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Full Length Research Paper

Immunoglobulin profile of Nigerian children with *Plasmodium falciparum* infection

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The immunoglobulin profiles of 126 Nigerian children infected with *Plasmodium falciparum* in their peripheral blood were investigated. The mean malarial parasitaemia was 4699.17 ± 3695.2 µl. The mean immunoglobulin profile of these infected children were 2.68 ± 0.019 mg/dl for IgA, 0.031 ± 0.01 mg/dl for IgD, 1358.29 ± 123.57 ng/dl for IgE, 19.09 ± 1.27 mg/dl for IgG and 2.80 ± 0.57 mg/dl for IgM. The relationship between the IgD and IgE were positively correlated with the ages of the volunteers at (r = 0.89 and r = 0.97, respectively). The levels of IgA, IgG and IgM were negatively correlated with the ages of the infected children (r = -0.96, r = -0.99 and r = -0.85, respectively). The relationship between the level of parasitaemia and IgA, IgD and IgM were negatively correlated (r = -0.82, r = -0.84 and r = -0.82, respectively). IgG correlated positively with the level of malarial parasitaemia (r = 0.99). We deduce that high IgE and low levels of IgA and IgM are associated with the high risk of *P. falciparum* malaria attack in our community.

Key words: IgA, IgD, IgE, IgG, IgM, *Plasmodium falciparum*, children, Nigeria.

INTRODUCTION

Malaria is ranked as a major tropical disease in Africa responsible for cases of morbidity especially in children. Among the children, those under 5 years of age are more vulnerable (WHO, 2000). The wide spectrum of clinical manifestations of malaria is due to the asexual blood stages of *Plasmodium falciparum* and attempts have consequently been made to identify asexual stage antigens that may be of importance in the development of protective immunity to the disease (Warrell, 1993). Of the antigens elaborated, the *P. falciparum* merozoite surface protein 1 (PfMSP1) is widely investigated (Dodoo et al*.,* 1999; Pitabut et al., 2007; Scopel et al., 2005; Omosun et al., 2005). The result from this investigation implicates PFMSP1 as a vaccine candidate.

Globally, the investigation on the humoral immune responses in *P. falciparum* infection exist (Desowitz 1989; Taylor et al*.,* 1998; Aucan et al*.,* 2002; Cavanagh et al., 2001, Diallo et al., 2002; Callisano et al., 2003). For instance, it has been documented that people living in malaria endemic regions develop elevated levels of IgE (Desowitz, 1989).

In Nigeria, there is dearth of information on humoral immune responses to *P. falciparum* infection despite the impact of this parasitic infection on our health. The only existing information (Omosun et al*.,* 2005) reports on the relationship between Anti-MSP-1_{19} and IgG among some Nigerians in a different location and of different ethnic group. In this communication, we therefore evaluate the immunological status of some children infected with *P. falciparum* in a different locality in Nigeria. We established the relationship between their ages, intensity of infection and the immunoglobulin classes.

MATERIALS AND METHODS

The study was conducted in Ekpoma, head quarters of Esan West Local government area of Nigeria. Ekpoma has an estimated population of 2 million and lies between latitude 6° 4′N and longitude 6° 4′E. It is an urban university town. There is a rainy season period of April to October which is followed by a dry season of November to March. Malaria transmission is perennial but highest during the rainy season.

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Table 1. Immunoglobulin profile and malarial parasitaemia of *P. falciparum* infection according to the age groups in years.

Age groups in years	IgM (mg/dl)	IgD (mg/dl)	IgE (ng/dl)	IgG (mg/dl)	IgA (mg/dl)	Malarial parasitaemia/µl	No (%) infected
1-5	3.18±1.31	0.014±0.015	1204.75±402.09	20.62±3.21	2.80±1.60	9560.4±2413.8	50 (39.7)
6-10	3.22±1.32	0.039±0.04	1362.78±394.11	19.13±1.46	2.64±1.04	3913.1±817.3	42 (26.9)
11-15	2.00±1.52	0.041±0.03	1507.33±447.07	17.52±1.53	2.59±0.73	624.0±56.6	34 (26.9)
Mean infected	2.80±0.57	0.031±0.01	1356.95±16.28	19.09±1.27	2.68±0.09	4699.17±3695.2	126
Mean for control	110.02±16.7	0.12±0.04	-	1205.15±13.53	210.01±11.5	-	-

One hundred and twenty six children who attended Faith Dome Medical centre, Ekpoma, Nigeria and had malaria attack based on *P. falciparum* parasitaemia in their thick blood smears stained by Giemsa stain participated in the study. The malaria parasitaemia was categorized as mild (<1000 asexual form of parasite /µl), moderate (1000 – 10,000 parasite/µl) and severe (>10,000 parasite/µl) as described earlier (Egwunyenga et al., 2004). They also had fever (axillary temperature of > 37.5°C) and clinical symptoms such as headache, vomiting, diarrhea, prostration, respiratory distress and other symptoms and signs of severe malaria as documented in WHO (2000). They were recruited after informed parental consent was obtained following thorough explanation of all procedures and the objectives of investigation. Ethical permission was obtained from Faith Dome Medical centre, Ekpoma and the Edo State ministry of Health, Benin City, Nigeria. Children with other detectable diseases such as HIV/AIDS, sickle cell anaemia, intestinal parasitic infections, and viral hepatitis B were excluded in this investigation using standard procedures and kits.

The Recombinant *P. falciparum* merozoite surface protein 1 ($PfMSP1_{19}$) was the antigen used in this investigation. The secreted recombinant protein $PfMSP1_{19}$ was purified as previously described (Egan et al*.,* 1995; Dodoo et al., 1999). Venous blood were obtained from the *P. falciparum* infected volunteers and the 30 control subjects. Their sera were separated by centrifugation using standard procedure. These were subjected to modified radial immunodiffusion assay of Mancini et al. (1965) for determination of the levels of immunoglobulins.

The data obtained in our study were subjected to statistical analysis using Microsoft Excel package

RESULTS

The immunoglobulin profile of *P. falciparum* infected volunteers and their control subjects according to their age groups in years are presented in Table 1. The mean immunoglobulin status were 2.68 ± 0.019 mg/dl for IgA, 0.031 ± 0.01 mg/dl for IgD, 1358.29 ± 123.57 ng/dl for IgE, 19.09 ± 1.27 mg/dl for IgG and 2.80 ± 0.57 mg/dl for IgM. These mean immunoglobulin of the infected volunteers were lower than their control subjects (IgA = 210.0 ± 11.5 mg/dl, IgD = 0.12 ± 0.04 mg/dl, IgG = 1205.15 ± 13.53 mg/dl, IgM = 110.02 ± 16.7 mg/dl) except IgE where the control subjects had 0 ng/dl. The mean malarial parasitaemia was 4699.17 ± 2985.22 µl. The IgD and IgE profile showed positive correlation with the ages of the *P. falciparum* infected volunteers (r = 0.89 and r = 0.97, respectively). The relationship between IgA, IgG and IgM were negatively correlated with the ages of the *P. falciparum* infected volunteers (r = -0.96, r = -0.99 and r = -0.85, respectively). The children within 1 - 5 years of age had the highest IgM, IgD and IgA as 3.18 ± 1.31 mg/dl, 20.62 ± 3.21 mg/dl and 2.80 ± 1.06 mg/dl, respectively. Conversely the highest IgD (0.041 ± 0.03 mg/dl) and IgE (1507.33 ± 447.07 ng/dl) occurred among the volunteers between 11 - 15 age group in years.

Table 2 showed the immunoglobulin profile of the *P. falciparum* according to the 3 levels of parasitaemia as mild (521.15 ± 95.41), moderate (3432.0 ± 1051.71) and severe (10,150 ± 1839.52). The relationship between total IgG and the levels of parasitaemia was positively correlated (r = 0.99). IgM, IgD and IgA had negative correlation with the intensities of the malarial parasitaemia (r = -0.99, r = -0.85 and r = 0.82) respectively.

DISCUSSION

The data on the malarial parasitaemia revealed that children within 1 – 5 years of age had the highest level of parasitaemia than the older children. This pattern of infection is expected considering the fact that these children get exposed repeatedly very early in life in our environment where adequate malarial control measure is lacking. Also severe malaria is seen most frequently in children less than 5 years of age, after which age dependent clinical immunity sets in. These observation accords earlier reports (WHO, 2000; Idro et al*.,* 2005).

We found a high level of IgE responses among the *P. falciparum* infected volunteers than their control subjects and a correlation of this IgE antibody with age which we consider immunologically significant as it supports the earlier deduction that it may play a role in protection against malaria (Calissano et al., 2003). This pattern of elevation of IgE and its importance in plasmodial infecton had been documented earlier (Perlmann et al., 1994; Daurte et al*.,* 2007; Desowitz 1989; Maeno et al., 1993). An important observation recorded in our present investigation is the lowest IgE status found among volunteers with severe malaria. This denotes that the lower IgE responses are associated with severity of infection. Also we found the IgE antibodies responses protective at the higher values which occurred among the older children with mild and uncomplicated malaria. These patterns of

Table 2. Immunoglobulin profile of *P. falciparum* according to the level of parasitaemia.

Intensity Parasitaemia(mean/µl)	No infected	IgM (mg/dl)	IgD (mg/dl)	IgE (ng/dl)	IgG (mg/dl)	IgA (mg/dl)
Mild (521.15±95.41)	42	3.58±1.41	0.044±0.046	1494±460.68	17.00±3.51	3.01±1.03
Moderate (3432±1051.71)	48	3.11±1.13	0.024±0.029	1345.67±398.25	18.57±2.94	2.60±1.24
Severe (10,150±1839.52)	36	1.79±1.34	0.019±0.015	1235.75±315.35	21.74±3.21	2.53±0.73
Mean for infected volunteer (4701.05±4459.52)	126	2.81±0.76	0.029±0.01	1358.22±106.12	19.10±1.56	2.71±0.21

presentation corroborates the documentation of Callisanno et al. (2003) which reported that anti malarial IgE may well be protective but may also contribute to the severity of the disease.

We observed a positive correlation between the level of malarial parasitaemia and total IgG profile. Also we reported the highest total IgG among the children within 1 – 5 years who had the highest parasite load and this implicates high IgG to be associated with severity of malaria in our locality. These observations appears to deviates from the assertion that IgG appears to play a role in protection against malarial diseases (Cohen et al., 1962; McGregor et al., 1963). The negative correlation between IgG and severity of infection accords the report of (Marsh, 1992; Miller et al., 1994).

The prevalence of antibodies namely IgA, IgG, and IgM were depleted in the *P. falciparum* infected volunteers compared to their control counterparts. This pattern of low prevalence had been documented in Gambia and Sierra Leone (Egan et al., 1996). The low antibodies prevalence reported in our present study may be in part be due to the short half life of anti-PfMP1$_9$ antigens which are mainly found after clinical episodes (Branch et al., 1998; Früh et al., 1991; Cavanagh et al., 1998) and low immunogenicity or lack of adequate T cell help for antibody production in these infected children (Egan et al., 1997; Hui et al., 1996; Udhayakumar et al., 1995).

Conclusion

The inadequate immune responses associated with these antibodies (IgA, IgG, and IgM) indicate that they do not provide sufficient protection in these children with *P. falciparum* infection. Therefore, these low IgA, IgG and IgM and high level of IgE antibodies are associated with high risk of *P. falciparum* infection in our locality.

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