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LIPID PROFILE AND DISEASE SEVERITY IN SICKLE CELL DISEASE PATIENTS IN LAGOS STATE. NIGERIA

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ABSTRACT

Background: Sickle cell anaemia is an autosomal recessive disorder that arises due to the substitution of glutamic acid with valine. This occurs at position 6 of the haemoglobin b chain, resulting in the synthesis of abnormal haemoglobin and the consequent production of the characteristic sickled red blood cells.

Studies have documented several alterations in lipid homeostasis in this population. Both hyper and hypolipidaemias are known to be associated with increased morbidity and mortality and it is therefore imperative to evaluate their relationships with sickle cell anaemia.

Aim: The aim of this study was to establish baseline serum lipid levels in sickle cell anaemia patients in LASUTH and correlate this with severity scores in the patients.

Subjects and Methods: Serum Total cholesterol (TC), Triglycerides (TG), Low-density lipoproteins (LDL), High-density lipoproteins (HDL) and Very low-density lipoproteins (VLDL) were measured in 57 Haemoglobin SS (HbSS) patients in steady state. All patients used had been fasting for at least 10 hours prior to sample collection. The LDL/HDL was also calculated. Their disease severity was calculated using an objective scoring method.

Results: Our results showed that there was no significant correlation between serum lipid levels and disease severity score.

Keywords: sickle cell disease, serum total cholesterol, low-density lipoproteins, high-density lipoproteins, triglycerides, disease severity.

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INTRODUCTION

Several biomarkers have been identified as predictors of clinical severity and prognosis of sickle cell anaemia. Some of the more common markers include foetal haemoglobin (HbF), white cell counts, nitric oxide (NO) levels and serum lactate dehydrogenase (LDH) (1-3)

Some researchers have suggested that serum lipid levels correlate with disease severity in patients with sickle cell anaemia and thus can be used as predictors of clinical severity in this group of patients. Studies have documented several alterations in lipid homeostasis in this population. Both hyper and hypolipidaemias are known to be associated with increased morbidity and mortality and it is therefore imperative to evaluate their relationships with sickle cell anaemia.

Low levels of total cholesterol (TC), high density lipoproteins (HDL-C) and low density lipoproteins (LDL-C) as well as hypertriglyceridaemia in sickle cell disease patients in both steady state and crises have been documented (4,5). Although the effects of disordered lipid metabolism on the course of sickle cell anaemia and its complications has yet to be clearly established, Zorca et al reported that hypertriglyceridaemia is a potential risk factor for pulmonary hypertension in sickle cell anaemia patients [4]. In their study, they established that a reduction in TC, LDL-C and HDL-C were significantly associated with severity of anaemia, whereas increased TG levels were associated with haemolysis, vascular dysfunction and increased prevalence of pulmonary hypertension.

Even though these alterations in lipid metabolism have been established by various workers, the mechanisms for these changes have remained largely unclear

It has been hypothesized that the low cholesterol seen in sickle cell anaemia may be due to an increase in the utilization of cholesterol during increased erythropoiesis and this was validated by Shalev et al. In their study, they demonstrated that the hypocholesterolaemia seen in sickle cell anaemia and other haemolytic anaemias was absent in patients with non-haemolytic anaemia. They thus concluded that hypocholesterolaemia is a specific manifestation of increased red blood cell synthesis (6). Other mechanisms may include defective liver function secondary to hyperferritinaemia, as well as defects in post absorptive plasma homeostasis of fatty acids (6, 7).

MATERIALS AND METHODS

Sixty (60) HbSS patients attending the Haematology Outpatient Clinic at Lagos State University Teaching Hospital who were in steady state were recruited for this study. However due to improper completion of questionnaires, 3 patients were excluded from the study and therefore 57 patients were used. Informed written consent was obtained from the subjects or their parents/caregivers before commencement.

Ethical approval for the study was obtained from the Hospital Ethics Committee (Ethics Registration Number LREC.06/10/949) Demographic information as well as transfusion history and disease complications were recorded.

Inclusion criteria

Adult HbSS patients with alkaline Haemoglobin electrophoresis showing HbSS phenotype and who 14 years are and above, with no history of crises in the past 3 months established by a careful history and complete physical examination and no history of blood transfusion in the past 3 months.

Exclusion criteria

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Patients with history of diabetes mellitus, hypertension, those on lipid lowering drugs and those with other identified risk factors for dyslipidaemias were excluded from the study. Non- consenting patients, patients with other haemoglobin phenotypes eg HbSC and those with a recent history of crisis were also excluded.

Evaluation of disease severity: Disease severity was determined using an objective score. A modification of the method described by Hedo et al (8) was used. Points were assigned for the number of blood transfusions received in the last one year, number of complications present, as well as the average crises rate/year Scores of 0-1 were considered mild disease, 2-3 moderate disease, and 4-6 severe disease.

Specimen Collection

5mls of blood was collected into a plain vacutainer bottle under aseptic conditions to obtain serum for lipid profile of all participants. The samples were analysed using the COBAS C111 auto-analyser manufactured by ROCHE DIAGNOSTICS GMBH SANDHOFER STRASSE 116 MANNHEIM which utilizes the enzymatic colometric method for assays.

Statistical analysis

Data obtained were analysed using Statistical Package for Social Sciences software package version 20 (SPSS Inc., IL, Chicago, USA). Pearson's and Spearman's correlation tests were used to determine the correlation between variables and P values ≤0.05 were considered as significant

RESULTS

Fifty-seven (57) HbSS patients in steady state were studied. There were 30 (52.6%) females and 27 (47.4%) males. Their ages ranged between 13 and 52 years with a mean age of 22.82 ± 8.4 years. The mean Body Mass Index (BMI) was 20.85 +3.34kg/m2. (Table 1).

Table 1: Socio-demographic variables and Body Mass Index of the patients

Sex	Age	BMI
Female30 (52.6%)	22.82 ± 8.4 years	20.85 ± 3.34 kg/m2
Male 27 (47.4%)		

The mean age of diagnosis of HbSS was 4.11 ± 4.19 years and all the patients were diagnosed using haemoglobin electrophoresis.

Most of the patients (68.4%) had been transfused with the number of units transfused ranging between 1 and 6 with a mean of 1.79 ± 1.28 units.

Of the 57 patients studied, 23 (40.4%) met the criteria for mild disease, 18 (31.6%) had moderate disease and 16 (28.1%) had severe disease. (Table 2).

Table 2: Disease severity among the patients

	Frequency (N)	Percentage (%)
Mild disease	23	40.4
Moderate disease	18	31.6
Severe disease	16	28
Total	57	100

The mean number of crises/year was 1.68 ± 0.69 (Figure 1) and the most frequent type of crises among the respondents was vaso-occlusive crisis (VOC). Figure 2. Only 9 out of the 57 patients had complications within one year but more than 3 months preceding the study. Figure 3. The mean TC, TG, HDL, and LDL

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were 3.25 ± 0.61mmol/L, 1.34 ± 0.60mmol/L, 0.92 ± 0.21mmol/L and 1.80 ± 0.56mmol/L respectively. The mean LDL/HDL was 2.05 ± 0.77mmol/L. Table 3. Lipid levels showed no significant correlation with disease severity scores. (Table 4)

Figure 1: Number of crisis/year

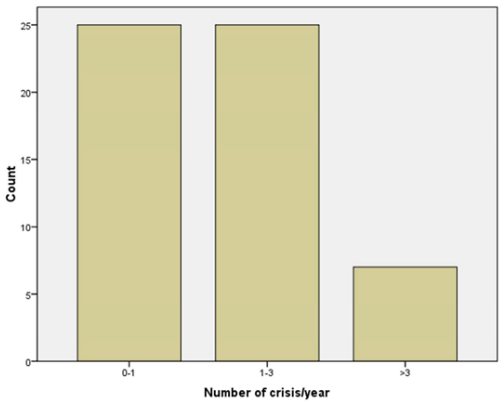


Figure 2: Most frequent type of crisis

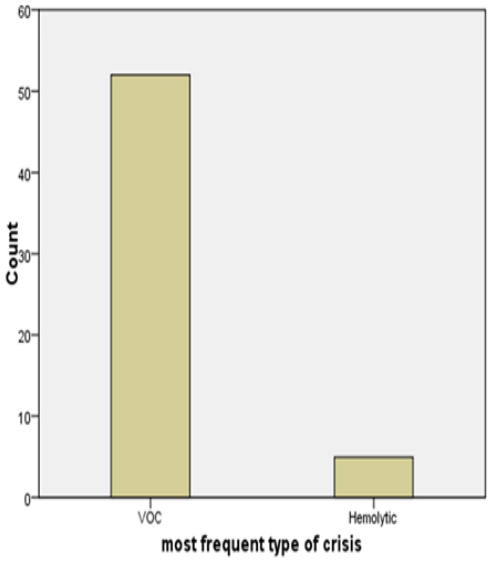
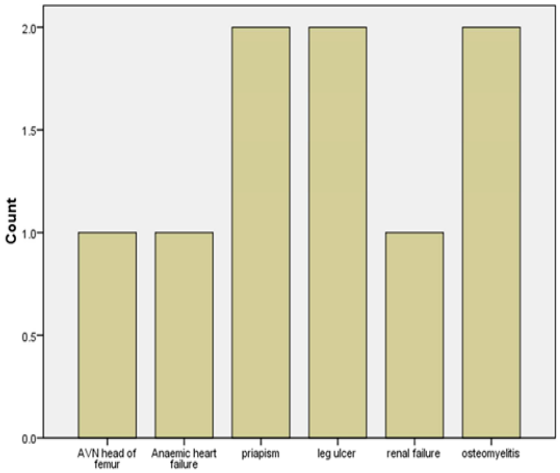


Figure 3: Complications in the past one year



Complications in the past one year but more than three months preceding the study.

Table 3: Mean Lipid levels of the patients

Variable	Mean (mmol/L)
Total cholesterol (TC)	3.25 ± 0.61
Triglycerides (TG)	1.34 ± 0.60
High Density Lipoproteins (HDL)	0.92 ± 0.21
Low Density Lipoprotein (LDL)	1.80 ± 0.56
LDL/HDL ratio	2.05 ± 0.77

Table 4: Correlation between lipid levels and disease severity

Variable	P value
Total Cholesterol	0.439
Triglycerides	0.292
HDL	0.288
LDL	0.456
LDL/HDL	0.406

Significant level P≤0.05

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DISCUSSION

This study was aimed at assessing the lipid profile of sickle cell disease patients in steady state, as well as to determine the relationship if any between lipid levels and disease severity scores.

The mean TC level was reduced when compared with HbAA controls used in other studies. Decreased TC and HDL-C levels in sickle cell disease have been well documented in previous studies (5, 9-13).

Studies have shown that hypocholesterolaemia is a significant predictor of death. In studying the effect of hypocholesterolaemia in a cohort of chronic hemodialysis patients, Kunitoshi and colleagues demonstrated that the 5 year survival rate was lowest in patients subgroup with serum cholesterol values of 140mg/dL (3.62mmol/L), and highest in the subgroup that had a serum cholesterol range of 200-219mg/dL (5.17-5.66mmol/L) (14).

There are several documented hypotheses explaining the hypocholesterolaemia seen in sickle cell disease. One of such hypothesis suggests that the low cholesterol level is due to utilization of cholesterol during increased erythropoiesis. In their study, Shalev et al showed that the hypocholesterolaemia seen in sickle cell disease as well as in other haemolytic anaemias was absent in non-haemolytic anaemias (6).

Increase in the exchange rate between plasma cholesterol and red blood cell membrane cholesterol may also be a causative factor in the low cholesterol levels seen in HbSS patients. It may also be that the hypocholesterolaemia in SCD may be induced by the HbS gene (15, 16). Low HDL-C levels (hypoalphalipoproteinaemia) are undesirable due to their direct relationship with coronary risk and incidence of atherosclerosis. According to The United States National Cholesterol Education Program (NCEP) Adult Treatment Panel III (ATP III) HDL-C levels < 40mg/dl (1.03mmol/L) constitute a coronary heart disease risk factor in both males and females. Hypoalphalipoproteinaemia is thought to accelerate the development of atherosclerosis due to impaired reverse cholesterol transport (17, 18). Our study recorded mean HDL-C value of 0.92$\pm$ 0.21mmol/L which is much similar to results of other studies, and much lower than results of HbSS controls used (9, 13, 19, 20). Also only 22 (38.6%) of the patients had HDL-C levels $\geq$ 1.03 mmol/L whilst the rest had values less than the minimum accepted. Not all studies have documented low HDL-C levels in SCD and possible reasons for this may include differences in age, diet, sex, weight and sample size between the studies. Available literature suggests that LDL/HDL values $\geq$3 are more likely to be associated with adverse cardiovascular events (21, 22). In this regard, only 7 (12.3%) of patients studied had a ratio $\geq$3. There are varying reports about triglyceride levels in SCD patients. While some researchers have reported elevated triglyceride levels in SCD with higher levels during crises when compared with steady state (5, 23, 24), others have reported normal levels (5). In our study, the mean serum Triglyceride level was within normal limits. Again, factors that may contribute to the discrepancies in results from different researchers may include differences in age of the study population, sex and weight. In addition, when compared to other dietary lipids, serum triglyceride level in an individual strongly depends on whether that individual is fasting or not. In a non-fasting individual, triglyceride levels will be strongly affected by the time interval between ingestion of dietary fats, as well as the type and quantity of fats consumed.

It has been shown that high TG levels are associated with vascular dysfunction and pulmonary hypertension (4). Various reports have shown an association between lipid levels and disease severity in patients with sickle cell

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anaemia (4, 25, 26). Akinlade *et al* reported that TC and LDL levels decreased progressively from controls, to sickle cell patients in steady state to sickle cell patients in vaso-occlusive crisis. Their study confirmed the widely reported defective lipid homeostasis in adults with sickle cell anaemia. They also showed that the alteration in lipid metabolism was more pronounced amongst patients in VOC compared with those in steady state (25).

In their study, Emokpae and Kuliya-Gwarzo found that SCD patients with decreased levels of HDL-C had more severe anaemia than those with normal HDL-C levels and thus concluded that the low HDL-C marker may assist in the prediction of adverse clinical events in these patients (29).

In contrast, our study failed to show significant correlation between the various lipid levels and disease severity in our cohort of patients. This may be due to the small sample size used in our study. It is not unlikely that if this study is repeated with a larger sample size, there may be a significant correlation between lipid levels and disease severity.

Even though the study failed to show significant correlation between the levels of the various lipids and disease severity, it showed the mean levels of all the serum lipids were lower compared with local laboratory reference ranges.

CONCLUSION

Our study documented low TC, LDL and HDL levels in SCD patients compared with local laboratory reference values. It however failed to show a significant correlation between lipid levels and disease severity in the patients.

Study Limitations

An important limitation of the study is the small sample size used. This would most likely lead to a type 2 error in which clinical significance does not reach statistical significance because of a small sample population.

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Conflict of interest:

The authors declare no conflict of interest

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