Assessment of the Relationship of Cd4 Count Levels with Lipid Profile and Atherogenic Index of Plasma among Newly Diagnosed HIV Patients in a Tertiary Hospital in Nigeria

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Abstract: *Lipid abnormalities have been recorded in both highly active antiretroviral therapy (HAART) ‑naïve HIV patients and those on HAART, and the presence of this dyslipidemia may lead to increased risk of cardiovascular disease (CVD). The atherogenic index of plasma (AIP) calculated as log of Tgl/HDL is currently being used as a screening tool for dyslipidaemia and is a strong predictor of CVD events. To determine the lipid profile pattern and atherogenic index of plasma among HAART ‑naïve HIV patients and relationship with CD$_4$ count levels. This was a prospective study of*
HAART‑naïve HIV patients attending the HIV clinic of the University of Benin Teaching Hospital (UBTH), Benin over a 4 month period from March to July 2011. Data generated was analysed using SPSS version 18. A total of 300 patients with significant higher mean age in males than females were recruited for the study (42.4±10.8 versus 35.8±8.4, p < 0.05). HIV- patients with CD$_4$ cells < 200/μl have significant lower age, BMI, and higher AIP with high risk category (AIP > 0.24). HDL is positively correlated with CD$_4$ cells, while TG and AIP are negatively correlated (p < 0.05). In conclusion, HIV- patients with CD$_4$ cells < 200/μl (lower immunity) have more lipid abnormalities and high risk AIP category.
Keywords: *Atherogenic index of plasma, Highly active antiretroviral therapy, HIV, Lipid profile.*

I. Introduction

Human immunodeficiency Virus (HIV) infection is pandemic worldwide [1]. HIV infection is a worldwide health problem that affects about 34 million men and women. Nigeria, the tenth most populous country in the world and the most populous country in sub-Saharan Africa, has the second highest population of people (about 3 million) living with HIV, after South Africa with about 5.6 million [2].

In the 2015 HIV/Acquired immunodeficiency syndrome (AIDS) estimates in Nigeria, the number of people living with HIV was put at 3.5 million while the prevalence rate of HIV in adults aged 15 – 49 years is 3.1% [3]. AIDS is the leading cause of death in Africa and the fourth leading cause of death worldwide [4]. However, death due to AIDS in Nigeria is 180,000 while 250,000 new HIV infections are recorded, with about 24% of adults on antiretroviral treatment [3].

 The availability of highly active antiretroviral therapy (HAART) has resulted in improved survival of HIV patients. Morbidity and mortality in these patients are therefore expected to come from organic diseases [5]. Metabolic abnormalities are often common among these HIV patients, and include dyslipidemia, diabetes, insulin resistance, altered fat distribution and endothelial dysfunction [6].

Various lipid abnormalities have been recorded in both highly active antiretroviral therapy (HAART)‑naïve HIV patients and those on HAART. Hypertriglyceridemia, low total cholesterol (TC), low high density lipoprotein (HDL-c) and reduced low density lipoprotein (LDL-c) levels have been observed in HAART‑naïve HIV patients [7] while HIV patients on HAART such as protease inhibitors and nevirapine, the lipid profile pattern showed elevated total cholesterol, triglyceride (TG), low density lipoprotein (LDL-c) and a normal or reduced HDL-c levels [8,9]. Low levels of high density lipoprotein (HDL) were also found to be common especially in those with low CD4 count [10].

In HIV infection, there is an increase in the activity of the cholesterol ester transfer protein which acts to transfer cholesterol from HDL to apo‑B‑containing proteins, hence resulting in a decrease in HDL [11].
 The ratio of triglycerides to HDL-cholesterol (Tgl/HDL-c) is said to correlate inversely with the level of small, dense LDL particles, and atherogenic index of plasma (AIP) calculated as log of Tgl/HDL is a strong predictor of CVD events [12].

A study in Nigeria showed that abnormal AIP is frequent in HAART‑naïve HIV patients and is inversely related to their level of immunity and CD$_4$ count was one of the predictor of AIP [13].

Hence, the present study aimed to evaluate the lipid profile pattern and atherogenic index of plasma among HAART‑naïve HIV patients and relationship with CD₄ count levels.

II. Materials And Methods

2.1. Ethical clearance
Approval for the study protocol was obtained from the ethics committee of the University of Benin Teaching Hospital, Benin City. Only patients who gave informed written consent were recruited.

2.2. Study setting
This was a cross – sectional study carried out in the adult HIV clinic and the medical wards of the University Of Benin Teaching Hospital (UBTH), Benin City. The UBTH is one of the major HIV treatment center in Nigeria that is largely supported by the United States presidential emergency fund for AIDS relief (PEPFAR) and receives referrals mainly from states in South – West and South – South Nigeria.

The study was conducted between March and July, 2011. Consecutive newly diagnosed HIV patients who were being enrolled into the HIV treatment program and admitted into the medical wards were recruited after meeting the study criteria and obtaining informed written consent. Patients who were on lipid lowering drugs, known hypertensive and diabetic patients and those with evidence of kidney disease were excluded. Significant history of alcohol use, cigarette smoking and exposure to ART were also part of the exclusion criteria.

Demographic parameters were obtained from the study participants using a structured pretested interviewer administered questionnaire. A history of alcohol use and smoking was also obtained. The weight and height of the patients were measured using a stadiometer with the participants wearing light clothing and without shoes. The body mass index (BMI) was then calculated using the formula: Weight (kg)/height (m2).

The blood pressure was measured with an Accoson sphygmomanometer and a standard sized cuff using the usual methods with the patient sitting quietly. The mean of two readings taken at least 5 minutes apart was recorded as the blood pressure.

2.3. Definition of variables
Dyslipidaemia is defined using the World Health Organization criteria as shown below [14]:
- Total cholesterol (TC) > 5.2mmol/l
- High density lipoprotein (HDL) cholesterol < 1.0mmol/l in females and < 0.9mmol/l in males
- Low density lipoprotein (LDL) cholesterol > 3.5mmol/l
- Triglyceride (TG) > 1.7mmol/l
  Also, the atherogenic index of plasma was classified as follows [15]:
- Low risk   -0.3 – 0.1
- Medium risk   0.1 – 0.24
- High risk   >0.24

2.4. Investigations
The HIV status of the patients was confirmed by Western blot and the CD4 cell count was measured using flow cytometry (Partec, Germany).

The blood for serum lipid profile was collected in plain bottles. Serum total cholesterol (TC) and triglyceride (TG) were determined by enzymatic estimation while high density lipoprotein cholesterol l (HDL-c) was determined by enzymatic estimation after precipitation. Low density lipoprotein cholesterol (LDL-c) was determined from the values of the aforementioned using the Friedewald's formula [16].
LDL= TCx HDL (TG/5)

2.5. Statistical analysis
Data collected was entered and analyzed using the Statistical Package for Social Sciences (SPSS) version 18.0 (Chicago, USA). Continuous variables were presented as means and standard deviation while categorical variables were recorded as frequencies and percentages. Chi‑square test was used to test for the difference between categorical variables while Student's t – test was used to compare means between two groups. One way ANOVA, Pearson's correlation coefficient and multiple linear regression were used as appropriate. $P < 0.05$ was considered to be significant.

III.   Results

Three hundred patients met the inclusion criteria of which 86 (28.7%) were males and 214 (71.3%) were females. The mean age of the patients was 37.7 ± 9.6 years. However, the mean age was significantly higher in males than in females (42.4 ± 10.8 Vs 35.8 ± 8.4, $P < 0.05$).

Table 1: Clinical Characteristics Of The Study Population

Parameters	Mean ±SD,(%)	P - value
Age Male Female	37.7 ± 9.6 42.4 ± 10.8 35.8 ± 8.4	< 0.0001
Sex Male Female	 86 (28.7) 214 (71.3)	

Table 2 showed that majority of the patients were females (79.0% Vs 21.0%), married (64.8% Vs 49.3%) and were in the 31 – 40 age category (40.8% Vs 49.3%) in both groups. Moreso, the mean age and BMI were significantly higher in HIV patients with CD_4 cells greater than 200/µl than those with CD_4 cells less than 200/µl (38.3 ± 10.2 Vs 35.8 ± 7.1, 24.4 ± 4.7 Vs 21.4 ± 3.5, $P < 0.05$).

Table 2: Socio – Demographic And Clinical Characteristics Of Patients

Variable	HIV- patients with CD_4 cells > 200/µl (n = 233) Mean ±SD, %	HIV- patients with CD_4 cells < 200/µl (n = 67) Mean ±SD, %	P – value
Age	38.3 ± 10.2	35.8 ± 7.1	0.007
Age category < 20 years 21 – 30 years 31 – 40 years 41 – 50 years 51 – 60 years > 60 years	 2 (0.9) 52 (22.3) 95 (40.8) 56 (24.0) 22 (9.4) 6 (2.6)	 1 (1.5) 18 (26.9) 33 (49.3) 14 (20.9) 1 (1.5) 0 (0.0)	 0.059
Sex Male Female	 64 (74.4) 169 (79.0)	 22 (25.6) 45 (21.0)	
Marital status Single Married Divorced Widows	 56 (69.1) 151 (64.8) 4 (1.7) 22 (9.4)	 25 (37.3) 33 (49.3) 0 (0.0) 9 (13.4)	 0.044
Body mass index (BMI)	24.4 ± 4.7	21.4 ± 3.5	< 0.0001
Waist-hip-ratio	0.89 ± 0.03	0.88 ± 0.03	0.331

In table 3, the mean HDL is significantly higher in HIV patients with CD_4 cells greater than 200/µl while the TG and AIP are significantly higher in HIV patients with CD_4 cells less than 200/µl ($P < 0.05$). Also, HIV patients with CD_4 cells less than 200/µl were more in the AIP high risk group as compared those with CD_4 cells greater than 200/µl but this was not statistically significant (61.2% Vs 46.4%, $P > 0.05$).

Table 3: Lipid Profile Pattern Based On CD_4 Count Levels

Variable	HIV- patients with CD_4 cells > 200/µl (n = 233) Mean ±SD, %	HIV- patients with CD_4 cells < 200/µl (n = 67) Mean ±SD, %	P - value
TC	3.8 ± 0.43	3.7 ± 0.56	0.196
HDL	0.9 ± 0.26	0.7 ± 0.22	< 0.0001
LDL	2.21 ± 0.37	2.15 ± 0.51	0.424
TG	1.57 ± 0.73	1.89 ± 0.86	0.006
AIP	0.19 ± 0.28	0.36 ± 0.30	< 0.0001
AIP risk Low risk Medium risk High risk	 101 (43.3) 24 (10.3) 108 (46.4)	 20 (29.9) 6 (9.0) 41 (61.2)	 0.09

Majority of the patients 149 (49.7%) and more females were in the high risk category. The mean HDL and LDL were significantly higher in the low risk category as compared to the medium and high risk categories (1.1 ± 0.23 Vs 0.9 ± 0.17 Vs 0.16, 2.3 ± 0.33 Vs 2.2 ± 0.27 Vs 2.1 ± 0.48 respectively, $P < 0.05$). However, the mean TC and TG were significantly higher in the high risk category as compared to the low and medium risk

categories (P < 0.05). The age is higher while the CD_4 count levels are lower in the high risk category compared with the other categories but not statistically significant (P > 0.05).

Table 4: Clinical And Biochemical Characteristics Of Patients According To AIP Risk

Variable	Low risk (AIP < 0.1) (n = 121)	Medium risk (AIP 0.1 – 0.24) (n = 30)	High risk (AIP > 0.244) (n = 149)	P - value
Age	36.4 ± 9.4	35.9 ± 8.1	39.1 ± 9.9	0.267
Sex Male Female	27 (22.3) 94 (77.7)	8 (26.7) 22 (73.3)	51 (34.2) 98 (65.8)	0.095
BMI	24.0 ± 4.8	24.5 ± 5.0	23.4 ± 4.3	0.152
WHR	0.89 ± 0.03	0.89 ± 0.03	0.89 ± 0.03	0.247
CD_4 count	423.9 ± 225.4	428.2 ± 269.5	356.0 ± 256.5	0.493
TC	3.8 ± 0.42	3.7 ± 0.37	3.9 ± 0.5	0.043
HDL	1.1 ± 0.23	0.9 ± 0.17	0.7 ± 0.16	0.003
LDL	2.3 ± 0.33	2.2 ± 0.27	2.1 ± 0.48	< 0.0001
TG	1.0 ± 0.26	1.3 ± 0.25	2.3 ± 0.6	< 0.0001

In the bivariate correlation analysis of CD_4 count levels with lipid profile, HDL is strongly positively correlated while TG and AIP are negatively correlated.

Table 5: Correlation Of CD_4 With Lipid Parameters

Variable	Correlation (R value)	P - value
TC	0.092	0.111
HDL	0.284[**]	< 0.0001
LDL	0.051	0.380
TG	-0.148[*]	0.010
AIP	-0.222[**]	< 0.0001

IV. Discussion

This study assessed the lipid profile and AIP in HAART – naïve HIV patients with a view of determining the relationship of these parameters with CD_4 count levels. Factors that may be associated with atherogenesis and cardiovascular disease risk in HIV infected patients include dyslipidemia either from the effect of HIV on lipid metabolism or worsen by the use of ART as lipid and AIP abnormalities have been observed in both HIV-infected patients on ARV therapy and ARV-naïve patients [13,17,18].

The study had more female participants but the mean age was significantly higher in males. Majority of the patients were in the younger age categories in both groups while the mean age and BMI were significantly lower in patients with more severity of HIV based on the CD_4 count levels.

The female preponderance in our study is similar to findings in studies on HIV patients in our environment [13, 19]. Also, the findings of younger patients in this study are similar to that from other studies [13, 18]. The mean age and BMI values in this study were similar to the values obtained in some previous studies on HAART- naïve HIV patients in Nigeria [13, 17, 18, 19].

Dyslipidaemia plays a prominent role as a risk factor for CVD, mainly characterized by increased TG, decreased HDL-c and accumulation of small dense LDL-c particles when the level of LDL-c are often normal. The mean values of TC, HDL-c and LDL-c in this study were low and these values were even lower in HIV patients with more severity of the disease based on the CD_4 count levels. These findings are similar to reports from previous study on HAART – naïve HIV patients in Eastern and Northern Nigeria [13, 19].

However, in our study the mean HDL-c value was significantly lower in HIV patients with CD_4 cell count < 200/µl. Also, the TG levels were significantly higher in HIV patients with more severity of the disease and depressed immunity and this is similar to previous reports from other studies [18, 20].

Moreso, it has been shown that increased TG tends to occur with profound immunosuppression [7]. It is postulated that in HIV, high levels of TG occur due to the presence of inflammation, resulting in elevation of interferon‑α which interferes with TG clearance [21].

A number of lipid related parameters have been used to predict the risk of coronary artery disease. According to Grover et al either the ratio of LDL-c/HDL-c or TC/HDL-c is the best related predictor of future cardiovascular events [22]. However, the log of TG/HDL-c was later shown to be a more accurate predictor of heart diseases, closely correlated with the LDL-c particle size and could serve as an indicator of the atherogenic lipoprotein phenotype [23].

The value of AIP indicates a balance between the actual concentration of plasma TG and HDL-c, which predetermine the direction of the cholesterol transport in the intravascular pool toward atherogenic LDL-c

or beneficial HDL-c [24]. Clinical studies have shown that AIP predicts cardiovascular risk and that it is an easily available risk marker and a useful measure of response to treatment [25].

In this study, the value of AIP is higher in HIV patients with lower CD$_4$ cell counts, while patients with high risk AIP had significantly low levels of HDL-c, LDL-c and high levels of TG. Also, lower CD$_4$ count is associated with higher atherogenic risk profile as shown by the high risk AIP. The findings in our study are similar to that in a previous study on HAART – naïve HIV patients in Eastern Nigeria [13].

Our study also showed that CD$_4$ count was closely related to atherogenic risk profile and index. In that, those who had lower CD$_4$ count (hence lower immunity) had a higher atherogenic risk profile. Hence, low HDL, high TG and AIP were predictive of the CD$_4$ count and this atherogenic profile can be used to identify HIV patients with high cardiovascular risk. There is a similar report from a study in Nigeria by Cajetan et al [13].

The AIP is a simple tool which can easily be used to assess atherogenic risk in patients who may have apparently normal lipid profile using other parameters.

This study is not without limitations, it was a cross – sectional single centre study and the participants were not followed up after the commencement of HAART.

Despite these limitations, our findings indicate that HAART – naïve HIV patients have variations in their lipid profiles based on the CD$_4$ count levels.

V.   Conclusion

Dyslipidemia is common in HAART‑naïve HIV patients in our environment. The AIP is more useful than single lipid parameters such as LDL-c or TC in identifying patients with atherogenic risk. In addition, reduced immunity is associated with increased atherogenicity. It is recommended that lipid assessment using lipid ratios such as the AIP should be carried out prior to initiation of therapy in patients with HIV infection.

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