ASSESSMENT OF LIPID PROFILE IN HIV INFECTED PATIENTS USING THE FIRST LINE ANTIRETROVIRAL DRUGS AT MUHIMBILI NATIONAL HOSPITAL

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By

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CERTIFICATION

The undersigned certify that he has read and hereby recommend for acceptance by Muhimbili University of Health and Allied Sciences a dissertation entitled; **ASSESSMENT OF LIPID PROFILE IN HIV INFECTED PATIENTS USING THE FIRST LINE ANTIRETROVIRAL DRUGS AT MUHIMBILI NATIONAL HOSPITAL,** in fulfillment of requirements for the degree of Master of Pharmacy (Hospital and Clinical Pharmacy) of Muhimbili University of Health and Allied Sciences.

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Prof. Appolinary Kamuhabwa

(Supervisor)

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I, **Walter Ombeni**, declare that this **dissertation** is my own original work and that it has not been presented and will not be presented to any other university for a similar or any other degree award.

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ACKNOWLEDGEMENT

DEDICATION

This dissertation is dedicated to my lovely parents, Mr. Ombeni Mnkeni and Fariji Lucas, my guardians; Mbonea Mnkeni and Marry Mnkeni who inspired me right from childhood to study hard.

ABSTRACT

Background: The use of antiretroviral therapy has been reported to cause dyslipidemia and increased risk of cardiovascular diseases (CVD) in HIV-infected patients in industrialized countries. The effects of antiretroviral drug**s** (ARVs) on lipid metabolism among sub-Saharan Africans, for whom access to antiretroviral therapy is expanding, remain largely unknown. Various factors have been found to be associated with poor lipid profiles. Therefore assessement of lipid profile as a marker for CVD in HIV-infected patients, is important in order to identify factors that are most likely to cause this problem among ARVs users. Identification of these factors is important in order to formulate appropriate interventions that will result in improved lipid profile and hence prevention of CVD among patients.

Objectives: The aim of this study was to determine the prevalence of dyslipidemia in HIV-infected patients using the first line ARVs triple regimen. The magnitude of the prevalence will help in addressing factors associated with dyslipidemia and discuss the treatment of choice for individuals experiencing dyslipidemia while using ARVs triple regimen.

Methodology: A hospital based cross - sectional study was conducted among HIV-infected patients aged 15 years and above attending Care and Treatment Clinic (CTC) at Muhimbili National Hospital (MNH). A systematic random sample of 231 patients using first line triple therapy of ARVs for at at least 24 weeks were enrolled over a period of 3 months from January 2014 to March 2014 to participate in the study. Patients data were collected from patient identification card (CTC1) & patient record form (CTC2) and interviews were done using a structured questionaire. Blood samples from patients **was** collected during their regular visits at the CTC and assayed for total cholesterol (TC), triglycerides (TG), high density lipoprotein (HDL), low density lipoprotein (LDL) cholesterol and CD4 cell counts. Data were described using means for continuous variables and proportions for categorical variables. Significance testing of proportions was carried out by using Chi-square test, where a probability (P) of less than 0.05 was considered to be statistically significant. Any factor with p- value of less or equal to 0.2 was considered for

binary logistic regression which was used to study the independent factors predicting poor lipid parameters.

Results: Majority of patients (75.9%) were females. The mean age of the patients was 42 ± 9.19 years and their median CD4 cell count was 346 cells/mm^3 (2-2600), after a median duration of 24 months (6-287) on ARVs. Proportion of patients using various first line ARVs, included 42.4%, 33.8%, 19.9% and 3.9% for zidovudine /lamivudine/efavirenz (AZT/3TC/EFV),   zidovudine/lamivudine/nevirapine   (AZT/3TC/NVP), tenofovir/emtricitabine/efavirenz (TDF/FTC/EFV) and tenofovir/lamivudine/efavirenz (TDF/3TC/EFV) respectively. The overall prevalence of dyslipidemia was 77.5%, the prevalence of hypercholesterolemia, hypertriglyceridemia, low HDLc and , incresed LDLc were 53.5%, 29.6%, 16.5%, and 59.8% respectively. Sex, age, CD4 cell count levels, body mass index (BMI) and duration of more than two years for ARVs use was significantly associated with poor lipid profiles.

Conclusion: Overall, the findings from the current study indicate that there is high prevalence of dyslipidemia among HIV-infected patients using first line ARVs attending CTC at MNH. We recommend that the National AIDS Control Program should institute mandatory laboratory monitoring of serum lipids for HIV infected patients at baseline before initiation of ARVs treatment and lipid profile monitoring during therapy.

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LIST OF ABREVIATIONS

AACE	American Association of Clinical Endocrinologists
AZT	Zidovudine
AIDS	Acquired Immunodeficiency Syndrome
ART	Antiretroviral Therapy
ARVs	Antiretroviral Drugs
ATP III	Adult Treatment Panel III
BMI	Body Mass Index
CAD	Coronary Arterial Disease
cART	combined Antiretroviral Therapy
CD4	Cluster of Differentiation 4
CDC	Center for Disease Control
CNS	Central Nervous System
CTC	Care & Treatment Clinic
CTC_1	Patient Identification Card
CTC_2	Patient Record Form
DAD	Data collection on Adverse Events of anti-HIV Drugs
DM	Diabetes Melitus
EFV	Efavirenz
EDTA	Ethylenediaminetetraacetic acid
FTC	Emtricitabine
HAART	Highly Active Antiretroviral Therapy

HDL	High Density Lipoproteins
HIV	Human Immunodeficiency Virus
IDL	Intermediate Density Lipoprotein
LDL	Low Density Lipoproteins
MUHAS	Muhimbili University of Health and Allied Sciences
NCEP	National Cholesterol Education Program
NNRTI	Non-Nucleoside Reverse Transcriptase Inhibitors
NRTI	Nucleoside Reverse Transcriptase Inhibitors
NVP	Nevirapine
PEP	Post Exposure Prophylaxis
PI	Proteases Inhibitors
RCF	Relative Centrifugal Force
SPSS	Statistical Package for Social Sciences
TC	Total Cholesterol
TG	Triglyceride
VLDL	Very Low Density Lipoprotein
WHO	World Health Organization
3TC	Lamivudine

CHAPTER ONE

1.1. INTRODUCTION

In the 1980s and 1990s, HIV infection led to a devastating burst of morbidity and mortality. However, the introduction of highly active antiretroviral therapy (HAART), a combination with at least three drugs that typically includes either a protease inhibitor (PI) or a non-nucleoside-analogue reverse-transcriptase inhibitor (NNRTI) and two nucleoside analogue reverse-transcriptase inhibitors (NRTIs) has rapidly decreased virus-linked mortality and morbidity (1–4). In developed countries, life expectancy has increased considerably as a result of antiretroviral therapy (ART), but cardiovascular disease has emerged as an important late concern (5). Several in vitro, ex vivo and in vivo experiments have since shown that some drugs from the two classes are directly toxic to adipose tissue and could act in synergy to produce complex clinical and biological alterations (3).

At the end of 2005, 40 million persons were living with HIV/AIDS, and nearly 5 million persons had become newly infected with HIV during the same year. Although access to HIV medications has been nearly universal to people in developed countries, only 1 in 7 Asians and 1 in 10 Africans who need HIV therapy were receiving HIV medications. Access has been gradually increasing in low- and middle-income countries, and leaders of the 2005 G8 Summit pledged to provide global access to HIV medications by 2010 (5-6).

Overall, 5.1% of Tanzanians age 15-49 are HIV-positive. HIV prevalence is higher among women (6.2%) than men (3.8%). HIV prevalence is higher in urban areas for both women and men than in rural areas (6). Sub-Saharan Africa is the world's most severely affected region. Though it is home to only 10% of the world's population, it shelters about two thirds of the total number of people living with HIV globally. One in 12 adults in this region is reported to be infected with HIV. Today in Tanzania as in most Sub-Saharan African countries, HIV and AIDS are recognized not only as a major public health concern, but also as a socio-economic and developmental problem (7).

Dyslipidemia is a common problem affecting HIV infected patients receiving ARVs (6,8,9). Alteration of lipid profile have been reported since the first year of HIV-epidemic and is observed in both therapy naive patients and those on treatment for a prolonged period of time. Its development involves multiple factors among which are those directly related to ARVs, inflammation, hormonal and other not well-defined genetic factors. Improved health status and health restoration may play a role in ART-related lipid disturbances (8,10).

The etiology of coronary heart disease (CHD) is multifactorial. Among other factors, high levels of low-density lipoprotein cholesterol (LDL-c) and low levels of high-density lipoprotein cholesterol(HDL- c) have been identified as risk factors for CHD in the general population (11,12). The importance of lipid disorders in patients with HIV infection is based on the increased cardiovascular risk associated with ART. Current trends suggest an increase in CVD in HIV infected patients due to aging and increased survival, so that the control of modifiable risk factors such as dyslipidemia should be a priority in the clinical managiment of HIV-infected patients (13–17).

Cardiovascular manifestations of HIV have been altered by the introduction of HAART regimens. On one hand, HAART has significantly modified the course of HIV disease, prolonged survival, and improved the quality of life of HIV-infected patients. On the other hand, the early data have raised concerns that HAART is associated with an increase in both peripheral and coronary arterial diseases (18,19). Earlier trials of lipid-lowering drugs in the primary prevention of coronary heart disease have demonstrated that lowering cholesterol levels in middle- aged men with hypercholesterolemia reduces the incidence of myocardial infarction (18,20).

Current antiretroviral drugs comprise of four classes. These include the following:
- **Nucleoside/nucleotide reverse transcriptase inhibitors**: zidovudine, lamivudine, emtricitabine, stavudine, didanosine, zalcitabine, abacavir, tenofovir.
- **Non-nucleoside reverse transcriptase inhibitors**: efavirenz, nevirapine, delavirdine.

- **Protease inhibitors**: amprenavir/fosamprenavir, atazanavir, indinavir, lopinavir/ritonavir, nelfinavir, saquinavir, tipranavir, ritonavir (not commonly used as the sole protease inhibitor in an antiretroviral regimen, but often used in low doses to "boost" the levels of other protease inhibitors).
- **Fusion inhibitors**, of which only enfuvirtide is approved by the US Food and Drug Administration (FDA) (21).

Highly active antiretroviral therapy

Highly active antiretroviral therapy (HAART) is a term used to distinguish aggressive multidrug regimens from earlier, less potent ones. Currently, antiretroviral therapy (ART) and HAART both refer to any potent combination of agents that can reduce the plasma HIV level to less than can be detected by polymerase chain reaction or b-DNA assay. These regimens most often consist of a protease inhibitor or non-nucleoside reverse transcriptase inhibitor in addition to a "backbone" of two nucleoside reverse transcriptase inhibitors (21).

Dyslipidemia, as a risk factor for CVD, is manifested by elevation or attenuation of plasma concentration of lipoproteins. Several methods have been used to classify the lipoproteins in respect to their density, physical, and chemical properties. Based on these classifications, different types of lipoproteins, including chylomicrones, IDL1, VLDL2, LDL3, and HDL4, and apolipoproteins (Apo), including Apo A, Apo B, Apo C, and Apo E, have been introduced. Generally, dyslipidemia is defined as the total cholesterol, LDL, triglycerides, apo B or Lp (a) levels above the 90th percentile or HDL and apo A levels below the 10th percentile of the general population (22).

Dyslipidemia is a primary, major risk factor for CAD and may even be a prerequisite for CAD, occurring before other major risk factors come into play. Epidemiologic data also suggest that hypercholesterolemia and perhaps coronary atherosclerosis are risk factors for ischemic stroke. Increasing evidence also points to insulin resistance, which results in increased levels of plasma triglycerides and low-density lipoprotein cholesterol (LDL-C) and a decreased concentration of high-density lipoprotein cholesterol (HDL-C)—as an important risk factor for peripheral vascular disease, stroke, and CAD (23).

American Association of Clinical Endocrinologists (AACE) recommends measurement of HDL-C as a screening test for dyslipidemia. Low HDL-C can act synergistically with other lipid risk factors to increase CAD risk. An HDL-C concentration greater than 60 mg/dl is an independent *negative* risk factor in both males and females (23). Increasing clinical evidence suggests that elevated triglycerides may be an independent risk factor for CAD; therefore, AACE recommends screening of triglycerides as a component of lipid screening. Triglycerides levels that are even moderately elevated (>150 mg/dl) may identify individuals at risk for the insulin resistance syndrome. Triglyceride levels of about 200 mg/dl or greater may indicate a substantial increase in CAD risk (23).

All adult treatment panels (ATP) reports have identified low-density lipoprotein cholesterol (LDL-C) as the primary target of cholesterol lowering therapy. Many prospective studies have shown that high serum concentrations of LDL-C are a major risk factor for coronary heart disease (CHD) (24). When serum LDL-C ranges from 160 to 189 mg/dL, introduction of a cholesterol- lowering drug is a therapeutic option in appropriate circumstances, such as when a severe risk factor is present. ATP III outlines several factors that can be taken into consideration to guide clinical judgment for this category (24).

Coronary artery disease in patients with HIV infection, once thought to be uncommon, has been less well described. Now as recent reports of myocardial infarctions in young HIV infected patients receiving antiretroviral therapy are surfacing, there is renewed interest in examining the associations between HIV infection, antiretroviral therapy, and coronary artery disease (25).

1.2 Literature review

HAART has changed the natural history of HIV infection, leading to a significant decrease in morbidity and mortality, and a notable extension of life expectancy. However, the benefits of antiretroviral combinations are tempered by a broad spectrum of side effects, including a wide range of laboratory and clinical disturbances (26). Armstrong et al performed a cross-sectional analysis in ART-naïve, non-fasting HIV-infected patients in Tanzania between November 2004 to June 2008 (27). Lipid parameters were assessed in 12,513 patients. Low HDL was prevalent in 67% and increased TG in 28%. High triglyceride and low HDL levels were associated with low CD4 counts. In this ART-naïve Tanzanian population, dyslipidemia was highly prevalent and associated with advanced disease (27).

Another study by Liu et al on First-line ART and changes in lipid levels over 3 years among HIV-Infected adults in Tanzania showed that in the first 6 months of ART, the prevalence of dyslipidemia decreased from 69% to 54%, TG decreasing from 127 mg/dL to 113 mg/dL and HDL cholesterol increasing from 39 mg/dL to 52 mg/dL (10). After 6 months, TG returned to its baseline level and increased to 139 mg/dL at 3 years; total cholesterol and low-density lipoprotein cholesterol continued to increase whereas HDL cholesterol leveled off. The prevalence of dyslipidemia increased to 73% after a 3-year follow-up period. In multivariate analyses, patients on zidovudine-containing regimens had a greater reduction in TG levels at 6 months and a lower increase at 3 years compared to patients on stavudine-containing regimens. On the other hand, patients on nevirapine-based regimens had a higher increase in HDL cholesterol levels at 3 years compared to those on efavirenz-based regimens. Their findings supported the latest World Health Organization guidelines on the substitution of stavudine in the first-line ART in resource-limited settings, and provided further evidence for selection of lipid-friendly ART for patients in sub-Saharan African countries (10).

Recently, Mgunya conducted a study in Tanzania to assess the risk factors for cardiovascular diseases among HIV infected patients. A total of 370 patients were analysed, with 69% being females. The mean age of the study subjets was 40 years with a range of 30-65 years. Seventy four percent of the patients were on HAART. The overall prevalence of dyslipidemia was 82%. The prevalence of hypertriglyceridemia, hypercholesterolemia, incresed LDL and low HDL were 36% ,42%, 53% and 50% respectively. HAART use was significantly associed with increased levels of total cholestrol, triglycerides, LDL, HTN, impared fasting blood glucose and DM (15).

Buchacz and colleagues conducted another study in Uganda to assess changes in lipid profile over 24 months among 987 adults HIV-infected patients on first-line HAART.   It was found that during 24 months of treatment, TC increased by a mean of 31 mg/dL, LDL-c by a mean of 26 mg/dL, and HDL-c by a mean of 19 mg/dL, whereas the TC/HDL-c ratio decreased from a mean of 4.6 to 3.4. TG levels initially decreased and then returned to baseline levels by 24 months. At baseline and 24 months, respectively, TC was ≥200 mg/dL for 2% and 10% of patients, LDL-c was ≥130 mg/dL for 1% and 6%, HDL-c was <40 mg/dL for 88% and 41%, and TG were ≥150 mg/dL for 23% and 20%. The study concluded that nevirapine- or efavirenz-based HAART experienced infrequent elevations in TC, LDL-c, and TG at baseline and after 24 months of therapy. Increases in HDL-c levels were substantial and proportionally greater than increases in TC or LDL-c levels. Thus the changes observed after 24 months of HAART seem unlikely to increase the risk of CVD (28).

A study conducted in Zambia to assess cardiometabolic risk factors among HIV patients on ART reported that from the baseline to 90 days after initiation of cART, the prevalence of low levels of high-density lipoprotein cholesterol significantly decreased (78.8% vs. 34.8%), total cholesterol was elevated (5.1% vs. 11.9%). The prevalence of TC:HDL-c ratio significantly decreased (44.9% vs. 6.8%). These changes in cardiometabolic risk markers were independent of the cART regimen. It was therefore concluded that short-term cART is associated with a cardioprotective lipid profile and a tendency towards insulin resistance regardless of the cART regimen (29).

A multination study by Nina et al was conducted in Europe, USA and Australia to determine the prevalence of risk factors for CVD among HIV-infected individuals, and to investigate any association between such risk factors, stage of HIV disease, and use of antiretroviral therapies (30). The results of this study showed increased prevalence of elevated total cholesterol (≥6.2mmol/l) among subjects receiving an NNRTI compared to the prevalence among ART-naïve subjects. Subjects who had discontinued ART as well as subjects receiving NRTIs had similar cholesterol levels to treatment-naïve subjects. Higher CD4 cell count, lower plasma HIV RNA levels, clinical signs of lipodystrophy, longer exposure times to NNRTI and PI, and older age were all associated with elevated total cholesterol levels (30). This study concluded that HIV-infected persons exhibit multiple known risk factors for CVD. Of specific concern is the fact that the use of NNRTI and PI classes of drugs (alone and especially in combination), particularly among older subjects with normalized CD4 cell counts and suppressed HIV replication, was associated with a lipid profile known to increase the risk of coronary heart disease (30).

A study conducted by Marc et al covering countries across Europe and America involving HIV-infected patients on nevirapine-containing antiretroviral therapy reported a striking increase in HDL-cholesterol (49%), apolipoprotein AI (19%), lipoprotein AI (38%) and HDL particle size (3%) in the nevirapine-treated patients at week 24 (31). Much less pronounced changes in these parameters were seen to a similar extent both in patients receiving lamivudine and indinavir. LDL-cholesterol also increased significantly both in the nevirapine and indinavir arms, but only in the nevirapine where the increase in LDL-cholesterol was offset by a significant reduction (14%) in total over HDL-cholesterol ratio. It was concluded that in HIV-1 infected patients treated with a regimen of stavudine, didanosine and nevirapine, changes in lipids and lipoproteins are associated with a sharp decrease in risk for CAD in other settings (31).

From the findings of the different studies that have been conducted in different parts of the world, it can be concluded that the prevalence of dyslipidemia in HIV patients using first line ARVs varies from one study to another and from one country to another. For the case of Tanzania, there **were** no sufficient published data in this area **and this** necessitates further studies.

1.3 Problem statement

Combination antiretroviral therapy has had a dramatic effect in reducing morbidity and mortality associated with human immunodeficiency virus infection. However, concern has been raised regarding the effect of such therapy on the risk of coronary heart disease. Previous findings from the Data Collection on Adverse Events of Anti-HIV Drugs (DAD) study indicated that the incidence of myocardial infarction increased with longer exposure to combination antiretroviral therapy (30).

HIV has been associated with dyslipidemia independent of antiretroviral therapy. Grunfeld et al found that HIV infection was associated with elevated triglyceride levels that worsened with progression of HIV-related disease (32). Antiretroviral therapy can also contribute to dyslipidemia. Dyslipidemia has been described as being more common and more severe in HIV patients receiving antiretroviral therapy than in patients not on therapy (13). The severity of the dyslipidemia and the typical pattern of the lipid profile differ among and within the classes of antiretroviral agents (11). Several studies have concluded that dyslipidemia is a high risk for CVD in HIV infected patient receiving antiretroviral therapy (22).

Although previous studies have reported a relationship between the use of protease inhibitors and CVD primarily due to dyslipidemia (26), there are few data on the risk for CVD associated with nucleoside and non- nucleoside reverse-transcriptase inhibitors (the first line combination) in Tanzania.

A study by Mgunya at Muhimbili National Hospital found that use of ARVs was significantly associated with increased levels of total cholestrol, triglycerides, LDL, HTN, impared FBG and DM. However, this study included patients who were on both the first line and second line ARVs regimen. In this study, we assessed patients who were using the first line regimen alone. Also in the study by Mgunya only patients aged ≥ 30 years were evaluated. Therefore there are no data of lipid profiles for patients of younger age (<30 years) who are using the first line ARVs in Tanzania.

1.4 Rationale of the study

This study was done to identify factors associated with dyslipidemia in HIV infected patients on first line ART. The risk factors for CVD have cumulative and interacting effects. While some risk factors such as male sex and family history are not modifiable, others including unhealthy diet, smoking, hypertension, dyslipidemia, diabetes, and physical inactivity can be modified. Aggressive treatment of dyslipidemia is key to managing and preventing CVD in patients with or without HIV infection.

Therefore this study has provided data on the lipid profile changes and its association to a particular ARV combination regimen. It is expected that, this information will be used to guide clinicians for routine assessement of HIV-infected patients using ARVs.

The findings of this study could form a base for discussion with regard to the choice of ARVs to be initiated among individual patients with co-existing CVD or elevated levels of TC, TG LDLc and low HDLc. This is of particular importance because pharmacological management of HIV-infected patients with dyslipidemia is particularly problematic due to potential for drug-drug interactions between ARVs and antidyslipidemia drugs (2).

1.5 Research questions

i. What is the prevalence of dyslipidemia among HIV-infected patients on first line ARVs?

ii. Are different antiretroviral drugs associated with different lipid profiles?

iii. What are the patients host factors that play a major role in development of dyslipidemia?

1.6 Research Objectives

1.6.1 Broad Objective:

To assess the lipid profile in HIV-infected patients on first line antiretroviral drugs

1.6.2 Specific objectives

i. To determine the prevalence of dyslipidemia by assesing the lipid profile in HIV-infected patients on the first line antiretroviral drugs.

ii. To asses the typical pattern of the lipid profile among and within the classes of antiretroviral drugs.

iii. To evaluate if the prevalence of dyslipidemia that has been found in adults (30yrs and above) can be comfirmed in young population.

iv. To asses host factors that play a major role in development of dyslipidemia.

CHAPTER TWO

2. RESEARCH METHODOLOGY

2.1. Study design

This study was cross sectional study through systematic sampling of HIV-infected patients at the CTC of the Muhimbili National Hospital (MNH) in Dar es Salaam. Data were obtained using questionnaire to assess patient's socio-demographic and clinical characteristics. Moreover data collection including a recent CD4 cell count, type of ARVs regimen used and duration on ARVs use were extracted from patients CTC_1 & CTC_2 cards. Blood samples were collected after interviewing the patients. Lipid profile was determined from blood samples collected from patients and assayed in MNH laboratory.

2.2. Description of the study area

The study was conducted at the CTC of the MNH in Dar es Salaam. MNH is the Tanzania's largest public and tertiary level referral hospital. It receives referred patients from district hospitals and from neighboring regions. It is located in the city of Dar es salaam which is also the largest commercial city in the country with a population of about 2.48 million. CTC at MNH, receives patients referred from district hospitals HIV clinics and those referred from the inpatient department of MNH. The MNH refferals are mainly those patients who are diagnosed to have HIV while admitted in the wards. The patients attend the clinic once in a month for clinical evaluation and prescription refill for ARVs. Currently there are about 6,500 HIV-infected outpatients receiving ART at the CTC. On average there are about 60 HIV infected patients attending CTC at MNH for ARVs prescription refill and clinical assessment per day.

2.3. Study population

The study population in this study included HIV infected patients who were on ARVs for ≥24 weeks. The reason for selecting this time for duration of ARV use is due to the fact that by this time the effect of ARVs in changing serum lipids has been established (31). The target patients were those at the age of 15 years and above attending CTC at MNH who are on ARVs triple combination therapy for ≥ 24 weeks. These patients were enrolled into the study after completing a written informed consent.

2.4. Study participants

2.4.1. Inclusion Criteria

   i. Age > 15 years

   ii. Known HIV-infection on first line ARVs triple combination therapy for at least 24 weeks

  iii. Outpatients

  iv. Not pregnant at the time of enrollment

2.4.2. Exclusion Criteria

   i. Critically ill patients

   ii. Patients receiving anabolic steroids, corticosteroids, or immune-modulating therapy

  iii. Patient receiving anti dyslipidemic drugs

  iv. Patients on second line ARVs regimen

2.5. Study period:

This study was conducted for three months from the beginning of January 2014 to end of March 2014.

2.6. Sample size calculation

The sample size for this study was determined based on the following formula:

$$n = \frac{Z^2 \, p(1-p)}{e^2}$$

n = sample size

Z = Value of constant at 95% confidence interval =1.96

P = Prevalence of dyslipidemia, which is based on a similar study that was conducted at MNH in which the prevalence of dyslipidemia among adult patients (> 30 years) using both first and second line ARVs was reported to be 82% (15). E = Level of error allowed = 5% = 0.05

Thus n = 1.96^2 x 0.82x0.18/0.05^2 = 226.8

Therefore a total of 230 patients were recruited into the study.

2.7. Sampling technique and study procedure:

Systematic sampling was used to recruit study participants using daily attendance at the CTC as sampling frame. The first patient was selected randomly by a physician after consultation and the consecutive participants were selected after every third patient until the end of that day. Patients were asked to participate in the study after they **had** been attended by the physician. They were approached and explained the purpose of the study while they were going to the pharmacy for prescription refill. Those who agreed to participate in the study were required to sign a consent form (Appendix III and IV). Those fulfilling the inclusion criteria were enrolled in the study until the sample size for the study was reached. In case a patient did not consent for the study, then the next third patient in a row was recruited.

2.8 Data collection and instruments

Data collected included sociodemographic characteristics, clinical data (AIDS events and known risk factors for CHD), laboratory markers (CD4 cell counts, LDLc, TC, HDLc, and TG levels), and treatment variables (antiretroviral treatment). The history of antiretroviral treatment and HIV infection were recorded from CTC1 and CTC2 cards.

2.8.1 Blood sample and assay

Cotton wool, methylated spirit, syringe, gloves and vacuntainer tubes were used for blood sample collection. Blood samples were collected from the study participants, coded, labeled and taken to MNH laboratory for assay. For analysis of TC, TG, HDL and LDL levels, blood samples were collected from patients into collecting tubes (serum and plasma tubes) and then frozen. The frozen specimens were then centrifuged at 10,000 RCF for ten minutes. The centrifuged specimens with lipid layer on top were then transferred into secondary tubes in the architect machine for lipid profiling (33). CD_4 cell count (appendix IC) was assayed for patients whose CD4 cell counts status for the past six months were not known. Blood samples were drawn from the patient into vacuntainer with EDTA (anticoagulant). CD4 cell counts were counted by using FACScalibur machine. This was done by incubating anti-coagulated whole blood containing monoclonal antibody (34). For those with known CD4 cell count within six months, the CD4 cell counts were recorded from CTC_1 or CTC_2 cards.

2.8.2 Demographic information

Data on socio-demographic and background information was collected by using a questionnaire (Appendix IA) and checklist (Appendix IB). This included age of the patient, sex, height, weight and Body mass index (BMI). Weight was measured by using a seca scale with the patient having no shoes. Weight was measured up to the nearest 100 grams. Height was measured using a standard height board with the participant having no shoes. The head piece was gradually lowered until it reached the patients head and was at a 90° angle with the measuring scale. The measurement was then taken to the nearest 1 cm. BMI was calculated as weight in Kg divided by height in meter square.

2.8.3 Study variables

i. Dependent variables for this study were serum TG, TC, HDL and LDL.

ii. Independent variables included age, sex, CD4 cell count, ARVs regimen, duration of ARVs use, and BMI.

2.8.4 Operational definitions

Serum lipids concentration were graded according to the criteria established by the American Association of Clinical Endocrinologists (AACE) (23), as shown in table 1.

Table 1: Classification of serum lipid levels as a risk for CVD according to American Association of Clinical Endocrinologists

Optimal/Near-Optimal, Borderline, and High-Risk Serum Lipid Concentrations			
Lipid (mmol/dl)	**Optimal/near-optimal serum concentration**	**Borderline serum concentration**	**High-risk/very high-risk serum concentration**
TC	<5.17	5.17-6.18	>6.21
HDLc	>1.04 (negative risk factor)	1.03-1.52 (men) 1.29-1.52 (women)	<1.03 men <1.29 women
LDLc	<3.3	3.36-4.11	4.14-4.89 high >4.91 very high
TG	<1.69	1.69-2.25	2.26-5.63 high >5.65 very high

TC= Total cholesterol, TG = triglyceride, HDLc = High density Lipoprotein cholesterol, LDLc = Low density Lipoprotein cholesterol

Dyslipidemia: *Atherogenic dyslipidemia* manifests in routine lipoprotein analysis and is defined as the presence of any of the following lipid abnomalities: hypercholestrolemia, hypertriglyceridemia, low HDLc, or increased LDLc. All of these abnormalities have been implicated as being independently atherogenic (23,35).

i. **Hypercholesterolemia** is defined as total serum cholesterol of more than 5.17mmo/l.

ii. **Hypertrigyceridemia** is defined as serum triglyceride of more than 1.69 mmo/l

iii. **Decreased HDLc** is defined as serum HDL levels of <1.04mmol/l.

iv.   **Increased LDLc** is taken as serum LDL levels of >3.3mmol/l (15,23).

BMI measures relative weight for height. BMI was calculated by dividing weight by height squared (kg/m^2). BMI is linearly related to the total cholesterol, LDL cholesterol and triglyceride concentrations and is, however, inversely related with HDL cholesterol. Dyslipidemia has been recognized to be strongly associated with overweight and obesity and its co morbid conditions. Overweight and obesity is conveniently determined from BMI (36).

Table 2: Categorization of body mass index

Category	Range (Kg/m^2)
Underweight	BMI < 18.5
Normal	BMI 18.5 – 25
Overweight	BMI 25 – 29
Obese	BMI $\geq$ 30

2.9 Statistical analysis

The outcomes of interest included the lipid parameters: TC, TG, HDLc and LDLc. In accordance with the United States National Cholesterol Education Program, Adult Treatment Panel III (NCEP-ATP III) guidelines, abnormal lipid profile was defined as TC > 5.17 mmol/l, HDLc < 1.04 mmol/l, LDLc > 3.3 mmol/l and TG > 1.69 mmol/l. Predictor risk factors for dyslipidaemia included: age, sex, CD4 cell count, ART regimen, duration of ART, and presence of comorbidity. Summary statistics were presented as proportions for categorical variables and as means (standard deviations) or medians (IQR: Inter-quartile Range) for continuous variables. Pearson Chi-square analyses were used to examine the differences in proportion of abnormal lipid parameters between the various categories of an explanatory variable. The relationships between the dependent variables (TC, HDLc, LDLc, and TG levels) and treatment group were studied by use of multiple logistic regression models. Associations were considered to be statistically significant at a

P value of ≤ 0 .05. All statistical analyses were performed by use of SPSS (Statistical Package for Social Sciences) version 20.

2.11. Ethical Considerations

Ethical clearance was sought from Muhimbili University of Health and Allied Sciences Senate, Research and Publications Committee. Permission to collect data was also sought from Muhimbili National Hospital management. Written informed consent was obtained from patients (Appendix IIa and IIb). Prior to data collection the CTC administration was visited to explain the purpose of the study. In addition, explanation was given to the participants on the objectives of the study. Confidentiality was observed (all the information provided was kept confidential) and no unauthorized persons had access to the data collected. Each study participant was assigned a study identification number, and these identifiers were not released outside the research group. Codes were used and no identification was made for the responders. All data collectors were asked to observe confidentiality for any patient information they receive.

CHAPTER THREE

3. RESULTS

3.1. Patients Characteristics

A total of 260 consecutive HIV-positive patients were screened, but 29 (11.2%) patients were excluded from the study. Out of those excluded, 4 patients declined consent, 15 failed to turn up for blood sample collection, 5 blood sample were unsuitable for analysis, and 5 ARVs treated patients had been using more than one regimen within 6 months. Thus the study included 231 patients receiving ART and returning to the clinic between January 2014 and March 2014 for their routine prescription refill and monitoring.

The socio-demographic characteristics of the study participants are summarised in (Table 3). Majority of patients (76.2%) were females. The mean age of the patients was 42 ± 9.19 years, ranging from 20 to 70 years. The mean body mass index (BMI) was 26 ± 5.8 kg/m^2 and the median CD4 lymphocyte count was 364 (2-2600) cells/mm^3.

Table 3: Socio-demographic characteristics of study participants attending Muhimbili National Hospital Care and Treatment Clinic (n=231)

Group Characteristic	Number	Percentage (%)
Sex		
Female	176	76.2
Male	55	23.8
Age group (years)		
15-30	22	9.5
31-40	84	36.4
41- 50	83	35.9
50 -70	42	18.2

The duration from the date of initiation of ART in study subjects ranged from six months to 287 months with a median of 24 months. First-line ARV regimens were a combinations of two nucleoside reverse transcriptase inhibitors (NRTIs) and one non nucleoside reverse transcriptase inhibitors (NNRTIs). The NRTIs consisted of zidovudine (AZT) and tenofovir (TDF), with lamivudine (3TC) constantly present in three of the first line triple combination antiretroviral regimens. AZT was the NRTI backbone in 176 (76.2%) cases while efavirenz (EFV) was used by 153 (66.2%) patients as the preferred non nucleoside reverse transcriptase inhibitor (NNRTI). Ninty eight patients (42.4%) were using a combination of zidovudine, lamivudine and efavirenz (AZT+3TC+EFV), while seventy eight patients (33.8%) were using zidovudine, lamivudine and nevirapine (AZT+3TC+NVP). The remaining fifty five patients (23.8%) were using TDF based regimens with 46 (19.9%) patients on tenofovir, emtricitabine and efavirenz (TDF+FTC+EFV), and only nine patients (3.9%) were using tenofovir, lamivudine and efavirenz (TDF+3TC+EFV). Propotion of patients on different ART regimen are summarised in table 4.

Table 4: Clinical characteristics of study participants (n=231)

Clinical Characteristic		Number	Percentage
Recent CD4 cell count (cells/mm^3)			
<350		112	48.5
350-500		45	19.5
>500		73	32
Body Mass Index (BMI) (kg/m^2)			
Underweight		20	8.7
Normal		95	41.1
Overweight		56	24.2
Obese		60	26
Patient's ARVs combination			
AZT+3TC+EFV		98	42.4
AZT+3TC+NVP		78	33.8
TDF+FTC+EFV		46	19.9
TDF+3TC+EFV		9	3.9
Duration of ARVs use (months)			
<12		36	15.7
12-24		81	35.2
>24		113	49.1
Lipid Subgroup			
TC	Normal	107	46.5
	Hypercholesterolemia	123	53.5
TG	Normal	162	70.4
	Hypertriglyceridemia	68	29.6
HDLc	Decreased HDLc	38	16.5
	Normal	192	83.5
LDLc	Normal	92	40.2
	Increased LDLc	137	59.8

ARVs = antiretroviral drugs, AZT = zidovudine, 3TC = lamivudine, TDF = tenofovir, EFV = efavirenz, NVP = nevirapine, TC= Total cholesterol, TG = Triglycerides, HDLc = High density lipoprotein cholesterol, LDLc = low density Lipoprotein cholesterol

3.2. Lipid profile of study participants

The mean serum concentration of total cholesterol was 5.34 ± 5.34 mmol/l with prevalence of hypercholesterolemia (>5.17mmol/l) of 53.5%. While majority (70.4%) of the patients had normal serum concentration of triglyceride (0-1.69mmol/l) with the median serum concentration of 1.2 mmol/l (0.44-6.61), (mean 1.5 ± 0.95), majority of the patients (59.8%) had elevated LDLc level (>3.3mmol/l) with the mean serum LDLc concentration of 3.63 ± 1.08 mmol/l. The proportion of patients with low HDLc (<1.04mmol/l) was 16.5% with mean serum concentration of 1.46 ± 0.48 mmol/l (Table 5).

Table 5: Lipid profile of the study participants (n=231)

Lipid Parameters	Serum concentration (mmol/l)
Total cholesterol	5.34 ± 5.34
Triglycerides, mediam (IQR)	1.2 (0.44 – 6.61)
HDLc	1.46 ± 0.48
LDLc	3.63 ± 1.08

Data are mean values (± standard deviation) unless otherwise indicated; IQR, interquartile range.

3.3.1. Prevalence of dyslipidemia among patients

Figure 1 shows lipid profiles of HIV-infected patients who were using ARVs. Dyslipidemia was detected in 179 patients (77.5%). Increased LDLc was the most prevalent (59.8%) with no significant differences between males and females (p= 0.293). The prevalence of hypercholesterolemia, hypertriglyceridemia, low HDL and incresed LDL were 53.5%, 29.6%, 16.5% and 59.8% respectively.
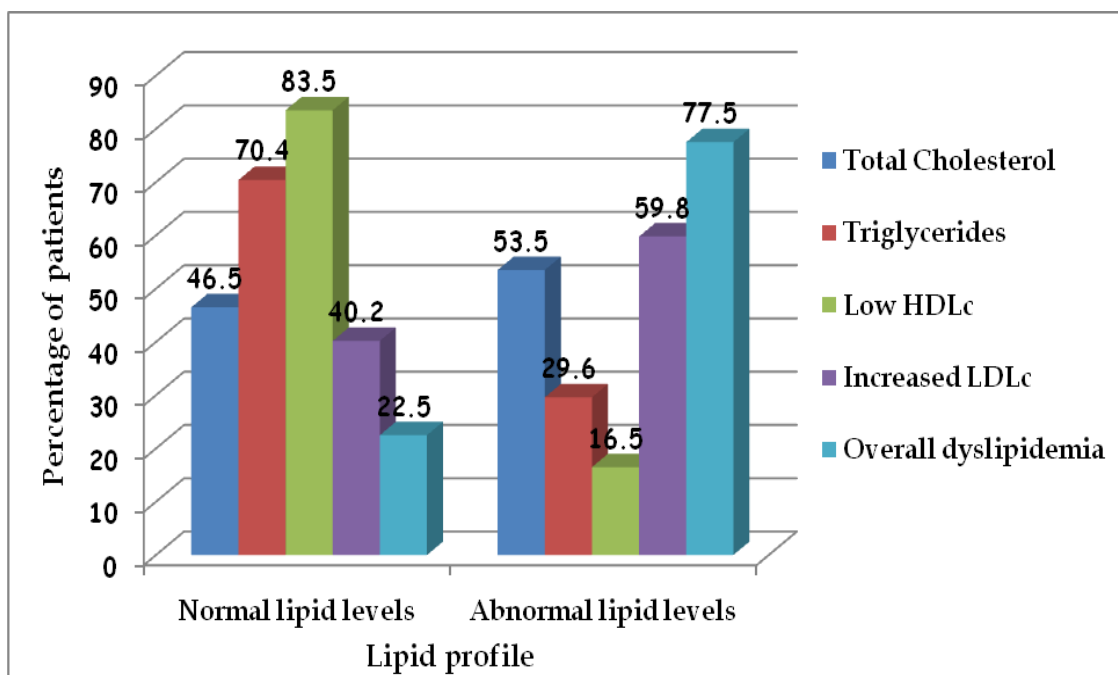
Figure 1: Prevalence of dyslipidemia in HIV-infected patients using ARVs.

3.4. Association between patients characteristics and dyslipidemia

3.4.1. Prevalence of dyslipidemia according to the type of ARVs regimen:

There were four first line triple ARVs combinations, which included (AZT+3TC+EFV), (AZT+3TC+NVP), (TDF+FTC+EFV) and (TDF+3TC+EFV). The prevalence of hypercholesterolemia, hypertriglyceridemia, and low HDLc were higher and fairly similar between (AZT+3TC+EFV) and (AZT+3TC+NVP) regimens, but lower in the (TDF+FTC+EFV) and (TDF+3TC+EFV) regimens. However the prevalence of increased LDLc was high and fairly similar among all four regimens (60.8%, 59%, 60% and 55.6% for (AZT+3TC+EFV), (AZT+3TC+NVP), (TDF+FTC+EFV) and (TDF+3TC+EFV) respectively) as shown table 6. In all four regimen the prevalence of dyslipidemia was not statistically different (Table 6).

Table 6: Prevalence of dyslipidemia according to ART regimen used by patients (n=231)

ART regimen	Lipid Abdomalities (mmol/l)			
	TC > 5.17 n(%)	TG > 1.69 n(%)	HDLc < 1.03 n(%)	LDLc > 3.3 n(%)
AZT+3TC+EFV	53(54.1)	30(30.6)	16(16.5)	59(60.8)
AZT+3TC+NVP	44(56.4)	27(34.6)	14(17.9)	46(59)
TDF+FTC+EFV	22(47.8)	11(23.9)	7(15.2)	27(60)
TDF+3TC+EFV	4(50)	0	1(11.1)	5(55.6)
P-value	0.824	0.20	0.948	0.980
NRTI based regimen				
AZT containing regimen	97(55.1)	57(32.4)	30(17.1)	105(60)
TDF containing regimen	26(48.1)	11(20.40	8(14.5)	32(59.3)
P-value	0.369	0.091	0.651	0.923
NNRTI based regimen				
EFV containing regimen	79(52)	41(27)	24(15.8)	91(60.3)
NVP containing regimen	44(56.4)	27(34.6)	14(17.9)	46(59)
P-value	0.523	0.229	0.676	0.850

ART = antiretroviral therapy, NRTI = nucleotide reverse transcriptase inhibitors, NNRTI = non-nucleotide reverse transcriptase inhibitor, AZT = zidovudine, 3TC = lamivudine, TDF = tenofovir, EFV = efavirenz, NVP = nevirapine

Figure 2 shows the prevalence of lipid abnormalities according to ART regimens. ART therapy was further classified into NRTI and NNRTI so as to asses the effect of individual drug within the regimen. Based on the classification of the major groups of ARVs, four subgroup were made which included two NRTI and two NNRTI based regimen. The two NRTI based regimen included zidovudine containing regimen and tenofovir containing regimen while the two NNRTI included efavirenz and nevirapine. The lipid derengement were slightly higher in the zidovudine containing regimen as compared to tenofovir containing regimen (hypercholesterolemia (55.1% Vs 48.1%), hypertriglyceridemia (32.4% Vs 20.4%), low HDLc (17.1% Vs 14.5%), increased LDLc (60% Vs 59.3%) respectively). In the NNRTI the lipid derengement were generally higher in the nevirapine containing regimen as compared to efavirenz containing regimen except for increased LDLc (hypercholesterolemia (56.4% Vs 52%), hypertrigyceridemia (34.6% Vs 27%), low HDLc (17.9% Vs 15.8%), increased LDLc (59% Vs 60.3%) respectively). However, there was no significant differences in the prevalence of lipid abnormalities in patients using ARVs regimens that included zidovudine as compared with those on regimens that included tenofovir. The prevalence of lipid abnormalities induced by nevirapine and efavirenz was also similar in patients on ART (Table 6).
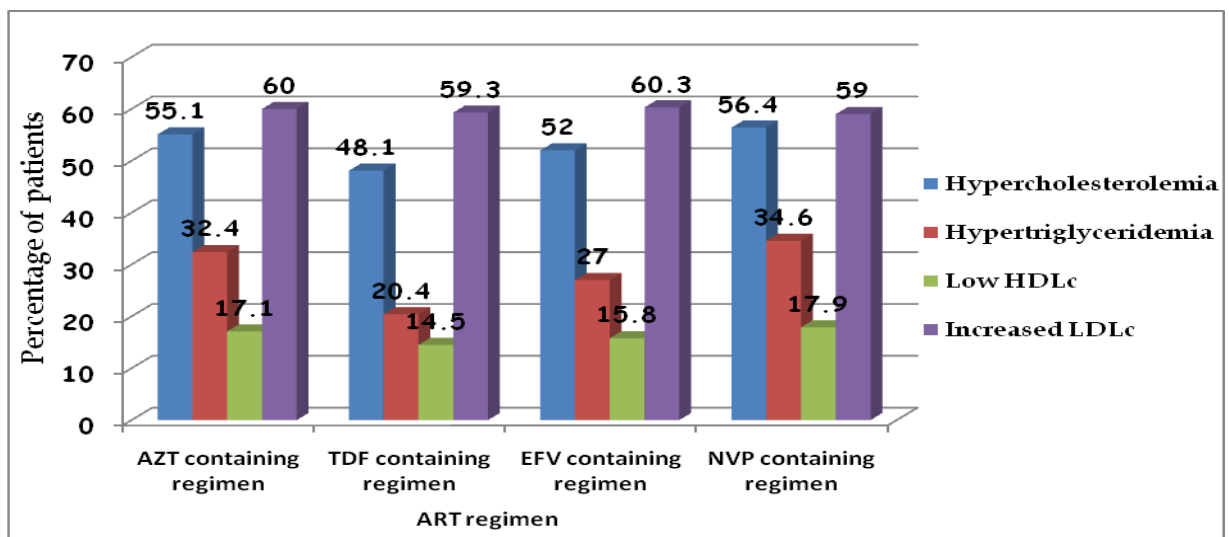
Figure 2: Prevalence of dyslipidemia according to the type of ART regimen used by patients (n=231)

3.4.2. Prevalence of Dyslipidemia with respect to the age of patients:

The prevalence of hypercholesterolemia, hypertriglyceridemia and increased LDLc was increasing with increasing age. Across all age groups, hypercholesterolemia and increased LDLc were most prevalent as compared to hypertriglyceridemia and low HDLc. The trend of the prevalence of dyslipidemia towards increasing age varied with the specific lipids. The prevalence of hypercholesterolemia was 31.6%, 51.2%, 48%, and 78.6% for the age groups of 15-30, 31-40, 41-50 and 51-70 years, respectively. Hypertriglyceridemia showed the prevalence of 21.1%, 22.1%, 26.5% and 54.8% respectively for the age groups of 15-30, 31-40, 41-50 and 51-70 years. The prevalence of increased LDLc were 47.4%, 55.8%, 55.4% and 81% for the age groups of 15-30, 31-40, 41-50 and 51-70 years. The differences in prevalence of dyslipidemia between the age groups were statistically significant with the p-value of 0.001 for hypercholesterolemia, 0.001 for hypertriglyceridemia and 0.017 for increased LDLc as shown in table 7. On the other hand, the prevalence of low HDLc did not show increasing trend towards increasing age and the differences between the age groups were not statistically significant (P = 0.316).

Table 7: Prevalence of dyslipidemia with respect to age of the patients (n=231)

Lipid parameters (mmol/l)	Age of the patients (years)				P value
	15-30 n(%)	31-40 n(%)	41-50 n(%)	51-70 n(%)	
Hypercholesterolemia	6(31.6)	44(51.2)	40(48.2)	33(78.6)	**0.001**
Hypertriglyceridemia	4(21.1)	19(22.1)	22(26.5)	23(54.8)	**0.001**
Low HDLc	4(21.1)	10(11.6)	18(21.6)	6(14.3)	0.316
Increased LDLc	9(47.4)	48(55.8)	46(55.4)	34(81)	**0.017**

Age groups were also classified into two groups, either ≤ 30 or > 30 years. This was done in order to asses whether the prevalence of dyslipidemia that has been confirmed in the age group of > 30 years (15), can also be confirmed in youg patients (≤ 30yrs). There was a statistically non-significant trend towards an increase in prevalence of dyslipidemia with

age of the study participants. Patients in the age groups of ≤ 30 and > 30 years had an overall prevalence of dyslipidemia of 63.6% and 76.1%, respectively (P= 0.201). The prevalence of hypercholesterolemia also showed a trend towards increasing with age, with corresponding prevalence of 31.8% for patients in the age group of ≤ 30 years and 55.8% in those who were > 30 years. The increase in the prevalence of hypercholesterolemia with age was statistically significant (P = 0.032). On the other hand, the prevalence of hypertriglyceridemia and elevated LDLc both showed similar trends as hypercholesterolemia, but there were no statistical significant differences (P = 0.219 and 0.148 respectively) between the two age groups. Hypertriglyceridemia showed prevalence of 18.2% for patients with the age of ≤ 30 years and 30.2% for patient who were > 30 years of age (P=0.219). On the other hand, elevated LDLc showed prevalence of 45.5% for patients with age of ≤ 30 years and 61.4% for patients who were >30 years of age (P = 0.148). However, the prevalence of decreased HDLc showed an inverse relationship with age as it decreased with increasing age, although there was no statistical significant difference (P = 0.410) as shown in table 8.

Table 8: Comparison of the prevalence of dyslipidemia among young patients (≤ 30 years) and older patients (> 30 years) (n=231)

Lipid abnormalities	Age group in years		P-value
	≤30 (n =)	>30 (n =)	
Overall prevalence	15(68.1)	164(77.5)	0.263
Hypercholesterolemia	7(31.8)	116(55.8)	**0.032**
Hypertriglyceridemia	4(18.2)	64(30.8)	0.219
Decreased HDLc	5(22.7)	33(15.9)	0.410
Elevated LDLc	10(45.5)	127(61.4)	0.148

3.4.3. Prevalence of dyslipidemia in relationship to the sex of the patients:

The prevalence of hypercholesterolemia and increased LDLc were higher in females (55.4% vs 61.5% respectively) than in males (47.3% Vs 54.5% respectively). On the other hand, the prevalence of hypertriglyceridemia and low HDLc were higher in male patients (34.5% Vs 25.5% respectively) compared to females (28% Vs 13.7%, respectively). In general, male patients had higher prevalence of dyslipidemia when compared to females. The prevalence of decreased HDLc were significantly higher among males as compared to females (P = 0.041). The differences in the prevalence of hypercholesterolemia, hypertriglyceridemia and increased LDLc between males and females were not statistically significant (Table 9).

Table 9: Prevalence of dyslipidemia in relation to the sex of patients (n = 231).

Lipid parameters	Sex of the patients		P-value
	Female n(%)	Male n(%)	
Hypercholesterolemia	97(55.4)	26(47.3)	0.27
Hypertriglyceridemia	49(28)	19(34.5)	0.353
Low HDLc	24(13.7)	14(25.5)	**0.041**
Increased LDLc	107(61.5)	30(54.5)	0.385
Overall dyslipidemia	135(76.7)	44(80)	0.389

3.4.4. Prevalence of Dyslipidemia in relationship to CD4 cell count of patients:

Majority of the patients (48.5%) had CD4 cell count of <350 cells/mm^3. The prevalence of hypercholesterolemia (64.9%), hypertriglyceridemia (44.5%), decreased HDLc (20.3%) and elevated LDLc (62.5%) were all higher in patients with CD4 cell count of >500cells/mm^3 as compared to the prevalence of dyslipidemia in patients with CD4 cell count of <350 cells/mm^3 and 350-500 cells/mm^3. The observed differences in the prevalence of hypercholesterolemia, low HDLc and increased LDLc across CD4 cell counts of <350 cells/mm^3, 350-500 cells/mm^3 and >500 cells/mm^3 **were** not statistically

significant (P = 0.047, 0.548 and 0.278, respectively). However, the prevalence of hypertriglyceridemia was significantly higher in patients with CD4 cell count of >500 cells/mm^3 (P= 0.003) than in the other categiries of CD4 cell counts (Table 10).

Table 10: Prevalence of dyslipidemia in relationship to CD4 cell counts of patients (n=231)

Lipid abnormalities	CD4 cell counts (cells/mm^3)			P-value
	< 350 n(%)	350-500 n(%)	>500 n(%)	
Hypercholesterolemia	52(46.4)	23(52.3)	48(64.9)	0.047
Hypertriglyceridemia	26(23.2)	9 (20.5)	33(44.5)	**0.003**
Low HDLc	17(15.3)	6(13.3)	15(20.3)	0.548
Increased LDLc	57(51.8)	28(62.2)	45(62.5)	0.274

3.4.5. Prevalence of Dyslipidemia in relationship to Body Mass Index of patients:

The prevalence of hypercholesterolemia, hypertriglyceridemia, and increased LDLc showed increasing trend towards obese patients. Poor lipid profiles were much higher in obese patients as compared to patients who were overweight, normal and underweight. Unlike other lipid abnormalities, the prevalence of low HDLc did not show increasing trend towards obese patients. Patients with normal body mass index had higher prevalence (17%) of low HDLc as compared to underweight (15%) and overweight patients (14.3%). Prevalence of hypercholesterolemia (P = 0.015), hypertriglyceridemia (P = 0.009), and elevated LDLc (P = 0.000) were significantly higher towards obese patients. However, the differences observed in the prevalence of decreased HDLc (P= 0.941) was not statistically significant across all the ranges of body mass index (Table 11).

Table 11: Prevalence of dyslipidemia in relationship to body mass index of patients (n=231)

Lipid Abnormalities	Body Mass Index (Kg/m^2)				P value
	Underweight	Normal	Overweight	Obese	
Hypercholesterolemia	9(45)	41(43.2)	32(58.2)	41(68.3)	**0.015**
Hypertriglyceridemia	3(15)	20(21.1)	19(34.5)	26(43.3)	**0.009**
Low HDLc	3(15)	16(17)	8(14.3)	11(18.3)	0.941
Increase LDLc	8(40)	44(47.3)	39(69.6)	46(76.6)	**0.000**

Figure 3 shows the distribution of body mass index according to the sex of patients. When comparing body mass index and sex of the patients, women were more obese (31.8%) than their male counterparts (7.3%). Majority (61.8%) of male patients had normal body mass index compared to female patients (34.7%).
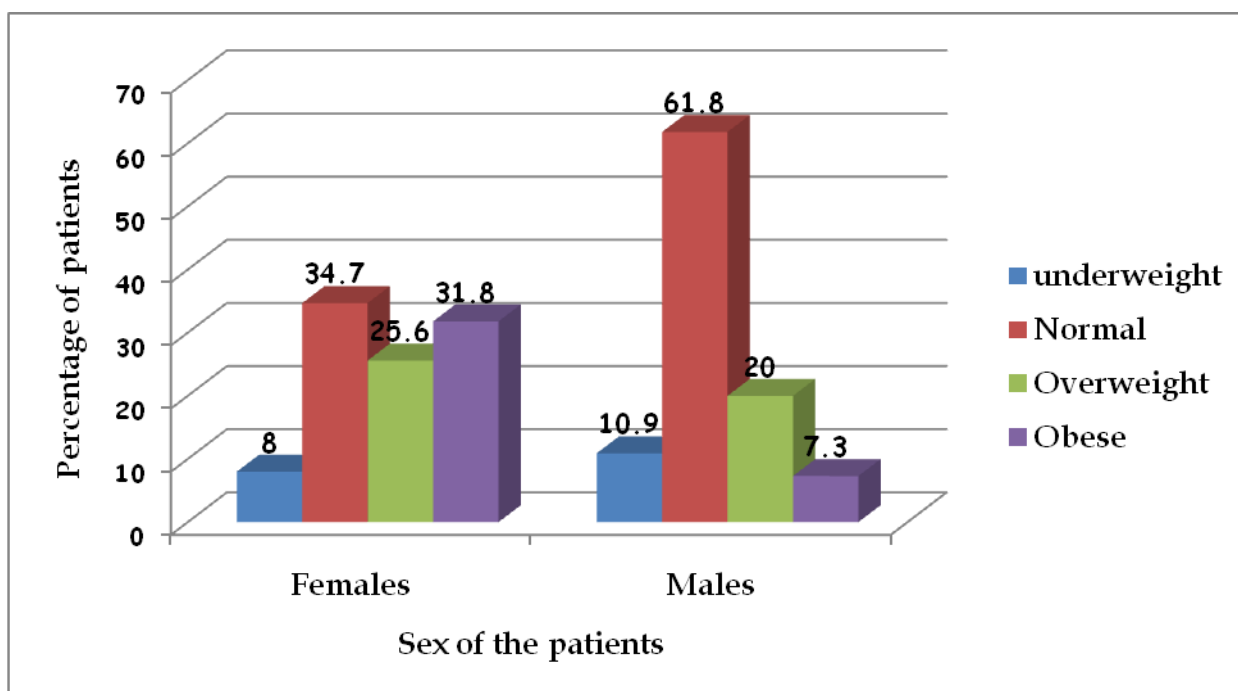
Figure 3: Comparison of body mass index categories according to sex of patients

3.4.6. Prevalence of dyslipidemia with respect to the duration of ARV use:

Duration on ARVs use was categorised into three groups. These included patients who had been using ARVs for less than one year, 1-2 years and >2 years. Patients who had been using ARVs for >2 years had varying prevalences for, hypercholesterolemia (64.6%), hypertriglyceridemia (35.4%), low HDLc (15.9%) and increased LDLc (71.4%), which were higher compared to patients who were using ARVs for less than a year (30.6%, 8.3%, 11.1% and 47.2% respectively). There was **a** statistically significant trend towards an increase in the prevalence of hypercholesterolemia, hypertriglyceridemia, low HDLc and elevated LDLc with duration of ARVs use. Patients who were using ARVs for <12 months, 12-24 months and >24 months showed varying prevalence, the differences of which were statistically significant.. Although the prevalence of low HDLc showed similar trend, the differences observed were not statistically significant (Table 12).

Table 12: Proportion of patients experiencing lipid abnormalities with respect to the duration of ARVs use (n=231)

Lipid deregiment	Time on ARVs (month)			
	<12 n(%)	12 -24 n(%)	>24 n(%)	P-value
Hypercholesterolemia	11(30.6)	38 (47.5)	73 (64.6)	**0.001**
Hypertriglyceridemia	3 (8.3)	25 (31.2)	40 (35.4)	**0.008**
Low HDLc	4 (11.1)	16 (20)	18 (15.9)	0.475
Increased LDLc	17 (47.2)	40 (50)	80 (71.4)	**0.003**
Overall dyslipidemia	23(63.9)	58(71.6)	97(77.4)	**0.004**

HDLc, high-density lipoprotein cholesterol; LDLc, low-density lipoprotein cholesterol; p, significance level

3.5. Multivariate Logistics Regression Analysis:

All factors with p-value of ≤ 0.2 on univariate analysis were considered for multivariate binary logistic regression analysis so as to control for confounding factors. Table 13 shows the factors associated with dyslipidemia on binary logistic regression analysis. In this analysis, age, normal BMI, overweight, obesity, CD4 cell counts, sex and duration of ARVs use were found to be predictors of poor lipid profiles. The types of ARVs regimen used did not show statistical significant association with dyslipidemia

3.5.1. Hypercholesterolemia

In multivariable logistic regression analysis (Table 13), there was evidence that patients who were using ARVs for more than two year were 3.46 times more likely to have hypercholesterolemia (aOR = 3.46 95% CI: 1.45 − 8.274 , p = 0.005) as compared to patients who were using ARVs for less than a year. On the other hand, patients with normal BMI were about 3 times less likely to have hypercholesterolemia (aOR = 0.38, 95% CI: 0.179 − 0.801 , p = 0.011) as compared to obese **patients**. Patients who were 50 years and above were 8.46 times more likely to have hypercholesterolemia(aOR 8.46, CI 2.329 − 30.711, P= 0.001) as compared to younger patients in the age group of 15 - 30 years as shown in (Table 13).

3.5.2. Hypertriglyceridemia

In this study patients who had CD4 cell counts of <350 cells/mm^3 were 2.3 times less likely to have hypertriglyceridemia (aOR 0.43, CI: 0.21 -0.897, P =0.024) as compared to those with CD4 cell counts of >500 cells/mm^3. Patients with CD4 cell counts of 350-500 cells/mm^3 were about 2.8 times less likely to have hypertriglyceridemia (aOR = 0.35, 95% CI: 0.137 − 0.892, p = 0.024) as compared to those who had CD4 cell counts of > 500 cells/mm^3. Patients who were using ARVs for 12 − 24 months were 4 times more likely to develop hypertriglyceridemia (aOR = 4.01, 95% CI: 1.03 − 15.621, p = 0.045) as compares to those who were using ARVs for less than a year. On the other hand, patients who were using ARVs for more than 2 years were 4.42 times more likely to develop hypertriglyceridemia (aOR 4.42, CI: 1.178 − 16.549, P =0.028). Moreover, patients with

normal BMI were 2.4 less likely to have hypertriglyceridemia (aOR = 0.41, 95% CI: 0.186 – 0.925, p = 0.032) as compared to obese patients (Table 13).

3.5.3. Decreased HDLc

In this study, we observed that male patients were 2 times more likely to have decreased HDLc (aOR = 2.15, 95% CI: 1.021 – 4.521, p = 0.044) as compared to females (Table 13).

3.5.4. Elevated LDLc

Patients who were using ARVs for 12-24 months were 2.9 times less likely to develop elevated LDLc (aOR = 0.347, 95% CI: 0.18 – 0.67, p = 0.002) as compared to those who were using ARVs for more than two years. Obese patienst were 3.43 times more likely to have elevated LDLc (aOR 3.43, 95% CI: 1.101 – 10.706, P = 0.034) as compared to underweight patients. In our study patients who were 50 years and above were 5 times more likely to have elevated LDLc (aOR 5.25, 95% CI: 0.011) as compared to patients who were 15 – 30 years as shown in table 13.

Table 13: Multivariate binary logistic regression analysis, showing factors associated with poor lipid profiles and odds of developing dyslipidemia among patients using first line antiretroviral drugs.

Outcome variables	Predictor variables	Group	OR	95% CI	P-value
TC $\geq$ 5.17 mmol/l	Age (years)	15- 30	1		
		31-40	2.53	0.825 – 7.779	0.104
		41-50	1.84	0.603 – 5.598	0.285
		51+	**8.46**	**2.329 – 30.711**	**0.001**
	BMI (Kg/m^2)	< 18.5	0.71	0.226 – 2.198	0.547
		18.5 - 24.99	**0.38**	**0.179 – 0.801**	**0.011**
		25 - 29.99	0.73	0.319 – 1.661	0.45
		30+	1		
	CD4 counts (cells/mm^3)	<350	0.59	0.3 – 1.16	0.126
		350-500	0.7	0.305 – 1.624	0.411
		> 500	1		
	Time on ARVs(month)	<12	1		
		12 - 24.	1.84	0.767 – 4.388	0.172
		> 24	**3.46**	**1.45 – 8.274**	**0.005**
TG $\geq$ 1.69 mmol/l	Age (years)	15 – 30	1		
		31 – 40	0.87	0.235 – 3.204	0.831
		41 – 50	0.96	0.263 – 3.522	0.954
		50+	3.7	0.951 – 14.385	0.059
	BMI (Kg/m^2)	< 18.5	0.36	0.084 – 1.563	0.173
		18.5 - 24.99	**0.41**	**0.186 – 0.925**	**0.032**
		25 - 29.99	0.69	0.300 – 1.599	0.389
		30+	1		
	CD4 cell counts (cells/mm^3)	**< 350**	**0.43**	**0.210 - 0.897**	**0.024**
		350 - 500	**0.35**	**0.137 – 0.892**	**0.024**
		500+	1		
	Time on ARVs(months)	< 12	**1**		
		12 -24.	**4.01**	**1.03 – 15.621**	**0.045**
		> 24	**4.42**	**1.178 – 16.549**	**0.028**
	NRTIs	AZT	1.21	0.484 – 3.007	0.688
		TDF	1		
	NNRTIs	EFV	1	0.496 – 2.035	0.99
		NVP	1		

HDLc <1.04 mmol/l	Sex	**Male**	**2.15**	**1.021 – 4.521**	**0.044**
		female	1		
LDLc <3.4 mmol/l	Age (Years)	15 -30	1		
		31-40	1.65	0.567- 4.784	0.360
		41 – 50	1.33	0.457 – 3.853	0.603
		50+	**5.25**	**1.468 – 18.771**	**0.011**
	BMI (Kg/m^2)	<18.5	1		
		18.5 - 24.99	0.94	0.326 – 2.688	0.902
		25 - 29.99	2.82	0.920 – 8.656	0.070
		30+	**3.43**	**1.101 – 10.706**	**0.034**
	Time on ARVs(months)	< 12	0.44	0.192 – 1.009	0.052
		12 - 24.	**0.347**	**0.180 – 0.670**	**0.002**
		> 24	**1**		

CHAPTER FOUR

4. DISCUSSION

The study has demonstrated a high prevalence of dyslipidaemia (77.5%) in HIV-infected patients receiving first-line ARVs. Hypercholesterolemia, hypertriglyceridemia, low HDLc, and increased LDLc were the most common forms of dyslipidaemia. Of these, elevated LDLc is most closely linked to cardiovascular risks, and usually is the primary target of therapeutic interventions. Though high, the prevalence of overall dyslipidaemia in this study is lower compared to the report of the previous study (82.7%) in patients using ARVs at Muhimbili National Hospital (15). The difference in the prevalence with the current study could be due to the fact that the previous study at Muhimbili National Hospital included patients who were using both the first line and second line ARVs (protease inhibitors (PIs)), while the current study assessed patients using first line ARVs regimen alone. It is already shown in other studies, (9,13,37) that PI containing regimen exacerbate dyslipidemia. However, the findings of this study are comparable to those observed in 12 Care and Treatment Clinics (CTCs) affiliated with Management and Development for Health (MDH) in Dar es Salaam, Tanzania where the prevalence of dyslipidemia was 76% in ART-naive patients (27). The similarity between the result of the two studies support the findings of the previous studies indicating that dyslipidemia is a result of metabolic effect of the HIV virus itself (11,30,38–40).

Apart from the effect of ARVs and HIV, occurence of dyslipidemia could also be explained in part by wrong eating habits among HIV affected patients. A common practice in the local population is to encourage HIV patients to 'over-feed' fat rich foods so that they maintain their weight and improve immunity. In the past, underweight patients were easily identified in the community as being HIV infected and hence the associated social stigma. Thus HIV patients strive to get nutritional support and use food supliments. This may lead to over weight and dyslipidaemia (15).

The prevalence of hypercholesterolemia in our study was comparable to that of the earlier study that was conducted at Muhimbili National Hospital in 2012 (15). In that study 370 patients were included of which 274 were on ARVs while 96 were ART naive. The results are also similar to that reported by Tadewos et al in Southern Ethiopian (40). In that study, 226 HIV infected patients were involved, 113 patients were ART experienced whereas 113 paients had not started using ARVs. However, the prevalence of hypercholesterolemia in this study is much higher compared to the previous report by Armstrong et al in Dar es Salaam where 12,513 ART naive patients were studed (27). The difference with the current study which constituted only patients who were on ART perhaps suggest an additional effect of ARVs in causation of hypercholesterolemia.

High triglyerides levels in HIV-infected patients have been postulated to be due to inflamation with subsequent cytokines release and decreased hepatic clearance related to a role of apolipoprotein (32). Estrogen treatment can aggravate hypertriglyceridemia by increasing very LDL secretion and reducing hepatic triglyceride lipase activity (41). From these findings it was expected that women would have had high triglyceride levels. However, the results of this study show that males had much higher prevalence of hypertriglyceridemia than females. This observation is supported by the report of the study by Erwin et al in the United States of America where the prevalence of hypertriglyceridemia was found to be higher in males than females (42). However, the prevalence of hypertriglyceridemia in our study is slightly lower compared to 36.4% which was reported by Mgunya (15). The reason for this difference could be due to the fact that the later study included both first line and protease inhibitor ARVs-containing regimen. The prevalence of hypertriglyceridemia in the present study is also much lower than that reported by Bekolo at al (51.8%) in rural Cameroonia population (43), and in a study by Pefura et al (43.5%) which included both ARVs experienced and ARVs naive patients in urban Cameroonian population (44).

Decreased HDLc occurred in 16.5% of the study subjects being significantly higher in males than females. A reduction of HDLc is recognized as an important risk factor for coronary artery disease. This is caused by mutation in the human lipoprotein lipase (LPL) gene which results in decreased LPL catalytic activity and hence low HDL cholesterol (45).

In the present study, the prevalence of elevated LDL cholesterol was 59.8%. This prevalence is much higher than 12% which was reported in a previous study in Dar es Salaam (27). The difference between the findings of the present study and that of Armstrong et al (27) could be due to the fact that in the later study both ARVs naive and ARVs experienced were studied. This could explain the effect additional effect of ARVs on lipid metabolism (46). Elevated levels of circulating cholesterol transported by apolipoprotein B100 (ApoB100)-containing LDL promote atherosclerosis and cardiovascular disease. Atherosclerosis gives rise to cerebrovascular disease and CAD through a slowly progressing lesion formation and luminal narrowing of arteries. Upon plaque rupture and thrombosis, these most common forms of cardiovascular disease manifest as acute coronary syndrome (ACS), myocardial infarction or stroke (47).

HDLc is positively associated with a decreased risk of CHD. As defined by the US National Cholesterol Education Program Adult Treatment Panel III guidelines, an HDLc of ≥60 mg/dl is a negative (protective) risk factor (39). In our study there was no significant difference in the prevalence of lipid abnormalities in patients on regimens that included zivodivudine as compared with those on regimens that included tenofovir. The prevalence of lipid abnormalities induced by nevirapine and efavirenz was also similar. This is in contrast with the previous reports where patients taking nevirapine based regimen were found to have higher levels of HDLc as compared to patients who were using efavirenz based regimen (31,48). In a study by Van Lethet at al, 706 HIV-infected were given stavudine and lamivudine with either nevirapine (417 patients) or efavirenz (289 patients). In that study, patients taking nevirapine-based regimen were found to have higher levels of HDLc as compared to patients who were using efavirenz-based regimen (48). These observed differences between the findings of our study and the later study could be due to the fact that the later study was a follow up study and also included stavudine and

lamivudine as NRTI backbone with either efavirenz or nevirapine. The inclusion of stavudine in the later study could be the main cause of increased serum lipid levels (2,49). In the current study, the nucleoside backbone used in the first line ARVs was either zidovudine or tenofovir.

In the current study, patients who were using ARVs for more than two years were significantly associated with a lipid derangement as compared to those who were using ARVs for less than a year. This is probably related to a combination of the effects of an ageing HIV-infected population coupled with improved health due to suppression of viral load and the effect of ARVs on lipid metabolism. The results in this study are comparable to those of the previous study where duration of ARVs use was positively associated with the prevalence of dyslipidemia. For instance the study in rural Uganda indicated that the mean TC, LDLc and HDLc levels were significantly higher at 24 months compared to baseline (28). Other studies conducted in Europe, United States of America and Cameroon showed that being on ARVs for more than two years was associated with poor lipid profiles (43,50). These two previous studies were follow up studies in which it was seen that the duration on ARVs use was associated with a cardio-protective lipid profile in the short term, because after initiation of ART, lipid levels return to baseline levels, but soon they rise above presero- conversion levels in the long term (28,50). This finding suggests that dyslipidemia develops with cumulative duration of exposure to ARVs.

In terms of the relationship between poor lipid profile and immune status of the patients, we observed that there was a trend of increasing prevalence of dyslipidemia towards increasing CD4 cell counts. This is probably because with ARVs use there is suppression of viral load, as a result CD4 cell counts increases which allows immune recovery leading to elevation of lipids in addition to the continued effect of ARVs in elevating lipid levels. As it has been reported from a previous study involving ART and ART naive participants by Pefura et al (44) in Cameroon, ART experienced patients are more likely to have high CD4 cell count and high body mass index indicating immune recovery and health restoration. However, patients with low CD4 cell counts also experienced poor lipid profile, indicating that low CD4 cell counts is associated with high viral load. Alterations in cholesterol metabolism that occur in HIV infected patients could be explained by lipid

peroxidation. The cytokine tumor necrosis factor (TNF)-a plays a role in plasma lipoprotein peroxidation in HIV-infected patients by stimulating the production of reactive oxygen species. This is in agreement with earlier reports indicating that dyslipidemia in HIV infected patients is partly due to the effect of the virus itself on lipid metabolism (8,10,51).

Obesity is known to be a predictor, an independent risk factor for cardiovascular disease and total mortality (38,52). Several studies have shown associations between obesity and hypercholesterolemia, hypertriglyceridemia, low HDLc and elevated LDLc in men and women and in diverse race and ethnic groups (44,52–54). The findings of the current study show that the prevalence of dyslipidemia increased significantly toward increasing body mass index. For instance obese patients were 3 times more likely to have elevated LDLc than underweight patients. Moreover, patients who had normal BMI were 2.4 times less likely to have hypertriglyceridemia compared to obese patients. These findings are comparable to the findings of a recent study by Tadewos et al which was conducted in Southern Ethiopia (40). In that study, overweight and obese patients who were using ARVs were found to be at increased risk for development of dyslipidemia.

As age increases, pro-atherogenic lipid parameters also increase (10,55,56). Levels of lipoproteins and therefore lipids, particularly LDLc, increase slightly as people age. Levels are normally slightly higher in men than in women, but levels increase in women after menopause. The increase in levels of lipoproteins that occurs with age can result in dyslipidemia and increase the risk of atherosclerosis. In our study, the prevalence of dyslipidemia increased significantly with advancing age. The findings of the current study show similar trend as the previous report by Wang et al in the general population in rural and urban China where the prevalence of dyslipidemia was found to be increasing with advancing age (57). However, the study participants in the later study had very low prevalence of dyslipidemia when compared to the prevalence of dyslipidemia in the current study. This could explain the additional effect of HIV itself and ARVs on lipid metabolism in the current study. On the otther hand, the observation that males had higher overall prevalence of dyslipidemia as compared to females could be due to the protective effect of oestrogen in pre-menpousal women (55,57).

Although our study had some limitations, the increased risk of atherothombotic cardiovascular disease associated with lipid derangement is well known and would therefore suggest that treatment with first-line ARVs may actually have harmful effects on the cardiovascular health of patients. The sample size was relatively small so much so that the precision of our odd ratios was low and we lacked the power to detect significant differences we hoped to. In addition, our study was cross-sectional **study** and inference about causal relationship could not be made. Cohorts' studies would be appropriate to monitor lipid profile alterations in patients using first-line ARVs, and their potential impact on cardiovascular health of people living with HIV in our settings.

CHAPTER FIVE

5.1 CONCLUSION

Dyslipidemia is a primary, major risk factor for CAD and may even be a prerequisite for CVD, occurring before other major risk factors come into play. Dyslipidemia is one of the modifiable risk factor for CVD. Optimal lipid level control reduces disease related complications, thereby improving patient outcomes and eventually decreasing health care cost. Overall, the findings from the current study indicate that there was high prevalence of dyslipidemia among HIV-infected patients using first line ARVs attending Muhimbili National Hospital.

Despite the importance of monitoring serum lipids and established association of ARVs use and increased prevalence of dyslipidemia in HIV-infected patients and their effect on cardiovascular complications, the unexpected finding in our study was that records of lipid profile measurements of patients were not available indicating that they were either not ordered or rather not recorded.

Male sex, CD4 cell counts of > 500cells/mm^3, age above 50 years, duration of ARVs use of >2 years and obesity were found to be associated with increased risk of lipid derangement among patients. Therefore, dyslipidemia among patients taking ARVs is related to multiple factors, including: individual patient characteristics, HIV infection itself and use of ARVs. The use of first line ARVs was associated with a lipid profile that is likely to increase the risk of cardiovascular disease. Therefore, these findings indicate the need to assess lipid profiles at baseline before initiation of ARVs treatment and monitoring the same during therapy to monitor any rising trends.

5.2. RECOMMENDATIONS

Dyslipidemia is one of the modifiable risk factor for CVD, thus we recommend an education program both to health professionals and to the patients that emphasize on the importance of physical exercise and weight management, and this would be of benefit in improving lipid profiles.

Different first-line ARVs regimen use have shown different levels of prevalence of dyslipidemia, thus we recommend another study to be done with relatively higher sample size, and a follow up study design so that causal relationship can be established.

Patients on first line ARVs shows improved health, indicated by general apperance, but also through rise of their CD4 cell counts and BMI. Thus because of this health improvement they are usually forgoten when it comes to assessment of risk for CVD especially dyslipidemia. However as it has been found in this study patients on first-line ARVs have shown a very high prevalence of dyslipidemia. In our setting lipid profile is performed only during initiation of second line ARVs regimen. The National guidelines for the management of HIV and AIDS put little emphasis on serum lipid assessment before initiation and during ARVs use (7). It is therefore, recommended that the National AIDS Control Program should institute mandatory laboratory monitoring of serum lipids for HIV infected patients during initiation of ARVs as well as monitoring during therapy.

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APPENDICES

Appendix I: Data collection tool - English version.

TITLE: **Assessment of lipid profile in HIV infected patients using the first line antiretroviral drugs at Muhimbili National Hospital.**

A: PATIENT INFORAMTION:

1. Patient code number ---------------------------

2. Name of CTC -----------------------------

3. File number -----------------------------

4. Sex: male ----------------- female ----------------- (tick)

5. Age of the patient ---------------------- years

6. Weight of the patient -----kg; Height ----- cm; Body Mass Index (BMI) -----kg/m^2

7. CD4 cell count ------------------ cells/mm^3

8. Duration of using ARVs ----------------- (months)

9. ARVs regimen which the patient is using. Indicate the names of the drugs;

   i. --

   ii. --

   iii. --

   iv. --

B: PATIENT INTERVIEW:

10. Do you have any other co-morbidity apart from HIV/AIDS? Yes ----- No- --(tick)

11. If yes to Q10 above, what is that co-morbid disease? ------------------ (mention)

12. Do you use any drug(s) to treat such co-morbid disease? Yes ------ No ------- (tick)

13. If yes to Q12 above what drug(s) do you use? (Mention below)

    i.   ------------------------------

    ii.   ------------------------------

C: LABORATORY RESULTS:

14. Serum TC level ------------------------- mmol/l

15. Serum TG level------------------------- mmol/l

16. Serum HDL level---------------------- mmol/l

17. Serum LDL Level ---------------------- mmol/l

Appendix II: Dodoso ya kukusanyia takwimu:

KICHWA CHA UTAFITI: Uchambuzi wa viwango vya cholesterol kwa wagonjwa wenye maambukizi ya virusi vya ukimwi ambao wanatumia dawa za ARVs wanaopata huduma ya matibabu katika Hospitali ya Taifa ya Muhimbili.

A: TAARIFA YA MGONJWA:

1. Namba ya utambulisho ya mgonjwa ------------------------------

2. Jina la kliniki ya uangalizi na matibabu (CTC) ------------------------------

3. Namba ya jalada -----------------------------

4. Jinsia: mme -------------- mke ------------------- (weka alama ya ''V'')

5. Umri wa mgonjwa (miaka) -----------------------

6. Uzito wa mgonjwa (kilo) --------------- ; Urefu (cm) -------------; Index ya mwili (BMI) ----------- kg/m^2

7. Kiwango cha chembechembe za CD4 kenye damu -------------------- cells/mm^3

8. Muda ambao mgonjwa amekuwa akitumia dawa za ARVs (miezi) ----------------

9. Mjumuisho wa dawa za ARVs anazotumia mgonjwa. Taja hizo dawa hapa chini

    i. -------------------------------

    ii. -------------------------------

    iii. -------------------------------

    iv. -------------------------------

B: USAHILI KWA MGONJWA

10. Je una ugonjwa mwingine wowote tofauti na VVU? Ndiyo ------ Hapana ------ (weka V)

11. Kama ndiyo kwa swali namba 10, ni ugonjwa gani? ----------------------- (utaje)

12. Je unatumia dawa zozote katipo hue ugonjwa? Ndiyo --------- Hapana --------- (weka V)

13. Kama ndiyo kwa swali namba 12, ni dawa gani unazotumia? Zitaje hapa chini

    i.   ------------------------

    ii.   ------------------------

C: MATOKEO YA MAABARA:

14. Kiwango cha 'TC' kwenye damu -------------------------- mmol/l

15. Kiwango cha 'TG' kwenye damu-------------------------- mmol/l

16. Kiwango cha 'HDL' kwenye damu------------------------ mmol/l

17. Kiwango cha 'LDL' kwenye damu ------------------------ mmol/l

Appendix III: Consent form - English version

TITLE: **Assessment of lipid profile in HIV infected patients using the first line antiretroviral drugs at Muhimbili National Hospital.**

My name is Walter Ombeni, a 2nd year resident doing master of pharmacy in hospital and clinical pharmacy at Muhimbili University of Health and Allied Sciences (MUHAS).

I am conducting a study with the above title as part of my study program.

Aims of the study:

The aim of the study is to determine the prevalence of dyslipidemia in HIV-infected patients on ARVs triple regimen. This study will further assess the factors associated with dyslipidemia and discuss the treatment of choice for individuals with dyslipidemia on ARVs triple regimen.

Participation in this study:

You can participate in this study if you are 15 years old or more and you have been on ARVs triple regimen for at least twenty four weeks.

The study mainly involves blood sample collection, assaying lipid profile, and responding to a questionnaire which has general questions about your demographic characteristics, and a section on questions related to ARVs regimen that you are using. In addition, weight and height will be measured by the nurse as part of routine care at the clinic before seeing the physician and these data will be used in the study.

If you choose not to participate in this study, you will continue to receive the normal care at the CTC.

Risks:
We do not anticipate any risks as a result of participating in the study.

Benefits:

By participating in this study, you will be able to know the status of your lipid profile in the body which is very important because if it is elevated is a risk factor for cardiovascular diseases. If serious abnormality of your lipid profile will be found, you will be referred to the doctor for detailed investigations and other treatment modality will be provided as per the care and treatment procedures at the hospital.

Confidentiality:

All the information collected during this study will be kept strictly confidential and will not be revealed to anybody outside the research team.

Cost:

You will not be required to make any payments to participate in this study and no payment will be made to you.

For further information, questions or queries, you can contact:

 The Principal Investigator,

Mr. Walter Ombeni

School of pharmacy,
MUHAS,
P. O. Box 65001,
Dar es Salaam.
Cell no: +255 713 133910
Email: walterombeni@gmail.com
The supervisor,
Prof. Appolinary Kamuhabwa
School of pharmacy,
MUHAS.
P. O. Box 65001
Dar es Salaam

In case you require more information about the conduct of the study, you can also contact Prof. M. Moshi, who is the Director of Research and Publications at Muhimbili University of Health and Allied Sciences, Tel 022 2152489.

I, _____, have read/been told of the contents of this form and have understood its meaning. I agree for to participate in this study.

Signature of participant _____

Signature of Researcher _____

Date _____

Appendix IV: Consent form - Swahili version

FOMU YA RIDHAA YA KUSHIRIKI KATIKA UTAFITI

KICHWA CHA UTAFITI: Uchambuzi wa viwango vya cholesterol kwa wagonjwa wenye maambukizi ya virusi vya ukimwi ambao wanatumia dawa za ARVs wanaopata huduma ya matibabu katika Hospitali ya Taifa ya Muhimbili.

Jina langu ni Walter Ombeni, mwanafunzi wa mwaka wa pili katika masomo ya uzamili kwenye program ya `Hospital and Clinical Pharmacy` katika Chuo Kikuu cha Afya na Sayansi Shirikishi Muhimbili (MUHAS). Ninafanya utafiti kama kichwa cha utafiti kinavyosomeka hapo juu.

Madhumuni ya utafiti:

Lengo la utafiti huu ni kuchunguza kiwango cha kuongezeka kwa cholesterol kitokanacho na matumizi ya dawa za ARVs, kwa wagonjwa wenye maambukizi ya virusi vya ukimwi.

Ushiriki katika utafiti:

Unaweza kushiriki katika utafiti huu kama una umri wa miaka 15 au zaidi na kama umekuwa ukitumia dawa za ARVs kwa muda usiopungua wiki 24 (miezi 6).

Utafiti huu unafanyika kwa kutolewa damu na kupima kiwango cha cholesterol. Utafiti huu pia utajumuisha kujibu maswali kuhusu wewe mwenyewe, na sehemu ya maswali kuhusu dawa za ARVs unazotumia. Pia muuguzi atakupima uzito na urefu wako. Vipimo hivi vitatumika kwenye utafiti tuu. Kama ukiamua kutoshiriki katika utafiti huu, utaendelea kupata huduma kama kawaida kwenye kliniki hii ya uangalizi na matibabu.

Madhara:

Hatutarajii madhara yeyeto yatokee kwa kushiriki katika utafiti huu.

Faida za utafiti:

Kwa kushiriki katika utafiti huu, utafahamu kiwango cha cholesterol katika mwili wako ambayo ni muhimu sana kwani inapokuwa katika kiwango cha juu ni kiashiria cha

magonjwa ya moyo. Kama tatizo lolote kubwa litagundulika, utapelekwa kwa Daktari kwa uchunguza wa kina na kupewa matibabu kutokana na taratibu za matitabu hospitalini hapa.

Usiri:

Taarifa zote zitakazokusanywa katika utafiti huu zitakuwa siri, hivyo ushiriki wako hautajulikana na mtu yoyote. Taarifa hizi zitajulikana kwenye timu ya watafiti tu.

Malipo:

Kwa kushiriki kwenye utafiti huu, hautalipwa wala hautalipa gharama yeyote.

Ukiwa na swali au tatizo lolote, unaweza kuwasiliana na wafuatao:

Mtafiti mkuu,

Bwana Walter Ombeni

Skuli ya Famasia,

MUHAS

S.L.P. 65001,

Dar es Salaam.

Simu: +255 713 133910

Barua pepe: walterombeni@gmail.com

Msimamizi wa utafiti

Prof. Appolinary Kamuhabwa

Skuli ya Famasia,

MUHAS.

S.L.P. 65001

Dar es Salaam

Kama utakuwa unahitaji taarifa zaidi juu ya utafiti huu unaweza kuwasiliana na Profesa M. Moshi ambaye ni Mkurugenzi wa Utafiti na Machapisho katika Chuo Kikuu cha Afya na Sayansi Shirikishi Muhimbili.

Mimi, _____, nimesoma/nimesomewa maelezo yote

yaliyomo kwenye fomu hii na nimeelewa. Nakubali kushiriki katika utafiti huu.

Saini ya mshiriki _____

Saini ya Mtafiti _____

Tarehe _____