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The Iron Status of Children with and without Sick Cell Disease at the University of Ilorin Teaching Hospital, Nigeria

L'état du fer chez les enfants avec et sans la maladie de la drépanocytose à l'Hôpital universitaire d'Ilorin, Nigéria

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ABSTRACT

BACKGROUND: Children with sickle cell disease (SCD) are at potential risk of iron overload from chronic transfusion and probable iron deficiency due to accelerated growth. However, only few studies on the iron status of children with SCD in Nigeria are available.

METHODOLOGY: A cross-sectional study compared the iron status of 109 children with sickle cell disease with 109 age- and sex-matched haemoglobin AA controls at the University of Ilorin Teaching Hospital. Parameters assayed were serum iron, ferritin, transferrin, and haemoglobin (Hb) concentrations. Considering the appropriate reference values for age and sex, these parameters were used to classify the children into high, normal or low iron status.

RESULTS: The median (interquartile range) serum ferritin level of 180.00 (237.50)ng/ml for the SCD subjects was significantly higher than 70.00 (120.00)ng/ml observed among controls, but the mean Hb and median serum transferrin levels were significantly lower in the subjects compared with the corresponding values in the controls, each showing statistical significance ($p < 0.05$). The median serum iron levels did not differ significantly between the SCD (112µg/dl) and non-SCD (128µg/dl), $p=0.309$.

A high proportion of subjects had low HB status (96.3%) compared with controls (56.9%), $p=0.001$. A significantly higher proportion of subjects (78%) had high ferritin status compared with the controls (48.3%; $p < 0.001$). Ten (9.1%) SCD children had low serum iron status compared to 28 (25.7%) HbAA controls, $p=0.002$. Thirty-four (31.2%) subjects had low transferrin status which was significantly higher than the corresponding number of controls (8; 7.3%; $p < 0.001$).

CONCLUSION: The children with SCD in the index study were iron sufficient. **WAJM 2023; 40(10); 1013-1020.**

Keywords: sickle cell disease; iron status; serum ferritin; children.

RÉSUMÉ

CONTEXTE : Les enfants atteints de la drépanocytose (SCD) sont potentiellement exposés à un excès de fer dû aux transfusions chroniques et à une éventuelle carence en fer due à la croissance accélérée. Cependant, seulement quelques études sur l'état du fer chez les enfants atteints de la SCD au Nigéria sont disponibles.

MÉTHODE: Une étude transversale a comparé l'état du fer de 109 enfants atteints de la drépanocytose à celui de 109 témoins de même âge et de même sexe porteurs d'hémoglobine AA à l'hôpital universitaire d'Ilorin. Les paramètres analysés étaient les concentrations de fer sérique, de ferritine, de transferrine et d'hémoglobine (Hb). En utilisant les valeurs de référence appropriées pour l'âge et le sexe, ces paramètres ont été utilisés pour classer les enfants en fonction de leur statut en fer, à savoir élevé, normal ou faible.

RÉSULTATS: Le niveau médian (plage interquartile) de ferritine sérique de 180,00 (237,50) ng/ml chez les sujets atteints de SCD était significativement plus élevé que les 70,00 (120,00) ng/ml observés chez les témoins, mais la concentration moyenne en Hb et le niveau médian de transferrine sérique étaient significativement plus bas chez les sujets par rapport aux valeurs correspondantes chez les témoins, chaque différence étant statistiquement significative ($p < 0,05$). Les niveaux médians de fer sérique ne différaient pas de manière significative entre les sujets atteints de SCD (112 µg/dl) et les témoins non atteints de SCD (128 µg/dl), $p=0,309$. Une proportion élevée de sujets présentait un faible statut en Hb (96,3 %) par rapport aux témoins (56,9 %), $p=0,001$. Une proportion significativement plus élevée de sujets (78 %) avait un statut élevé en ferritine par rapport aux témoins (48,3 % ; $p < 0,001$). Dix (9,1 %) enfants atteints de SCD avaient un faible statut en fer sérique par rapport à 28 (25,7 %) témoins HbAA, $p=0,002$. Trente-quatre (31,2 %) des sujets avaient un faible statut en transferrine, ce qui était significativement plus élevé que le nombre correspondant de témoins (8 ; 7,3 % ; $p < 0,001$).

CONCLUSION: Les enfants atteints de la SCD dans cette étude étaient suffisamment approvisionnés en fer. **WAJM 2023; 40(10); 1013-1020.**

Mots-clés: maladie de la drépanocytose ; statut en fer ; ferritine sérique ; enfants

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INTRODUCTION

Sickle cell disease (SCD) is an inherited chronic haemolytic anaemia with clinical manifestations arising from the tendency of the haemoglobin molecule (HbS) to polymerize and deform red cells into sickle shapes.¹ The characteristics of the disease include lifelong severe haemolytic anaemia, attacks of painful crisis, and chronic organ damage.²

In SCD, the chronic haemolysis increases iron availability from red cell destruction. Also, the increased iron absorption from the gastrointestinal tract and repeated blood transfusions imply that iron deficiency is unlikely in those with SCD.^{3,4} However, some studies have shown that iron deficiency does occur in children with SCD with a prevalence of 8.5%, 13.0% and 13.3%, as reported from Jamaica,⁵ Tanzania,⁶ and Yemen,⁷ respectively. In Nigeria, studies on iron status amongst children with sickle cell disease are few, with findings of iron deficiency⁸⁻¹⁰ or sufficient iron states.¹¹

Iron status can be measured using a combination of haematological parameters and biochemical indices. These parameters include haemoglobin concentration, mean corpuscular volume, red cell distribution width, serum levels of iron, transferrin, transferrin receptor and ferritin, transferrin saturation, and erythrocyte protoporphyrin. Each parameter of iron status reflects changes in different body iron compartments and how they are affected at varying levels of iron depletion.¹² Using a single indicator of iron status can be misleading when assessing iron deficiency because of the overlap between normal and abnormal values. Indeed, no single indicator monitors the entire spectrum of iron status. Hence, the recommendation of a battery of tests.^{12,13}

Considering the high prevalence of iron deficiency anaemia reported amongst Nigerian children (27.5% in under-fives),¹⁴⁻¹⁶ there is a clear need to know the iron status of those with SCD; hence, the study aimed to determine and compare the iron status of children with SCD and those with haemoglobin AA.

MATERIALS AND METHODS

Study design and site: A comparative cross-sectional study was conducted at the University of Ilorin Teaching Hospital (UIITH). The University of Ilorin Teaching Hospital is in Ilorin East Local Government Area of Kwara State, Nigeria. The state has an estimated population of 2.4 million using the 2006 census figure adjusted at an annual 2.83% growth rate.¹⁷ The hospital has an Emergency Paediatric Unit and a Specialist Consultation Clinic for sickle cell disease that holds once a week (Monday mornings). The average attendance for each clinic day is 25 patients, and the age range of children attending the clinic is 6 months to 18 years. The well-child clinic runs every weekday except Wednesdays. The well-child clinic also serves as a follow-up clinic for children managed for acute illnesses on the ward after discharge. These children are assessed to be well and subsequently discharged from the well-child clinic. The study was carried out over four months (February to June 2012).

Sample size determination: The minimum sample size was estimated by the formula [18]: $n = 1 + 2C(s/d)^2$, where s is the standard deviation calculated from the mean serum iron of subjects from a pilot study (98.1 µg/dl); d is the difference to be detected from the difference from the mean serum iron of subjects and controls (168.46 - 122.96 = 45.50); C is a constant dependent on the value of α and β selected with a power of 90% and a significance level of 5%, such that the minimum calculated sample size was 99. With a 10% attrition rate, the sample size was 109. Hence, 109 subjects and controls each were enrolled.

Study Population: Children with sickle cell disease aged 1-18 years in the steady state attending the sickle cell disease clinic were recruited as subjects. The controls were Hb AA children attending the well-child clinic.

Inclusion criteria for the subjects- Children aged 1-18 years with Genotype SS or SC who had no blood transfusion in the preceding three months and were in a

steady state. Steady state was defined as when the patient had neither fever nor was in crisis at presentation and at least in the previous four weeks; and was not on any iron-containing medication apart from routine folic acid, vitamin C, and proguanil or pyrimethamine.

Exclusion criteria of subjects - Those with a history of admission in the preceding four weeks, history suggestive of chronic liver disease, haematuria, or use of steroids or contraceptives in the preceding four weeks.

Inclusion criteria for controls - Children aged 1-18 years with Genotype AA, with no blood transfusion in the preceding three months, and had no fever at presentation.

Exclusion criteria for controls - Similar criteria as for subjects.

Subject Recruitment: Consecutive patients with sickle cell disease (in steady state) who came for routine follow-up clinic, satisfied the inclusion criteria, and signed the informed consent form, were recruited as subjects. In contrast, consecutive age- and sex-matched children with haemoglobin AA attending the well child clinic served as controls.

An interview-based semi-structured questionnaire was used to collect information on the child's socio-demographic characteristics and the clinical and laboratory findings. The questionnaire was pre-tested among mothers of children with sickle cell disease in the children's ward at Children Specialist Hospital, Ilorin, to ensure the validity and reliability of the instrument. These children were excluded in the main study.

Specimen Collection and Analysis: Utilizing an aseptic procedure, five millilitres of blood was collected from each subject and control via venepuncture.

Full blood count analysis was done using an automated blood analyzer (model: Symex KX 21®) for the red blood cell (RBC) count, haemoglobin concentration, haematocrit and red blood

cell indices (mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), and mean corpuscular haemoglobin concentration (MCHC)), white blood cell (WBC) count (total and differential), and platelet count.

Serum ferritin concentration was analyzed using the Nova Path TM ferritin kit which is a quantitative test based on the principle of a solid phase enzyme-linked immunosorbent assay. The human transferrin assay was a two-site immuno-enzymatic assay. Serum iron was assayed by adding a serum sample to a reagent buffered at an acid pH. The reagent included a reducing agent which aided in dissociation and reduced ferric iron to ferrous iron. At this acid pH, ferric iron dissociated from transferrin. A chromogenic reagent was then added and the chromogen complexes with ferrous iron formed a coloured complex. The coloured complex was measured by spectrophotometry.

The reference standards were calibrated against the International Committee for Standardization in Haematology (ICSH) Expert Panel on iron for the various tests.^{19,20}

Interpretations of results: All the serum ferritin, transferrin, and iron values obtained were compared with the standard paediatric reference range (Table 1)^{4,12} and subsequently used to categorize the patients. Subjects and controls with levels below, within or above the reference range were classified into low, normal and high iron status, respectively.

DATA ANALYSIS

The data obtained were analyzed using the Statistical Package for the Social Sciences (SPSS) software package version 21. Means and standard deviations (SD) were derived for the continuous variables with normal distribution, and median (interquartile range) for those without normal distribution. The Student's t-test and Mann-Whitney U tests were used to compare the means and medians, respectively. For simple proportions,

frequency tables were generated and presented. The Chi-square test (χ^2) was used to compare categorical variables and, when cell values were less than five, the Yates exact test was used. A *p*-value of less than 0.05 was regarded as statistically significant.

Ethical considerations: The study was approved by the Ethics and Research Committee of the University of Ilorin Teaching Hospital. Written informed consent was obtained from parents and assent from children older than seven years after adequate explanations.

RESULTS

Socio-demographic characteristics of the subjects and controls

Table 1 shows no significant differences in the age, gender and parental educational levels of the children with SCD and the controls. Of the 109 children with SCD, 98 (89.9%) subjects had haemoglobin SS while 11 (10.1%) had haemoglobin SC. A significantly higher proportion of children with SCD (67, 61.5%) had been previously transfused compared to 36 (33.0%) children with HbAA, *p* < 0.001. None of the participants had been transfused

within three months preceding the study.

The mean RBC count, haematocrit, and haemoglobin values were significantly lower in the children with SCD compared with the controls without SCD (*p* < 0.05 for all 3 variables). The mean values of the MCH and WBC of the children with SCD were significantly higher than in the controls (*p* < 0.05 for both variables). Other haematological parameters are shown in Table 2.

Iron status indices of the children with and without SCD

Table 3 shows the median serum ferritin level of subjects was significantly higher than that of controls, but the mean haemoglobin and median serum transferrin levels were significantly lower in the subjects compared with the corresponding values in the controls (each *p* < 0.05).

Iron status indices of children with SCD and controls based on reference standards.

Table 4 compares the iron indices of the patients and controls. A high proportion of those with SCD had low HB status

Table 1: Some laboratory tests and normal reference range for the indices of iron status in children^{4,12}

Test	Age (years)	Reference range
Serum iron	1 - 5	50 - 180 µg/dl
Transferrin	>1	203 - 60 mg/dl
Serum Ferritin (SF)	1 - < 2	4.0 - 405 ng/ml
	2 - < 10	9.3 - 58.7 ng/ml
	10 - < 14	10.1 - 62.9 ng/ml
	14 - 18	12.4 - 78.9 ng/ml
Haemoglobin (Hb)	1 - 2	10.5 - 13.5 g/dl
	3 - 6	11.5 - 13.5 g/dl
	7 - 12	11.5 - 15.5 g/dl
	13 - 18	13.0 - 16.0 g/dl
Mean Corpuscular Volume (MCV)	1 - 2	70 - 86fl
	3 - 6	75 - 87fl
	7 - 12	77 - 95fl
	13 - 18	78 - 98fl

Table 2: Socio-demographic and Haematologic Parameters of the subjects and controls

Variables	SCD Children n (%)	Non-SCD Children n (%)	χ^2	p
Age (years)				
<5	26 (23.9)	36 (33.0)	4.21	0.239
5-<10	25 (32.1)	37 (34.0)		
10-<15	36 (33.0)	30 (27.5)		
≥15	12 (11.0)	6 (5.5)		
Gender				
Male	61(56.0)	51 (46.8)	1.84	0.175
Female	48 (44.0)	58 (53.2)		
Mothers' Educational status				
None	13 (11.9)	6 (5.5)		
Primary	21(19.3)	17 (15.6)		
Secondary	28 (25.7)	38 (34.9)	4.53	0.210
Tertiary	47 (43.1)	48 (44.0)		
Fathers' Educational status				
None	5 (4.6)	6 (5.5)		
Primary	12 (11.0)	3 (2.8)		
Secondary	28 (25.7)	38 (34.9)	7.04	0.070
Tertiary	64 (58.7)	62 (56.9)		
Haematologic Parameters	Mean ± SD	Mean ± SD		
RBC ($\times 10^3$ cell/mm ³)	2.82 ± 0.56	4.2±0.90	-13.59	0.001
Haemoglobin (g/dl)	8.455±2.30	11.15±1.66	-9.92	<0.001
Haematocrit (%)	25.39±4.22	35.73±4.52	-17.46	<0.001
MCV (fl)	81.63±7.04	79.77±8.73	1.73	0.085
MCH (pg)	26.35±2.87	25.58±2.54	2.07	0.039
MCHC (g/dl)	32.08±1.91	32.58±4.51	-1.07	0.288
Platelet count ($\times 10^3$ / μ L)	318.29±111.55	308.08±102.98	0.70	0.483
WBC ($\times 10^9$ cell/l)	14.12±4.32	6.90±2.10	15.70	<0.001
Neutrophils (%)	59.4±7.50	57.73±10.15	1.38	0.169
Lymphocyte (%)	46.21±12.53	48.48± 12.20	1.36	0.170

(96.3%) compared with controls (56.9%), $p < 0.05$. A low proportion of children with SCD (10.1%) had low MCV (microcytosis) compared with the controls (21.1%) while 93 (85.3%) SCD participants had normal MCV compared to 80 (73.4%) of the HbAA participants, but the difference was not statistically significant, $p = 0.071$. No subject had low ferritin status but 13.8% of the controls had low ferritin status, and a significantly higher proportion of subjects (78%) had high ferritin status compared with the controls (48.3%) $p < 0.001$. Eighty-three (76.1%) controls had high transferrin status while a lower proportion of subjects (39, 35.8%) had high transferrin status. About one-third of subjects (31.2%) had low transferrin status which was significantly higher than the 7.3% of controls with a low transferrin status, $p < 0.001$.

Iron indices based on age among the children with and without SCD

The mean haemoglobin levels for all four age groups were significantly lower among the SCD children than the non-SCD controls. Table 5 shows the median serum ferritin levels were significantly higher in the children with SCD compared to controls for all the age groups except those aged 15 years and above. The median transferrin levels in the three age categories less than fifteen years were significantly lower than the subjects compared with the controls (Table 5).

Table 3: Comparisons of iron status indices of the subjects and controls

Iron indices	SCD children	Non-SCD children	z/t	P
Iron (μg/dl)				
Median (IQR)	112.00 (185.0)	128.00 (217.00)	-1.018*	0.309
Ferritin (ng/ml)				
Median (IQR)	180.00 (237.50)	70.00 (120.00)	-6.30*	<0.001
Transferrin(mg/dl)				
Median (IQR)	277.50 (454.25)	710.00 (2143.00)	-6.29	<0.001
HB (g/dl)				
Mean±SD	8.00 ± 1.54	11.15±1.66	-14.46	<0.001
MCV (fl)				
Mean±SD	81.63 ± 7.05	79.77±8.73	1.73	0.085

* = z value

Iron indices based on gender among the children with and without SCD

Table 6 shows the median ferritin levels were significantly higher in male cases than in male controls; however, the transferrin and haemoglobin levels were significantly lower amongst the male SCD subjects compared to the corresponding values in males without SCD (each $p < 0.001$). Similarly, this trend was identified by comparing the iron indices of the females with and without SCD (each $p < 0.001$) (Table 6).

Table 4: Comparison of iron status indices of the subjects and controls

Iron status indices based on references ranges for age	Subjects N=109 n (%)	Controls N=109 n (%)	χ^2	<i>p-value</i>
Haemoglobin(g/dl)				
Low	105 (96.3)	62 (56.9)	47.38	<0.001
Normal	4 (3.7)	45 (41.3)		
High	0 (0.0)	2 (1.8)		
MCV(fl)				
Low	11 (10.1)	23 (21.1)	5.303	0.071
Normal	93 (85.3)	80 (73.4)		
High	5 (4.6)	6 (5.5)		
Serum Iron(μg/dl)				
Low	10 (9.2)	28 (25.7)	12.22	0.002
Normal	65 (59.6)	45 (41.3)		
High	34 (31.2)	36 (33.3)		
Ferritin(ng/ml)				
Low	0 (0.0)	15 (13.8)	26.87	<0.001
Normal	24 (22.0)	41 (37.6)		
High	85 (78.0)	53 (48.6)		
Transferrin(mg/dl)				
Low	34 (31.2)	8 (7.3)	37.96	<0.001
Normal	36 (33.0)	18 (16.5)		
High	39 (35.8)	83 (76.1)		

Table 5: Bivariate analysis of serum iron indices levels based on age group between SCD and non-SCD children

Variable	SCD children	Non-SCD children	Mann U/T-test	p-value
Iron(μg/dl)				
Median (IQR)				
<5 years	111.50 (195.8)	146.00 (184.50)	461.5	0.926
5-<10 years	99.00 (202.00)	104.00 (189.50)	557.0	0.308
10-<15 years	119.50 (201.80)	62.00 (143.80)	240.0	0.075
≥ 15 years	112.00 (177.80)	457.00 (96.00)	7.0	0.006
Ferritin(ng/ml)				
Median (IQR)				
<5 years	139.00 (216.30)	90.00 (104.30)	318.5	0.033
5-<10 years	150.00 (270.00)	23.00 (111.5)	246.0	<0.001
10-<15 years	192.50 (233.80)	25.00 (76.00)	180.0	<0.001
≥ 15 years	166.50 (178.80)	340 (153.8)	16.0	0.060
Transferrin(mg/dl)				
Median (IQR)				
<5 years	351.00 (500.73)	550 (1347.50)	317.0	0.031
5-<10 years	333.5 (481.50)	2175 (5095)	251.0	<0.001
10-<15 years	253.25 (293.75)	2000 (3592)	175.0	<0.001
≥ 15 years	310.5 (799.38)	147 (268.75)	16.0	0.059
Hb (g/dl)				
Mean (SD)				
<5 years	8.13 (2.02)	10.14 (1.74)	-4.193*	<0.001
5-<10 years	7.45 (1.25)	11.60 (1.65)	-13.603*	<0.001
10-<15 years	7.73 (1.26)	11.70 (1.48)	-10.538*	<0.001
≥ 15 years	8.85 (1.57)	11.77 (1.45)	-4.014*	0.001
MCV(fl)				
Mean (SD)				
<5 years	78.81 (7.86)	76.72 (11.27)	0.811*	0.421
5-<10 years	81.84 (7.09)	80.49 (5.27)	0.925*	0.358
10-<15 years	82.91 (6.18)	83.10 (7.76)	0.113*	0.910
≥ 15 years	83.28 (6.56)	77.00 (7.90)	1.794*	0.094

* = t-values

DISCUSSION

The present study showed that all children with SCD had either normal or high serum ferritin levels as a significantly higher proportion of subjects (78%) had high ferritin status. This study finding of mainly normal or elevated serum ferritin levels is similar to the reports from Philadelphia, USA, which reported normal or high serum ferritin amongst 104 African-American children with SCD aged 0.5-17 years who had never received a blood transfusion.⁴ The index study's finding of high ferritin levels compares with findings from Congo with a prevalence of 21.4%²¹ and 35.0%,²² and the prevalence of 48.0% in Yemen in children with SCD.²³ Furthermore, the higher serum ferritin levels among children with SCD compared with those with HbAA in the current study were similarly reported by other authors in Nigeria from Lagos,²⁴ and Ife.²⁵ Also, in 2015, a similar study conducted in Ilorin, but among those aged 16-62 years, had reported higher serum ferritin levels in those with SCD compared to those without SCD.²⁶ The high ferritin level in SCD compared to controls reflects high iron body stores which may be attributable to the chronic haemolysis in the former group. Serum ferritin reportedly reflects mainly reticulo-endothelial iron stores.²⁷ Contributors to the raised serum ferritin in sickle cell disease may be increased iron in the reticulo-endothelial cells because of the constant breakdown of Hb, subnormal circulating mass of Hb, and the inflammatory reaction which is common in SCD patients due to infections.²⁷ However, this mechanism alone cannot explain the very high serum ferritin levels found in some of the patients. The most obvious source of excess body iron might be blood transfused because 61.5% of the patients in the present study had been previously transfused. It is noteworthy that almost half (48.6%) of the children with HbAA had high ferritin status in the study. While the explanation for this is not completely clear, about one-third of the children had been previously transfused in the past and

Table 6: Bivariate analysis of the serum iron indices levels based on gender between subjects and controls

Iron indices	SCD subjects		Non-SCD controls		P1	P2
	Male (n = 61)	Female (n = 48)	Male (n = 51)	Female (n = 58)		
Iron µg/dl, Median	114.00	104.00	129	128.00	0.296	0.696
(IQR)	(188.50)	(173.00)	(210)	(161.50)		
Ferritin ng/ml,	163.00	180.00	78.00	70.00 (86.30)	<0.001	<0.001
(Median (IQR))	(240.00)	(238.75)	(134.00)			
Transferrin mg/dl,	278.00	274.00	641.00	710.00	<0.001	<0.001
Median (IQR)	(492.75)	(390.35)	(1950.00)	(2822.50)		
Haemoglobin g/dl,						
Mean (SD)	8.06 (1.44)	7.91 (1.68)	11.31 (2.01)	11.01 (1.28)	<0.001	<0.001
MCV fl, Mean (SD)	80.61 (7.28)	82.91 (6.59)	77.02 (9.53)	82.20 (7.21)	0.026	0.601

P1=compares male scd and male non-scd

P2= compares female scd and female non-scd

some of them had been treated for infections such as severe malaria from which they had just recovered. The recovery process and transfusion may therefore have contributed to the high ferritin status noted as one of the inclusion criteria was lack of fever at presentation. A normal serum C-reactive protein (CRP) level in the control group would have shown that the child had fully recovered from the initial infection before enrollment; however, this was a limitation of this study as the CRP was not measured.

The present study, wherein none of the children had iron deficiency based on serum ferritin levels, contrasts with reports emanating from Enugu, South-East Nigeria and Calabar, South-South Nigeria of a prevalence of iron deficiency anaemia (IDA) in children with SCD of 47%,⁸ and 53%,¹⁰ respectively. The index study also contrasts with the reports from Jamaica, Tanzania, and India wherein IDA was found in 8.5%,⁵ 13.0%,⁶ and 67.7%,²⁸ respectively. The differing results obtained from the other Nigerian studies may be due to dietary iron deficiency, infections, and growth spurt, as well as socio-economic factors such as household size and income, which play a role in dietary iron intake.¹⁰ The Jamaican and Tanzanian studies were

limited by age group enrolled to under-fives⁵ and ten years.⁶ The Indian study was community-based, involving four poor, remote states in India where most of the population had low iron status.²⁸ Also, zinc protoporphyrins/haem (ZPP/haem) was used in the determination of iron status which could be falsely elevated in the presence of malaria parasitaemia.²⁸ In addition, a false elevation of protoporphyrins could be seen in patients with elevated reticulocyte count due to an accumulation of protoporphyrins in reticulocyte-rich blood. These variables were not sought in the present study. The heterogeneity found in the reported results seems to stem from the diversity of the groups studied in terms of being community or facility-based, age, sex, and transfusion status.^{5,8,28}

Haemoglobin or haematocrit determination has been more widely used as a laboratory index of iron status than any other iron parameter.²⁹ Any assessment of iron status should usually include the haemoglobin concentration because it defines a more advanced stage of iron lack, and it is the only laboratory assay that provides a quantitative measure of the severity of iron status once anaemia has developed.³⁰ As expected, the haemoglobin levels were low in all patients with SCD in the

present study compared to 28% of the controls. This finding could be explained by the recurrent haemolysis associated with SCD. The major limitation of haemoglobin determination is that it lacks both sensitivity and specificity.³⁰ The wide overlap in the frequency distribution of haemoglobin values between normal and iron-deficient populations severely limits the sensitivity of haemoglobin as a measure of anaemia.³⁰ The specificity of haemoglobin measurements as an index of iron status is also very low because many factors other than iron lack limit red cell production. Anaemia due to chronic infection, protein-calorie malnutrition, and haemoglobinopathies may be wrongly attributed to iron lack.³⁰ These limitations of isolated haemoglobin determinations can be readily circumvented by combining the measurement with more specific indices of iron status as was done in the present study.

The children with SCD had higher MCV compared to the controls, and this also holds true when disaggregated by sex in the current study. This finding has been similarly reported by Akodu et al²⁴ and Sani et al²⁶ but contrasts with the findings of lower MCV reported by Odunlade et al²⁵ and Akinbami et al.³¹ Moreover, the current study reported increasing MCV

with age which was similarly reported by Odunlade et al²⁵ in Ile-Ife. The mean corpuscular volume increases with age and is thus higher in children of an older age group compared to the younger age group. The different MCV results may be attributed to the different age groups studied in the various studies which were under-fives,²⁴ 1-15 years,²⁵ 14-57 years,³¹ and 16 to 62 years,²⁶ respectively.

The MCH level was higher in the SCD subjects than the HbAA controls which was similarly reported by the authors in Ile-Ife,²⁵ India,²⁸ and Jamaica.³² Studies have shown that serum ferritin concentration significantly increases when MCV and MCH levels increase and vice versa.^{33,34} In children with SCD, an increase in the demand of erythropoiesis resulting from chronic hemolysis may cause a deficiency state and macrocytosis depending on the vitamin B12 and folate balance.²⁵ Thus, the findings of high MCV and MCH in the children with SCD with high ferritin levels may be a reflection of increased iron uptake and hemoglobin synthesis by immature erythroid cells.³³

The haematologic response to therapeutic iron supplementation is the gold standard for diagnosing iron deficiency in those with SCD.³⁵ However, the potential risk of iron overload from iron supplementation or treatment in children with sickle cell disease should be a consideration. Moreover, iron indices are also influenced by inflammation or the high reticulocyte count associated with sickle cell disease. In the index study, the children who showed decreased MCV and low serum iron did not have low ferritin levels or high transferrin. The combination of normal or high ferritin with normal or low transferrin likely excludes the presence of clinically significant iron deficiency in these children with sickle cell disease. This variation in iron status indicators was also reported by Stettler et al.⁴ The findings were attributed to intra- and inter-individual variability and diurnal variation in the iron status indicators.⁹

A major limitation and potential

confounder of the study is that we did not rule out acute inflammation in these children, which could have been done with a biochemical assay such as C-reactive protein.

CONCLUSION

The present study has demonstrated that the children with sickle cell disease were iron sufficient. Children with SCD had high or normal iron status while 15% of the controls had low iron status based on serum ferritin. The use of serum ferritin may provide a comprehensive assessment of iron status in screening programmes.

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