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Elevated anti-malarial IgE in asymptomatic individuals is associated with reduced risk for subsequent clinical malaria

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Abstract

Immunological characteristics were assessed for prospective risk of clinical malaria in a longitudinally followed population in a holoendemic area of Tanzania. Baseline characteristics including crude *Plasmodium falciparum* extract-specific IgE and IgG; total IgE; and parasitological indices, e.g. number of *P. falciparum* clones, were investigated among 700 asymptomatic individuals. Cox regression analysis estimated the risk of succumbing to a new clinical episode during a 40 weeks follow up. High anti-*P. falciparum* IgE levels were associated with reduced risk of acute malaria in all age groups independently of total IgE levels. Statistically significant reduced odds ratio of 0.26 (95% CI, 0.09–0.72, $P = 0.010$) and 0.44 (95% CI, 0.19–0.99, $P = 0.047$) for the two highest fifths, respectively was observed after adjustment for age, sex, total IgE, numbers of parasite clones per infection and HIV-1 seropositivity. In contrast, high levels of malaria specific IgG or total IgE were not associated with reduced risk to succumb to a new clinical episode. A protective effect of asymptomatic multiclonal *P. falciparum* infections was also confirmed. For the first time, anti-malarial IgE levels in asymptomatic individuals in endemic area are found to be associated with reduced risk for subsequent malaria disease. Specific IgE antibodies may play role in maintaining anti-malarial immunity, or indicate other aspects of immune function relevant for protection against malaria.

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1. Introduction

Malaria remains a major threat to global health and yet the mechanisms for protective immunity are still incompletely understood. In areas of high malaria transmission, young children are the most vulnerable to disease and death due to their lack of protective immunity. Prolonged exposure to new inoculations and repeated infections, results in successive acquisition of protective immunity and part of this effect may be associated with controlled persistence of parasites without clinical symptoms.

Mechanisms for malaria immunity are highly complex with involvement of several components of immune

function. Antibody-dependent mechanisms are presumed to play an important role in protection, with a wide range of antigen-specific antibodies as well as polyclonal-antibody production. Studies on passively transferred immunoglobulins from immune adults have suggested that IgG antibodies are important in reducing parasite density during clinical malaria disease (Cohen et al., 1961; Sabchareon et al., 1991). There is also accumulating evidence of a protective role for certain IgG subclasses (Aribot et al., 1996; Taylor et al., 1998; Aucan et al., 2000; Ndungu et al., 2002). Although both total and malaria-specific IgE levels are elevated in individuals living in areas of high endemicity (Desowitz, 1989; Perlmann et al., 1994), the role of IgE in malaria is not as clearly established as for other parasites (Hussain and Ottesen, 1986; Hagan et al., 1991; Rihet et al., 1991). The elevated IgE levels seen in severe acute malaria

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patients indicate a pathogenic role of these antibodies (Perlmann et al., 1994, 1997, 1999, 2000). A protective role for *P. falciparum*-specific IgE has also been proposed (Perlmann et al., 1997, 1999) but this has not been previously demonstrated.

Here, we investigated if plasma levels of the antibody isotypes IgG and IgE are predictive of risk for subsequent acute malaria episodes. In a longitudinal study of a population in Nyamisati village, a highly endemic area the Rufiji Region, coastal Tanzania (Rooth and Bjorkman, 1992), a prospective malariometric survey measured several baseline characteristics, and subsequently recorded episodes of acute malaria over 40 weeks of follow-up. To avoid potential confounding through acute disease activity or treatment, only the individuals with no clinical malaria episodes during the baseline survey, 4 weeks before or 1 week after the survey were included in the analysis. Baseline characteristics included plasma levels of anti-*P. falciparum* (crude) IgE and IgG, total IgE as well as genetic diversity of *P. falciparum* defined as number of clones per infection.

2. Materials and methods

2.1. Study area and population

The study was conducted in Nyamisati village, situated by the Rufiji River Delta, coastal Tanzania. The area is holoendemic for malaria, with perennial transmission and some seasonal fluctuation. A research team, which also provides health care including antimalarial drugs, has lived in the village since 1985, and studied the malaria epidemiology with longitudinal follow up of the population of about 1000 individuals (Rooth and Bjorkman, 1992). In March–April 1999, preceding the heavy rain period, 890 villagers, 1–84 years of age, participated in a malariometric survey including analysis of parasite densities and clinical status.

2.2. Clinical data

Clinical episodes of malaria were detected by a passive case detection system, which is well established in this population with presentation of all fever episodes to the research unit. After diagnosis of malaria by microscopy, free treatment is administered and the episode registered. A clinical malaria episode was defined as >400 parasites/ μ l, together with reported or confirmed fever within the last day and without signs of other cause of fever (Rooth and Bjorkman, 1992). More than one episode within a period of 2 weeks was considered as a single episode.

At the time of survey 700 individuals were asymptomatic, i.e. there was no record of a clinical malaria episode 4 weeks before and 1 week after the survey. The time period was chosen due to possible confounding effect of

sulfadoxine/pyrimethamine (Fansidar[®]) treatment within 4 weeks prior to survey or potential ongoing disease activity of a clinical episode within the following week.

2.3. Sample collection

Venous blood was collected in EDTA sterile tubes; after centrifugation and separation, plasma and packed cells were stored frozen. All samples were obtained after informed consent of the participants and/or their guardians. The study was approved by the National Institute for Medical Research in Tanzania and by the Ethical Committee at Karolinska Institute (Dnr 00-084).

2.4. Detection of *P. falciparum* infections

Giemsa stained thick films were analysed by light microscopy at $1000\times$ magnification. Number of parasites per μ l of blood was enumerated in 200 microscopic fields corresponding to 0.2 μ l blood.

Plasmodium falciparum infections were also detected by nested PCR amplification of the highly polymorphic block 3 region of merozoite surface protein 2 (*msp2*) marker which provides an estimate of the number of concurrent clones (Snounou et al., 1999). DNA was purified from frozen packed red blood cells by phenol–chloroform extraction. Detection of *P. falciparum* infection by this PCR method, i.e. detection of one *msp2* allele or more, yields a higher sensitivity than microscopy with detection of 1–10 parasites per μ l whole blood.

2.5. Antibody analysis

Lysates of infected erythrocytes, (*P. falciparum* laboratory line F32), were prepared for determination of anti-malarial IgE and IgG (Perlmann et al., 1994). Briefly, microtiter plates were coated with 50 μ l of crude parasite antigen solution (20 μ g/ml) overnight at 4 °C, and then blocked with 100 μ l of 0.5% BSA in coating buffer for 3 h at 37 °C. For determination of malaria-specific IgE the test plasma were diluted 1:100 and incubated at room temperature overnight for optimal binding. The test plasma, diluted 1:1000 for total IgE and anti-malarial IgG were added to the wells and incubated at 37 °C for 1 h. The secondary antibodies were biotinylated goat anti-human IgE (Vector Laboratories, USA) (1:8000) or goat anti-human IgG (Mabtech, Nacka, Sweden) conjugated to alkaline phosphatase (1:2000), with 1 h incubation at 37 °C. ALP-conjugated Streptavidin (Mabtech, Nacka, Sweden) diluted 1:2000 were added to the biotinylated antibody and incubated under the same conditions. Sera from African donors with high antibody levels and sera from Swedish donors not exposed to malaria were used as positive and negative controls, respectively. The plates were washed four times between each incubation step. The bound secondary

antibody was quantified with p-nitrophenylphosphate (Sigma Diagnostics, USA). The optical density (OD) at 405 nm was determined in a Vmax microplate reader (Molecular Devices, Menlo Park, USA).

2.6. Statistical analysis

Our data was analysed using SPSS software (version 11.5). The analyses excluded individuals with a clinical episode of malaria 4 weeks prior and 1 week after the baseline survey. As we made no assumptions about linearity, the antibody levels were grouped into quintiles of their distributions and the middle fifth was used as the comparison group. Clinical episode of malaria was the dependent variable in Cox-regression analyses. Simultaneous mutual adjustment was made for age, sex, number of *msp2* genotypes, HIV status, antibody levels for specific IgG and IgE, total IgE, as well as parasite density (coded in fifths of the density distribution and a separate, sixth category for no parasites), modelled as fixed covariates. These measures were all coded as series of binary dummy variables. As most symptomatic infections occur in individuals aged 0–16 years, a specific analysis was conducted to confirm the finding among only younger individuals. This included recalculation of the fifths estimating immunoglobulin distributions in this age group.

3. Results

Baseline characteristics were analysed in the 700 individuals (1–84 years, median 23 years) who were asymptomatic at the time of survey. The *P. falciparum* prevalence, including results from microscopy and PCR, was highest in children 0–16 years (59%) and decreased in adults with 32% of individuals over 16 years infected with *P. falciparum*. Including the results from PCR analysis increased *P. falciparum* prevalence by 25% in all age groups. The mean number of clones detected by the PCR genotyping was 2.4, with a range of one to seven clones

per isolate (SE = 0.09 clones per infection). More detailed description of the genotyping data will be presented elsewhere.

Prospective risk for malarial disease was based on the first clinical episode after the baseline survey and was assessed using Cox regression analysis. Some 60 individuals experienced at least one clinical episode during the follow up. The clinical malaria episodes were scattered during the 40 weeks period. Table 2 shows the odds ratios from Cox regression for individual baseline characteristics: age, sex, total and anti-*P. falciparum* IgE, number of infecting *msp 2* clones and HIV-1 seropositivity. The odds ratios for risk for clinical malaria episode were highest at younger ages, independent of the other measures. The risk for malarial disease was not statistically significantly associated with sex.

Antibody levels in plasma increased with age but with differing patterns. The overall median value was 1.6 ng/ml for malaria specific IgE, 15.6 µg/ml for specific IgG and 1700 ng/ml for total IgE. Antibody level (median) in negative controls was: 0.8 ng/ml for malaria specific IgE, 1.5 µg/ml for specific IgG and 41 ng/ml for total IgE. Total IgE and anti-*P. falciparum* IgG and IgE levels were grouped into fifths of their distributions (Table 1) and comparisons were made with the middle fifth. Malaria-specific IgE above the middle fifth was associated with a reduced risk for a clinical episode of malaria (Table 2). Statistical significance of these estimates increased after adjustment for the total IgE with odds ratios of 0.26 (95% CI, 0.09–0.72, $P = 0.010$) and 0.44 (95% CI, 0.19–0.99, $P = 0.047$) for the fourth and fifth levels, respectively (Table 2—adjusted values).

When measures of anti-*P. falciparum* IgG and anti-*P. falciparum* IgE were included in the same model with adjustment for all of the potential confounding factors, IgE, but not IgG, was significantly associated with risk of subsequent malarial disease. As anti-malarial IgG was not independently associated with risk of malarial disease it was excluded from the final model to avoid confounding due to co-linearity between IgE and IgG isotypes.

Table 1
Antibody levels grouped into fifths of their distribution

Fifth ^a	Anti- <i>P. falciparum</i> IgE		Total IgE		Anti- <i>P. falciparum</i> IgG	
	ng/ml ^b	Age ^c	ng/ml ^b	Age ^c	µg/ml ^b	Age ^c
1	0.50 (<0.71)	19	196 (<454)	25	3.1 (<6.0)	12
2	0.93 (0.71–1.25)	23	806 (454–1232)	23	9.0 (6.0–11.9)	17
3	1.54 (1.26–1.93)	26	1701 (1233–2316)	22.5	15.7 (12.0–21.2)	23.5
4	2.47 (1.94–3.42)	22	3150 (2317–4423)	19	28.0 (21.3–40.5)	27
5	5.22 (3.43–20.57)	21	6676 (4424–19275)	21	62.9 (40.6–212.5)	34

Antibody levels (median) in negative controls (Swedish not exposed to malaria) were: anti-*P. falciparum* IgE, 0.8 ng/ml; total IgE, 41 ng/ml; anti-*P. falciparum* IgG, 1.5 µg/ml.

^a Fifths (140 subjects in each), where 1 is the lowest and 5 is the highest.

^b Antibody levels, median and range.

^c Median age (years), within each fifth of the antibody distributions.

Table 2

Baseline characteristics in 700 asymptomatic individuals and prospective risk for clinical malaria episode

Age (years)	Episode after survey ^a		Unadjusted ^b		Adjusted ^c	
	No, <i>n</i> (%)	Yes, <i>n</i> (%)	Odds ratio	95% CI	Odds ratio	95% CI
≤1	1 (100)	0 (0)	*	*	*	*
2–4	15 (62)	9 (38)	9.95	4.53–21.87	9.37	4.08–21.55
5–7	44 (79)	12 (21)	4.85	2.37–9.92	5.30	2.52–11.14
8–10	55 (87)	8 (13)	2.75	1.21–6.25	3.66	1.52–8.85
11–13	57 (84)	11 (16)	3.52	1.68–7.38	4.43	2.01–9.75
14–16	52 (100)	0 (0)	*	*	*	*
≥17	416 (95)	20 (5)	1.00		1.00	
Sex						
Male	295 (90)	31 (10)	1.00		1.00	
Female	345 (92)	29 (8)	0.83	0.50–1.37	0.96	0.57–1.64
Anti- <i>Pf.</i> IgE ^d						
1	123 (88)	17 (12)	1.01	0.52–1.98	1.05	0.51–2.20
2	129 (93)	10 (7)	0.59	0.27–1.28	0.65	0.29–1.47
3	124 (88)	17 (12)	1.00		1.00	
4	135 (96)	5 (4)	0.29	0.11–0.77	0.26	0.09–0.72
5	129 (92)	11 (8)	0.65	0.31–1.39	0.44	0.19–0.99
Total IgE ^d						
1	127 (91)	13 (9)	0.92	0.43–1.96	1.03	0.46–2.31
2	133 (95)	7 (5)	0.49	0.20–1.21	0.53	0.21–1.34
3	126 (90)	14 (10)	1.00		1.00	
4	128 (91)	12 (9)	0.85	0.39–1.83	1.27	0.56–2.91
5	126 (90)	14 (10)	1.01	0.48–2.11	1.91	0.83–4.38
Anti- <i>Pf.</i> IgG ^d						
1	114 (81)	26 (19)	2.97	1.39–6.33		
2	131 (94)	9 (6)	0.98	0.39–2.47		
3	131 (94)	9 (6)	1.00			
4	129 (92)	11 (8)	1.23	0.51–2.96		
5	135 (96)	5 (4)	0.55	0.18–1.63		
Nr. clones ^e						
0	396 (91)	39 (9)	0.77	0.39–1.55	0.82	0.40–1.68
1	78 (89)	10 (11)	1.00		1.00	
2–3	107 (96)	5 (4)	0.37	0.13–1.08	0.31	0.10–0.94
≥4	59 (91)	6 (9)	0.79	0.29–2.17	0.46	0.16–1.34
HIV-1						
Seronegative	626 (92)	58 (8)	1.00		1.00	
Seropositive	14 (87)	2 (13)	1.52	0.37–6.21	2.28	0.52–9.96
Total	640	60				

*Not estimated because of empty cells.

^a Number of individuals with clinical malaria episodes registered within 40 weeks after the survey.^b Unadjusted odds ratios from Cox regression analysis.^c Odds ratios adjusted for age, sex, anti-*P. falciparum* and total IgE, nr. clones and HIV-1 seropositivity.^d Anti-*Plasmodium falciparum*(anti-*Pf.*) and total antibody levels grouped into fifth on their distribution, where 1 is the lowest and 5 is the highest.^e Number of parasite clones identified as *msp2* alleles.

In a separate analysis limited to children (0–16years), the two highest fifths of anti-*P. falciparum* IgE levels produced similar adjusted odds ratios of 0.26 (95% CI, 0.08–0.83, $P = 0.024$) and 0.34 (95% CI, 0.12–0.98, $P = 0.046$), respectively. The association between malaria-specific IgE levels and risk for disease was non-linear with a greater protective association for the top two fifths of the distribution. If modelled as an ordinal term in the adjusted model, the trend across the fifths produces an odds ratio of 0.79 (0.64–0.98, $P = 0.030$), despite

the non-linearity of the association. Total IgE was not statistically significantly associated with risk for malarial disease when the levels were compared (Table 2) or when trend across levels was estimated, with an odds ratio of 1.20 (0.96–1.52, $P = 0.105$).

The lowest fifth of malaria-specific IgG was associated with an increased risk for a clinical episode of malaria (Table 2-unadjusted values), with odds ratio of 2.97 (95% CI, 1.39–6.33, $P = 0.005$). However, this result no longer remained significant following adjustment for age, with an

odds ratio of 1.91 (95% CI, 0.87–4.20, $P = 0.108$) for the first compared with the middle fifth indicating confounding by age rather than an independent effect. Mutual simultaneous adjustment for the potential confounding factors did not result in any statistically significant associations with clinical malaria. Using Cox regression we found no evidence for statistically significant trend across the groups, either in unadjusted or adjusted models. The unadjusted model produced an odds ratio for trend across the groups of 0.88 (0.71–1.10, $P = 0.253$).

When number of *P. falciparum* clones was assessed, the risk of subsequent episode was found to be lower in individuals with multiclonal infections. The odds ratios were of 0.31 (95% CI, 0.10–0.94, $P = 0.038$) for two to three clones and 0.46 (95% CI, 0.16–1.34, $P = 0.154$) for four or more clones, compared with having one single clone. The risk was, however, also reduced in subject with no clones, i.e. absence of detectable parasites, with an odds ratio of 0.82 (95% CI, 0.40–1.68, $P = 0.588$) compared with having one clone (Table 2). In the children (age under 17 years) the odds ratios are 0.26 (95% CI, 0.08–0.90, $P = 0.033$) for two to three clones, 0.32 (95% CI, 0.10–1.06, $P = 0.062$) for four or more clones and 0.66 (95% CI, 0.28–1.58, $P = 0.354$) in subjects with no clones.

The risk for malarial disease was not statistically significantly associated with HIV-1 seropositivity (Table 2). However, a separate analysis limited to children (0–16 years) revealed an increased risk for HIV-1 seropositives, producing odds ratio of 11.65 (95% CI, 1.12–121.01, $P = 0.040$).

Parasite density among these asymptomatic individuals, in neither the univariate nor the multivariate analysis, was statistically significantly associated with risk of subsequent clinical malaria and adjustment for parasite density did not statistically significantly alter any of the other associations with risk for malarial disease.

4. Discussion

In this study we investigated factors among subjects free of acute malaria, associated with the risk of subsequent acute malaria episodes. After adjustment for multiple potential confounding factors, we observed independent associations of older age and higher levels of anti-*P. falciparum* IgE with a reduced risk of clinical malarial disease. The non-linear association of malaria-specific IgE with disease risk may reflect antagonistic influences.

The importance of anti-malarial IgE in protective malaria immunity has long been proposed (Perlmann et al., 1997, 1999) but not previously demonstrated. IgE antibody levels have mainly been investigated during the acute phase of disease, comparing patterns in mild and severe, e.g. cerebral malaria (Perlmann et al., 1994, 1997, 2000), or correlating *P. falciparum*-IgE levels and placental parasitemias (Desowitz et al., 1993; Maeno et al., 1993).

To our knowledge, the present study is the first to prospectively assess IgE antibody levels in asymptomatic subjects and describe its association with the risk for subsequent malarial disease.

Several methodological considerations, in addition to the large sample size, represent advantages in this assessment of predictive markers for anti-malarial immunity. Firstly, baseline measurements of antibody levels and other factors were collected in asymptomatic individuals: thus, these measures were unaffected by ongoing or recent malarial disease or treatment, which may result in temporally affected serum levels. While this was necessary to avoid confounding by disease activity, some of the most susceptible individuals may be excluded from the analysis, such that our results could underestimate some associations. Secondly, the baseline measurements were collected prospectively and risk of subsequent disease was studied with recording of each clinical episode of malarial disease in the entire study population, so retrospective recording did not introduce error. Thirdly, we used Cox regression analysis, providing a sensitive measure of risk taking duration of follow-up into account. Multiple simultaneous adjustment were made for factors, such as age, sex, HIV status and parasite densities, thus eliminating many of the problems associated with residual confounding that may influence results.

Our data revealed that high levels of malaria specific IgE were associated with reduced risk for subsequent clinical episodes of malaria. The effect was most prominent in the two highest fifths of the distribution, when compared with the middle levels. Even though a statistically significant trend across the categories was established, this relationship was non-linear, which may reflect that the two lowest levels lie around the threshold of detection of the assay. The non-linearity even seen within the highest anti-*P. falciparum* levels may reflect opposing elements of protection against malaria (discussed below). Among children, who are most vulnerable to infection, higher levels of malaria-specific IgE were associated with a statistically significantly reduced risk of acute malaria. This indicates that the association of IgE with protection from malaria may occur in all age groups, but this is also evidence of its potential importance during the years of greatest vulnerability to acute malarial disease. Total IgE was not associated with reduced risk for disease, and the association found for anti-*P. falciparum* IgE was not solely a function of high total IgE levels. When modelled together with the malaria-specific IgG and the other potential confounding factors, specific IgE was statistically significantly associated with risk of malarial disease, while specific IgG was not.

In univariate analysis, the risk for subsequent malarial disease was highest in subjects with the very low levels of malaria-specific IgG. However, this association is due to confounding by age as demonstrated by the elimination of this association by adjustment for age alone. This is because there is an almost linear increase of malaria-specific IgG

levels with age and the risk of malarial disease decreases with age. Levels of IgG specific to crude *P. falciparum* lysates were not found to be predictive of subsequent malaria episodes, even where trend across the categories was estimated to increase statistical power. The reasons for not finding an association between anti-malarial IgG and protection may be because these antibodies contain many more antigenic specificities which can not be distinguished in our ELISA using a crude *P. falciparum* antigen extract. Another factor could be the distribution of different IgG subclasses. Several studies have reported associations between different IgG subclass responses to specific antigens and clinical protection, e.g. IgG1 (Ndungu et al., 2002), IgG2 (Aucan et al., 2000) and IgG3 (Taylor et al., 1998; Aucan et al., 2000; Oeuvray et al., 2000). The overall malaria specific IgG levels were however not found to be protective (Aribot et al., 1996; Ndungu et al., 2002), which is consistent with our findings in the present study. Analysis of total IgG levels was not included in the assessment since these measurements may produce poor estimates due to aggregation of antibodies at very high concentrations. A potential effect of different IgG subclasses and antigenic specificities needs further investigation in this population.

Persistence of *P. falciparum* infections even at low levels may be an important feature in acquisition of protective immunity in individuals in malaria endemic areas (Smith et al., 1999). Presence of *P. falciparum* parasites per se or total number, i.e. parasite density was not a confounder for the associations of antibody levels with risk for subsequent malaria disease. Fluctuations in number of parasites in the peripheral blood at different time points may however restrict the value of single measurements of parasite densities in asymptomatic individuals (Farnert et al., 1997; Delley et al., 2000). In contrast, the genetic diversity of *P. falciparum*, i.e. multi-clonal infections, was associated with reduced risk for subsequent malaria disease, consistent with previous reports (Al-Yaman et al., 1997; Farnert et al., 1999; Smith et al., 1999), suggesting the importance of polyclonal antigen stimulation. The association between number of *P. falciparum* clones and risk of subsequent acute malaria was however not straightforward, with both no detectable clones and multiple clones associated with protection, when compared with a single clone infection. This may be because anti-malarial immunity is based on an equilibrium, where controlled maintenance of parasites is associated with protection. This may help to explain the non-linear association with other factors such as specific IgE level: if IgE is protective against infection this is a benefit, but too high a level may prevent an adequate level of controlled parasite persistence to fully maintain functional immunity. The different elements of anti-malarial immunity do not operate in unison and this highlights the complexity of malaria immunity.

Longitudinal data of malaria-specific IgE levels over time within asymptomatic individuals has to our knowledge not been reported. In other parasitic infections, e.g.

Dracunculus medinensis (Bloch and Simonsen, 1998) and *Wuchereria bancrofti* (Jaoko et al., 2001), levels have been shown to be stable within the individual, with no seasonal or patency-dependent variation in the levels of parasite-specific IgE. The IgE antibodies detected in our assay correspond to the free fraction of IgE with short half-life (about 2.5 days) which may fluctuate over time, thus the one-time measurement may rather underestimate an association between malaria-specific IgE and protection against acute malarial disease.

The potentially protective mechanisms of anti-malarial IgE need to be clarified. They might act in concert with cell types equipped with receptors for IgE and thereby mediate effector mechanisms as shown for helminthic infections (Pirron et al., 1990; van der Heijden, 1993). However, cell bound IgE can also be involved in presenting antigen to T cells (Heyman, 2002). In addition IgE, in the form of immune complexes, is a potent inducer of tumor-necrosis factor (TNF) in human monocytes (Dugas et al., 1995; Perlmann et al., 1997), a factor involved in both immune protection and pathogenesis of malaria (Kwiatkowski et al., 1990; Perlmann et al., 1996). The lack of association between total IgE and reduced risk for subsequent infection, favours an antigen-specific protective mechanism. How can anti-malarial IgE be involved in both protection and pathogenesis of malaria? The result of IgE responses is probably a combination of induction and host genetic factors (Rosenwasser et al., 1995; Hill, 1999; Perlmann et al., 1999; Troye-Blomberg, 2002). If one of the role of IgE is induction of TNF, over-producers of TNF may have an increased risk of succumbing to severe disease while in individuals with low TNF production IgE antibodies might help to protect against disease (Kwiatkowski et al., 1990; Knight et al., 1999; McGuire et al., 1999). Moreover, the role of IgE is probably different in the maintenance of protective immunity in asymptomatic individuals, compared to the immune activation in acute clinical malaria. Further studies are thus needed to clarify the role of anti-malarial IgE in immune reactions against malaria.

Asymptomatic individuals with higher levels of anti-malarial IgE were less likely to exhibit a clinical episode of malaria, independent of age and other potential confounding factors. Neither high levels of total IgE, nor malaria-specific IgG were found to be predictive of protection. Malaria-specific IgE antibody levels may represent a useful and readily available indicator of susceptibility to subsequent malarial disease in asymptomatic individuals.

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