

IMPACT OF MALARIA ON THE CONCENTRATION OF SELENIUM IN PREGNANT WOMEN IN NNEWI (SOUTH EAST NIGERIA)

ABSTRACT

Malaria during pregnancy continues to be a major health problem in endemic countries, with clinical consequences including death of both mother and child and attendant derangements in trace elements.

This

study is aimed at evaluating the relationship between the trace element antioxidant; selenium and malaria density in pregnant women with malaria. A total of 460 subjects were recruited for the study, comprising 160 pregnant women with malaria attending the ante natal clinic of Nnamdi Azikiwe University Teaching Hospital Nnewi, Anambra, South East, Nigeria. The controls are 100 pregnant women without malaria attending the same clinic, 100 non-pregnant women with malaria and 100 non-pregnant women without malaria. The concentration of selenium was determined by atomic absorption spectrophotometry while the malaria density was determined by counting the parasites against white cells. From the results selenium showed a significant decrease in pregnant women with malaria $55.68 \pm 16.69 \mu\text{g/L}$ ($p < 0.05$) compared to non-pregnant women without malaria $64.83 \pm 13.27 \mu\text{g/L}$ and a significant increase compared to pregnant women without malaria $32.94 \pm 14.41 \mu\text{g/L}$ and non-pregnant women with malaria $36.44 \pm 9.59 \mu\text{g/L}$ ($F = 42.91$; $p < 0.05$). Selenium showed a weak positive correlation with parasite density ($r = 0.27$; $p = 0.004$).

KEY WORDS: Selenium, Malaria, Pregnancy, Antioxidants, Malaria parasite density, Trace elements.

INTRODUCTION

Malaria is an important public health problem in developing countries. *Plasmodium falciparum* a pathogenic agent remains a major cause of morbidity and mortality to mother and child (Jeffrey and Pia, 2012). There are about 300 million cases of malaria each year, 9 of 10 cases occur in Africa. Women and children are most at risk (WHO, 2011). About 30 million African women are pregnant yearly, for these women; malaria is a threat both to themselves and their babies (Menendez *et al.*, 2000; WHO, 2011). In Malaria endemic areas, malaria

during pregnancy may account for up to 15% of maternal anemia, 5-14% of low birth weight, 30% of preventable low birth weight (WHO, 2011). Pregnant women are particularly vulnerable to malaria as pregnancy reduces a woman's immunity to malaria infection and increasing the risk of illness, severe anemia and death for the unborn child. Maternal malaria increases the risk of spontaneous abortion, still birth, premature delivery and low birth weight (WHO, 2003).

Pregnancy is a period of increased metabolic demands with changes in a woman's physiology and requirements of a growing fetus (Broughton, 2007). Insufficient supplies of essential vitamins and micronutrients can lead to a state of biological competition between the mother and conceptus which can be detrimental to the health status of both (King, 2003).

Some authors have associated malaria acquisition and its severity to the concentration of micronutrients in pregnant mothers, the protection against acute infection through a moderated deficiency in iron (Nyakeriga *et al.*, 2004); the reduction of risk of fever and clinical malaria episodes through a zinc supplementation (Zeba *et al.*, 2008) and the copper associated with zinc, which the reduction of the ratio copper/zinc is an

increasing factor of the oxidative stress (Mezzetti *et al.*, 1998).

Deficiencies of specific antioxidant activities associated with the micronutrients iron, selenium, copper, zinc and manganese can result in poor pregnancy outcomes including fetal growth restriction (Fall *et al.*, 2003), pre eclampsia and associated risk of diseases in adulthood, including cardiovascular diseases and type 1 diabetes (Lykke *et al.*, 2009). Another consequence of oxidative stress resulting from antioxidant deficiency is the development of malaria anemia (Kremsner *et al.*, 2000). Micronutrients are known to be integral part of antioxidants and have been found to influence host cellular and humoral immunological functions (Spallhoiz *et al.*, 1990). Cell mediated immunological response to malaria is found to decrease during pregnancy (Riche *et*

51 *al.*, 2000). These antioxidants have been shown to provide protection against oxidative stress induced by malaria
52 (Adelekan *et al.*, 1997).

53 Selenium is incorporated into proteins to make seleno-proteins including the glutathione peroxidase antioxidant
54 enzymes. Selenium is essential for the production of active thyroid hormones and for normal thyroid function.

55 Trace elements are known to be an integral part of antioxidant and have been found to influence host cellular
56 and humoral immunological functions. These essential factors are very important in the body in order for the
57 immune system to cope with the challenges imposed by infectious agents.

58 This study is therefore aimed at evaluating the relationship between the trace element selenium and malaria
59 density in pregnant women.

60 **Aim and Objectives**

61 To determine the relationship between the trace element selenium and malaria density in pregnant women
62 in

62 Nnewi. The objectives are as follows:

- 63 - To determine the relationship between selenium concentration and malaria density in pregnant women.
- 64 - To determine the level of selenium in pregnant women with malaria

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67 **MATERIALS AND METHODS**

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69 This study was conducted at Nnamdi Azikiwe University Teaching Hospital, Nnewi. Ethical approval for
70 this study was issued by the ethical committee of Nnamdi Azikiwe University Teaching Hospital, Nnewi.
71 Four Hundred and sixty women were used for the study, out of this, One Hundred and Sixty pregnant
72 women served as the test subjects, One Hundred pregnant women without malaria, One Hundred women
73 without malaria, One Hundred women with malaria served as control. These women were selected using
74 simple random sampling technique. The pregnant women among them were selected from their clinic while
75 the non-pregnant women were apparently healthy women within Nnewi town. The scope, nature, aims and
76 objectives of the study were explained to the participants for their consent. Women with malaria were later
77 grouped according to parasite density (Melaine *et al.*, 2010).

78 Women with established medical risk factors for oxidative stress such as AIDS, diabetes, tuberculosis, smoking
79 and alcohol consumers were excluded from the study.

80 A volume 6ml of venous blood was collected from each of the participants, 2ml was dispensed into an EDTA
81 container for total white cell count, a drop of blood from the syringe was placed on a clean grease free slide that
82 has been labeled for a thick film while the remaining blood was dispensed into a plain tube. It was allowed to
83 clot at room temperature for approximately one hour and then centrifuged at 2500 RPM for 10 minutes to
84 separate the serum. The serum samples were analyzed for, selenium. The thick film was left to air dry before
85 staining.

86 **Statistical Analysis**

87 This was done using graph pad prism version 5. The results were presented as mean $\pm$ standard deviation. The
88 statistical methods utilized for the analysis were one way analysis of variance, students "t" test, and correlation

89 **RESULTS**

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91 **Selenium in Pregnant Women with Malaria and Control subjects (Mean $\pm$ SD)**

92 The mean serum level of selenium in pregnant women with malaria is $55.68 \pm 16.69 \mu\text{g/L}$ while that in pregnant
93 women without malaria, non-pregnant women with malaria and non-pregnant women without malaria is
94 $32.94 \pm 14.41 \mu\text{g/L}$, $36.44 \pm 9.59 \mu\text{g/L}$ and $64.83 \pm 13.27 \mu\text{g/L}$ respectively. The result shows a statistically significant
95 difference between the means ($F = 42.91$; $p < 0.0001$). Further analysis shows significant increased level in
96 pregnant women with malaria compared to pregnant women without malaria ($p < 0.0001$), significant higher level
97 in pregnant women with malaria when compared with non-pregnant women with malaria ($p < 0.0001$) and a
98 significant decrease in pregnant women with malaria when compared to non-pregnant women without malaria
99 ($P = 0.0151$) (Table 1)

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Table 1: Selenium in Pregnant Women with Malaria and Controls subjects (mean±SD)

	SELENIUM µg/l
Pregnant Women With Malaria n=160	55.68±16.69
Pregnant Women Without Malaria n=100	32.94±14.41 ^a
Non-Pregnant Women With Malaria n=100	36.44± 9.59 ^a
Non Pregnant Women Without Malaria n=100	64.83±13.27 ^{a,b,c}
F-Value	42.91
P-Value	< 0.0001**

NB: a; p<0.05 compared with pregnant women with malaria
b; p<0.05 compared with pregnant women without malaria c;
p<0.05 compared with non-pregnant women with malaria

Selenium and Parasite Density in Pregnancy (Mean±SD)

Selenium in pregnant women with parasite density of <2000/µl is 48.90±21.38µg/L, between 2000-10000/µl is 53.46±23.17µg/L while pregnant women with parasite density of >10000/µl has selenium level of 62.19±25.43. The result of shows no significant difference between the means though there is a progressive increase in selenium level as the malaria parasite density increases (F=2.301; p> 0.05). (Table 2).

Table 2: Selenium and parasite density in pregnancy (Mean±SD)

	SELENIUM µg/l
<2000/µl n =44	48.90±21.38
2000-10000/µl n =96	53.46±23.17
>10000/ µl n =20	62.19±25.43
F-Value	2.301
P-Value	0.1035

NB: **; significant difference between the means (p<0.05)
a; p<0.05 compared with parasite density <2000/µl

Selenium And Parasite Density In Non-Pregnancy (Mean±SD)

Selenium level in non-pregnant women with parasite density of <2000/μl is 32.18±11.28μg/L, between 2000-10000/μl is 39.67±10.08μg/L while non-pregnant women with parasite density of >10000/μl has selenium level of 33.83±1.93. The result shows a significant difference between the means (F=6.850; P=0.0016). Parasite density between 2000-10000/μl has higher selenium level than <2000 and >10000 (Table 3).

Table 3
Selenium and parasite density in non-pregnancy (Mean±SD)

	SELENIUM μg/l
<2000/μl n =23	32.18±11.28
2000-10000 n =51	39.67±10.08 _a
>10000 n =26	33.83±1.926 _b
F-Value	6.850
P-Value	0.0016**

NB: **, significant (P<0.05) difference between the means

a; p<0.05 compared with parasite density <2000/μl

b; p<0.05 compared with parasite density 2000-10000/μl

Selenium and Parasite Density in Pregnant and Non Pregnant Women (Mean±SD)

Pregnant women with parasite density <2000/μl has selenium level of 48.90±21.38μg/L while non-pregnant women have 32.18±11.28μg/L. There is a statistically significant higher level of selenium in pregnant women than in non-pregnant (P<0.0001). At parasite density level between 2000-10000/μl, pregnant women (53.46±23.17μg/L) has a statistically significant higher level of selenium compared to non-pregnant women (39.67±10.08μg/L), (P<0.0001). At parasite density level >10000/μl, pregnant women (62.19±25.43μg/L) has a statistically significant higher level compared to non-pregnant women (33.83±1.926μg/L) (P<0.0001), (Table 4.).

While non-pregnant women has 2.057±0.2874μmol/L. There is a statistically significant higher level of manganese in pregnant women than in non-pregnant (P<0.0011). At parasite density level between 2000-10000/μl, pregnant women (9.218±4.23μmol/l) has a statistically significant higher level of manganese compared to non-pregnant women (2.077±0.3027μmol/L), (P<0.0048). At parasite density level >10000/μl, pregnant women (4.532±2.643μmol/L) has a statistically significant higher level compared to non-pregnant women (2.331±0.7059μmol/L), (P<0.0016). (Table 4).

	<2000/μl		2000-10000/μl		>10000/μl	
	Pregnant women	Non pregnant women	Pregnant women	Non pregnant women	Pregnant women	Non pregnant women
Selenium	48.90±21.38	32.18±11.28	53.46±23.17	39.67±10.08	62.19±25.43	33.83±1.94
P-Value	<0.0001**		<0.0001**		<0.0001**	

Selenium and Parasite Density in Pregnancy

Selenium has a weak positive correlation with parasite density ($r = 0.27$; $p = 0.004$). (Table5). This implies that selenium level increases with an increase in parasite density

Trace Element	R	P
Selenium	0.27	0.004

Correlation is significant at the 0.05 level.

Table 5

DISCUSSION

Selenium is a major component of glutathione. Glutathione peroxidase is a major antioxidant defense system within the body. Wide variations in selenium levels occur in humans. Geographical location, soil content, intake of selenium in diet and its bioavailability significantly affect selenium status (Wasowiaz *et al.*, 2003).

In this study, it was observed that there is a significant higher level of selenium in non-pregnant women without malaria compared with pregnant women with and without malaria. There have been varying reports on the selenium status during pregnancy with various studies showing lower level in pregnancy (Mihailoviae *et al.*,

2000). Nwagha *et al.*, (2011) also reported a lower selenium level in pregnancy. Active transfer of selenium from maternal blood to the tissues of the developing fetus has been advocated. Indeed, haemodilution due to increased plasma volume in

pregnancy further depletes selenium concentration. Additionally, inadequate intake and storage in the maternal tissue and increased demand by the fetus invariably leads to low maternal level during pregnancy (Olife *et al.*,

2007). From this study, there is a significant higher level of selenium in malaria during pregnancy.

Pregnant women with malaria have a significant higher level of selenium compared with pregnant women without malaria. During pregnancy, there is also an increase in level of selenium with an increase in malaria parasite density though not statistically significant. Arinola *et al.*, (2008) in their study reported a higher level of selenium in pregnant women with malaria infection. This is attributed to induction of oxidative stress by malaria parasites. The destruction of malaria parasites by white blood cells through oxygen dependent mechanism and engulfing of kupffer cells by larva stage of malaria parasite induces oxidative stress. This will require antioxidant for removal, thus the need for increased production of antioxidants. Selenium is an integral part of enzymatic antioxidants and this account for the increase of selenium in malaria infection (Langhom, 1989; Hasib, 2000).

Conclusion

From this study, serum level of selenium is reduced in pregnancy but increases in malaria infection during pregnancy. A further increase was observed with an increase in malaria parasite density.

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