

Nutritional status of parasitemic children from malaria endemic rural communities in eastern Nigeria.

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Abstract

The impact of malaria infection on the health of the sufferers especially infants and young children is enormous, causing wide range of biochemical and haematological changes. We investigated the effect of malaria infection on the nutritional status of these children, from two rural communities of Ebonyi State, Nigeria, by measuring their serum proteins and iron levels as well as the haemoglobin and packed cell volumes, using standard methods. Our results showed that there was initial significant increase in the total protein and albumin levels in low and moderate malaria densities ($p=0.0026$ and 0.0277 for total protein, and $p< 0.0001$ and $= 0.0006$ for albumin). These decreased as the malaria density increased. There was positive correlation between serum iron and malaria density ($r=20.93$; $p< 0.0001$), while other parameters showed negative correlation at $p\leq 0.0025$. From the results we are of the opinion that iron supplementation should be avoided during malaria treatment, not minding the degree or severity of anaemia/parasitemia unless in the presence of other complicating condition(s) that may cause iron deficiency. Our results also support the use of albumin infusion in place of other colloidal solutions as a good intervention in severe malaria.

Key words: Nutritional status, parasitemic children, malaria endemic areas.

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Introduction

In Africa, about 90% of deaths are mainly due to malaria. This is more prevalent in children below 5 years of age [1]. Those who survive the attack usually suffer one disability or the other especially brain damage and learning impairment [2], while recurrent episodes are known to lead to cerebral malaria. About 10 - 20% of children who develop cerebral malaria do not survive for long while 7% of them will develop neurological complications even when treated adequately [3]. The major cause of some of these disabilities may be aggravation of accumulated effects from infancy, especially those who were infected in utero, or severity of the prevailing infection or both. Infant malaria, which is mainly due to

utero-placental infection, is common among infants of primigravidae and secundigravidae. This is because immune suppression associated with pregnancy seems to be more in these classes of pregnant women than in the multigravidae [4,5].

Malaria infections result in 75,200 - 200,000 low birth weight babies each year, probably due to combination of preterm delivery and fetal restriction [6, 7] from high placental parasitaemia as seen in high malaria transmission areas [8]. It is possible that the infants who survive this utero-placental transmission may have reduced immunity to face subsequent infections early in life, making them more susceptible to frequent infections. Although rare

symptomatic infections have been reported in some endemic areas with high utero-placental transmission, this was thought to be due to many reasons including innate mechanisms (Fetal Hb and PABA-deficient breast milk), cultural practices (swaddling of newborns), trans-placental transfer of protective antibodies and priming of neonatal responses by trans-placental transfer of parasites or their products [7, 9, 10, 11]. Whichever is the reason, gives way as the infant grows, thus making it less protected with increase in age. Moreover, it has been established that absence of malaria symptoms, particularly fever, does not exclude malaria infection [12]. Hence, asymptomatic but infected infants may later develop serious complications with subsequent infection because of accumulation of effects.

Malaria causes alterations in some biochemical and haematological parameters in both adults and children, including hypoglycaemia, lactic acidosis, dislipidaemia, raised serum transaminases, anaemia etc [2, 13]. Interestingly, poor nutrition and micronutrient deficiencies play important roles in the pathogenesis of malaria and malarial anaemia [14], thus in rural areas of poor socio-economic burden, with inevitable nutritional deficiencies, the parasitization of these vulnerable children is of alarming rate. This will go further to cause more biochemical changes. We had earlier reported changes in lipids and lipoproteins in these parasitized children [2]. We now present further investigation of the nutritional indices of these children including serum proteins and serum iron concentrations, haemoglobin levels and packed cell volumes. Ethical clearance was obtained from the Ethical and Research Committee of Ebonyi State University Teaching Hospital, Abakaliki, Nigeria, while additional written consents were sought and obtained from the parents/guardians and Headmasters/Headmistresses of the children.

Materials and Methods

A total of 60 children aged between 5 and 12 years were recruited for the study. They comprised of 33 males and 27 females. Another 28 age-matched apparently healthy and afebrile children, whose blood samples were negative for malaria parasites at the time of the study, were used as controls. They comprised of 16 males and 12 females.

Our study areas were the rural Izzi in Ohaukwu Local Government Area and Onueke in Ezza East Local Government Area, all of Ebonyi State, Nigeria. These areas are known to be endemic for malaria, giving as high as 59.9% among pregnant women [15]. The patients were from the Health Centres of these areas while the controls were from the Primary Schools. Few asymptomatic primary school children whose blood samples were found to be positive for malaria parasites were excluded from the control groups. Febrile patients whose blood samples were nega-

tive for malaria parasites were also excluded from the study. The later were found to have been treated in the house but could not get better due to other prevailing conditions, particularly, typhoid/enteric fever.

A total of 5.0ml of venous blood was collected from each subject. About 2.0ml of this was dispensed into EDTA-anticoagulated haemogram container while the remaining was put into a chemically clean plain test tube. The last drop that remained in the syringe was used to make a thick film on the grease free microscope slide for malaria parasite identification. The anticoagulated sample was used for the estimation of haemoglobin level (Hb) and packed cell volume (PCV), while the sample in the plain test tube was allowed to clot and retract and then centrifuged at 3000rpm for 5 minutes. The serum obtained was used for the estimation of serum proteins and iron concentrations.

The films were stained with diluted Giemsa stain, air-dried and examined microscopically using x100 objective lens (oil immersion lens). The malaria parasite densities were graded as earlier reported [15]). The asexual forms of the parasites were counted in 200 leucocytes. The number of parasites per microlitre of blood was calculated using the total leukocyte count, thus;

$$\text{Parasites}/\mu\text{L} = \frac{\text{No of asexual parasites} \times \text{Total WBC count}}{200}$$

The degree of parasitemia or malaria density was therefore graded according to the number of parasites per microlitre thus, 1 – 999 as low (or +), 1000 – 9999 as moderate (or ++) and >100000 as high (or \geq +++). Hb levels were estimated using cyamethaemoglobin method while PCVs were determined using haematocrit centrifuge [16]. Total serum protein was estimated using Biuret method [17], albumin was estimated by dye-binding method using Bromocresol green - BCG [18], while globulin was calculated as the difference between the two. Serum iron was estimated using bathophenanthroline method [19].

Results

Table 1 shows the means (\pm) SEM of all the parameters in the different malaria densities and the control. There were decreasing values of Hb, PCV and serum proteins as the malaria densities increased while the serum iron concentration showed steady increase as the densities increased. When the parameters in each malaria density were compared with the control group (table 2), there was significant difference in each except three; there was no significant difference ($p=0.7366$) between albumin level in high malaria density and control group, and there were no significant differences ($p=0.3726$ and $p=0.1837$ respectively) between globulin levels in low and moderate malaria densities and control group. Correlation studies using one way ANOVA showed positive correlation between serum iron

and malaria density (f=20.93, p=0.0001) while other parameters showed negative correlations (Table 3).

Table 1.: Mean (\pm) SEM of all parameters in different malaria densities and control.

Density	Low	Moderate	High	Control
Iron (mg/L)	9.55(0.54)	16.68 (1.79)	27.82 (3.30)	6.34 (0.42)
Tot Protein (g/L)	78.95(1.10)	75.63 (0.73)	66.29 (0.67)	72.45 (1.22)
Albumin (g/L)	50.41(0.50)	50.08 (0.78)	44.64 (2.12)	45.36 (1.04)
Globulin (g/L)	28.36(0.90)	25.63 (0.68)	21.86 (1.88)	27.18 (0.95)
Hb (g/100ml)	9.33(0.26)	9.53 (0.23)	8.27 (0.20)	11.27 (0.16)
PCV (%)	27.00(0.75)	27.42 (0.71)	23.43 (0.72)	33.36 (0.54)

Table 2. Comparison between the parameters in different density groups and control.

Parameter	Iron	Total Protein	Albumin	Globulin	Hb	PCV
Control	6.34 (0.72)	72.45 (1.22)	45.36 (1.04)	27.18 (0.95)	11.27 (0.16)	33.36 (0.56)
Low Density	9.55 (0.54)	78.55 (1.10)	50.41 (0.50)	28.36 (0.90)	9.32 (0.27)	27.00 (0.75)
P – Value	<0.0001	0.0026	<0.0001	0.3726	<0.0001	<0.0001
Moderate Density	16.68 (1.79)	75.63 (0.73)	50.08 (0.78)	25.63 (0.68)	9.53 (0.23)	27.42 (0.17)
P – Value	<0.0001	0.0277	0.0006	0.1837	<0.0001	<0.0001
High Density	27.82 (3.30)	66.29 (0.67)	44.64 (2.12)	21.86 (1.88)	8.27 (0.20)	23.43 (0.72)
P – Value	<0.0001	0.0006	0.7366	0.0086	<0.0001	<0.0001

Table 3. Correlation of the effect of the malaria densities on the parameters.

	Low	Moderate	High	F	P-Value	Significance
Iron	9.55 (9.54)	16.68 (1.79)	27.82 (3.30)	20.93	<0.0001	Sig.
Total Protein	78.56 (1.10)	75.63 (0.73)	66.29 (0.57)	40.57	<0.0001	Sig.
Albumin	50.41 (0.50)	50.08 (0.78)	44.64 (2.12)	7.62	0.0012	Sig.
Globulin	28.36 (0.90)	25.63 (0.68)	21.86 (1.88)	8.13	0.0008	Sig.
Hb	9.33 (0.26)	9.53 (0.23)	8.27 (0.20)	6.73	0.0025	Sig.
PCV	27.00 (0.75)	27.42 (0.71)	23.43 (0.72)	6.57	0.0024	Sig.

Discussion

Our results showed significant differences between the serum total proteins in the control group and those of low and high malaria densities (p=0.0026 and 0.0006 respectively). The differences are in line with changes in one of the protein fractions - albumin. Serum albumin in control group was significantly lower than that in low and moderate malaria densities (p=0.0001 and 0.0006 respectively), while there was no significant difference between the values in control and high malaria density (p=0.7366). It is expected that malaria infection with its attendant pathologies will decrease the serum albumin at the onset. Therefore, the mechanism involved in this initial increase as observed in this study is not well understood. However, it

is known that albumin is synthesized in the liver; hence it is possible that the initial inflammation of the liver may have increased its production, probably, in the same way the production of C-reactive proteins is influenced. Again, in children particularly, one of the initial symptoms of malaria is vomiting, which was noticed in over 70% of our subjects. This could have increased haemo-concentration (resulting from hypovolemia), leading to initial increase in serum proteins. Thus, as the condition progressed, there was reduced functioning of the liver, and increased adaptation to the condition (including reduction in vomiting). At the same time, there was continuous breakdown of the already produced albumin due to high fever. All these will lead to progressive reduction in serum protein level, especially albumin.

Albumin has a range of physiological effects. For instance, it exerts direct effects on vascular endothelium by binding to the endothelial glycocalyx to maintain normal permeability [20] and also exerts complex influences on erythrocytes [21]. Based on these properties, it is recognized that albumin may be of particular importance in the pathophysiology of malaria especially cerebral malaria where adherence of parasitized red cells to endothelium, aggregation of red cells and impairment of red cell deformity can be influenced by the highly negative charge of the albumin molecule [22, 23]. Therefore, we are of the opinion that the increase in albumin at low parasitemia may have accounted for the non-severity of the fever and other symptoms of malaria at that level. Thus, the decrease in albumin level subsequently gave way to high fever and other complications of malaria as observed clinically. The effects of albumin on these complications may be one of the mechanisms by which albumin infusion achieves faster recovery from hypovolemia due to malaria than synthetic colloidal infusions [24]. On the other hand, globulin fraction did not show any significant difference between the control group and low and moderate malaria densities ($p=0.3726$ and 0.1837 respectively). However, high density parasitemia had significantly lower globulin level than the control group ($p=0.0086$). This decrease in globulin level may give rise to lower immune status in this group of subjects.

Unlike the serum proteins, serum iron showed steady increase in concentration as the malaria density increased. The serum iron concentration in control group was significantly lower than those of the three density groups ($P<0.0001$ in all), showing positive correlation between serum iron and malaria density ($r=20.93$; $p<0.0001$). Iron is one of the commonest elements in the earth's crust, yet iron deficiency is probably the most common nutritional disorders in the world, particularly in developing countries [25]. It is involved in the constitution of enzymatic systems like catalase, peroxidases and cytochrome that play essential roles in cellular respiratory mechanisms in mitochondrial respiratory channel [26].

Hence its deficiency has serious nutritional adverse effects. On the other hand, iron overload has its own burden. It is known to cause oxidative stress-one of the complications of malaria parasitemia [27]. The tolerable upper intake level of iron in children between the ages of 0 to 8 years (about 22kg) is 40ug/day [28]. Even though this may not be exceeded as a supplement during the treatment of malaria in our study group (5 - 12 years), the finding that more iron is produced in parasitemia is a danger to these patients. This expected overload may be one of the causes of untoward reactions noticed in some patients before and during therapy. This supports the earlier caution on the use of iron supplements in malaria endemic areas as this may increase the likelihood of the subject developing potent malaria [29].

Some classical clinical symptoms of malaria in many patients, particularly children, include nausea, vomiting, abdominal pain, diarrhea hematemesis and melena. These may be as a result of initial toxic effect of iron on GIT. This toxic effect is due to direct corrosive action of iron on the mucosa causing, among other things, mucosal edema, infarction, ulceration and haemorrhage [30]. Apart from the initial action of iron on the mucosa, some anti-malaria drugs may also interact with iron, producing some untoward effects. Hence, some patients who were relatively stable before commencement of treatment (barring fever and headache), suddenly developed severe reaction including vomiting, abdominal cramps and dizziness, as soon as treatment was initiated. This may possibly be as a result of oxidative stress caused by iron overload or interaction between iron and the drug or both. Therefore, there is need for further evaluation of iron supplementation during malaria treatment.

In all the density groups, Hb and PCV showed significant decrease ($p<0.0001$ in all) when compared to the control group. This result is consistent with our earlier finding in pregnant women [15]. The significant decrease can be attributed to increasing breakdown of red cells, thus making malaria anaemia one of the anaemic states associated with iron overload. Iron overload in the presence of anaemia in some conditions like hypoplastic anaemia and anaemia due to chronic renal failure is, usually, due to multiple blood transfusions, which causes iron to accumulate in the RES [31], while in parasitemic anaemia iron overload is due to liberation from the red cells. To avoid oxidative stress in parasitemic individuals, we advocate that only multivitamins should be used in correcting the anaemic states (especially in children), since this study has ruled out iron deficiency in these patients. It is also necessary that iron status of pregnant women in particular and adults in general be re-evaluated to establish the place of iron supplementation in these groups. This is particularly to avoid the systemic actions of excess unbound iron that evidently affect the cardiovascular system, general metabolic functions, the liver and central nervous system [30].

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