Hemoglobin and Ferritin Concentrations in Subjects with Metabolic Syndrome

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ABSTRACT

BACKGROUND: Metabolic syndrome (MetS), a clinical condition characterized by insulin resistance, glucose intolerance, dyslipidemia, hypertension, and obesity, has been linked with raised levels of serum ferritin (Sfr) concentrations.

OBJECTIVES: This study was carried out to compare hemoglobin (Hb) and Sfr concentrations in patients with MetS, regular donors and first-time donors.

MATERIALS AND METHODS: A total of 102 subjects who were between 18 and 60 years were enrolled for the study. They were divided into three groups. The first group ($n = 20$) was made up of 5 males and 15 females, all who met the criteria that define MetS. The second group ($n = 52$; $M = 34$, $F = 18$) were regular donors, while the last group ($n = 30$; $M = 16$, $F = 14$) were first-time donors or those who had not donated before. Following an overnight fast, 20 mL of venous blood was drawn from each subject. About 5 mL of this was put into sodium ethylenediaminetetraacetate (EDTA) specimen bottles for the full blood count parameters with Sysmex KX-21N hematology analyzer (made in Japan). The remaining 15 mL had serum separated for Sfr assay using enzyme-linked immunosorbent assay (ELISA) with a commercial assay kit manufactured by Teco Diagnostics.

RESULTS: Significant difference was found in the mean Sfr concentration of subjects with MetS (163 ± 136.92 ng/mL) and regular donors (41.46 ± 40.33 ng/mL), $P = 0.001$. The mean Sfr concentrations of subjects with MetS (163 ± 136.92 ng/mL) were also higher than that of first-time donors (102.46 ± 80.26 ng/mL), but it was not statistically significant, $P = 0.053$. The Hb concentrations of the three groups were not significantly different.

CONCLUSION: Sfr concentrations of regular donors were lower than that of subjects with MetS and first-time donors. The difference between regular donors and subjects with MetS was statistically significant. However, there is no significant difference in the Hb concentrations in the three groups. MetS is not associated with anemia or hyperferritinemia.

KEYWORDS: metabolic syndrome, ferritin, insulin resistance, dyslipidemia

CITATION: Adediran et al. Hemoglobin and Ferritin Concentrations in Subjects with Metabolic Syndrome. *Nutrition and Metabolic Insights* 2015:8 15–19 doi:10.4137/NMI.S23302.

RECEIVED: January 7, 2015. **RESUBMITTED:** February 23, 2015. **ACCEPTED FOR PUBLICATION:** February 25, 2015.

ACADEMIC EDITOR: Joseph Zhou, Editor in Chief

TYPE: Original Research

FUNDING: Authors disclose no funding sources.

COMPETING INTERESTS: Authors disclose no potential conflicts of interest.

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Introduction

Metabolic syndrome (MetS) is a clinical condition characterized by insulin resistance, glucose intolerance, dyslipidemia, hypertension, and obesity.[1] Most patients with this condition are old, sedentary, and have a degree of insulin resistance. Major predisposing factors include stress, genetics,[2] endocrine disorders like polycystic ovary syndrome, and excess caloric intake.[3]

MetS is known to affect 25% of western adults.[4] It is closely linked to insulin resistance and an increased cardiovascular risk,[5] and has been shown to accelerate the course of atherosclerosis.[6] This is because of overproduction of very low-density lipoprotein.[7]

Though the pathogenesis of this condition is incompletely understood, recent reports have suggested the role of oxidative stress catalyzed by accumulation of iron in excess of physiologic requirements.[8]

Iron, an element essential to the life of all mammalian organism, plays a key role in oxygen transport and in enzymes involved in mitochondrial respiration, DNA biosynthesis, and the citric acid cycle.[9] An important property of iron is derived from the ease with which it is reversibly oxidized and reduced. However, this characteristic also renders excess iron detrimental—mostly via the formation of reactive oxygen species (ROS), which may lead to severe organ damage.[3]

Furthermore, iron plays an important role in promoting lipid peroxidation, and iron overload may increase the risk of ischemic cardiovascular events through acceleration of development of atherosclerosis.[10]

Many years ago, a close relationship between the total amount of stored iron and the serum ferritin (Sfr) concentration in normal individuals was established.[11] Leggett et al also found out that Sfr concentration decreases with blood

donation.[12] We have also recently reported that regular blood donation has lowering effect on Sfr concentrations with no adverse effect on hemoglobin (Hb) status.[13]

MetS alterations have been well established as a clinically important differential diagnosis underlying hyperferritinemia, once inflammatory conditions or true iron overload syndromes have been excluded.[14]

The association of higher iron stores with diabetes and insulin resistance has been repeatedly confirmed by many investigators.[15–17] Ferritin levels were found to predict a higher rate of diabetes in prospective studies and case–control cohorts.[16] Furthermore, Sfr was positively associated with body mass index (BMI), visceral fat mass,[17] serum glucose levels, insulin sensitivity,[18,19] and cholesterol levels.[20]

Current evidence suggests that elevated body iron stores exert a detrimental effect on the clinical course of obesity-related conditions and that iron removal improves insulin sensitivity and delays the onset of type 2 diabetes mellitus.[9]

This study was therefore carried out to compare Hb and Sfr concentrations in patients with MetS and regular donors. If Sfr concentrations in patients with MetS were higher than in regular donors, regular blood donation may be protective against development of MetS or may ameliorate the clinical condition. This may also be a campaign tool to encourage regular blood donation in our society where regular blood donation is still not well accepted by most people.

Materials and Methods

A case–control study was carried out at Lagos University Teaching Hospital, Idi-Araba, Lagos, and Lagos State University Teaching Hospital, Ikeja, Lagos, after an ethical approval. All subjects gave their written, informed consent to participate in the research, which was conducted in accordance with the Declaration of Helsinki. Three groups of subjects who were between 18 and 60 years included those with MetS with at least three of the following features: (1) abdominal obesity, defined as the presence of waist circumference ≥94 cm in men or ≥80 cm in women; (2) fasting plasma glucose ≥100 mg/dL or drug treatment for elevated blood glucose; (3) serum triglycerides ≥150 mg/dL or drug treatment for elevated triglycerides; (4) serum HDL cholesterol (HDL-C) <40 mg/dL in men and <50 mg/dL in women or drug treatment for low HDL-C; or (5) blood pressure ≥130/85 mmHg or drug treatment for elevated blood pressure. The second group was regular donors who had donated for at least two times in one year, at least four times in two years, or at least six times in three years, and the third group was first-time donors or those who had not donated before. Menstruating women and participants with history and clinical features suggestive of hereditary hemochromatosis were excluded from the study.

Specimen collection and preparation. Following an overnight fast, 20 mL of venous blood was drawn from each subject. About 5 mL of this was put into sodium ethylenediaminetetraacetate (EDTA) specimen bottles for the full blood count with Sysmex KX-21N hematology analyzer (made in Japan). This was analyzed within six hours of collection. The remaining 15 mL of the blood was transferred to new plain screw-capped disposable plastic tubes and allowed to stand at room temperature until clotted (after about two hours), and the clot retracted. The serum was aliquoted and stored at −72°C until analysis was done for Sfr using enzyme-linked immunosorbent assay (ELISA) method with a commercial assay kit manufactured by Teco Diagnostics.

Statistical assessment. Data entry and analysis was performed using the SPSS version 16 (SPSS Inc.). The descriptive data were given in percentages and as mean ± standard deviation (SD). Chi-squared test was used for the analytic assessment. The differences were considered statistically significant when the *P* value obtained is less than 0.05.

Results

A total of 102 subjects who were between 18 and 60 years in three groups were enrolled into this study. The first group ($n = 20$) was made up of 5 males and 15 females, all who met the criteria that define MetS. In the second group ($n = 52$; $M = 34$, $F = 18$) were regular donors, while the last group ($n = 30$; $M = 16$, $F = 14$) were first-time donors or those who had not donated before.

As shown in Table 1, there was a statistical difference in the *M*:*F* ratio, mean age, and mean BMI between subjects with MetS (group 1) and regular donors (group 2). The *M*:*F* ratio of subjects with MetS (group 1) was 1:3 as against 2:1 of regular donors (group 2). The ages of subjects with MetS (group 1) was between 36 and 60 years with a mean age of 52.85 ± 6.62 years, while the age range of regular donors was 22–53 years and their mean age was 35.19 ± 7.94 years, $P = 0.001$ The BMI range of subjects with MetS was 22.58–38.20 kg/m^2 with a mean BMI of 31.27 ± 5.29 kg/m^2 and that of regular donors was 21.20–33.10 years and 26.50 ± 4.2 years, respectively, $P = 0.002$.

In Table 2, we compared the demographic pattern and BMI of subjects with MetS (group 1) with those of first-time donors (group 3). The *M*:*F* ratio of first-time donors was 1:1 as against 1:3 of subjects with MetS. There was a significant difference in the mean age of both groups. While the mean age of subjects with MetS was 52.85 ± 6.62 years, that of first-time donors was 33.71 ± 9.15 years, $P = 0.001$. There was also a significant difference in the mean BMI of both groups (MetS, 31.27 ± 5.29 kg/m^2; first-time donors, 26.30 ± 4.0 kg/m^2), $P = 0.004$.

Hb and Sfr concentrations of subjects with MetS (group 1) and regular donors (group 2) are shown in Table 3. The Hb range of subjects with MetS and regular donors was 8.20–16.50 g/dL and 10.9–15.8 g/dL respectively. The difference in the mean Hb concentration of subjects with MetS (12.99 ± 7.84 g/dL) and that of regular donors (13.47 ± 2.36 g/dL) was not statistically significant. Differences were found in the

Table 1. Sociodemographic data of subjects with MetS and regular donors.

PARAMETER	SUBJECTS WITH METABOLIC SYNDROME *n* = 20	REGULAR DONORS *n* = 52	*P*-VALUE
Sex			
Male:Female ratio	1:3	2:1	
Age (years)			
Range	36–60	22–53	
Mean (±SD)	52.85 ± 6.62	35.19 ± 7.94	0.001
BMI (Kg/m²)			
Range	22.58–38.20	21.20–33.10	
Mean (±SD)	31.27 ± 5.29	26.50 ± 4.2	0.002

Abbreviation: BMI, body mass index.

range of Sfr concentrations of both groups. The range of Sfr concentrations of subjects with MetS was 10.00–550.20 ng/mL, while that of regular donors was 1.10–81.90 ng/mL. Furthermore, statistically significant difference was found in the mean Sfr concentrations of both groups. The mean ferritin concentration of subjects with MetS was 163.00 ± 139.92 ng/mL, while that of regular donors was 41.46 ± 40.33 ng/mL, $P = 0.001$.

As shown in Table 4, though the Hb range of subjects with MetS (8.20–16.50 g/dL) was different from that of first-time donors (11.5–15.20 g/dL), the difference in their means (MetS, 12.99 ± 7.84 g/dL; first-time donors, 12.98 ± 1.3 g/dL) was, however, not statistically significant.

Lastly, mean Sfr concentration of subjects with MetS (163.23 ± 136.92 ng/mL) was insignificantly higher than that of first-time donors (102.46 ± 80.26 ng/mL), $P = 0.053$.

Discussion

The finding of more women than men in this study agrees with a report by Beigh and Jain[21] who found out that the prevalence of MetS was higher in women (29%) than in men (23%) and with that of Park et al[22] who reported a prevalence of 39.4% for women and 37.6% for men. Our finding is, however, at variance with a report by Novak et al[23] in a study conducted among Swedish population in which MetS was more prevalent in men than in women. It has been established that there is close interrelationship between sex hormones and some pathways involved in the MetS, and the circulating concentrations of hormones may determine the degree of overall pathological alterations in the syndrome.[24] There is a strong inverse correlation between body fat and testosterone levels in men.[25] The presence of low testosterone and/or low sex hormone-binding globulin (SHBG) predicts the development of MetS and type 2 diabetes.[26] Furthermore, obesity is a proinflammatory state resulting in increased release and secretion of proinflammatory cytokines and adipokines, free fatty acids, and estrogens from adipose tissue.[26] This goes to show that the prevalence of MetS is expected to be higher in women than in men.

We observed that the mean BMI of subjects of MetS (31.27 ± 5.29) was significantly higher than other groups (regular donors, 26.30 ± 4; first-time donors, 26.50 ± 4.2). This difference did not affect erythropoiesis in the subjects studied. This is in agreement with Ghadiri-Anari et al[27] who concluded that BMI has no correlation with Hb in subjects with MetS in their study.

Though the range of Hb concentrations in subjects with MetS (8.20–16.50 g/dL) was wider than the two other groups,

Table 2. Sociodemographic data of subjects with MetS and first-time donors.

PARAMETER	SUBJECTS WITH METABOLIC SYNDROME *n* = 20	FIRST-TIME DONORS *n* = 30	*P*-VALUE
Sex			
Male:Female ratio	1:3	1:1	
Age (years)			
Range	36–60	18–59	
Mean (±SD)	52.85 ± 6.62	33.71 ± 9.15	0.001
BMI (kg/m²)			
Range	22.58–38.20	20.20–30.00	
Mean (±SD)	31.27 ± 5.29	26.30 ± 4.0	0.004

Table 3. Hb and ferritin concentrations of subjects with MetS and regular donors.

PARAMETER	SUBJECTS WITH METABOLIC SYNDROME *n* = 20	REGULAR DONORS *n* = 52	*P*-VALUE
Haemoglobin (g/dL)			
Range	8.20–16.5	10.9–15.8	
Mean Hb (±SD)	12.99 ± 7.84	13.47 ± 2.36	0.689
Ferritin (ng/mL)			
Range	10.00–550.20	1.10–81.9	
Mean (±SD)	163.23 ± 136.92	41.46 ± 40.33	0.001

reflecting various pathological stages such as renal dysfunction and fatty liver that may compromise erythropoiesis in some of the subjects, the mean Hb concentrations of the three groups (MetS, 12.99 ± 7.84 g/dL; regular donors, 13.47 ± 2.36 g/dL; first-time donors, 12.98 ± 1.30) were normal and not significantly different from each other, indicating that MetS or regular donation may not predispose to anemia.

That the mean value of Hb concentration of subjects with MetS (12.99 ± 7.84) is not significantly different from first-time donors (12.98 ± 1.30) is at variance with report by Päivi Hämäläinen et al[28] from Finland who observed that subjects with MetS have elevated Hb concentrations in their study.

We had earlier reported health benefits of regular blood donation in preventing the accumulation of body iron, which can cause free radical formation and result in lowering blood lipids, reducing the risk of cardiovascular accident.[13] Several reports have established that MetS being a chronic inflammatory disorder is usually associated with increased levels of Sfr and other acute-phase proteins.[29] That means Sfr concentration of subjects with MetS (163.00 ± 136.92 ng/mL) is insignificantly higher than that of first-time donors (102.46 ± 80.26 ng/mL) may also reflect various pathological states beyond the scope of this study. This notwithstanding, there are reports linking iron with risk of development of MetS.[1] In this study, we found out that the mean Sfr concentration of subjects with MetS (163.00 ± 136.92 ng/mL), though within the reference range (15–300 ng/mL), was significantly higher than in regular donors (41.46 ± 40.33 ng/mL) but with no appreciable difference in Hb concentration, emphasizing the health benefits of keeping body iron at minimal levels.

Furthermore, using Sfr concentration of 100 ng/mL as cutoff for adequate iron storage in subjects with chronic inflammatory disease as compared to 15 ng/mL for normal population,[30] we have found out that MetS may not be associated with iron overload or poor iron storage in the subjects studied.

It should be noted that iron overload is not a prerequisite for iron to mediate either diabetes or its complications. Important in its pathophysiology is the availability of the so-called catalytic iron or iron that is available to participate in free radical reactions.[31] Therefore, a reliable and sensitive method needs to be developed to precisely measure the free/catalytic iron that participates in oxidative injury.

A randomized, controlled, single-blind clinical trial carried out by Houschyar et al[1] on 64 patients with MetS who were randomly assigned to iron reduction by phlebotomy resulted in consecutive reduction of body iron stores, lowered BP, and improvements in markers of cardiovascular risk and glycemic control. They concluded that blood donation may have beneficial effects for blood donors with MetS. In fact, simple and inexpensive therapies, such as bloodletting and iron chelators, are emerging as alternative and effective treatments for insulin resistance.[32] That the mean age of the subjects with MetS is significantly higher than that of regular blood donors provides an opportunity for us to follow up the regular blood donors overtime to see if and how many of them would develop MetS as they grow older, while still remaining regular donors.

Table 4. Hb and ferritin concentrations of subjects with MetS and first-time donors.

PARAMETER	SUBJECTS WITH METABOLIC SYNDROME *n* = 20	FIRST-TIME DONORS *n* = 30	*P*-VALUE
Haemoglobin (g/dL)			
Range	8.20–16.5	11.5–15.20	
Mean Hb	12.99 ± 7.84	12.98 ± 1.30	0.994
Ferritin (ng/mL)			
Range	10.00–550.20	15.72–409.23	
Mean	163.23 ± 136.92	102.46 ± 80.26	0.053

In conclusion, mean Sfr concentration of regular blood donors was lower than that of subjects with MetS and first-time donors with no significant difference in the Hb concentrations in the three groups. Regular blood donation may have a beneficial effect in the clinical management of MetS. Though concerted effort is being made to provide alternative to blood donation such as Hb substitutes,[33] to the best of our knowledge, the effort is yet to yield fruitful results. It is therefore heartwarming to welcome the emergence of many beneficial effects of blood donation. This report presents another campaign tool in this direction.

Limitations of the Study

Serum iron, serum transferrin, erythrocyte sedimentation rate (ESR), and C-reactive protein (CRP) concentrations could not be evaluated.

Author Contributions

Conceptualization/study design/study supervision/manuscript write-up: AAdediran. Study design/study supervision/manuscript review: UE. Study design/data analysis/manuscript review: AAkinbami. Sample collection/literature search: DA. Study design/manuscript review: WT. Study design/manuscript review: DD. Study design/literature search/manuscript review: AS. Literature search/study design/manuscript review: OO.

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