

Serum ferritin levels in adults with sickle cell disease in Lagos, Nigeria

Akinsegun A Akinbami¹
Adedoyin O Dosunmu¹
Adewumi A Adediran³
Olajumoke O Oshinaike²
Vincent O Osunkalu¹
Sarah O Ajibola³
Olanrewaju M Arogundade¹

¹Department of Haematology and Blood Transfusion, ²Department of Medicine, Lagos State University, College of Medicine, Ikeja, Nigeria; ³Department of Haematology and Blood Transfusion, Faculty of Clinical Sciences, College of Medicine, Idiaraba, Nigeria

Background: Serum ferritin is considered to be one of the most important tools in the measurement of iron balance in steady-state sickle cell disease. Increased gastrointestinal absorption of iron has been reported in sickle cell disease because of the associated chronic hemolysis, and it is also thought that repeated red cell transfusion consequent to chronic hemolysis and anemia causes excessive iron levels. The aim of this study was to determine overall and gender-specific mean ferritin levels in patients with steady-state sickle cell disease in order to establish the prevalence of iron deficiency and overload.

Methods: This was a cross-sectional study in homozygous patients with sickle cell disease attending the sickle cell clinic at Lagos State University Teaching Hospital, Ikeja. A 5 mL blood sample was collected in plain bottles from consenting participants during steady-state periods. The serum was separated and analyzed for ferritin by enzyme-linked immunosorbent assay. Another 5 mL sample was collected for a full blood count, done on the same day of collection, to determine red blood cell indices, ie, mean cell volume, mean cell hemoglobin concentration, and mean corpuscular hemoglobin concentration. The Pearson Chi-square test was used for statistical analysis. The differences were considered to be statistically significant when P was <0.05 .

Results: In total, 103 patients were recruited for this study and comprised 58 women (56.40%) and 45 men (43.70%). The overall mean ferritin concentration was 93.72 ± 92.24 ng/mL. The mean ferritin concentration in the women was 92.00 ± 88.07 ng/mL and in men was 96.41 ± 99.80 ng/mL. Only eight (7.76%) of the 103 patients had a serum ferritin level < 15 ng/mL, while two subjects (1.94%) had a serum ferritin level > 300 ng/mL. Ninety-three subjects (90.29%) had serum ferritin within the normal reference range of 15–300 ng/mL.

Conclusion: In this study, 90% of subjects with sickle cell disease had normal iron stores; serum ferritin was higher in men than in women, and iron deficiency was more common than overload in the disease.

Keywords: serum ferritin levels, sickle cell disease

Introduction

Sickle cell disease results from an inherited abnormality of hemoglobin structure. The abnormal hemoglobin has valine replacing glutamine at position 6 of the beta-globin chain.¹ The hemoglobin variant produced polymerizes at low oxygen tension, causing the characteristic sickle deformity of red blood cells, increased mechanical fragility, shortened survival, and chronic hemolytic anemia.²

The normal iron level in men is 50 mg/kg and 40 mg/kg in women, and the normal serum ferritin range is 15–300 μ g/L, being higher in men (mean 100 μ g/L) than in premenopausal women (mean 30 μ g/L).³ Ferritin is the primary iron storage protein in

Correspondence: Akinsegun A Akinbami
Department of Haematology and Blood Transfusion, Lagos State University, College of Medicine, Ikeja, Nigeria
Tel 23 4180 2306 4925
Email ajoke_clinic@yahoo.co.uk

tissues, and ferritin is also an acute phase reactant, with elevated serum levels in the presence of chronic inflammation, infection, and liver disorders associated with sickle cell disease in crisis. In the absence of deranged iron status, conventional laboratory assessments of iron status, such as mean cell volume, mean cell hemoglobin, mean corpuscular hemoglobin concentration, and red cell distribution width, may be abnormal in patients with sickle cell disease due to the chronic inflammation and hemolysis associated with the disease.^{4,5}

It is also difficult to quantify the contribution of iron deficiency to anemia in chronic inflammatory conditions. This is especially true in conditions such as sickle cell disease where proinflammatory cytokines may induce an anemia of inflammation and alter the biochemical indicators of iron status commonly used to diagnose iron deficiency.^{6,7} Despite all these limitations, and in the absence of other pathology, serum ferritin is generally a reliable indicator of the presence of iron overload.³ Serum ferritin is also considered one of the most important tools in measurement of the state of iron balance in steady-state sickle cell disease.⁸ In the event of increased gastrointestinal absorption of iron due to chronic hemolysis associated with sickle cell disease,⁹ it is also expected that repeated red cell transfusions consequent to chronic hemolysis and anemia leads to excessive iron levels.¹⁰ Several studies have reported iron overload in sickle cell disease.^{11–13} However, other studies have reported iron deficiency in sickle cell disease,^{10,14} despite the risk of increased gastrointestinal absorption. This study aimed to determine both overall and gender-specific mean serum ferritin levels in patients with steady-state sickle cell disease in order to establish the prevalence of iron deficiency and overload in steady state.

Materials and methods

This was a cross-sectional study in homozygous patients with sickle cell disease attending the sickle cell clinic of Lagos State University Teaching Hospital, Ikeja, between October and December 2011. Approval for the study was obtained from the research and ethics committee at the hospital. Consenting participants were helped to complete a structured questionnaire, consisting of demographic information, history of smoking, consumption of alcohol, drug use apart from the usual routine medications for sickle cell disease, history of blood transfusion, and frequency of bone pain crises. Inclusion criteria were hemoglobin phenotype SS, no history of crises in the previous three months as established by a careful history and complete physical examination, no previous history of surgery, and no history of blood transfusion in the previous

three months. Exclusion criteria were a history of blood transfusion in the past three months, hemoglobin phenotype SC, previous history of surgery, and human immunodeficiency virus infection. Treatment with hydroxyurea is not a routine and standard practice at the center where the study was carried out. Participants were routinely treated with folic acid, vitamin B complex, and paludrine.

Collection of samples

A 5 mL blood sample was collected from each subject into a nonanticoagulated bottle during a period of steady state. The serum was separated and stored at -20°C on the day of collection until analysis for ferritin. Another 5 mL sample was collected from each subject for a full blood count, done on the same day of collection, to determine mean cell volume, mean cell hemoglobin, and mean corpuscular hemoglobin concentration.

Testing

An enzyme-linked immunosorbent assay kit (Biotec Laboratories Ltd, Ipswich, UK) was used for determination of ferritin. After incubation, bound/free separation was performed by simple solid-phase washing. The ferritin concentration in the sample was calculated based on a standard curve. The kit has a control and six standards (S_1 – S_5) with concentrations of 0, 5, 20, 100, 400, and 1000 ng/mL, respectively. The standards were calibrated against the World Health Organization Human Liver Ferritin International Standard 80/602. Assay of the control, standards, samples, and blanks was performed in duplicate, using the mean value for each parameter. Values of the absorbance of the standards against concentration were plotted to obtain a standard curve using the best-fit points. Absorbance values for the samples were interpolated on the standard curve to obtain the corresponding concentrations expressed in ng/mL. The mean reference values and ranges quoted by the manufacturer are 53 ng/mL; 6–180 ng/mL and 175; 20–400 ng/mL for both premenopausal women, and men, respectively.

Analysis of data

The descriptive data are shown as the mean \pm standard deviation. The Pearson chi-square test and *t*-test were used for analytical assessment. The differences were considered to be statistically significant at $P < 0.05$.

Results

In total, 103 patients were recruited for the study, comprising 58 (56.40%) women and 45 (43.70%) men. The mean age for

the entire group was 24.08 ± 7.84 (range 14–144) years. The mean age for the women was 25.13 ± 8.47 years and for the men was 22.82 ± 6.89 years. The majority (77, 74.8%) had previously received blood transfusions, while 26 (25.2%) gave no history of blood transfusion (Table 1).

Overall mean hemoglobin, cell volume, cell hemoglobin, and corpuscular hemoglobin concentrations were 7.92 ± 1.49 g/dL, 80.32 ± 11.30 fl, 26.40 ± 3.19 pg, and 32.42 ± 1.05 g/dL, respectively. The minimum and maximum hemoglobin concentrations were 4.6 g/dL and 11.8 g/dL, respectively; the minimum mean cell volume, mean cell hemoglobin, and mean corpuscular hemoglobin concentration values were 62.9 fl, 16.5 pg, and 29.9 g/dL, respectively, and the maximum values were 103.0 fl, 34.30 pg, and 34.40 g/dL, respectively. Mean hemoglobin, cell volume, cell hemoglobin, and corpuscular hemoglobin concentration values for men were 8.11 ± 1.53 g/dL, 81.80 ± 14.12 fl, 26.56 ± 3.70 pg, and 32.44 ± 1.01 g/dL, respectively; and for women were 7.78 ± 1.42 g/dL, 79.09 ± 6.39 fl, 26.27 ± 2.48 pg, and 32.41 ± 1.10 g/dL, respectively (Table 1).

The overall mean ferritin concentration was 93.72 ± 92.24 ng/mL, the minimum was 5 ng/mL, and the maximum was 480 ng/mL. The mean ferritin concentration for women was 92.00 ± 88.07 ng/mL and for men was 96.41 ± 99.80 ng/mL. The minimum ferritin concentration for women was 5 ng/mL and the maximum was 430 ng/mL. The minimum for men was 10 ng/mL and the maximum was 480 ng/mL. The difference in mean ferritin concentration between men and women was not statistically significant

using the unpaired *t*-test (0.24 , $P = 0.81$, Table 2). The mean ferritin concentration of subjects who had a history of blood transfusion was 91.72 ± 98.21 ng/mL and for those who had not been previously transfused was 100.83 ± 68.75 ng/mL. This was also not statistically significant ($P = 0.662$, *t*-test = 0.44, Table 2). Only eight (7.76%) of the 103 patients had serum ferritin < 15 ng/mL. While the majority (95, 92.23%) had serum ferritin > 15 ng/mL, only two (1.94%) had serum ferritin > 300 ng/mL. In total, 93 (90.29%) had serum ferritin within the normal reference range of 15–300 ng/mL (Table 1).

No statistically significant relationship was found between hemoglobin and corresponding ferritin concentration ($P = 0.877$), nor between patient age and ferritin concentration ($P = 0.187$, Table 1).

Discussion

Blood transfusion in patients with sickle cell anemia serves two major functions, ie, increasing the oxygen-carrying capacity of the blood¹⁵ and replacing abnormal red cells with normal ones, thereby alleviating symptoms and preventing complications.^{16,17} Over 70% of the patients recruited for this study had a previous history of blood transfusion. This highlights the fact that the practice of blood transfusion is common in the sickle cell anemia population, and almost all adult patients may have had multiple transfusions. However, this study did not show a statistically significant correlation between previous history of blood transfusion and ferritin concentration.

Table 1 Demographic distribution, history of blood transfusion, red blood cell indices, and ferritin levels

	Gender			
	Male	Female	Total	
Mean age (years)	58 (56.40%)	45 (43.70%)	103 (100%)	
Previous history of blood transfusion				
	Yes	No		
	77 (74.8%)	26 (25.2%)		
Red blood cell indices	Minimum	Maximum	Mean	
			Male	
			Female	
Hemoglobin concentration	4.6	11.8	8.11 ± 1.53	7.78 ± 1.42
Mean cell volume (femtoliter)	62.9	103.0	81.80 ± 14.12	79.09 ± 6.39
Mean cell hemoglobin (picogram)	16.5	34.30	26.56 ± 3.70	26.27 ± 2.48
Mean cell hemoglobin concentration (g/dL)	29.9	34.40	32.44 ± 1.01	32.41 ± 1.10
Ferritin concentration levels	Male	Female		
Mean \pm standard deviation (ng/mL)	96.41 ± 99.80	92.00 ± 88.07		
15–300 ng/mL	93 (90.29%)			
<15 ng/mL	8 (7.76%)			
>300 ng/mL	2 (1.94%)			
Hemoglobin versus ferritin levels	$P = 0.877$			
Age versus ferritin levels	$P = 0.187$			

Table 2 Correlation between mean ferritin concentration, gender, and history of blood transfusion

Parameters	n	Mean ferritin	P value	t-test
Male	45	96.41 ± 99.80	0.812	0.24
Female	58	92.00 ± 88.07		
	Yes	No	P value	t-test
Blood transfusion	77	22	0.662	0.44
Mean ferritin	91.72 ± 98.21	100.83 ± 68.75		

Mean cell volume, mean cell hemoglobin, and mean cell hemoglobin concentration are all reduced in anemia of chronic disease. Expectedly, in this study, mean hemoglobin concentration, mean cell volume, and mean cell hemoglobin were higher in men than in women. Increased erythropoiesis due to androgens in males, and iron loss or blood loss in females during menstruation may be responsible for higher levels of the red cell indices in males. However, reference ranges for erythropoietin are not different between the men and women.¹⁸

The mean ferritin concentration obtained for women was much higher than the nonsickle cell value quoted by the manufacturer of the kit used for the study. However, a lower value was obtained in the men. As expected, the mean ferritin value obtained in this study was higher in men than in women.³

The majority (90%) of the patients had serum ferritin within the normal reference range of 15–300 ng/mL. This was an unexpected result, given that over 70% of the patients recruited for this study had a previous history of blood transfusion, and also considering that, given the high rate of chronic hemolysis seen in patients with sickle cell disease, increased gastrointestinal absorption of iron due to chronic hemolysis would have been expected.⁹ It has been shown that repeated red blood cell transfusion consequent to chronic hemolysis and anemia provides excessive iron levels and ferritin values above the normal reference range.¹⁰ This study reported only about 2% of patients with a ferritin concentration above the normal reference range of 300 ng/mL, while 7.76% had ferritin levels below the lower limit of the normal reference range (15 ng/mL). A similar result was obtained by Aken'ova et al who reported a prevalence of 6% for iron deficiency in adult patients with sickle cell disease in Ibadan, Nigeria.¹⁹ Iron deficiency may be more common in sickle cell anemia than is widely believed.^{14,20} Iron deficiency is thus more common than iron overload in patients with sickle cell disease, which is not consistent with the numerous reports of iron overload in patients with sickle cell disease.^{11–13} Major confounders in our study were the other causes of anemia

not evaluated, including malaria, hookworm infestation, and folate/vitamin B12 deficiency, but unless associated with loss of iron they would not impact on the results.

Conclusion

The majority of patients with sickle cell disease have normal iron stores, mean serum ferritin is higher in men than in women, and iron deficiency is more common than iron overload in sickle cell anemia.

Acknowledgment

We are grateful to S Adeyoloja for performing the ferritin enzyme-linked immunosorbent assay.

Disclosure

The authors report no conflicts of interest in this work.

References

- Sergent GR. Nomenclature and genetics of sickle cell disease. In: *Textbook of Haematology on Sickle Cell Disease*, 1st ed. New York, NY: Oxford University Press; 1985.
- Isaacs WA, Hayhoe FG. Steroid hormones in sickle cell anemia. *Nature*. 1967;215:1139–1142.
- Hoffbrand AV, Lewis SM, Tuddenham ED, et al, editors. *Iron. Postgraduate Haematology*, 4th ed. New York, NY: Oxford University Press; 2001.
- Burhman E, Mentzer WB, Lubin BH. The influence of plasma bilirubin on zinc protoporphyrin measurement by a hematofluorimeter. *J Lab Clin Med*. 1978;91:710–716.
- Koerper MA, Stempel DA, Dallman PR. Anemia in patients with juvenile rheumatoid arthritis. *J Pediatr*. 1978;92:930–933.
- Stoltzfus R. Defining iron-deficiency anemia in public health terms: a time for reflection. *J Nutr*. 2001;131:565S–567S.
- Jurado RL. Iron, infections, and anemia of inflammation. *Clin Infect Dis*. 1997;25:888–895.
- Brownell A, Lowson S, Bronzovic M. Serum ferritin concentrations in sickle cell crises. *J Clin Pathol*. 1986;39:253–255.
- Erlanson ME, Walden B, Stern G, Hilgartner MW, Wehman J, Smith CH. Studies on congenital hemolytic syndromes, IV. Gastrointestinal absorption of iron. *Blood*. 1962;19:359–378.
- O'Brien RT. Iron burden in sickle cell anemia. *J Pediatr*. 1978;92:579–582.
- Oluboyede OA, Usang EA, Lukanbi FA, Ajayi OA. Evaluation of serum ferritin levels and other hematological parameters in a Nigerian population. *J Natl Med Assoc*. 1983;75:885–889.
- Hussein MA, Davis LR, Lulicht M, Hoffbrand AV. Value of serum ferritin estimation in sickle cell anemia. *Arch Dis Child*. 1978;53: 319–321.
- Brownell A, Lowson S, Bronzovic M. Serum ferritin concentrations in sickle cell crisis. *J Clin Pathol*. 1986;39:253–255.
- Rao KRP, Patel AR, McGinnis P, Patel MK. Iron stores in adults with sickle cell anemia. *J Lab Clin Med*. 1984;103:792–797.
- Charache S, Bleecker ER, Bross DS. Effects of blood transfusion on exercise capacity in patients with sickle-cell anemia. *Am J Med*. 1983;74:757–764.
- Lusher JM, Haglight H, Khalifa AS. A prophylactic transfusion program for children with sickle cell anemia complicated by CNS infarction. *Am J Hematol*. 1976;1:265–273.
- Green M, Hall RJC, Huntsman RG, Lawson A, Pearson TC, Wheeler PC. Sickle cell crisis treated by exchange transfusion. *JAMA*. 1975;231:948–950.

18. Krafte-Jacobs B, Williams J, Soldin SJ. Plasma erythropoietin reference ranges in children. *J Pediatr*. 1995;126:601–603.
19. Aken'ova YA, Adeyefa I, Okunade M. Ferritin and serum iron levels in adult patients with sickle cell anaemia at Ibadan, Nigeria. *Afr J Med Med Sci*. 1997;26:39–41.
20. Davies S, Henthorn J, Brozovic M. Iron deficiency in sickle cell anemia. *J Clin Pathol*. 1983;36:1012–1015.

Journal of Blood Medicine

Dovepress

Publish your work in this journal

The Journal of Blood Medicine is an international, peer-reviewed, open access, online journal publishing laboratory, experimental and clinical aspects of all topics pertaining to blood based medicine including but not limited to: Transfusion Medicine; Blood collection, Donor issues, Transmittable diseases, and Blood banking logistics; Immunohematology; Artificial and alternative

blood based therapeutics; Hematology; Biotechnology/nanotechnology of blood related medicine; Legal aspects of blood medicine; Historical perspectives. The manuscript management system is completely online and includes a very quick and fair peer-review system. Visit <http://www.dovepress.com/testimonials.php> to read real quotes from published authors.

Submit your manuscript here: <http://www.dovepress.com/Journal-of-blood-medicine-journal>