α^+-thalassemia is limited to severe manifestations of disease [11, 12].

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Severe malarial anemia can develop rapidly as a result of hemolysis during acute malaria episodes [13, 14] but may also be the cumulative result of smaller reductions in hemoglobin concentration during repeated episodes of uncomplicated malaria or impaired erythropoiesis during chronic asymptomatic infections [15]. The notion that severe malarial anemia can develop gradually is supported by observations from community-based surveys and hospital studies that many cases occur with only mild or no symptoms, no reported history of fever, and relatively low parasite densities [16–19], suggesting that adaptation to low hemoglobin concentrations has taken place over time.

Recently, it has been hypothesized that the protection afforded by α^+-thalassemia is restricted to severe malarial anemia resulting from acute hemolysis, with no protection conferred against gradual reductions in hemoglobin concentrations during chronic or repeated infections [6]. This was supported by findings from a large cohort study in Kenyan children, which found no evidence that α^+-thalassemia influences the degree of anemia associated with episodes of uncomplicated falciparum malaria [8].

By contrast, on the basis of a birth cohort study in Melanesia, Oppenheimer et al. [20] suggested that malarial anemia is relatively mild in children with α^+-thalassemia, although this effect was detected only at the age of 6 months and was no longer present at 12 months. Among nonhospitalized Nigerian children, malaria-associated anemia seemed less pronounced in heterozygous children than in children with a normal genotype and was virtually absent in homozygotes, although the difference in the effect of infection on hemoglobin concentration was not statistically significant [21]. Similarly, a study in Ghana among predominantly asymptomatic pregnant women showed that the effect of *Plasmodium falciparum* infection on hemoglobin concentration was less pronounced in women with α^+-thalassemia than in their counterparts with a normal genotype [22].

We hypothesized that α^+-thalassemia exerts its protective effect against severe malarial anemia by preventing the gradual decline in hemoglobin concentrations during milder or asymptomatic *P. falciparum* infections. The present study aimed to investigate to what extent α^+-thalassemia influences the degree of anemia associated with *P. falciparum* infection among afebrile children recruited during community-based surveys.

METHODS

Study area and population. We used data from 2 community-based surveys. One survey was conducted in a lowland area in northern Tanzania, around the Segera and Kwedizinga wards in Handeni District, in May–July 2006. The other survey was conducted in Marafa, the hinterland of Malindi District, in the coastal lowlands of Kenya, in May 2004. Malaria transmission is intense and perennial in both areas, with virtually all infections due to *P. falciparum*. In both areas, poor farmer families predominate, and access to health care is limited. The present study was approved by the responsible ethics review committees in The Netherlands, Tanzania, and Kenya; informed consent was obtained from community leaders, local government officials, and parents or guardians.

Sampling methods and eligibility criteria. For the Tanzanian survey, a census list was made listing all resident children aged 6–72 months in the study area. By means of this list, 16 children were randomly selected from 19 communities, resulting in a total of 304 children. Children were eligible to participate in the survey if they had no signs of severe febrile disease or severe malnutrition at the time of assessment. Not all selected children participated; nonparticipating children were on average younger than their participating peers. To avoid bias on replacement, we resampled children from within the same age category as the nonparticipants (the age categories were 6–18, 18–36, and 36–72 months) until 304 participating children were obtained.

Of 325 children selected, 21 did not participate in the study. Two children died between the time of the census and the start of the study, 3 children were temporarily absent, the parents of 10 children refused consent, 4 children did not show up for unknown reasons, and 2 children were not eligible because they were sick and were referred to the hospital on the day of recruitment.

The Kenyan survey ($n = 516$) was conducted in 4 schools, among all children who were enrolled in nursery or the first year of primary school, as a baseline assessment for an intervention trial (details have been reported elsewhere) [23]. Three weeks before the baseline assessment of the trial, 528 children were screened for eligibility. We excluded 7 children with hemoglobin concentrations <70 g/L and 5 who migrated, were absent during this 3-week period, or were likely to drop out during the subsequent trial.

Field procedures. All children were examined by a clinical officer, who also measured axillary temperature. Anthropometric measurements were collected from all children. Venous blood was collected in containers with sodium heparin as anticoagulant (Becton Dickinson). Children were treated free of charge for common childhood infections and anemia according to the guidelines of the Kenyan and Tanzanian Ministries of Health.

Laboratory procedures. In Tanzania, parasitemia was detected by rapid immunochromatographic assay (OptiMAL; Flow). Contrary to most other types of rapid dipstick tests, this test detects *P. falciparum*–specific lactate dehydrogenase (pLDH), which is produced only by live parasites. The test result becomes negative rapidly after parasite clearance [24, 25]. In Kenya, parasitemia was detected by conventional microscopy. Whole-blood hemoglobin concentrations were measured using hematology analyzers (Beckman Coulter and KX-21 [Sysmex] for samples collected from Tanzania and Kenya, respectively).

Plasma was separated by centrifugation, transferred to cryotubes, and stored in liquid nitrogen. We preserved red blood cells, including the buffy coats (90 μL), for subsequent α^+-thalassemia genotyping, at 4°C in microcentrifuge tubes containing a DNA-stabilizing buffer (AS1; Qiagen). As indicators of iron status and inflammation, plasma concentrations of ferritin and C-reactive protein were measured using a Behring nephelometer (BN ProSpec; Dade-Behring) in The Netherlands (Meander Medical Centre). DNA was extracted using the QIAamp DNA Blood Mini Kit (Qiagen), in accordance with the manufacturer's instructions. The $-\alpha^{3.7}$ deletion type of α^+-thalassemia was determined by polymerase chain reaction assays, as described by Liu et al. [26]. Other types of α^+-thalassemia, including the $-\alpha^{4.2}$ type, occur only sporadically in Africa [1, 21].

Statistical analyses. We used the following definitions: current infection, the presence of at least 1 asexual *P. falciparum* parasite on microscopic examination of blood smears from Kenyan children or a positive result for the pLDH test in blood from Tanzanian children; inflammation, plasma C-reactive protein concentration >10 mg/L [27]; anemia, hemoglobin concentration <110 or <115 g/L for children aged <5 or ≥5 years, respectively; and iron deficiency, plasma ferritin concentration <12 or <15 μg/L for children aged <5 or ≥5 years, respectively [28]. Because it is difficult to interpret estimates of iron deficiency in a population with a high prevalence of both malaria and inflammation (both affect plasma ferritin concentrations independently of iron status), we restricted this analysis to children without inflammation.

All data were entered into a dedicated Microsoft Access database and were cleaned and analyzed using SPSS for Windows (version 13.0; SPSS). Anthropometric *z* scores were calculated using Epi Info software (version 3.3.2; http://www.cdc.gov/epiinfo). For normally distributed variables, we calculated means, SDs, and 95% confidence intervals (CIs). For variables that were not normally distributed, we calculated the geometric mean. We used the normal approximation of the binary distribution to obtain prevalence differences and their corresponding 95% CIs. For variables that could not be normalized by log transformation, we assessed group differences by Mann-Whitney *U* or Kruskal-Wallis tests.

We assessed the effect of *P. falciparum* infection on hemoglobin concentrations in a multivariate linear regression model that adjusted for study site, age, height-for-age *z* score, and iron deficiency. In this model, we evaluated whether the effect of *P. falciparum* infection depended on genotype by examining interaction terms (dummy variables for α^+-thalassemia genotype times infection status). We did not adjust for the presence of inflammation because we considered that the effect of infection on hemoglobin concentration may be mediated at least in part through inflammation. Thus, we repeated the above analysis stratifying children with *P. falciparum* infection by inflammation status. The groups thus formed were mutually exclusive and were compared with the reference group of children without malaria and without inflammation. For each of the groups, we used multivariate analysis to evaluate directly whether the effect of infection on hemoglobin concentration depended on genotype.

RESULTS

In Kenya and Tanzania, 10 and 9 children, respectively, were febrile at the time of examination and were excluded from the analysis. The characteristics of the remaining 801 children are shown in table 1. Eight Tanzanian children were afebrile but were reported sick by the clinical officer (5 were weak without localized signs but with a history of fever, 1 had signs of an upper respiratory tract infection, 2 had signs of a gastrointestinal infection, and 1 was completing a course of quinine for malaria). Mothers of the Tanzanian children reported the following symptoms at the time of examination: history of fever during the last 24 h (23%), cough (43%), vomiting or diarrhea (16%), ear problems (5%), and skin lesions (19%). For Kenyan children, none of the children included in the study were reported sick by the clinical officer, but mothers were questioned about the history of illness during the previous 2 weeks. The following symptoms were reported: history of fever (8%), cough (18%), gastrointestinal symptoms (11%), ear problems (1%), and skin problems (5%).

The Tanzanian children were younger than the Kenyan children and had lower hemoglobin concentrations. There were no children with a hemoglobin concentration <50 g/L. In Kenya and Tanzania, the prevalence of *P. falciparum* infection was similar, but the prevalence of inflammation was higher in Tanzania (78/295 [26%]) than in Kenya (35/506 [7%]). Children with *P. falciparum* infection had a higher prevalence of inflammation than did those without infection (21% vs. 8%; difference, 13% [95% CI, 9% to 18%]) and higher plasma concentrations of C-reactive protein ($P < .001$; Mann-Whitney *U* test). We found no evidence that parasite density was different between the study sites ($P = .11$; Mann-Whitney *U* test), but the frequency of inflammation among children with *P. falciparum* infection was lower among the children from Kenya (25/265 [9%]) than among those from Tanzania (58/134 [43%]). In the following paragraphs, data presented are from both study sites combined, unless otherwise specified.

For 47 children, α-globin genotyping results were not available because samples were unclearly labeled or because of technical problems. Of the remaining children, 12% were homozygous, 46% were heterozygous, and 42% had a normal genotype. Compared with their Tanzanian peers, the children in Kenya had a higher prevalence of both heterozygous and homozygous α^+-thalassemia. There was no evidence that parasite density was associated with genotype ($P = .67$; Kruskal-Wallis test).

Table 1. Characteristics of the study population, by $\alpha^{+3.7}$ genotype and study site.

	Tanzania				Kenya			
Characteristic	Normal	Heterozygous	Homozygous	Undetermined	Normal	Heterozygous	Homozygous	Undetermined
Indicated genotype, % (no.)	62.1 (169)	33.5 (91)	4.4 (12)	. . . (23)	30.7 (148)	52.7 (254)	16.6 (80)	. . . (24)
Age, years	2.7 ± 1.6	2.9 ± 1.5	2.3 ± 2.0	2.6 ± 1.6	5.9 ± 1.5	5.9 ± 1.5	5.8 ± 1.5	5.9 ± 1.3
Hemoglobin concentration, g/L	105.3 ± 17.2	107.6 ± 17.1	94.2 ± 14.3	104.3 ± 19.2	113.6 ± 11.8	111.9 ± 10.3	104.8 ± 11.5	114.2 ± 8.1
Anemia, % (proportion)[a]	55.6 (94/169)	48.4 (44/91)	100 (12/12)	52.2 (12/23)	39.9 (59/148)	59.4 (151/254)	80.0 (64/80)	45.8 (11/24)
Plasma ferritin concentration, geometric mean, μg/L[b]	19.7	22.0	21.5	13.8	23.8	22.6	27.3	21.3
Iron deficiency, % (proportion)[a,b,c]	13.2 (16/121)	16.7 (12/72)	16.7 (1/6)	22.2 (4/18)	16.8 (23/137)	17.8 (43/241)	5.6 (4/72)	23.8 (5/21)
Inflammation, % (proportion)[a]	28.4 (48/169)	20.9 (19/91)	50.0 (6/12)	21.7 (5/23)	7.4 (11/148)	5.1 (13/254)	10 (8/80)	12.5 (3/24)
Plasmodium falciparum infection, % (proportion)[a]	49.1 (83/169)	39.6 (36/91)	50.0 (6/12)	39.1 (9/23)	48.6 (72/148)	53.5 (136/254)	56.3 (45/80)	50 (12/24)
Height-for-age *z* score	−1.76 ± 1.27	−1.64 ± 1.23	−1.57 ± 0.89	−1.41 ± 0.87	−0.77 ± 0.72	−0.76 ± 0.72	−0.85 ± 0.72	−0.58 ± 0.59

NOTE. Data are mean ± SD values, unless otherwise indicated.

[a] Anemia, iron deficiency, inflammation, and *P. falciparum* infection were defined as specified in the main text.

[b] For heterozygotes in Tanzania, $n = 90$ because of 1 missing value for plasma ferritin concentration.

[c] The results for iron deficiency are restricted to children without inflammation.

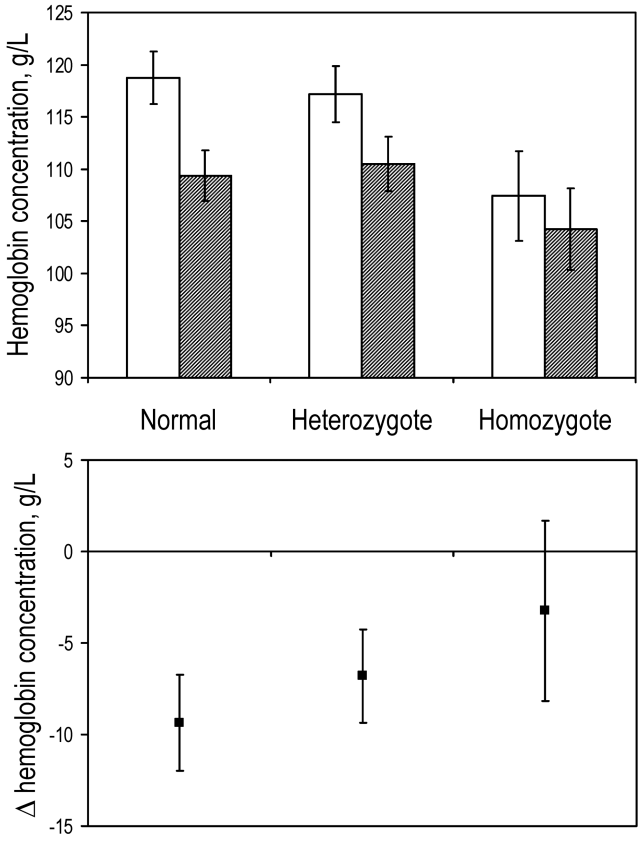

Figure 1. Associations between hemoglobin concentration and malarial infection, by α^+-globin genotype. Hemoglobin concentrations indicated are group means *(top)* or differences in means *(bottom),* as obtained by multivariate regression analysis that adjusted for study site, age, height-for-age *z* score, and iron deficiency. White bars represent children without *Plasmodium falciparum* infection, and dark gray bars represent children with *P. falciparum* infection. Error bars show 95% confidence intervals.

In children with normal, heterozygous, and homozygous genotypes, *P. falciparum* infection was associated with reductions in hemoglobin concentration of 9.3, 6.7, and 3.2 g/L, respectively (figure 1). Compared with the effect observed in children with a normal genotype, this reduction appeared smaller in heterozygotes (difference, 2.6 g/L [95% CI, −1.0 to 6.3 g/L]) and was clearly smaller in homozygotes (difference, 6.1 g/L [95% CI, 0.5 to 11.7 g/L]). This pattern of protection by α^+-thalassemia was seen consistently within the populations from both study sites, although it seemed less pronounced in the Kenyan children (data not shown).

Among children without inflammation, *P. falciparum* infection was associated only with small reductions in hemoglobin concentration, and the effects were similar across α-globin genotypes. By contrast, the reduction in hemoglobin concentration associated with *P. falciparum* infection plus inflammation was larger and strongly depended on genotype (figure 2); among children with a normal genotype, this reduction was 21.8 g/L (95% CI, 18.0 to 25.6 g/L), compared with 16.7 g/L (95% CI, 11.9 to 21.9 g/L) in heterozygotes and only 4.6 g/L (95% CI, −3.5 to 12.7 g/L) in homozygotes. Relative to children with a normal genotype, this difference in effect was 5.1 g/L (95% CI, −1.0 to 11.1 g/L) and 17.2 g/L (95% CI, 8.3 to 26.2 g/L) for heterozygotes and homozygotes, respectively. These estimates were adjusted for study site, age, height-for-age *z* score, and iron deficiency. There were only 27 children with inflammation but without malaria. Among these children, we found no evidence of protection conferred by α-thalassemia, but the estimates in this group were imprecise (data not shown).

Among children without infection and without inflammation, homozygotes had lower hemoglobin concentrations than did children with a normal genotype (difference in means, −11.1 g/L [95% CI, −7.0 to −15.2 g/L]) (figure 2). The opposite was observed among children with both *P. falciparum* infection and inflammation; among these children, homozygotes tended to have higher hemoglobin concentrations than children with a normal genotype (difference, 6.2 g/L [95% CI, −1.8 to 14.2 g/L]).

Reanalysis of these data by study site led to similar results and conclusions, although the estimates obtained were less precise owing to smaller numbers. Both in Tanzania and Kenya, the *P. falciparum*–associated hemoglobin reduction in the presence of inflammation was significantly smaller in homozygous children than in children with a normal genotype (difference in effect for children from Tanzania, 24.2 g/L [95% CI, 2.8 to 45.7 g/L]; difference for children from Kenya, 13.6 g/L [95% CI, 2.6 to 24.5 g/L]).

As a last step, we evaluated plasma concentrations of C-reactive protein among children with *P. falciparum* infection by genotype. In the presence of *P. falciparum* infection, plasma C-reactive protein concentrations were lower ($P = .002$; Mann-Whitney *U* test) and inflammation occurred less frequently in heterozygous children (14% vs. 29%; difference, 15% [95% CI, 10% to 20%]) than in their counterparts with a normal genotype. Similarly, among children with *P. falciparum* infection, inflammation occurred less frequently in homozygotes than in their peers with a normal genotype (18% vs. 29%; difference, 11% [95% CI, 2% to 20%]), but the group was small and we found no support for a difference in C-reactive protein concentrations ($P = .10$; Mann-Whitney *U* test). Because C-reactive protein concentrations could not be normalized by log transformation, we could not conduct multivariate analyses to adjust for potential confounding by study site or age.

DISCUSSION

The data from our survey of afebrile children from communities with intense malaria transmission support our hypothesis that α^+-thalassemia protects against the decline in hemoglobin concentration associated with mild or asymptomatic *P. falciparum*

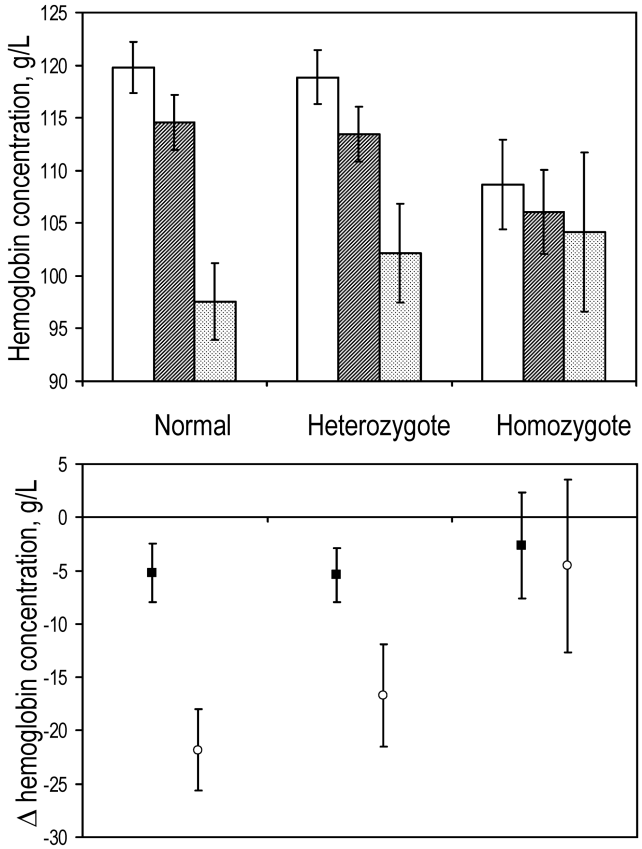

Figure 2. Hemoglobin concentration in groups defined by malarial infection, inflammation, and α^+-globin genotype. Hemoglobin concentrations indicated are group means *(top)* or differences in means *(bottom),* as obtained by multivariate regression analysis that adjusted for study site, age, height-for-age *z* score, and iron deficiency. White bars represent children without *Plasmodium falciparum* infection and without inflammation (plasma C-reactive protein concentration $\leq$10 mg/L), dark gray bars represent children with *P. falciparum* infection but without inflammation, and light gray bars represent children with *P. falciparum* infection and inflammation. Error bars show 95% confidence intervals. Sample sizes are as follows (from left to right): $n = 147$, $n = 164$, $n = 36$, $n = 110$, $n = 148$, $n = 42$, $n = 45$, $n = 24$, and $n = 9$. The no. of children with inflammation but without *P. falciparum* infection was not sufficient for meaningful analysis; these data were therefore left out.

infection. The protective effect was more pronounced in homozygotes than in heterozygotes and was observed when infection was accompanied by inflammation. In children without inflammation, *P. falciparum* was associated only with small reductions in hemoglobin concentrations that were similar across genotypes.

Children in Kenya were older and may have had more protective immunity against malaria than their peers in Tanzania. They also may have differed in other, unmeasured characteristics. Reanalysis of the data presented in figures 1 and 2 by study site, however, yielded similar results. In addition, the estimates were obtained from a multivariate analysis that adjusted for study site and several other factors that may have been associated with anemia. Thus, it is unlikely that our results can be explained by differences in population characteristics between the study sites. Because data were collected cross-sectionally, we can provide no insight into the dynamics of the decline in hemoglobin concentration or the preceding duration of infection at the point of measurement. We have no reason to assume, however, that the duration of infection differed between the genotype groups, and we do not think that the cross-sectional design affected the validity of our results.

Our data show that the protection conferred by α^+-thalassemia is not confined to the severe forms of malarial anemia, as has been suggested by others [6, 8]. A recent large study conducted at the Kenyan coast found that reductions in hemoglobin concentrations due to α^+-thalassemia were similar in children recruited from community-based surveys ("steady state") and those with uncomplicated malaria, suggesting no interaction between *P. falciparum* infection and α^+-thalassemia during uncomplicated disease episodes. Among children admitted to the hospital with severe malaria, however, heterozygotes and homozygotes had higher mean hemoglobin concentrations than did children with a normal genotype [8]. This suggested that the protection conferred by α^+-thalassemia mainly prevents the decline in hemoglobin concentration during the progression from episodes of uncomplicated malaria to severe malarial anemia and does not play a role during milder infections. It should be noted, however, that the group of children in the steady state probably included both children with and without (asymptomatic) malarial infection and that their infection status was not considered in the analysis. Moreover, children in this group were on average 5 years older than the patients with uncomplicated malaria. By contrast, our findings show that α^+-thalassemia already protects against malaria-associated anemia in children with *P. falciparum* infections that are accompanied by inflammation but not by febrile illness. This is compatible with our hypothesis that α^+-thalassemia protects against severe malarial anemia by preventing the gradual decline in hemoglobin concentration during repeated or chronic infections.

Because the protective effect was observed particularly when an infection was accompanied by inflammation, our findings suggest that, even in a spectrum of relatively mild infections, the severity of *P. falciparum* infection may differ between children with and those without α^+-thalassemia. This was supported by the finding that plasma concentrations of C-reactive protein were lower in infected heterozygotes and also seemed lower in homozygotes, although this analysis was not adjusted for age and study site. Nevertheless, similar findings were reported in a study conducted among predominantly asymptomatic pregnant Ghanaian women with *P. falciparum* infection, in whom α^+-thalassemia was associated with reduced frequency of inflammation [29].

It remains unknown how α^+-thalassemia can modify the inflammatory response and how it confers protection against ma-

larial anemia. α^+-Thalassemia results in an imbalance in the production of α- and β-globin chains, but there is no evidence that the resulting changes in red blood cell characteristics limit progression to severe anemia by restraining parasite development or the number of destroyed red cells [1, 11]. Whether α^+-thalassemia can modify the bone marrow response or the degree of immune-mediated destruction of nonparasitized cells during *P. falciparum* infection should be investigated. Urban et al. [30] recently suggested that activation of dendritic cell subsets during malaria may be faster, more profound, or prolonged in children with α^+-thalassemia than in their peers with a normal hemoglobin genotype, thus enhancing the acquired immune response to infected red blood cell antigens. An alternative hypothesis is that other associated polymorphisms—for example, that of red cell complement receptor 1—may contribute to the protection [31].

It should be noted that the protection conferred by α^+-thalassemia against the inflammation-associated decline in hemoglobin concentration observed in this study may not be specific for *P. falciparum* infection. In areas in which *P. falciparum* is highly endemic and a large proportion of children are asymptomatically infected, an increase in plasma concentrations of C-reactive protein cannot always be attributed to parasitemia [32]. In inflammatory processes, such cytokines as tumor necrosis factor–α, interleukin (IL)–1, IL-6, and interferon-γ limit the supply of iron to the erythron [33] and limit the proliferation of erythroid progenitor cells by suppressing the production of and responsiveness to erythropoietin [34]. Thus, any inflammatory process may exacerbate anemia by interfering with the host's ability to compensate effectively for the loss of red blood cells due to malaria. Several studies have suggested that α^+-thalassemia may restrict the consequences of inflammation in general. In a study in Papua New Guinea, α^+-thalassemia not only reduced hospitalizations due to severe malaria but also—and to a similar extent—reduced the risk of hospitalization due to other infectious diseases [5]. Similarly, other observations suggest that α^+-thalassemia protects against severe nonmalarial anemia [8, 12]. Because our sample size was insufficient for us to evaluate the protective effect among children with inflammation but without parasitemia, it remains unknown whether α^+-thalassemia protects against anemia due to inflammatory processes other than malaria. Moreover, such analysis would have been difficult to interpret, because bone marrow suppression and anemia may persist after an infection has been cleared [35–37].

In conclusion, our data show that the protective effect of α^+-thalassemia against malaria-associated anemia is not confined to severe malaria cases but is also present in mild, predominantly asymptomatic *P. falciparum* infections. This protective effect is evident only when the infection is accompanied by inflammation. This observation, in combination with the finding that the protection conferred by α^+-thalassemia seems to be restricted to the anemic forms of disease, may help direct the search for the protective mechanisms at work in α^+-thalassemia.

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Note added in proof. A recent study among Tanzanian children found that α^+-thalassemia was associated with protection against episodes of uncomplicated malaria, particularly in those aged >5 years: Enevold A, Lusingu JP, Mmbando B, et al. Reduced risk of uncomplicated malaria episodes in children with α^+-thalassemia in northeastern Tanzania. Am J Trop Med Hyg **2008**; 78:714–20.