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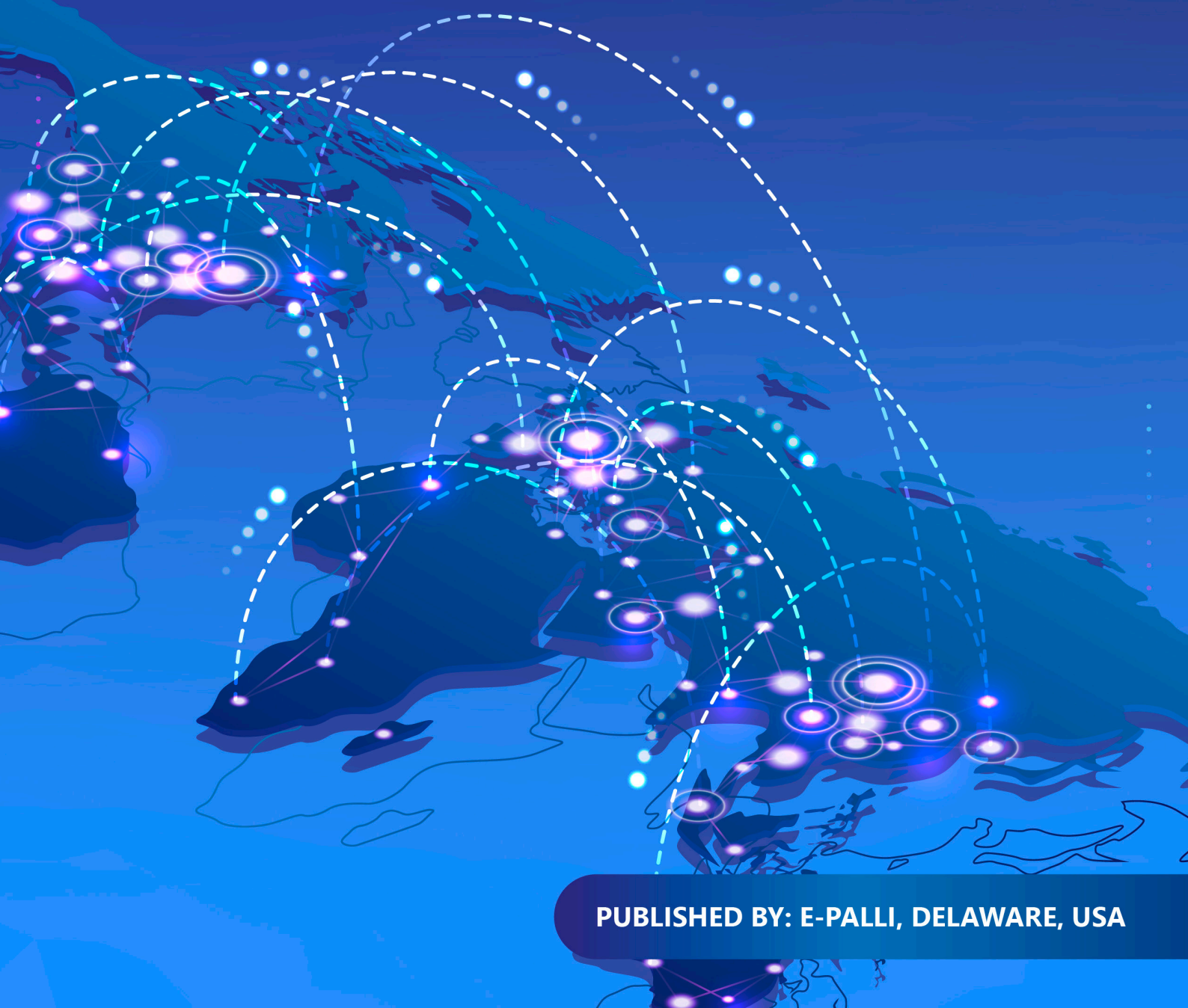

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Establishing the Nexus Between Iron Status and Markers of Immune Functions Among School Age Children in Ogun State, Nigeria

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ABSTRACT

Iron deficiency as a public health problem is based on the seriousness of its consequences on human health. It suggests that certain indices of iron status are associated with anthropometric measures used independently as markers of immune functions. This study established the relationship between the iron status, and markers of immune functions among school-age children in selected Local Government Areas (LGAs) of Ogun state, Nigeria. A multistage sampling technique used to select 312 school-age children from the three senatorial districts of Ogun State. Blood samples of the children were analyzed for biochemical [serum ferritin (SF), C-reactive protein (CRP), Heamoglobin (Hb), reticulocyte count] parameters. Selected immune function markers and parasitic infections were also measured using standard procedures. Data were analyzed using frequency counts, percentages, means, standard deviations, correlation, T-test and Chi-Square. Results showed that 36.5% of the respondent families earned less than two hundred thousand naira annually. Also, 43.0% and 62.0% of the mothers had secondary and tertiary education respectively. WASH practices showed that 61.1% of the respondents did not have a place for handwashing. The study further revealed that the prevalence of iron deficiency was 23.7%, aneamia was 16.3% while 13.1% of the anaemic children were due to iron deficiency aneamia with significant sector (p=0.003) and gender (p=0.032) differences. Malaria (17.6%) and helmintes (18.3%) infection also showed a significant difference in the sector (p=0.020, p=0.042). The range of CD4 counts, Neutrophils, Lymphocytes, Monocytes, Basophils and Eosinophils were 708.85 to 727.11, 3.31 to 3.88, 3.03 to 4.38, 0.53 to 0.59, 0.03 to 0.04, 0.33 to 0.38 respectively. CRP positively correlated with household size (r= 0.155), Heamoglobin(r= 0.238), serum ferittin (r= 0.101), and PCV (r= 0.103). CD4 also positively correlated with mothers age and education (r= 0.252; r= 0.142) but negatively related to PCV(r= -0.102)while malaria and Household size (r=0.109) also shows a positive relationship. This study concluded that a significant relationship exists between socio-economic status, makers of iron status and infections in the children. Hence, appropriate investigations for iron status and inflammation/infection screening need to be integral in the evaluation of anaemia, and interventions that target the multifactorial nature of anaemia in school-aged children need to be strengthened

INTRODUCTION

Iron deficiency (ID) is the most prevalent micronutrient deficiency worldwide, resulting in adverse health outcomes, including anemia, impaired muscle function, poor immune function, delayed psychomotor development, and impaired cognitive performance in children in the short and long term (Beard, 2001; Sachdev et al, 2005). Due to the morbidity and mortality associated with iron deficiency, the World Health Organization (WHO) has deemed it to be a public health issue since 2004 (WHO, 2011; WHO, 2019). Poor nutrition especially iron deficiency in school-aged children is associated with retardation of growth and poor cognitive development. The school age children are at risk of iron deficiency because of an expanding red cell and muscle mass (Herbert et al., 1997). It has been estimated that about 40% of the world's population (more than 2 billion individuals) suffer from anaemia with a prevalence of 48% in school-aged children (WHO, 1996;Shell-Duncan et al., 2005). The key nutrient deficiency observed among school-aged children is iron deficiency anaemia (SCN, 2002; Onimawoet al.,2010).

The results from the study of Onimawo et al (2010) showed that the prevalence of anaemia among the school children was 82.6%, while iron deficiency was 77.8%. The average daily iron intake was 30% below the recommended allowance. There was a high prevalence of inflammatory disorders as indicated by CRP among the children.Anemia lowers immunity, making children more vulnerable to communicable diseases and putting them at risk of dying, and anemia's consequences have an effect on a country's social and economic growth (Mulugeta et al., 2018). Moreover, its consequences are severe in children as their bodies develop, and it has been linked to stunted growth, reduced psychomotor growth, and decreased social, emotional, and cognitive functioning in youngsters (Abebe et al., 2018).

Iron is also necessary for normal development of the immune system. Its deficiency affects the capacity to have an adequate immune response as it is necessary for immune cell proliferation and the generation of specific response to infection (Beard, 2001). Both experimental, and some clinical studies have emphasized the importance of iron in the integrity of the immune system especially the innate immunity (decreased bactericidal effect and respiratory burst of neutrophils) and the cellular component system

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of cell-mediated immunity (CMI) (decreased lymphocyte proliferation and delayed hypersensitivity responses) (Beard, 2001 &Ekizet al., 2005).

The Standing Committee on Nutrition (SCN) reported that the prevalence of iron deficiency anaemia among schoolchildren in Africa stood at 49.8% (SCN, 2002). Iron deficiency with or without anemia is associated with increased susceptibility to infection owing to impaired immune function (Onabanjo et al; 2012). Anaemia is one of the most widespread public health problems, especially in developing countries, and has important health and welfare, social, and economic consequences. These include impaired cognitive development, reduced physical work capacity, and in severe cases increased risk of mortality (Nestle and Davidson, 2002). Thus, this study will comprehensively determine the relation of iron status to measures of immune function among the school age children in south west, Nigeria.

Study Population

The study population consisted of primary school children (7-12 years old) in selected schools in both urban and rural local governments in Ogun states.

Inclusion Criteria

School age children between 7-12 years attending the schools selected for the study. School children from primary two to six. Children whose parents give permission for them to participate in the study. Children giving ascent participated in the study. Children who are present at the day of data collection. Children not on medication that can affect the data analyses.

Children with no known case of infection or inflammation

The three senatorial districts in Ogun states are selected for the study (Ogun Central, Ogun west and Ogun East) and Three hundred and twelve (312) school children were considered. Two local governments randomly selected in each senatorial districts (one urban and one rural). Four schools randomly selected in each LGA and total of twenty four (24) schools in all the LGAs. Pupils in each school were stratified into groups according to their class starting from primary two to six. Simple random sampling was used to select the required number from each class

Procedure and Method of Data Collection

Prior to the commencement of the research, submission of the research proposal were made at the State Hospital Ijaye, Abeokuta and selected local governments in the three senatorial districts and also to the headmaster of each school. Ethical consent was sought from the State hospital, Ijaye, Abeokuta and also meeting with parents with the assistance of the school management for their verbal consent. Trained fieldworkers with the principal investigator engaged in data collection and Medical laboratory scientist assisted in blood sample collection and analysis.

A pretested, structured interviewer administered questionnaire was used to collect information from the respondents. The questionnaire was used to collect information on the respondent's bio-data and socio-economic characteristics.

Blood Collection (Including Analytical Procedures)

Venous blood samples (10ml) were collected and delivered in two containers as follows: (i) 4ml blood collected in EDTA- containing tube for hemoglobin (Hb) and full blood count; (ii) 6ml blood collected in coagulant free tubes and centrifuged for the estimation of serum ferritin (SF) and C-reative protein (CRP). PCV, CD4 count, Reticulocyte count were also measured.

Iron Status Indices

Iron status was determined by measurement of hemoglobin, PCV and serum ferritin. Since hemoglobin, PCV and serum ferritin may be altered in the presence of infection, C- reactive protein (CRP) was used to identify individuals with inflammation and infection.

Heamoglobin: Was measured in situ by means of the direct cyanmet hemoglobin method (Ames Mini-Pak Hb test pack &AmesTM Minilab), using Drabkins solution and a standard photometer.

Serum ferritin (indication of iron-stores): Was determined using ELISA (Randox kits, UK)

C-reactive protein: an acute phase protein, and an indicator of acute infection, was determined using a turbidity method from Bayer Corporation (Tarrytown, NY, USA). It was spectrophotometrically measured with a Technicon RA-1000 automated system.

Immune Function Determination

The relevance of iron in the make-up of the immune system particularly in the innate immunity by decreasing the anti-bacterial activity as well as the respiratory burst of polymorphonuclear cell, cellular component and delay hypersensitivity have being emphasized (Beard, 2001).

White blood cell count and differentials were determined (neutrophil, lymphocyte, monocytes, eosiniphil and basophil) using the method of Ghai (2001) and Abu syed (2014).

Reticulocyte count was performed on the blood sample according to Lewis et al. (2001).

Parameters such as microcytosis, macrocytosis, spherocytes, anisocytosis and Rouleaux formation was determined by monicalcheesbrough method (2000).

The level of CD4 lymphocytes count using monoclonal antibodies was verified by cytoflow method (Marti et al., 2001).

Parasitological Test

Stool Samples

Containers were given to each child and they were asked to bring a sample of their faeces to school the next day. Stool samples were microscopically examined for helminthic infection within one hour of staining using a modification of formol-ether and ethyl acetate

concentration technique (Lim et al., 2009). Hookworms were counted.

Malaria Screening

Thick blood smears were prepared on glass slides within 12 hours of blood collection for the determination of malaria parasites. The slides were fixed and stained with Field stain (A and B), allowed to dry and observed under a microscope for malaria parasite using oil immersion objectives (Ogbuileet al., 2000). The presence or absence of malaria was reported as well as malaria parasite count.

Ethical Considerations

Ethical approval to carry out the research was sought from Ogun State Hospital, Ijaiye, Abeokuta, Nigeria. Verbal informed consent was collected from mothers and caregivers.

Statistical Analyses

Changes in biochemical indicators were calculated. Children were defined as: (i) iron deficient if serum ferritin< 15 μg L-1; (ii) anaemic if haemoglobin is (Hb) < 11.5 g dL-1; and (iii) Iron deficient anaemic if serum ferritin< 15 μg L-1 and Hb< 11.5 g dL-1. The mean value for immune parameters like CD4 count, White blood cell count and differentials, Reticulocyte count were calculated and T-test used to determine the difference while the severity of both stool and malaria parasite were measured and changes within groups were examined with a paired t- test. SPSS was used for all statistical calculations and a p-value < 0.05 was considered significant.

RESULTS

Socio- Demographic Characteristic of Respondents

Table 1 and 2 shows the socio-demographic characteristics of the children and family sampled. The age of the children ranged between 7-8years, 9-10years and 11-12years but majority of the children were within the ages of 11-12years. It shows that 21.1% (urban) and 19.2% (rural) of the children were within the age range of 7-8years while 35.4% (urban) and 36.4% (rural) were within 9-10years of age and the remaining 43.5% (urban) and 44.4%(rural) were within the age range of 11-12years . The table also shows age range of parents of the children. The mothers within the age range of 0-20years were 3.1% and 6.0% in urban and rural LGAs respectively and between 21-30years of age were 13.7% and 22.5%, while within 31-40years were 34.8% and 29.8% in urban and rural LGAs respectively. Majority of the fathers were above 40years of age in both urban (46.6%) and rural (43.0%) LGAs. It also shows that 1.9% in urban and 4.6% in rural LGAs were within 0-20years. More than half of the mothers had tertiary education (61.5%) in urban LGAs but 45.0% in rural LGAs. Considering the size of the family, the percentage of household within one to four members was 53.4% in urban and 29.8% in rural LGAs, while the size of the household within five to eight members was 31.7% in urban but 51.0% in rural LGAs. Above eight members were 14.9% and 16.2% in urban and rural LGAs respectively. The average annual income of the family ranges from less than a hundred thousand to five hundred thousand and above

Table 1: Socio-demographic Characteristics of the Children

Characteristics	**Sector**			
	Urban		Rural	
	Frequency	%	Frequency	%
AGE(YRS)				
7-8	34	21.1	29	19.2
9-10	57	35.4	55	36.4
11-12	70	43.5	67	44.4
Total	161	51.6	151	48.4
Fathers age (years)				
0-20	3	1.9	7	4.6
21-30	32	19.9	32	21.2
31-40	51	31.7	47	31.1
Above 40	75	46.6	65	43.0
Total	161	51.6	151	48.4
Mothers age(years)				
0-20	5	3.1	9	6.0
21-30	22	13.7	34	22.5
31-40	56	34.8	45	29.8
Above 40	78	48.8	63	41.7
Total	161	51.6	151	48.4
Educational level of the mothers				
No formal education	4	2.5	10	6.6
Primary education	15	9.3	32	21.2
Secondary education	43	26.7	41	27.2
Tertiary education	99	61.5	68	45.0
Total	161	51.6	151	48.4
Religion of the fathers				

Islam	56	34.8	65	43.0
Christianity	99	61.5	77	51.0
Traditionalist	6	3.7	9	6.0
Total	161	51.6	151	48.4
Religion of the mothers				
Islam	51	32.1	63	41.7
Christianity	101	67.7	81	53.6
Traditionalist	9	0.4	7	4.6
Total	161	51.6	151	48.4
Household Size				
1-4	86	53.4	45	29.8
5-8	51	31.7	77	51.0
Above 8	24	14.9	29	16.2
Total	161	51.6	151	48.4

Table 2: Water, Sanitation, Hygiene of the Households and Household Income

Characteristics		Sector		
	Urban		Rural	
	Frequency	%	Frequency	%
Primary Water Source				
Pond/lake	0	0	11	7.3
Spring/river	1	0.6	20	13.2
Well	20	12.4	72	47.7
Borehole	85	52.8	36	23.8
Pipe borne water	55	34.2	12	7.9
Total	161	51.6	151	48.8
Primary method of waste disposal				
Bush	10	6.2	30	19.9
Refuse dump	56	34.8	60	39.7
City service	42	26.1	15	10.0
Burning	53	32.9	46	30.5
Total	161	51.6	151	48.8
Type of toilet				
Pit latrines	44	27.3	92	61.0
Water closet	108	67.1	41	27.2
Bush	9	5.6	18	11.9
Total	161	51.6	151	48.8
Sharing of the toilet				
With other households	56	34.8	72	47.7
General Public	23	14.3	46	30.5
None of the two	82	51.0	33	21.9
Total	161	51.6	151	48.8
Average Annual Income of the Family				
<100, 000	9	5.6	18	11.9
100,000-199,000	23	14.3	36	23.8
200,000-299,000	20	12.4	46	30.5
300,000-399,000	55	34.2	30	19.9
400,000-499,000	31	19.3	15	10.0
500,000 above	23	14.3	6	4.0
Total	161	51.6	151	48.8

with 5.6% and 11.9% earned less than one hundred thousand, 14.3% and 23.8% earned less than two hundred thousand, 12.4% and 30.5% earned less than three hundred thousand, 34.2% and 19.9% earned less than four hundred thousand, 19.3% and 10.0% earned less than five hundred thousand, 14.3% and 4.0% earned five hundred above.

Biochemical Indices of Children

Table 3 shows the mean biochemical indices of the children. The mean values were: haemoglobin concentration for boys (12.14±1.39 g/dl), serum ferritin (21.52±20.38µg/l), CD4 (741.71±347.20 cells/µl), C-reactive protein (3.32±5.06 mg/l), PCV (36.73±3.60), reticulocyte (0.83±0.37), WBC(7.02±2.21(100/µl) and

neutrophils, lymphocytes, monocytes, Eosinophils, Basophils were (3.51±1.63), (3.85±4.45), (0.57±0.15), (0.36±0.31), and (0.04±0.05), respectively. The mean values are: haemoglobin level for girls (11.73±0.97 g/dl), serum ferritin (19.64±16.50µg/l), CD4 (695.13±345.12cells/µl), C-reactive protein (3.92±5.62 mg/l), PCV (36.10±3.92), reticulocyte (0.89±0.41), WBC (7.49±2.99 (100/µl) and neutrophils, lymphocytes, monocytes, Eosinophils, Basophils were (3.69±3.29), (3.51±1.52), (0.55±0.19), (0.34±0.35), and (0.04±0.03) respectively.

Table 3: Biochemical Indices of Respondents according to gender (N=312)

Variable	Male (n=155)	Female (n=157)	Total (N=312)	Cut-off Range	p-Value
Hb(mg/dl)	12.14±1.39	11.73±0.97	11.93±1.20	11.5-13.0	0.000*
SF (µg/l)	21.52±20.38	19.64±16.50	20.57±18.52	12-150	NS
CD4 (cells/µl)	741.71±347.20	695.13±345.12	718.27±346.39	365-1571	NS
CRP(mg/l)	3.32±5.06	3.92±5.62	3.62±5.34	3-10	0.002*
PCV(%)	36.73±3.60	36.10±3.92	36.42±3.76	>34	NS
Reticulocyte	0.89±0.41	0.83±0.37	0.86±0.39	0.5-1.5	NS
WBC(100/ µl)	7.02±2.21	7.49±2.99	7.26±2.64	4.0-11.0	NS
Neutrophils (1000/µl)	3.51±1.63	3.69±3.29	3.60±2.59	2.0-7.5	NS
Lymphocytes (1000/µl)	3.85±4.45	3.51±1.52	3.68±3.32	1.5-4.0	0.003*
Monocytes (1000/µl)	0.57±0.15	0.55±0.19	0.56±0.17	0.1-1.5	0.04*
Eosinophils (1000/µl)	0.36±0.31	0.34±0.35	0.35±0.33	0.04-0.4	NS
Basophils (1000/µl)	0.04±0.05	0.04±0.03	0.04±0.04	0.0-0.1	NS

Hb- Haemoglobin, SF- Serum Ferritin, CD4 – Cluster of differentiation 4, CRP- C-reactive protein, PCV – Packed cell volume, WBC- White blood cells

Table 4: Biochemical Indices of Respondents according to sector(N=312)

Variable	Urban(n=161)	Rural (n=151)	Total (n=312)	Cut-off Range	p-Value
Hb(mg/dl)	12.15±1.23	11.71±1.15	11.93±1.21	11.5-13.0	0.004*
SF (µg/l)	20.51±20.59	20.63±16.08	20.57±18.52	12-150	NS
CD4(cells/µl)	727.11±338.59	708.85±355.39	718.27±346.39	365-1571	0.002*
CRP(mg/l)	3.24±5.09	4.05±5.71	3.63±5.40	3-10	0.001*
PCV(%)	36.65±3.50	36.04±3.86	36.36±3.68	>34	NS
Reticulocyte	0.87±0.38	0.85±0.41	0.86±0.39	0.5-1.5	NS
WBC(100/µl)	7.28±2.19	7.24±3.05	7.26±2.64	4.0-11.0	NS
Neutrophils (1000/µl)	3.88±2.94	3.31±2.14	3.60±2.59	2.0-7.5	NS
Lymphocytes (1000/µl)	3.03±0.97	4.38±4.57	3.68±3.32	1.5-4.0	0.002*
Monocytes (1000/µl)	0.59±0.15	0.53±0.18	0.56±0.17	0.1-1.5	NS
Eosinophils (1000/µl)	0.33±0.25	0.38±0.39	0.35±0.33	0.04-0.4	NS
Basophils (1000/µl)	0.04±0.03	0.04±0.05	0.04±0.04	0.0-0.1	NS

Hb- Haemoglobin, SF- Serum Ferritin, CD4 – Cluster of differentiation 4, CRP- C-reactive protein, PCV – Packed cell volume, WBC- White blood cells

Iron Status of The Children

Figure1 showed the Iron status of the respondents, it shows that 23.7 were iron deficient, 16.3% were anaemic while 13.1% had iron deficiency anaemia and the remaning 46.7% are iron sufficient. Iron status of the respondents is classified based on their sector and gender in Table 5,it shows that 20.6% of the male were iron deficient and 12.9% of iron deficient respondents were anaemic while 11.0% of the anaemia among the male is as a result of iron deficiency (IDA). Females that are iron deficient were 26.8%, while 19.7% of them were anaemic and 15.3% of the anaemia among the female is as a result of iron deficiency (IDA). In urban LGAs, 18.6% of the children were iron deficient and 7.5% of iron deficient respondents were anaemic while 13.7% of the anaemia among the children is as a result of iron deficiency (IDA). In rural LGAs, the percentages of children that are iron deficient were 29.1%, while 25.8% of them were anaemic and 12.6% of the anaemia among the rural children is as

a result of iron deficiency (IDA).

The overall iron status of the respondents shows that 74(23.7%) of the respondents were deficient in iron and 51(16.3%) were anaemic while iron deficiency anaemia was noticed in 41(13.1%) of them. One hundred and forty six (46.7%) are sufficient in iron.

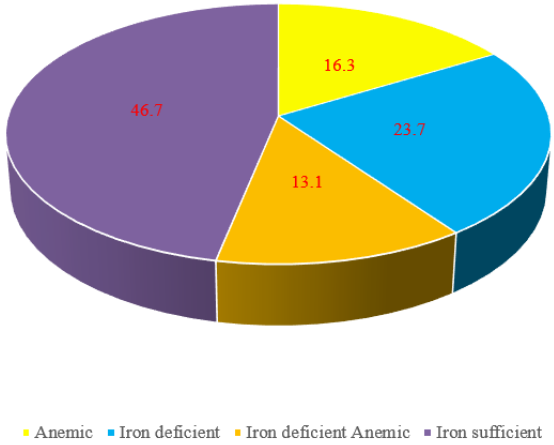

Figure 1: Iron status of the children

Severity of Some Biochemical Indices Among the Children

Table 6 shows the level of some of the indicators of iron in both urban and rural LGAs and this classification is necessary to show the actual status of the indicators. It shows that 80.0% of the male had a normal haemoglobin level and 12.3% are mildly deficient while 5.8% and 1.9% are moderately and severely deficient in haemoglobin respectively. As for the females, 71.3% had normal level of haemoglobin and 19.1% are mildly deficient while 5.7% and 3.8% are moderately and severely deficient in haemoglobin respectively. In urban LGAs, 83.2% had a normal haemoglobin level and 10.6% are mildly deficient while 4.3% and 1.9% are moderately and severely deficient in haemoglobin respectively. As for the rural LGAs, 67.5% had normal level of haemoglobin and 21.2% are mildly deficient while 7.3% and 4.0% are moderately and severely deficient in haemoglobin respectively. The overall haemoglobin status shows that75.6% had a normal level and 15.7% are mildly deficient while 5.8% and 2.9% are moderately and severely deficient in haemoglobin respectively. There is a significant difference in heamoglobin status across the sector (p=0.032) and gender (p=0.040).

Table 5: Classification of school children according to iron status(N=312).

Variables	Sector			p-value	Gender			p-value
	Urban	Rural	Total		Male	Female	Total	
Anaemia (Hb<11.5g/dl)	12(7.5)	39(25.8)	51(16.3)	0.003*	20(12.9)	31(19.7)	51(16.3)	0.032*
Iron Deficient (Hb≥11.5g/dl & SF <12µg/l)	30(18.6)	44(29.1)	74(23.7)	0.022*	32(20.6)	42(26.8)	74(23.7)	0.041*
Iron Deficiency Anaemia (Hb<11.5g/dl & SF <12µg/l)	22(13.7)	19(12.6)	41(13.1)	0.013*	17(11.0)	24(15.3)	41(13.1)	0.022*
Iron Sufficient (Hb≥ 11.5g/dl & SF≥12µg/l)	97(60.2)	49(32.5)	146(46.7)	0.012*	85(55.5)	60(38.2)	146(46.7)	0.013*
Total	161	151	312		155	157	312	

Parked cell volume (PCV) was at low level in 19.4% of the males and 34.4% females while 21.1% and 33.1% had a low level of PCV in urban and rural LGAs respectively, with a significant difference in both the sector (0.024) and gender (0.032).Serum ferittin was deficient in 25.2% of the males and 34.4% females while in the LGAs, deficiency of ferittin was noticed in 22.4% of the children in urban and 37.7% in rural LGAs.Reticulocyte which is an immature red blood cell was low in 18.7% and 24.2% of male and female respondents and noticed to be high in 14.2% and 17.2% of male and female respondents respectively. It also shows that 16.1% and 27.2% have a low level of reticulocyte while it was high in 14.3% and 17.2% in urban and rural LGAs.

Severity of Markers of inflammation and immune function of the subjects

Table 7 shows the maker of inflammation and immune function of the respondents. In the LGAs, the CRP was at low risk in 72.7% of urban and 68.2% of the rural respondents while mild risk in 13.7% urban and 17.9% of the rural but at high risk in 13.7% of urban and 13.9% of the rural children. As for the gender, it shows that 71.0% males and 70.1% females were at low risk while mild risk was noticed in 17.4% males and 14.0% females but 11.6% males and 15.9% females were at high risk without any significant difference in both gender and sector. The CD4 count which is a determinant of immune status appeared to be low in 17.4% and 15.9% in urban and rural local government area while the respondents with low CD4 are 16.8% males' and16.6% females.

Prevalence of parasitic infections and Film appearance of the red blood cell status among the children

Table 8 shows the prevalence of parasitic infections among the respondents by location and gender. As for stool examination in urban LGAs, Ascarislumbricides was detected in 13.7% and Stronglyloides in 7.5%. No parasite was seen in 78.9% of the children in urban local government area. The stool examination in rural LGAs

Table 6: Severity of some biochemical indices among the children

Variables	Sector			p-value	Gender			p-value
Heamoglobin (mg/dl)	Urban	Rural	Total	0.032*	Male	Female	Total	0.040*
Severe	3(1.9)	6(4.0)	9(2.9)		3(1.9)	6(3.8)	9(2.9)	
Moderate	7(4.3)	11(7.3)	18(5.8)		9(5.8)	9(5.7)	18(5.8)	
Mild	17(10.6)	32(21.2)	49(15.7)		19(12.3)	30(19.1)	49(15.7)	
Normal	134(83.2)	102(67.5)	236(75.6)		124(80.0)	112(71.3)	236(75.6)	
PCV(%)				0.024*				0.032*
Low	34(21.1)	50(33.1)	84(26.9)		30(19.4)	54(34.4)	84(26.9)	
Normal	127(78.9)	101(66.9)	228(73.1)		123(80.6)	103(65.6)	228(73.1)	
	161	151	312		155	157	312	
Ferittin(ng/ml)				0.021*				0.051
Deficient	36(22.4)	57(37.7)	93(29.8)		39(25.2)	54(34.4)	93(29.8)	
Normal	125(77.6)	94(62.3)	219(70.2)		116(74.8)	103(65.6)	219(70.2)	
Reticulocyte				0.001*				0.122
Low	26(16.1)	41(27.2)	67(21.5)		29(18.7)	38(24.2)	67(21.5)	
Normal	112(69.6)	84(55.6)	196(62.8)		104(67.1)	92(58.6)	196(62.8)	
High	23(14.3)	26(17.2)	49(15.7)		22(14.2)	27(17.2)	49(15.7)	
	161	151	312		155	157	312	

[a]WHO, 2001; [b]Tatala et al, 2004; [C]Thurnham et al, 2010 and [d]Thurnham et al, 2010

Table 7: Severity of Markers of inflammation and immune function of the subjects

Variables	Sector			p-value	Gender			p-value
CRP	Urban	Rural	Total	0.211	Male	Female	Total	0.121
Low risk	117(72.7)	103(68.2)	220(70.5)		170(71.0)	110(70.1)	220(70.5)	
Mild risk	22(13.7)	27(17.9)	49(15.7)		27(17.4)	22(14.0)	49(15.7)	
High risk	22(13.7)	21(13.9)	43(13.8)		18(11.6)	25(15.9)	43(13.8)	
	161	151	312		155	157	312	
CD4				0.113				0.342
Low	28(17.4)	24(15.9)	52(16.7)		26(16.8)	26(16.6)	52(16.7)	
Normal	133(82.6)	127(84.1)	260(83.3)		129(83.2)	131(83.4)	260(83.3)	
	161	151	312		155	157	312	

shows that 23.2% were affected by Ascarislumbricides while Stronglyloides was detected in 11.3%. No parasite was seen in 65.6% respondents in rural LGAs respectively. As for gender, Ascarislumbricides was detected in 18.7%males and Stronglyloides in 10.3% of them. No parasite was seen in 71.0% of the male children. The stool examination among female children shows that 17.8% were affected by Ascarislumbricides while Stronglyloides was detected in 8.3%. No parasite was seen in 78.9% of the female children. The result showed a significant difference only in the sector (p=0.023).

Malaria was moderate in 28.6%, high in 20.5% but negative in 51.6% in urban LGAs. In rural LGAs, malaria was moderate in 36.4, high in 28.5% but negative in 35.1%. Malaria was moderate in 33.5% of the male children, high in 25.2% but negative in 41.3% of the male respondents. Among the female children, malaria was moderate in 30.6%, high in 23.6% but negative in 45.9% of them. There is a significant difference in the sector (p=0.023) but not in gender.

Table 8 also shows the film appearance used to determine the nature of the red blood cell of the respondents. Macrocyte is when the red blood cell is bigger than the normal while dacroctyte (tear drop cells) as to do with variability in RBC shapes, one side of the RBC is pinched and it forms a tear drop like shape and thus appear to be smaller than the normal counterparts. Poikilocyte can refer to an increase in abnormal red blood cell of any shape where they make up ten percent or more of the total population and normocyte is the normal red blood cell.

In urban LGAs, it shows that the nature of the RBC was macrocyte in 7.5%, and dacrocyte was the nature in 4.3%, while poikilocyte was noticed in 6.2% and normocyte was the nature of RBC in 82.0% of the childrenin urban local government areas. In rural LGAs, it shows that the nature of the RBC was macrocyte in 18.5%, and dacrocyte was the nature in 7.3%, while poikilocyte was noticed in 11.3% and normocyte was the nature of RBC in 62.9%.It also shows that the nature of the RBC was macrocyte in 13.5% of the male children, and dacrocyte was the nature in 6.5% of them, while poikilocyte was noticed in 8.4% and normocyte was the nature of RBC in 71.6% of the male children. For females, it shows that the nature of the RBC was macrocyte in 12.1%, and dacrocyte was the nature in 5.1%, while poikilocyte was noticed in 8.9% and normocyte was the nature of RBC in 73.9% of the female respondents. Only significant across the sector(p=0.030) and not gender (p=0.153).

Table 9 shows the correlations between socio economic status, markers of iron status, white blood differentials and makers of infection.CRP positively correlated

Table 8: Prevalence of parasitic infections and Film appearance of the red blood cell status among the children (N=312).

Variables	Sector			p-value	Gender			p-value
STOOL	Urban	Rural	Total	0.042	Male	Female	Total	0.121
Ascarislumbricides	22(13.7)	35(23.2)	57(18.3)		29(18.7)	28(17.8)	57(18.3)	
Stronglyloides		12(7.5)	17(11.3)	29(9.3)		16(10.3)	13(8.3)	29(9.3)
No parasite seen	127(78.9)	99(65.6)	226(72.4)		110(71.0)	116(78.9)	226(72.4)	
	161	151	312		155	157	312	
MALARIA				0.023				0.103
High	33(20.5)	43(28.5)	76(24.4)		39(25.2)	37(23.6)	76(24.4)	
Moderate	45(28.0)	55(36.4)	100(32.1)		52(33.5)	48(30.6)	100(32.1)	
Negative	83(51.6)	53(35.1)	136(43.6)		64(41.3)	72(45.9)	136(43.6)	
	161	151	312		155	157	312	
FILM APPEARANCE				0.030				0.153
Macrocyte	12(7.5)	28(18.5)	40(12.8)		21(13.5)	19(12.1)	40(12.8)	
Dacrocyte	7(4.3)	11(7.3)	18(5.8)		10(6.5)	8(5.1)	18(5.8)	
Poikilocyte	10(6.2)	17(11.3)	27(8.7)		13(8.4)	14(8.9)	27(8.7)	
Normocyte	132(82.0)	95(62.9)	227(72.8)		111(71.6)	116(73.9)	227(72.8)	
	161	151	312		155	157	312	

Table 9: Pearson's Correlations between socio economic status, markers of iron status, white blood differentials and makers of infection.

Variable	Childs age	Mothers age	Mothers Education	Annual Income	HH Size	Hb	SF	PCV
CRP	0.107	0.105*	0.070	-0.040	0.155*	0.238**	0.101*	0.103**
CD4	-0.120	0.252*	0.142**	0.005	0.144	-0.051	0.131	-0.102*
RETICS	0.012	-0.156	0.027	0.116	0.103	0.009*	-0.088	0.106
WBC	0.127*	0.030	0.110*	0.109	0.163*	-0.022	0.067	-0.036
NEUT	0.101*	-0.011	0.175**	-0.014	0.173	0.588**	0.124	0.126
LYMP	-0.078	0.202**	-0.043	0.511**	-0.049	0.109	-0.109*	0.115
MONO	-0.026	0.071	0.002	-0.020	0.008	0.103**	-0.010	0.017
EOSINO	0.171	0.080	-0.027	-0.007	0.101	-0.098	0.039	0.206
BASO	-0.008	0.080	0.030	-0.007	0.048	0.008	0.144	-0.104
MALARIA	0.342*	-0.045	-0.104*	-0.207**	0.003	0.112	0.109	-0.133*
STOOL EXAM.	0.014	0.157*	0.123	0.114**	0.168	-0.027	-0.036*	0.178
FILM APPEAR.	0.011	0.137	-0.058	0.003	0.112*	0.352	0.288	0.045*

Hb- Heamoglobin , SF- Serum Ferittin, PCV- Packed Cell Volume, CRP- C-reactive protein

***Correlation coefficient is significant at the 0.01 level (2- tailed*

**Correlation is significant at the 0.05 level (2- tailed)*

with mothers age (r= 0.105, p= 0.05), household size (r= 0.155, p= 0.05), Heamoglobin(r= 0.238, p= 0.01), serum ferittin (r= 0.101, p= 0.05), and PCV (r= 0.103, p= 0.01). CD4 positively correlated with mothers age and education (r= 0.252, p= 0.05; r= 0.142, p= 0.01) but negatively related to PCV(r= -0.102, p= 0.05). There was no significant correlation between reticulocyte counts and socio demographic parameters but positively related to Hb (r= 0.09, p= 0.05). There were significant relationships between white blood differentials and socio demographic parameters. Neutrophills related positively with childs age, and mothers education (r= 0.101, p= 0.05; r= 0.175, p= 0.01) and Hb (r= 0.588, p=0.01). Lymphocyte correlated positively with mothers age (r= 0.202, p= 0.01) and negatively related to SF (r=-0.109, p=0.05) while monocyte related positively to Hb (r=0.103, p= 0.05) but eosinophills and basophills has no significant correlation with the socio demographic and iron parameters. Malaria correlated positively to childs age (r=0.342, p= 0.05) and SF(r=-0.109, p= 0.05), but correlated negatively to mothers education (r=-0.104, p= 0.05), annual income(r=-0.207, p= 0.05), and PCV (r= -0.133, p= 0.05). Stool examination negatively correlated to SF (r=-0.036, p= 0.05) while film appearance positively related household size (r=--0.112, p= 0.05) and PCV (r= 0.045, p= 0.05).

DISCUSSION

According toWorld Health Organization (WHO) (2008), it has been reported that anaemia is a global public health problem, with one in four people being affected. Iron deficiency was the commonest cause of anaemia and contributed further to the anaemias of sickle cell disease and protein-energy malnutrition (Commeyet al., 1995). Irondeficiency has been reported to be the most prevalent nutritional deficiency in the world (Biesalski& Erhardt, 2007). A considerable number of children are anaemic in this study but it is not surprising to also note that more than one third is as a result of iron deficiency. This high rate of anaemia and iron deficiency anaemia in the

present study may be indicative of the fact that the diet of the school age most especially in the rural LGAs are not adequate for their iron needs. Observations and brief interviews of the people living in this rural areas during the data collection revealed that "garri" (cassava flakes) is their most common staple food with no formof animal protein. Farming is the major occupation and only crops that are likely to yield some income are planted. The level of income of family breadwinners is also low, judging by their houses and the yield from their farms. Their financial access to meat and other good animal sources of iron is therefore very limited. Considering other biochemical parameters under study, it can be easily concluded that though anaemia affect both urban and rural LGAs but the majority of those affected in urban may likely not be as a result of diet but other factor like parasitic infections and all these factors come into play in the rural LGAs. It is worthy to note that the public health effects of iron deficiency and anemia include reduced work capacity and mental performance, poor growth development, impaired regulation of body temperature, impairments in behavior and intellectual performance, and decreased resistance to infections (WHO, 2003; Yip, 2001). The values reported in this study are higher than the report of Taljaard (2011) which reported iron deficiency anaemia among black African school children aged 6-11 years in the Klerksdorp area of North West, South Africa. The prevalence of anemia and ID by Sanaet al. (2016) in the study conducted in Mexico, US and Colombia shows that 12% and 18% of children in Mexico were anaemic and iron deficient respectively. However, the values of aneamia and iron deficiency in this study arecloser to that reported byAchouriet al. (2015). Other studies (Sukanya et al., 1996, Onabanjo et al., 2014), have reported higher prevalence of anemia and iron deficiency among children. According to other study, school age children are at risk of iron because of an expanding red cell and muscle mass (Herbert,1992). Although the prevalence of anemia was more in girls than boys,and significantly higher in rural than in the urban LGAs in this study.Various studies have established the fact that girls are more likely to be anaemic than boys and an example of such is the study of Izolda et al (2006) of which the same trend was also observed in the present study. In contrast, some other studies found anaemia prevalence to be more in boys than girls and a good example is the study of Barduagniet al. (2004). Onimawo reported 82.6% anaemia among school age children in Abia state butsex specific anaemia rates were not significantly different for boys and girls (Onimawoet al.,2010). A study by Alom et al. (2022) showed that there is a positive growth rate among school children in the different age level .

In other to show the severity of the iron deficiency in the subjects, the heamoglobin concentration was further classified into severe, moderate, mild and normal according to World Health Organization classification (WHO, 2007). The result indicated that a good number of the children were severely and moderately anaemic with more in rural areas and female gender. The value reported in this study was lower than what was reported on the severity of anaemia among schoolchildren (6–15 years) in rural Nigeria by Rufina et al.(2015) but consistence with the study of Onimawo and colleagues where the overall prevalence of anaemia was 82.6% with rates of mild, moderate and severe anaemia being 9.6%, 71.6% and 1.4%, respectively and all the subjects that had severe anaemia were females while none of the males had severe anaemia (Onimawoet al.,2010). Thando et al. (2017) also reported 51.2% and 41.9% for mild and moderate forms of anaemia respectively, while severe anaemia was 2.3%. A jointly sponsored study in 2001 by WHO/UNICEF/UNN reported varying degrees of anaemia with38.0% mild anaemia, 31.8% were moderately anaemic and 0.8% was severely anaemic (WHO/UNICEF/UNN, 2001). However, low haemoglobin may not be totally a specific indicator for anaemia because it is also influenced by blood depleting parasites, chronic infections and haematological conditions (Hallet al., 1998&Flemming, 1981) and that is the reason for considering other parameter like PCV. It is however, interesting to note that majority of the children that are severely anaemic were females. This suggests that anaemia may not only have occurred as a result of low haemoglobin but may have been influenced by other factors like monthly menstrual period.

Serum ferritin (SF) concentration has been identified as the most specific biochemical test that correlates with relative total body iron store, hence is a precondition for iron deficiency in the absence of infection (Shell-Duncan et al., 2005). Low levels of SF were used to indicate iron depletion (Parachaet al., 2000). The result of the study shows that almost one third of the children had a low level of ferittin indicative of low iron stores in the body. There was significant difference in serum ferritin values in rural LGAs and urban LGAs but more female as compared to male were anemic.Thisresults obtained in this study is at variance with the studies of Thando et al.(2007) and that ofOnabanjoet al. (2014) that assessed the anthropometric and iron status of Adolescents inOgunState. The serum ferritin values reported by Onimawoet al. (2010) are lowerthan the present study. In the study, serum ferritincorrelatewith malaria parasites (r=0.109, p=0.05), indicative of infections in the study population. This conclusion is confirmed by a study conducted in Lagos, which showed that ferritin levels were significantly higher in subjects with high densities of malaria parasites (Odunukweet al.,2001).

C-reactive protein (CRP) is a marker of infection or inflammation in the body. It is released into the blood by the liver shortly after the start of an infection or inflammations as an early indicator of these problems and its levels can rise quickly. CRP and Serum ferritin when used in combination showed the best agreement with body iron stores (Thando et al., 2007). In the present study, result shows that CRP was elevated in a very close number of the subjects both in urbanand rural LGAswhich could be due to inflammation or infectionand

also more female gender than the males were affected. Elevated level of CRP (above 20mg/l) in this study is a pointer to the fact that malaria is still endermic in most of the areas featured in the study and other parasites like hookworm still affects good number of subjects in both urban and rural settlements.The overall percentage of the subjects with elevated CRP in this study is close to the value reported by John et al. (2008)in which raised CRP levels was identified in 16% of the children. The results obtained in this study is similar to works of others studies, (Cook et al., 2000; Ford, 2003; Thierfelderet al., 2007&Soldinet al., 2004) that found CRP to be higher in girls than in boys. Conversely, Rodooet al. (2013) and Colantonioet al. (2012) reported no gender differences in CRP values of children.

Reticulocyte are young, anucleate erythrocytes, which are released from bone marrow into the blood in increased numbers as a response to anemia caused by hemolysis (destruction) or loss (hemorrhage) of erythrocytes. Detection and identification of immature anucleate RBCs verifies whether the bone marrow is responding to the anemia by increasing RBC production in a regenerative response. Reticulocyte hemoglobin content is a reliable and early indicator of bone marrow iron status and may detect functional iron deficiency with more sensitivity than biochemical parameters (Brugnara, 2003). Low values indicate a low production of red cells possibly due to nutrient deficiency, whereas high values indicate a high production of reticulocytes to replace lost blood and healthy hematopoiesis. The result shows that more subjects in the rural LGAs have low level of reticulocyte as compared to urban subjects and this may likely be as a result of nutrient deficiency and this result is in line with the study of Foyet al. (2015) that reported reticulocyte hemoglobin content to be significantly lower in the blood donor group in their study, but it is not surprising that there are also subjects from urban LGAs that are anaemic because it reflected from the result that good number of subjects from both urban and rural LGAs had higher number of reticulocyte than normal range which is very likely to be a result of other factors like infections apart from diet. The result of reticulocyte heamoglobin content is a pointer to the fact that the rate of anaemia recorded in this study is not solely as a result of inadequate diet but other factors also come into play, more subjects had a low level of reticulocyte in rural which is suggestive of inadequate diet but also an increase in the level of reticulocyte was noticed in almost the same number of children in both urban and rural LGAs which is indicative of other parasitic infections and this is supported by the result of CRP in this study that reflects almost the same number of children in both urban and rural with elevated level of CRP. The implication of this is that the high level of anaemia in this study is not only as a result of diet but also infection plays a major role because both diet, malaria, hookworm infestation really affects rural children and surprisingly both malaria and hookworm infestation also affects urban children but their diet may be adequate. Several other factors such as intestinal parasitic infections and malaria parasitehave been showed to affect the iron status of the children apart from inadequate diet (James et al., 2015).

Malaria is one of the main public health problems in several developing countries affecting especially children, a particularly vulnerable population with the highest morbidity and mortality burdens associated with this disease (WHO, 2012 and Gethinget al., 2010). Malaria usually co-exists with other diseases and poor socioeconomic status, further impairing the development of the affected populations. Malaria currently accounts for nearly 110 million clinically diagnosed cases per year, 60 percent of outpatient visits, and 30 percent hospitalizations (FMOH, 2009) with prevalence of 76 per cent in the country (Nigeria) (WHO, 2007).Malnutrition is one of the most common and worrying conditions, impairing child development and the severity of other health conditions (Custodioet al., 2009). In this study the prevalence of malaria parasitaemia reported was high(43.2%). Other authors have also reported same percentage of overall malaria prevalence to be 43.1% during the follow-up study conducted by Alexandre et al. (2015) among the children but higher than the 27% prevalence of malaria parasite reported by Ademowo (1995) among school children from a rural village in western Nigeria and the study of Maketa that reported prevalence of malaria to be 30.9% and 14.3% in Cite Pumbu and Kindele health areas respectively (Maketaet al., 2015). Also, Isaac and colleaguesin their study on assessment of iron status among preschool children (6 to 59 months) with and without malaria in Western Province, Kenya reported a smaller percentage ofmalaria prevalence of 6.8% (Isaac et al., 2015). However, thevalues reported in this studywas lower when compared to 80% malaria parasite prevalence reported among school children in the malaria-endemic village of Erunmu in southwest Nigeria (Adeyemo, 1999) and also lower than 60% reported by Fuseniet al. (2009) in Ghana. The present study has shown that Plasmodium infections might appear to be more common among males than in the female subjects. Numerically, more males than females were infected despite the fact that more females than males were examined in the present study and it is comparable to the report of Okeahialamet al. (1972) and Uzoegwu and Onwurah (2003) for children and adults respectively. The present result conforms to the recorded higher prevalence of Plasmodium infection in male than in female school children in Ebonyi and Edo States, Nigeria as reported by Ani (2004). Also the Abah et al study shows that Out of the 300 samples, 63.3% were found to be positive with malaria parasite at varying degrees of parasiteamia. Sex related infection showed that more males were infected than females (Abahet al., 2015). The gender differences may be attributed to the fact that males expose their bare bodies more than females especially during hot weather. Thus, such males are more likely to be bitten by mosquitoes. Females, on the other hand, are usually not exposing most parts of

their bodies and tend to stay indoors, helping out with household chores thus reducing their contact with the mosquito vector as pointed out.Also, studies have shown that females have better immunity to parasitic diseases which is attributable to genetic and hormonal factors Zuket al., (1992).

Moreso, the percentage prevalence of Malaria in rural LGAs is far more as compared to the urban LGAs but the result clearly shows that there are good numbers of pupils from urban LGAs that are also affected and the result disagrees with the report of Kimbiet al, (2005).Also, the endemicity is high in rural than urban communities of Ebonyi State as reported by Adeyemo et al. (1999). This result suggests that the rural environment offers adequate conditions for breeding of mosquitoes. The socio–economic status of parents in rural communities also helps in the transmission of malaria, as most of these parents cannot afford screening of their homes, insecticides and treated bed nets for mosquito control. Illiteracy and ethnic beliefs among the rural dwellers have further encouraged the transmission of malaria parasites. In some cases, the infected children are not fully treated with the correct doses of antimalaria drugs. Angyoet al. (1996) in a similar study at Jos Nigeria reported a parasite rate of 70.5 % among children. In this study, older children are therefore less susceptible to malaria attack because they seem to have developed their own active immunity against malaria parasite (Angyoet al., 1996). Immunity has important effects on the transmission of the disease by reducing the level of parasitaemia after infective bites and increasing ten folds the rate of clearance of parasitaemia (Ademowo, 1995). With repeated infections in areas of intense transmission, the level at which parasitaemia stabilizes falls and the threshold for symptoms rises. Consequently, parasitaemia becomes asymptomatic and the risks of unrestrained parasitic multiplication to lethal burdens decline (Ademowo, 1995). The intensity of parasitaemia was high and the fact that children with such parasite densities were healthy and able to go to school means that malaria is well tolerated in the areas under study just as in many other parts of the sub- Saharan Africa. This is in consonance with the report of Ogunrin, (2001) who noted that children in endemic areas might tolerate very high levels of parasitaemia without severe symptoms.The result of this study is a pointer to the fact that the malaria prevalence in both urban and rural LGAs is high and can be a factor that predispose some of the pupils to anaemia in later years if that are not well taken care of.

Intestinal parasitic infections were one of the 17th neglected tropical diseases listed by the World Health Organization (WHO, 2008). Neglected tropical diseases accounts for the top 4th leading cause of communicable diseases and constituted 46–57 million disability adjusted life years lost (Jeremiah et al., 2012 and Hotezet al., 2006).The effect of intestinal parasites was not limited to morbidity and mortality; their impact extends to cognitive impairment, malnutrition, anemia, decreased school attendance, increased susceptibility of the host for infections like HIV/AIDS and tuberculosis, and impairing the growth of the child and adult productivity. They also impose serious socioeconomic impact on the development of a nation (Jeremiah et al., 2012; Bethonyet al., 2006; Hotezet al., 2009; Nokeset al., 1994; Hadidjajaet al., 1998 and Nyaruhuchaet al., 2006).Worldwide, approximately more than 2 billion people are infected with helmets infection (Jeremiah et al., 2012; Savioliet al., 2002 and Hotezet al., 2009). In Africa more than 173 million people are infected with Ascaris lumbricoides, more than 162 million cases of trichiasis, and more than 198 million cases of hook worm (Crompton et al.,2010; Molyneux et al., 2005 and Brooker et al., 2006). In developing countries 12% of the global disease burdens due to intestinal worm were estimated to occur on children 5–14 years old (Awasthi et al., 2003). Intestinal parasitic infections were the leading cause of infection in sub-Saharan Africa resulting in avoidable death. The results of this study shows that the burden of intestinal parasitic infection on the children inboth rural and urban areas to be high but more in the rural areas, calling for proper implementation of primary healthcare components in urban and rural areas. There is a little difference in the number of male and female affected if compared.This finding is similar to the report of Berhanu (2016) where no gender differences were observed in the number of male and female school children. Other studies report more of female been affectedby intestinal parasites than male; in that category is the work of Dinku (2017) that out of the total examined children, 50.81% were found to be infected with one or more intestinal parasites and higher infection prevalence was in femalesthan in males. The result of this study also shows that more children were affected in rural LGAs as compared to urban. The total prevalence of intestinal parasitic infection was high, This magnitude was higher when compared to finding from Malaysia and Philippines (Ahmed et al., 2012 and Janice,2014), but lower when compared to a study conducted by Berhanu (2016) that reported 61.7% prevalence of intestinal parasite among school children and also the study of Mumtaz et al. (2009). Also lower than the result of the study was the report of Ghiwotet al. (2014) in which the prevalence of intestinal helminth infection among the study participants was 15.5%.

There are several socio economics characteristics that contributed to the high prevalence of parasitic infections in both urban and rural LGAs in the study, some of which includespurifying water before drinking which is essential to prevent parasitic infestations. There are over 500 waterborne pathogens of potential concern in drinking waters, identified by the US Environmental Protection Agency (EPA) through its Candidate Contaminant List (Solleret al., 2010). Almost half of the children in this study did not drink boiled or filtered water and the implication of this is that unclean water is an avenue for transmission of many pathogenic organisms and the result is similar to the results obtained from a comparative

study done by Osten (Ostenet al., 2007). Hand washing with soap removes potentially pathogenic organisms and was found in this study that majority of infected children did not wash their hands regularly with soap.Proper hand washing with soap can reduce the output of infective stages in feces that results in the contamination of the environment, and hence can reduce infection transmission in the community (Harhayet al., 2010; Andersonet al., 2013) and the most important way to reduce parasitic infections is improved hygiene, especially adequate hand washing which reduces mortality due to diarrhea by as much as 50%, contributing more lives saved than vaccination or other medical intervention (Pittet, 2006). Similar results were obtained in study among Turkish school children (Ostanet al., 2007).

It also show from the study that high family size increases the risk of intestinal parasitic infection, the odds of intestinal parasitic infection for high family sized were 2-fold higher than children with low family size and it is in line with the study of Faleke (2016).Personal hygiene greatly reduces the burden of intestinal parasites. The prevalence of intestinal parasitic infection in school age children that keep their personal hygiene was low. This finding agrees with finding from different parts of the world (Ziegelbaueret al., 2012). This is due to the reason that proper personal hygiene breaks the chain of intestinal parasite transmission.The presence of intestinal parasitic infections may have multiple effects among children including physical and mental developments. The presence of chronic and heavy intestinal parasitic infection cause intestinal bleeding, malabsorption of nutrients, nutritional deficiency, destruction of cells and tissues and other associated effects. The overall effect of these results in growth retardation reduced mental development, school absenteeism, low academic performance, susceptible to malnutrition and infection (Brooker, 2010).

Packed Cell Volume is the proportion of the blood volume occupied by RBCs and is determined by cell number and size. Concentrations below the reference range may indicate abnormal cell development. In this study, about one third of the children had values below the reference range for PCV using WHO criteria but more of the children with low PCV are from rural LGAs and also more female subjects were affected. The results obtained in this study are similar to the finding of other study that reported 87.1% of the subjects been anaemic using PCV and also went further to categorized PCV values into mild anaemia, moderate anaemia and lastly severe anaemia(Onimawo et al. (2010).The result showed that PCV is positively correlated to CD4 (r= 0.05) and the implication is that when the immune system is been compromised there is every likelihood that the PCV level becomes low.

The CD4 count is like a snapshot of how well the immune system is functioning. CD4 cells (also known as CD4+ T cells, T-lymphocytes, or helper cells) are white blood cells that fight infection. It gives an indication of the healthy immune system and is the body natural defense system against pathogens, infections and illnesses. Inthis study, about one quarter of the children had a low level of CD4 with close values for male and female. This observation is similar to the report of Olga et al (2010) that assessed immune status and enzymes activity in blood lymphocytes in adult patients where CD4+-lymphocytes content was lower with increased Ig M and Ig G concentration. It is interesting to note that several studies have stated the link between low level of CD4 and anaemia may not necessarily be diet but majorly due to infection.Some of the studies include that of Hughes et al. (2009) which reported that total white blood cell and lymphocyte counts in peripheral blood are not decreased in malnourished children, and granulocytes are frequently elevated. Likewise, T-lymphocytes and CD4 counts appear normal in malnourished children. Their levels seem to be determined more by infections than by nutritional state, and do not reflect the degree of malnutrition-related immune deficiency, as high infectious mortality is seen in malnourished children, despite unaffected white blood cell counts (Hughes et al.,2009). Other studies in support of this finding is the studies by Keuschet al, (2013) and Campbell et al. (2003) which associated a decline in CD4 to result from high pathogen load rather than nutrient deficiencies, and thus primarily a cause of malnutrition, particularly of stunting. The result of the study clearly shows a weak correlation between immune status (CD4) and iron deficiency anaemia which implies that most of the subject that are anaemic in this study may not necessarily have a compromised immune system due to inadequate diet but because certain infection might have set in due to other factors. However, when data for anaemic participants were analyzed, there was a weak positive correlation between Hb concentration and CD4 count (r = 0.249). Results from this present study therefore suggest that Hb concentration may not be a suitable predictor of immune status (based on CD4 count). Results of more recent studies have supported the non-suitability of Hb concentration for predicting CD4 count (Emuchayet al., 2014; Sen et al., 2011; Alaviet al., 2009). Some other studies have however reported that Hb concentration can increase the sensitivity of total lymphocyte count in predicting CD4 count (Spaceket al., 2003), but this was not explored in the study.Nevertheless, food availability is still of vital importance in the study of nutritional status and their relationship with low CD4 cell outcomes since hunger is often a barrier (Johannessen et al., 2008); perhaps, the association between the nutritional status and immune status should be uniquely observed in populations where extreme malnutrition is persistent and thus stronger correlations can be deduced, but this study has convincingly showed that there is relationship between the iron status of the children and the markers of infection considered.

CONCLUSION

The following can be concluded from this study:

The socio-economic status and WASH practices were very poor among the children who participated in the study, particularly in rural areas.

The study revealed high prevalence of iron deficiency (23.7%) and aneamia (16.3%) with 13.1% of the prevalence of aneamia caused by iron deficiency among the children. This is also significant ($p<0.05$) for gender and sector differences in the iron status.

From this study, both malaria and helminthes infection are highly prevalent affecting 17.6% and 18.3% of the children respectively.

Data on immune markers (CD4 and white blood differentials) showed that more females had lower values than males. About 16.7% of the children had a low level of CD4 though more in rural with no significant difference among male and female ($p =0.015$).

The result of Reticulocyte (21.5%), ferittin (29.8%), C-reactive protein (13.8%) and PCV (26.9%) also buttress the fact that some of the children health status has been affected which could in turn leads to a compromised immunity.

The study revealed significant relationship between socio economic status, markers of iron status, white blood differentials and makers of infection in the children that participated in the study.

RECOMMENDATIONS

Based on the findings of the study, the following recommendations were made;

i. Appropriate investigations for iron status and inflammation/infection screening, need to be integral in the evaluation of anaemia and its causes before anaemia control interventions are implemented. Interventions that target the multifactorial nature of anaemia in school-aged children need to be strengthened. Additionally, regular screening of anaemia in school-aged children from disadvantaged communities is recommended.

ii. Better data are needed on the burden of malaria and other parasitic infections in school-age children to inform global policy makers and funders of the increasing importance of malaria and others in this age group and to ensure appropriate interactions between educational and health providers at national level. A standardised approach would improve the ability to monitor progress in this special group, and to document any changes in the risk of clinical malaria. Systems to capture episodes of clinical and fatal infections in school-age children need not be school-based, but should summarize data for this specific risk group.

iii. Operational research is needed to determine how best to raise awareness of the importance of malaria and other infections in school-age children and on how to improve the use of established control measures such as ITNs in this age group.

iv. A national policy for improved sanitation in rural zones would increase the quality of drinking water. Combining such a policy with better access to basic social services, including education and healthcare, would reduce the prevalence of parasitic infections and anemia and decrease the disease burden on these vulnerable remote communities, particularly among women and children.

v. At the household level, the promotion of community-based sanitation in the sedentary communities and of good hygiene, including regular hand washing will enhance the effects of such efforts. Access to safe water and environmental sanitation is particularly challenging among rural dwellers. Interventions should aim to reduce surface water consumption by providing access to modern wells, pipe borne water or locally adapted water treatments.

vi. Culturally sensitive behavior modifications to reduce open defecation, such as defecation far from water points and burial of feces, would decrease the level of human pathogens in surface water. Another approach should be directed toward improved personal hygiene, such as fostering hand washing schemes in schools.

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