THE EFFECT OF METHADONE MAINTENANCE TREATMENT ON BODY MASS INDEX AND LIPID PROFILE AMONG CLIENTS ATTENDING MMT CLINIC AT MUHIMBILI NATIONAL HOSPITAL IN DAR ES SALAM, TANZANIA

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Department of Biochemistry and Molecular Biology

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By

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A Dissertation Submitted in (Partial) Fulfillment of the Requirements for the Degree of Master of Science (Biochemistry) of

Muhimbili University of Health and Allied Sciences
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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; **‘‘The Effect of Methadone Maintenance Treatment on Body Mass Index and Lipid Profile among clients attending MMT clinic at Muhimbili National Hospital in Dar es Salam, Tanzania’’,** in (partial) fulfillment of the requirements for the degree of Master of Science (Biochemistry) of Muhimbili University of Health and Allied Sciences.

_____

Dr. Teddy F. Mselle

(Supervisor)

_____

Date

DECLARATION AND COPYRIGHT

I, **Haji Ally Sangu**, 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.

Signature…….. **Date**…….....….....

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ABSTRACT

Background: Methadone, a full mu-opioid receptor agonist, reduces illicit drug use and the spread of infections such as HIV and HBV, which are common among drug abusers due to injection sharing. However, it is also associated with changes in body mass index and lipid profile, an area with limited studies in Tanzania.

Objective: The objective of this study was to examine the effect of methadone maintenance treatment on body mass index and lipid profile among clients who visit MMT clinic in MNH at Dar es Salaam, Tanzania.

Materials and Methods: This was a retrospective cohort study with a cross-sectional element included 100 clients from an MMT clinic at MNH. Body weight and height were measured during data collection to calculate post-treatment BMIs, while baseline BMIs were determined from baseline data collected from treatment files. Fasting blood samples were collected for lipid analysis. The BMI before and after MMT and lipid profiles were determined and analyzed by age, sex and prior heroine use using t-test analyzing approach.

Results: Ninety-two percent of participants were male, the mean age of study participants was 42 ± 9.3 years and 73% had education below secondary level. Paired t-test analysis showed a significant increase in mean BMI from 19.3 ± 2.6 kg/m^2before treatment to 20.3 ± 3.10 kg/m^2 after treatment), $p<0.0001$, $t(100) = 4.35$ (2-tailed), 95% CI= 0.56 to 1.50kg/m^2. We noticed that participants on MMT for more than 24 months had significantly higher mean BMI (21.28±2.5 kg/m^2) compared to those on MMT for less or equal to 24 months (20.05±2.5 kg/m^2),$p=0.0480$, $t=2.00$, two tailed, 95%CI =0.01075 to 2.443 kg/m^2. The mean (± SEM) increase in BMI was 1.227 ± 0.6128 kg/m^2, indicating that on average BMI is more likely to increase by 1.227kg/m^2 after two years of methadone treatment. Overall, 54% had one or more abnormal lipid levels. Decreased levels of HDL were most prevalent, rating 40% followed by elevated levels of TG, rating 21%.

Conclusions: Methadone use is associated with an increase in BMI, with longer durations of therapy leading to higher BMI. Over 50 percent of participants had abnormal lipid profile, particularly HDL (low) and TG (high).

Recommendation: Because of benefits of MMT we recommend expansion of MMT clinics to other urban centers in Tanzania, consider determining lipid profile of clients at enrolment and during follow up, conducting a follow-up study comprising of large sample size for a longer period to investigate the long effects of methadone on lipid profile.

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LIST OF ABBREVIATIONS

AFHU	Age at first heroin use
ARV	Antiretroviral
BMI	Body mass index
CVD	Cardiovascular diseases
HDL-C	High density lipoprotein cholesterol
HIV	Human immunodeficiency virus
HU	Heroin use
LDL-C	Low density lipoprotein cholesterol
MAT	Medically assisted therapy.
MORs	Mu-opioid receptors
MMT	Methadone maintenance treatment
MNH	Muhimbili National Hospital
MUHAS	Muhimbili University of Health and Allied Sciences
NCEP ATP III	National cholesterol education program adult treatment panel III
TC	Total cholesterol
TG	Triglyceride
WHO	World Health Organization

DEFINITION OF TERMS

Body mass index; is a ratio of weight (in kg) to a square of height (in meters)

Client: as used in this document refers to a person who needs maintenance treatment due to a drug addiction.

Drug addiction: is characterized by obsessive drug seeking and use, regardless of negative consequences. It is a chronic, recurrent condition**.**

Dyslipidemia: Refers to suboptimal concentrations of triglycerides (TG), total cholesterol (TC), low-density lipoprotein (LDL), high-density lipoprotein (HDL), and apolipoproteins.

Lipid profile: Laboratory blood test to measure the amount of lipid molecules in the blood. The test includes total cholesterol (TC), low density lipoprotein (LDL), high density lipoprotein (HDL) and triglycerides.

Methadone: Is a medication given to a client as a treatment of drug use disorder. It acts as a mu-opiod receptor agonist, to reduce opioid craving.

Mu opioid receptors: are class of receptors that bind to opioids and neuromodulate various physiological activities, including temperature, respiration, stress, endocrine activity, gastrointestinal activity, memory, mood, and motivation in addition to primary nociception

Opioid: Opioids are class of drugs such as heroin and prescription pain killers derived from poppy opium plant. They can be highly addictive and cause significant harm if misused.

CHAPTER ONE

INTRODUCTION

1.1 Background

Methadone maintenance treatment (MMT) is a widely recognized intervention for managing opioid dependence, aiming to reduce illicit opioid use and associated risks, including the transmission of infectious diseases. Despite its benefits, MMT has been associated with various metabolic side effects, including changes in body mass index (BMI) and alterations in lipid profile. These metabolic changes can increase the risk of cardiovascular diseases, necessitating regular monitoring and management. In Tanzania, particularly in urban settings like Dar es Salaam, the prevalence of opioid use and the subsequent need for MMT are growing public health concerns. However, there is limited data on the metabolic effects of MMT among this population. Understanding the impact of MMT on BMI and lipid profiles is crucial for developing comprehensive care strategies that address both addiction and its associated health complications. This study aims to fill this gap through examining the effect of MMT on BMI and lipid profiles among clients attending MMT clinic in Dar es Salaam will provide insights into the metabolic health of this population and informing future healthcare practices and policies.

1.1.1 Substance addiction

Addiction to substances, sometimes known as drug addiction, is a neuropsychiatric condition marked by a persistent want to use drugs despite the negative effects on one's health and socioeconomic status (1). It is a major health concern worldwide with an estimate of 15.6 million people affected globally (2). In Tanzania, cannabis is the most commonly used illegal substance, followed by khat, heroin, and cocaine, as well as over-the-counter drugs from hospitals that have narcotic and psychotropic effects, like tramadol and benzodiazepines (3).

In March 2023, Tanzania reported experiencing a spike in drug trafficking during the 66th meeting of the Commission on Narcotic Drugs (CND) in Vienna, Australia. The drugs included 176.05 tons of cannabis, 130.21 tons of khat, 3,311.97 kg of heroin, and 38.186 kg of cocaine

that were seized between June 2018 and December 2022 (3). This demonstrates the severity of the issue facing the nation and highlights one of the main causes of youth drug abuse: the accessibility of narcotics.

Drug usage has a number of detrimental effects on a person's psychological, emotional, and social well-being (4). It raises the risk of contracting infectious diseases (5) and other ailments, such as mental health conditions, cancer, stroke, and diseases of the liver, lungs, and cardiovascular system (6).Infectious illnesses including hepatitis B virus and HIV are more likely to be contracted by drug addicts due to needle sharing (7). According to estimates, the prevalence of HIV among drug injectors in Dar es Salaam was between 42% and 51%, which was higher than the estimated 4.5% prevalence of adult population in Tanzania (8). Drug addiction, then, has a number of negative effects on a person's health and socioeconomic status, which collectively is a burden to a country.

Individuals who consume substances were also reported to have poorer eating habits and nutritional health (9,10). Most studies generally indicated that malnutrition (10–12) was linked to reduced intake, poor dietary practices (4,13) and anthropometrics below standard values (10,14–16), all of which increased an individual's risk of contracting infectious diseases. Conversely, drug users who have been institutionalized and are on methadone maintenance tend to have higher weights and body mass indices (BMIs) (9,17,18), the area that has limited studies in Africa, particularly in Tanzania. Also, studies showed that opium consumption was associated with dyslipidemia such as increased blood concentration of total cholesterol (TC), triglycerides (TG), low density lipoprotein (LDL) and decreased blood concentration of high density lipoprotein (HDL) (19). Dyslipidemia is a component of metabolic syndrome, thus a risk factor for cardiovascular diseases (CVD) (20).

1.1.2 Methadone maintenance treatment (MMT)

MMT is the best evidence based and most effective opioid substitution treatment that is being used by many countries (14,21). Tanzania has adopted it for more than 10 years and Muhimbili National Hospital (MNH) which is the largest hospital in the country, was the first hospital to provide such treatment (7).As a mu-opioid receptor agonist, methadone is the first-line treatment

for opioid use disorders, including heroin(22) . This is because it has been shown to be effective in keeping patients on treatment while also reducing opioid use, risk behaviors for HIV and hepatitis, drug-related criminal activity, and fatal overdoses while enhancing health-related quality of life (23).

MMT is an extended course of methadone treatment for opioid drug dependence, and it may have negative physiological repercussions (21). Nonetheless, there is evidence linking the clinical use of methadone to a higher risk of overdose-related respiratory depression and heart rhythm abnormalities due to prolongation of the QT interval (24). Additionally, methadone has been linked to changes in the endocrine system and metabolism (25).According to studies, insulin resistance and other anomalies in glucose metabolism, as well as an increased risk of diabetes, have been associated with methadone treatment and heroin use (26,27).Also dependency on methadone is linked to metabolic derangements (28) such as changes in body mass index (29) and lipid profile (25). Data on body mass indices and lipid profile among individuals on MMT in Africa are limited despite having centers for MMT as preventive measures to help drug addicts.

1.1.3 Methadone induced lipid metabolism

Lipid metabolism is one of the liver's many activities, making it a significant organ (30). Methadone, like other drugs, is processed by the liver using a variety of intricate enzyme systems; including the cytochrome P450 group (31,32).Methadone is therefore thought to have an impact on liver function (14). There are several factors that may affect lipid profile of an individual. Factors such as diet, activity, smoking and co-morbidities like diabetes, hyperthyroidism, liver and kidney diseases are known to contribute dyslipidemia. Medications used to treat variety of diseases may, however, cause adverse effects on lipid metabolism (33).

Methadone being the case of this study has been related to have effects on lipid profile and body mass index of an individual, the area that has limited studies in Tanzania. The results of a study conducted in Barcelona, Spain, on metabolic syndrome in people with heroin use disorder receiving methadone therapy showed a significant prevalence of low HDL cholesterol and hypertriglyceridemia (51.6% and 36.8% respectively) (2).In a study which compared the

metabolic and addiction indices in patients on opioid medication assisted treatment concluded that more methadone exposure duration was associated with elevated triglycerides and low HDL levels (28).

1.2 Problem statement

In Tanzania, there are several studies conducted on clients undergoing MMT, however, most of these studies investigated the impact of methadone use in the reduction of infectious diseases and improvement of quality of life. Infectious diseases such HIV and hepatitis A, B and C virus were found to be common problem among drug addicts. This has helped to improve and modify treatment modality of these clients where as of now HIV and hepatitis screening is being done regularly.

However, there are limited studies conducted in Tanzania on clients undergoing MMT to associate methadone with metabolic syndromes. Metabolic syndromes are cluster of conditions including hypertension, diabetes, dyslipidemia and obesity which increase individual's risks of heart attack and stroke. Studies in other countries revealed that methadone is associated with metabolic derangements such as increased body weight following initiation and alteration of lipid profile (dyslipidemia). In Tanzania, the lipid profile status among clients on MMT is unknown despite the existence of the MMT service for more than 10 years and no lipid screening tests are conducted during enrolment and on progressive monitoring of MMT individuals.

Ethnic differences between Europeans and Africans can influence variations in metabolism, which, in turn, affect body mass index and lipid profiles. These differences stem from genetic factors (34,35), dietary patterns(36), environmental influences (37), and physiological variations. Certain gene variants linked to lipid metabolism, insulin sensitivity, and obesity are more prevalent in either African or European populations. For example, variations in genes such as APOB**,** LDLR**,** and PCSK9(35)can impact on lipid levels differently depending on ethnicity. Therefore, this study aimed to examine the effect of MMT on body mass index and lipid profile of individuals visiting MMT clinic at Muhimbili National Hospital in Dar es Salaam, Tanzania.

1.3 Conceptual framework

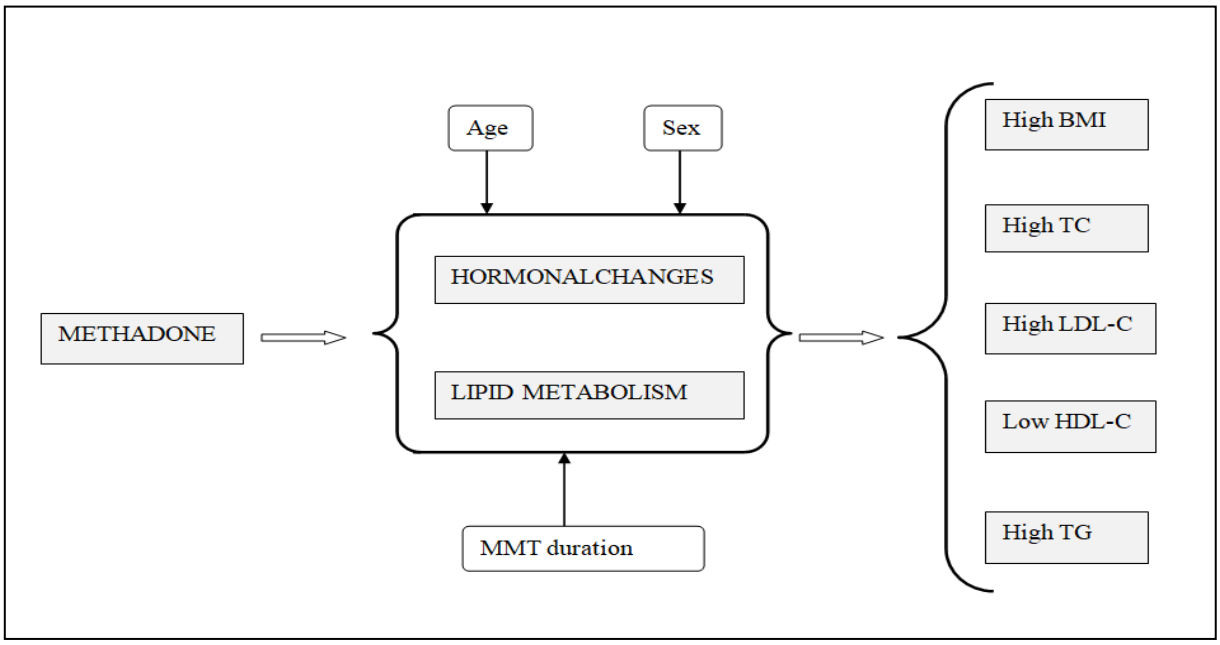


Figure 1: The way duration on methadone use, age and sex affect metabolic and hormonal pathways leading to changes in BMI and lipid profile.

1.4 Significance of the Study and Rationale

This study has for the first time in Tanzania demonstrated the role of methadone in significantly increasing the BMI of patients on MMT. Their mean lipid levels also observed to be different among age, sex and time on MMT. Higher BMIs and high levels of TC, LDL and TG are risk factors for many non communicable diseases such hypertension, atherosclerosis, liver and kidney diseases. The risks can be reduced through change in lifestyles. Awareness of weight gain induced by methadone is important to clinicians and care givers in providing the best care possible to patients with increased body weight and lipid disorders. Therefore the current study provides clues on the role of nutrition in modifying weights among patients on MMT. Thus, this study may be used by clinicians to educate clients on MMT on the importance of modifying their lifestyles such as food preferences and exercises (activities) as preventive measure to lower weights and cholesterols thereby, reducing the risks of atherosclerosis and hypertension.

1.5 Research questions

What is the effect of Methadone Maintenance Treatment on body mass index and lipid profile of patients visiting MMT clinic in Dar es Salaam, Tanzania?

1.5.1 What is the body mass index before and after Methadone Maintenance Treatment of patients who visit MMT clinic at Muhimbili National Hospital in Dar es Salaam, Tanzania?

1.5.2 What is the level of TC, LDL, HDL and TG o patients visiting MMT clinic at Muhimbili National Hospital in Dar es Salaam, Tanzania?

1.6 Research Objectives

1.6.1 Broad Research objective

To explore the impact of MMT on BMI and lipid profile of clients attending MMT clinic in Dar es Salaam, Tanzania.

1.6.2 Specific Research Objectives

1.6.1 To examine the effect of MMT on BMI and its associated factors of clients attending MMT clinic at Muhimbili National Hospital in Dar es Salaam, Tanzania.

1.6.2 To determine the effect of MMT on levels of TC, LDL, HDL and TG of clients attending MMT clinic at Muhimbili National Hospital in Dar es Salaam, Tanzania.

CHAPTER TWO

LITERATURE REVIEW

2.1 Introduction

Methadone maintenance therapy (MMT) is a well-researched treatment for opioid dependence that offers a number of benefits, such as a decrease in the use of illegal opioids, a decrease in criminal activity, and an increase in social functioning. In order to prevent more severe impacts from opioid use, treatment is usually a long-term program aimed at improving the patient's social and physical functioning in addition to reducing or eliminating opioid usage (39). Studies carried out in Dar es Salaam, Tanzania, have investigated the effectiveness of methadone in improving patients' health-related quality of life in the early stages of the medication(7). However, MMT is also linked to a number of adverse consequences, including as alterations in body mass index (BMI) and lipid profiles, which raise the possibility of metabolic and cardiovascular disorders. Tanzanian research has been scarce in this area. Comprehending these impacts is essential to maximizing the patients' long-term health results while on MMT. This study will examine BMIs and lipid profiles among clients on MMT in Dar es Salaam.

2.2 Body mass index

The body mass index (BMI) is a value that is calculated using a person's height and weight. It is calculated by dividing height in meters by weight in kilogram. The BMI is expressed in kg/m^2 units(40). Based on a person's body weight in relation to their height, the BMI offers a straightforward and popular way to classify individuals. The BMI is classified as underweight if it is less than 18.5 kg/m^2, normal if it ranges 18.5 to 24.9 kg/m^2, overweight if it ranges 25 to 29.9 kg/m^2 and obesity if it is more than 30 kg/m^2 (40).

BMI is a fast screening method for determining a person's nutritional state. It facilitates the identification of underweight, overweight, and obesity—three vital markers of an individual's general health and nutritional status (41). A high BMI is linked to a higher chance of developing a number of chronic illnesses, such as heart disease, type 2 diabetes, hypertension, and several

types of cancer, while a low BMI could be a sign of malnourishment or underlying medical issues.(42).

2.3 Methadone induces change in BMI

Methadone induces change in BMI through several mechanisms such as increased appetite particularly sugar cravings leading to weight gain(18,43), reduced energy expenditure due to sedation effects of methadone(43,44). Methadone also reduces basal metabolic rate, making it easier to gain weight even with normal caloric intake.(44)

Several studies in Europe, America and Asia have reported significant change in BMI among patients undergoing MMT. After six months of methadone treatment, a substantial increase in BMI was observed in 96 patients who were enrolled in MMT, according to a retrospective study done in the United States (45) .

A study conducted in Canada that examined the effects of methadone treatment on body composition over the course of a year, tracking 274 patients, found that all BMI categories had significantly increased(46).A study in Lebanon pointed out drug users with substance use disorders generally have poor eating habits and nutritional status, which can result in malnutrition. After being referred for treatment, hyperphagia and unhealthy lifestyle choices that cause weight gain were observed (29).. However, such studies are limited in Africa despite presence of MMT clinics in African countries. Tanzania in particular, the MMT clinic for heroin users began in 2011 at Muhimbili National Hospital (MNH). Since then it has registered a number of clients (39). Data on the effects of methadone on metabolic syndromes does not exist. Hence, the essence of this study. The present study will establish whether age distribution, sex-specific, duration on MMT and early age on first heroin consumption are significant contributors to metabolic changes.

2.4 Absorption of lipids

Lipids, like cholesterol or triglycerides, are absorbed from the intestines and transported throughout the body via lipoproteins for energy, steroid production, or bile acid formation. Key

players in these processes include cholesterol, low-density lipoprotein cholesterol (LDL-C), triglycerides, and high-density lipoprotein (HDL). Any imbalance in these factors, whether due to organic or nonorganic causes, can result in dyslipidemia(47). Dyslipidemia is a disorder of lipoprotein metabolism, including lipoprotein over production (hyperlipidemia) or deficiency (hypolipidemia). Hyperlipidemia may be manifested by high levels of TC, LDL and TG concentration and low level of HDL concentration in the blood (48) and is associated with atherosclerosis in the process of causing CVD (49).

2.5 Determination of lipid profile

Lipid profile also known as lipid panel is laboratory blood test to measure the amount of lipid molecules in the blood. The test includes total cholesterol (TC), low density lipoprotein (LDL), high density lipoprotein (HDL) and triglycerides (50). The four fundamental lipid parameters are assessed from a fasting blood sample to determine the serum lipid profile. When someone fasts, they completely restrict their diet for 12 to 14 hours over night, excluding only water and prescription (51). The majority of reference values for serum lipids are developed on fasting blood samples, which is why fasting blood samples are preferred over non-fasting blood samples in lipid testing. Triglycerides are known to be elevated for several hours following a meal (52).

In clinical laboratories, the fasting triglycerides number is utilized in Friedewald's equation to calculate LDL (mg/dL). The Friedewald's formula (FF) is presented as LDL = (TC) - (HDL) - (TG/5).

2.5.1 Cholesterol

Cholesterol is a steroid with a secondary hydroxyl group in third -carbon position. There are numerous tissues that produce it, but the liver and intestinal walls are the main locations. In roughly 25% of cholesterol comes from food consumption, while 75% is produced in the body (53). Assays for measuring cholesterol are used to screen for the risk of atherosclerosis, diagnose and treat lipid and lipoprotein metabolic diseases, and diagnose conditions involving increased cholesterol levels(54). Burchard (1889) followed Liebermann (1885) as the first to report on cholesterol analysis. The Liebermann-Burchard reaction involves the formation of a blue-green

dye by cholesterol from polymeric unsaturated carbohydrates in a solution containing strong sulfuric acid, acetic acid, and acetic anhydride (55).

A variety of techniques have been developed to determine cholesterol levels. These techniques fall into three main categories: 1) Abell-Kendall protocol-based classical chemical procedures; 2) fluorometric and colorimetric enzymatic assays, which are widely used for automated plate readers and test kits; and 3) analytical instrumental approaches like mass spectrometry or gas and liquid chromatography. Every one of those approaches has unique advantages. Although they take multiple steps, classical chemical processes are very easy and inexpensive to perform. The employment of expensive enzymes is a requirement for enzymatic assays, despite often low limits of detection (LOD). The most precise and sensitive techniques are mass spectrometry and chromatography, but they need expensive equipment and a thorough sample preparation (56). This study is interested with enzymatic assays method.

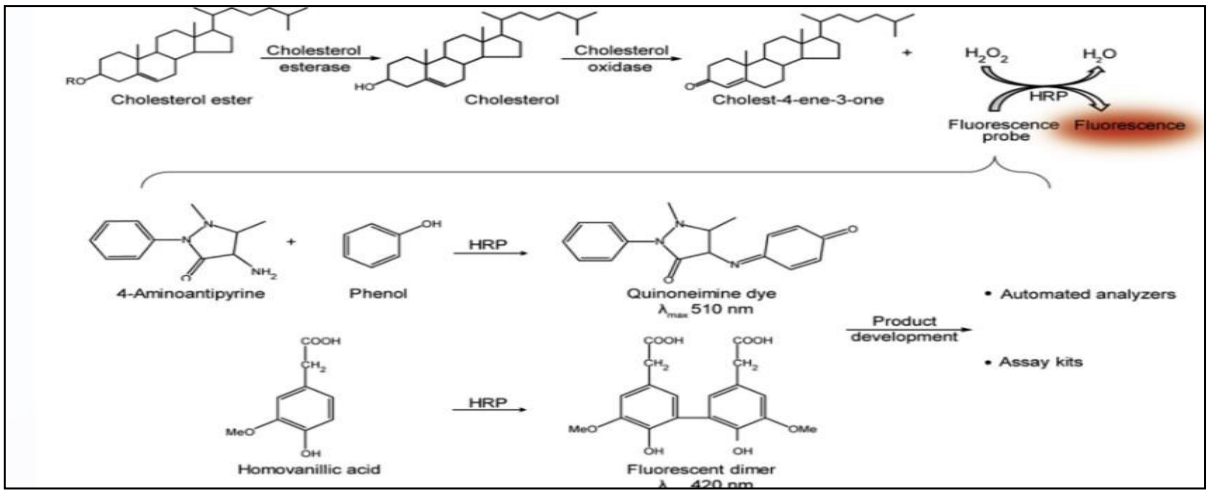


Figure 2: The scheme of the enzyme-coupled reaction of fluorometric enzymatic assay. (From Li H et al, 2019) (56)

Enzymatic assays use the enzyme-coupled reaction as its foundation to measure total cholesterol levels. In 1974, the first enzymatic test to quantify serum cholesterol was introduced (56). From that point on, enzymatic tests became readily available, and now they are utilized extensively for both automated analyzers and assay kits (57).

The enzymatic, colorimetric method of cholesterol determination rely on the following test procedures: first, cholesterol esterase hydrolyzes esterified cholesterol to release cholesterol (see Figure 2.) (58,59). Cholesterol oxidase then converts the free cholesterol to cholest-4-en-3-one. This reaction yields hydrogen peroxide as a byproduct, which is readily detectable using highly sensitive colorimetric or fluorometric probes. Since several fluorometric probes have been created, enzymatic tests are now a widely used method. The rise in absorbance at 512 nm is used to measure the color intensity of the dye produced, which is directly proportional to the cholesterol concentration (54).

2.5.2 Low density lipoprotein cholesterol

Low density lipoproteins (LDL) are a major contributor to the development of atherosclerosis and its progression (60). A variety of lipolytic enzymes work to convert VLDLs (Very Low Density Lipoproteins) high in triglycerides into LDLs, which are then produced in the liver. Specific LDL receptors on liver parenchymal cells are the primary means by which LDL is removed from plasma. Increased LDL concentrations in blood, along with a longer residence time and a higher rate of biological modification, lead to the breakdown of endothelial function and increased uptake of LDL-cholesterol by smooth muscle cells in vessel walls and the monocyte/macrophage system.(61).

LDL is the primary source of cholesterol that is accumulated in atherosclerotic plaques. When it comes to coronary atherosclerosis, the LDL-cholesterol value is the most potent clinical predictor of all the individual indicators. Therefore, the main goal of lipid reduction therapy is to lower LDL cholesterol, which is subsequently translated into improved endothelial function, prevention of atherosclerosis and slowing down the disease's course, and prevention of plaque rupture (60,62,63).

Numerous techniques, such as ultracentrifugation, lipoprotein electrophoresis, HPLC, and precipitation procedures, can be used to measure LDL-cholesterol. Precipitation is the process of employing polyvinyl sulfate, dextran sulfate, or polycyclic anions to precipitate LDL cholesterol that contains apolipoprotein B. Although precipitation methods are time-consuming and prone to interference, Lipid Research Clinics advise combining these techniques using

polyanions in the presence of divalent cations (64).An approach that has gained popularity recently measures LDL-cholesterol after immunoadsorption and centrifugation of the material (65,66).

Friedewald's formula, which is frequently used to calculate LDL-cholesterol content, can produce deceptively low values even when chylomicrons or aberrant lipoproteins are present in modest numbers. This underscores the need for a more straightforward, trustworthy procedure that does not require any prior processes (61). The homogeneous enzymatic colorimetric assay is a blood LDL-cholesterol measurement technique that is automated. It raises cholesterol reactivities in the following order: HDL < chylomicrons < VLDL < LDL (66). It does this by interacting with lipoproteins through a nonionic detergent and sugar component. The presence of Mg++ reduces the reaction.

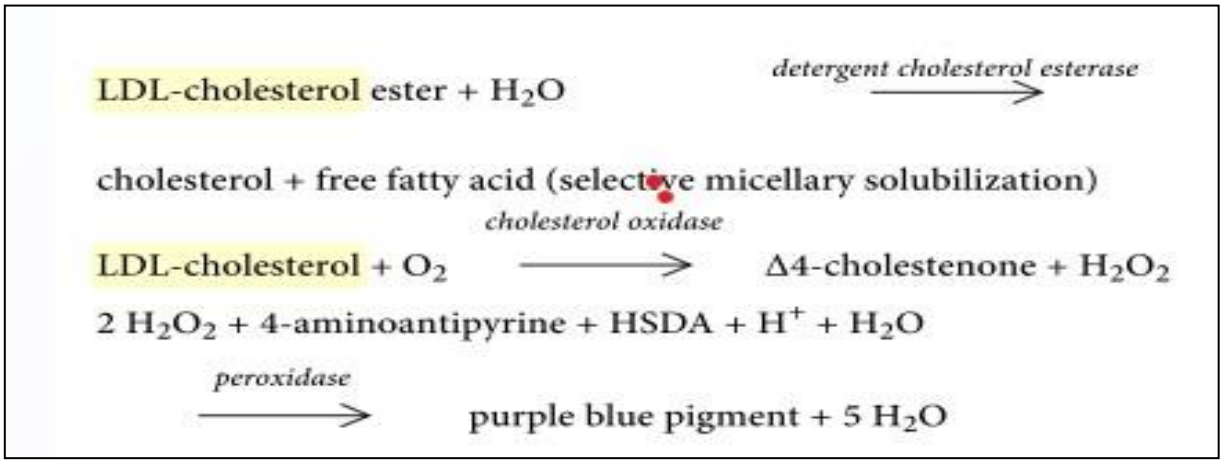


Figure 3: Represents a series of reactions to determine LDL (Taken from Gen E, 2017) (64).

Using an enzyme approach that uses cholesterol oxidase and cholesterol esterase in the presence of surfactants that solubilize just LDL, cholesterol esters and free cholesterol in LDL are quantified (see Figure 3) (64). Surfactants and a sugar molecule block the enzyme responses to lipoproteins other than LDL. The amount of cholesterol present in chylomicron, VLDL, and HDL is not determined. LDL-cholesterol concentration is directly correlated with the color intensity of the blue quinoneimine dye that forms. Measurement of the increase in absorbance at 583 nm is used to determine LDL (61).

2.5.3 High density lipoprotein

High density lipoproteins (HDL) transport cholesterol from peripheral cells to the liver, converting it into bile acids and excreted into the intestine. Monitoring serum HDL-cholesterol concentration is crucial as it correlates negatively with atherosclerotic disease risk (67).

There are numerous techniques for calculating HDL-cholesterol, including direct approaches, precipitation-based techniques, HPLC, electrophoresis, and ultracentrifugation. Among these, the direct approaches are frequently employed (68). Numerous methods have been suggested for the direct detection of HDL-cholesterol in serum, such as the employment of polyethylene glycol (PEG) with anti-apoprotein B and anti-apoprotein CIII antibodies and magnetically responsive particles as polyanion-metal combinations (69). PEG-modified enzymes and dextran sulfate are used in this automated approach for the direct measurement of HDL-cholesterol in serum and plasma. The PEG modification of cholesterol esterase and cholesterol oxidase enzymes results in specific catalytic activities directed at lipoprotein fractions. Theorder of increasing reactivity is as follows: LDL < VLDL ≈ chylomicrons < HDL(70). Homogeneous enzymatic colorimetric test is used to determine the serum HDL concentration. In this method, first, dextran sulfate preferentially combines with LDL, VLDL, and chylomicrons to form water-soluble complexes that are resistant to PEG-modified enzymes when magnesium sulfate is present. Then, enzymatic measurements of HDL-cholesterol concentration are made using cholesterol esterase and cholesterol oxidase in conjunction with PEG to the amino groups. Cholesterol esterase quantitatively breaks down cholesterol esters into free cholesterol and fatty acids.When oxygen is present, cholesterol oxidase oxidizes cholesterol to Δ4-cholestenone and hydrogen peroxide while peroxidase catalyses the reaction between hydrogen peroxide with 4-amino-antipyrine and sodium N-(2-hydroxy-3-sulfopropyl)-3,5-dimethoxyaniline reagents HSDA to form a purple-blue dye. The color intensity of this dye is positively correlated to the cholesterol concentration and isdetermined photometrically (67,68,70,71).

2.5.4 Triglycerides

Triglycerides (TG) are the main lipids in human plasma, formed in the intestinal mucosa by esterification of glycerol and free fatty acids. They constitute 95% of adipose tissue lipids and are associated with high risk of severe atherosclerosis, diabetes, nephrosis, and metabolic disorders (72). Enzymatic colorimetric method with glycerol phosphate oxidase and 4-aminophenazone (GPO/PAP is used to determine serum TG in the laboratory.

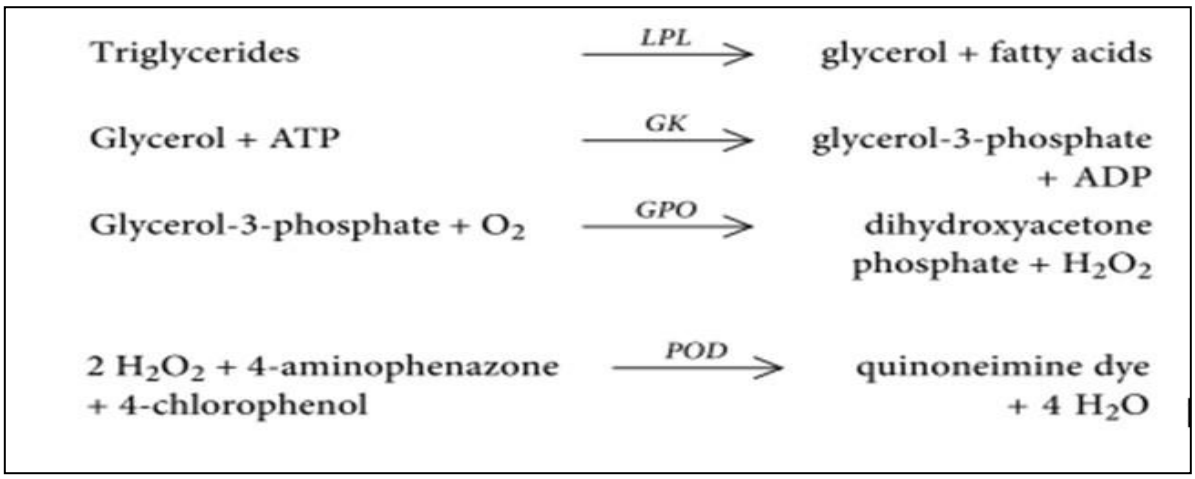


Figure 4: Catalytic reactions in determining triglycerides. (Taken from Cassette C, 2017) (72)

As shown in figure 4, the determination of triglyceride levels involves a series of enzymatic reactions, starting with lipoprotein lipase (LPL) hydrolyzing triglycerides into glycerol and fatty acids. Glycerol kinase (GK) then phosphorylates glycerol to glycerol-3-phosphate, which is oxidized by glycerol phosphate oxidase (GPO) to produce dihydroxyacetone phosphate and hydrogen peroxide (H_2O_2). The H_2O_2 enables the oxidative coupling of 4-chlorophenol and 4-aminophenazone, catalyzed by peroxidase (POD), forming a red quinoneimine dye whose absorbance at 512 nm correlates with the triglyceride concentration (72).

2.5 Clinical interpretation of lipid profile

The criteria for clinical interpretations of lipid determination values is set by the United States National Cholesterol Education Program Adult Treatment Panel III (NCEP ATP III) (73) established criteria for determining the lipid profile. Table 1 shows the classification of total

cholesterol (TC), low density lipoprotein cholesterol (LDL), high density lipoprotein cholesterol (HDL), and triglycerides (TG).

Table 1: Classification of lipid profile for clinical interpretation

Lipid profile	Clinical interpretation	Range of values mg/dL	Lipid disorder
TC	Desirable	<200	
	Borderline High	200-239	
	High	≥240	Yes
LDL	Optimal	<100	
	Near/above optimal	100-120	
	Borderline high	130-159	
	High	160-189	Yes
	Very high	≥190	Yes
HDL	Low (undesirable)	<40 (males)	
	Low (undesirable)	<50 (females)	
	High	≥ 60 (both)	Yes
TG	Low	<150	
	Borderline high	150-199	
	High	200-299	Yes
	Very high	≥ 500	Yes

This table adopted from Fremont R et al, 1963 (49)

2.6 Methadone and Lipid Metabolism

Methadone is narcotic analgesic drug, once used modifies the nerve system's and brain's reaction to pain. It has been used since the second world war and in the maintenance treatment of opiate addicts (74). Methadone retains certain pharmacological effects such as antitussive activity and it is being used because has little effects on the individual compared to other opioids (32). The fact that methadone is metabolized by the liver(32,74) and it is being used as first line treatment among individuals with opioid use disorders has found to impair lipid profile.

There are several ways in which methadone maintenance treatment (MMT) affects lipid metabolism, and these effects may involve BMI alterations. The effects of methadone on lipid metabolic pathways and, in turn, body mass index are explained in depth as follows:

2.6.1 Mechanisms how methadone influence lipid metabolisms

(i) Opiod Receptor interaction and Hormonal Modulation

Methadone primarily acts on mu-opioid receptors (MORs) in the central nervous system, which triggers downstream effects on metabolic pathways by influencing the hypothalamic-pituitary-adrenal (HPA) axis(75). This activation leads to changes in the secretion of hormones such as cortisol and insulin, which are key regulators of lipid metabolism, affecting lipid synthesis, storage, and mobilization(76).

***(*ii) Insulin and Glucose Metabolism**

Methadone may lead to insulin resistance, affecting glucose uptake and increasing circulating lipid levels(26). This resistance impairs the body's ability to use glucose effectively, causing excess glucose to be converted into triglycerides. Elevated insulin levels can also promote lipogenesis (fat synthesis) in the liver, contributing to dyslipidemia (an imbalance in lipid levels) (76).

(iii) Effects on Lipid Enzymes and Transporters

Methadone can alter the activity of enzymes involved in lipid metabolism, such as lipoprotein lipase (LPL) and hormone-sensitive lipase (HSL)(14). LPL is responsible for hydrolyzing triglycerides into free fatty acids for tissue uptake, and methadone can reduce LPL activity, leading to elevated triglycerides in the blood(76). Additionally, HSL, which plays a role in breaking down stored fat (adipose tissue) into fatty acids, can have its activity altered by methadone, affecting lipid mobilization and storage.

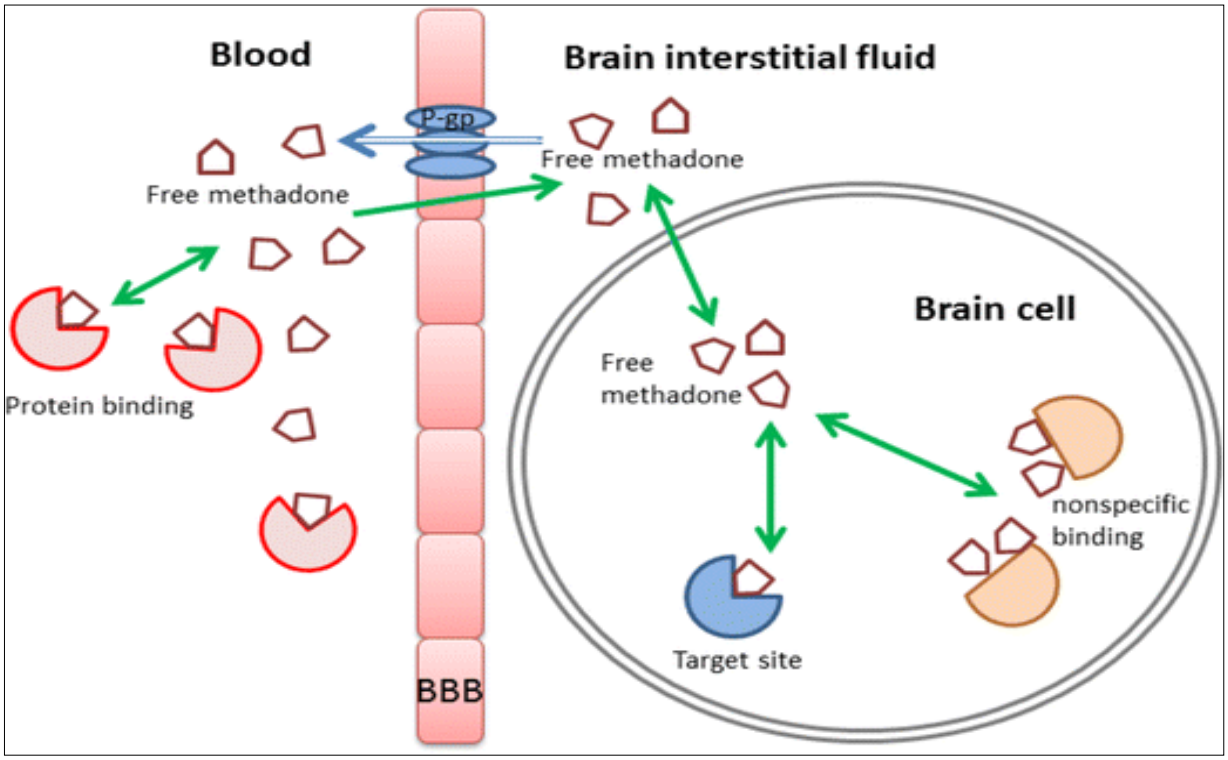


Figure 5: Pathway of methadone exchange within the brain cell. (Source: Holm K et al, 2016) (75).

(iv) Effects on Cholesterol and Lipoproteins.

Methadone treatment has been associated with elevated levels LDL cholesterol and reduced levels of HDL cholesterol(2), creating an atherogenic lipid profile(77). This shift can lead to increased central adiposity, with fat accumulating around the abdomen and contributing to an increased BMI(44).

A study of individuals with heroin use disorder undergoing MMT in Barcelona (Spain), found the prevalence of low HDL and high TG to be 51.6% and 36.8% respectively (2).

Furthermore, longer methadone use was associated with low HDL and high triglyceride levels, in a study evaluating metabolic and addiction indices in patients receiving opioid medication assisted treatment (28). Drug abuse itself is associated with dyslipidemia.Opium consumption

is associated with high TC, high TG, high LDL and low HDL and the average prevalence rates is 36% (19).

The duration where the effects of methadone on lipid profile can be diagnosed is still controversial, as it depends on factors such diet, smoking and presence or absence of other co-morbidities. However, metabolic changes can be detected after six months of methadone initiation, by this time the individual also gains weight (46). Studies on lipid profile among clients undergoing MMT in Africa are scarce and their lipid profile status of this population is not known.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Type of study

This was a retrospective cohort with a cross-sectional element and hospital-based study. Clients with heroin use addiction who were attending MMT clinic from January 2014 to January 2024 at Muhimbili National Hospital (MNH) in Dar es Salaam were recruited in the study. Data were collected from participants based on their exposure duration to methadone.

3.2 Study population

Patients visiting MMT clinic at Department of Psychiatry and Mental Health at Muhimbili National Hospital (MNH). This population is at risk of experiencing effects of methadone treatment on BMI and lipid profile.

3.3 Study area

The study was conducted at the Department of Psychiatric and Mental Health at MNH in Dar es Salaam. It is an out-patients cost free clinic where medically assisted therapy and methadone as an opioid agonist drug available are being provided to clients with heroin use addictions under supervision of medical personnel's. Clients attend the clinic daily to receive methadone orally. MNH is the largest and oldest hospital to provide MMT services in Tanzania, with an estimate of about 3000 clients benefited since its establishment on 2011. On average 900-1000 clients attend clinic every day.

3.4 Sample size

The predetermined sample size for this study was 100 participants and selected based on their exposure time on methadone. The reason was to investigate if prolonged exposure to methadone is associated with changes in BMI and lipid profile among clients on MMT. The following formula was used to determine sample size for this study (78).

$$n = N \times \frac{\frac{Z^2 \times p \times (1 - p)}{e^2}}{\left[N - 1 + \frac{Z^2 \times p \times (1 - p)}{e^2}\right]}$$

Where

n= required sample size

N = population size. In this study, refers to the total number of clients attending MMT at MNH, which is estimated to be 1000 clients.

Z =this refers to the level of confidence that the results revealed by this study will be accurate. For 95% confidence level Z = 1.96.

p = Sample proportion from previous surveys. The exact data for this study is not known so 50% will be used.

e = Margin of error (the level of precision, sampling error). This is the range in which the true value of the population is estimated to be. 10% sampling error is estimated.

Substituting data into the formula, we get;

n = 1000 x[$(1.96^2$ x 0.5 x0.5)/0.1^2]/[900 + $(1.96^2$ x 0.5 x0.5)/0.1^2)] = 96

Therefore, the sample size for this study is 96 (approximately 100participants)

3.5 Sampling technique

Simple random sampling technique was used to select study participants who met inclusion criteria until saturation. Purposive technique was also used to minimize biasness of age, gender and education and duration on methadone use

3.6 Inclusion and Exclusion criteria

The study included adult individuals over the age of 18 years irrespective of sex, addicted with heroin use and had been registered on MMT at MNH from January 2014 to January 2024. Participants excluded from the study were critically ill, tested HIV positive and using ARVs,

diabetic patients using ant diabetic drugs, pregnant clients, clients using antidyslipidemic drugs and clients did not sign written informed consent. These individuals were excluded because of the likelihood of being confounders on BMI and lipid profile.

3.7 Study variables

In this study, the predictor variable assessed to associate with body mass index and lipid profile was time on MMT. The interest of this study was to observe the relationship between exposure duration on methadone treatment with changes in BMI and lipid levels. Other factors that were also checked were socio-demographic characteristics of the study participants, included variables such as age, sex, educational level, early age at first heroin use (FHU), and years spent on heroin use. These variables were determined via questionnaires used to interview a participant during data collection.

The outcome variables of the study were change in BMI and serum lipid profile parameters of the study participants following initiation of methadone therapy. The whole spectrum of the serum lipid panel (TC, TG, HDL and LDL) were determined in the MUHAS Clinical Research Laboratory at the department of Immunology and Microbiology by analyzing fasting blood serum samples of each participant using Cobas integra 400 machine model as per the manufacturer instructions.

3.8 Data collection method

In this study two types of data were collected: data for BMI and data for lipid profile

3.8.1 Data on body mass index

Weight and height of clients at baseline (before treatment) were extracted from the patients file (MAT file) kept in the Department of Psychiatry and Mental Health. These data were used to calculate baseline BMI (BMI before treatment) of each study participant.

During recruitment, weight and height of each client were measured using seca scale and standard board respectively, to determine post MMT BMI (BMI after treatment). The baseline

BMI and BMI after treatment were calculated through BMI calculator software. The results were used in statistical analysis.

3.8.2 Data on lipid profile

About 5mls of fasting blood samples was drawn on red top vacutainer from the antecubital vein of study participant after signing a consent form. The blood samples were kept temporarily in a portable storage container before transferred into the refrigerator in the laboratory. There after s separated serum samples were stored at -20 degrees centigrade at MUHAS clinical research laboratory until the day of the analysis. Lipid tests were carried in the labaratory using Cobas integra 400 machine Roche Diagnostics (Basle, Switzerland). In brief, about 2µl of serum samples were placed on crystal micro-tubes and thereafter stored at -8 degree centigrade.

3.9 Data analysis

Summary statistics was shown as means (standard deviation) or median (interquartile range) to describe continuous variables such as age and durations on heroin and methadone use, whereas proportions were used to describe categorical variables such as gender and educational level.

In this study, paired and unpaired t-test analysis was used. Paired t test (Dependent Two sample tests) was used to compare mean BMI of individuals calculated before and after treatment. Unpaired t-test (Independent two samples tests) was used to compare mean values of two different groups. For categorical variables such BMI classifications a Chi-square analysis was used to test the association between variables. The analysis was done using Graph Pad Prism (version 8) software. The significant level of 0.05 (i.e if $p<0.05$) was used to interpret the difference between two groups were statistically significant.

3.10 Ethical issues

The permission to conduct this study was obtained from Muhimbili University of Health and allied Sciences (MUHAS) Institutional Review Board (IRB). There after permission was obtained from the MNH management and the Department of Psychiatry and Mental health, to interact with the study participants. The MNH through its Clinical Research, Training and

Consultancy Unit granted permission to collect data. The study participants were provided a written informed consent form before taking any procedures required for this study. Only those who signed consent form were part of the study. Every piece of information provided by the study participants was kept private and participants were identified not by names but by study identification number that was only known by a researcher.

3.11 Limitations and mitigations

The challenges of this study were lack of baseline data on lipid profile of clients on MMT that led to difficulties in comparing lipid profile before and after MMT. This could be reduced through conducting long term follow-up study with periodic measurements of lipid levels of clients on MMT. The effect of gender on BMI and lipid profile was not well analyzed among heroin users because women are relatively fewer in number on heroin use.

CHAPTER FOUR

RESULTS

4.1 Socio-demographic characteristics of the study population

A total of 100 individuals on MMT attending the routine methadone clinic at MNH, Dar es Salaam were recruited into this study. As shown in Table 2, the vast majority 92 (92%) of the study participants were males and the mean age was 42 ± 9.3 years, ranging from 19 to 62 years. Seventy-eight percent of the study participants were below the age of fifty years and most of study participants (73%) education was below secondary level.

Table 2: Socio-demographic characteristics of the study participants N = 100

Variables	Frequency	Percent (%)
Age group (years)		
18-34	21	21
35-49	57	57
≥ 50	22	22
Mean age (M ± SD) years 42±9.3		
Sex		
Male	92	92
Female	8	8
Education level		
Below secondary	73	73
Secondary and above	27	27

MMT= methadone maintenance therapy, M =mean, SD= standard deviation,

4.2 Duration on heroine and methadone treatment

As shown in Table 3, study participants reported to have started using heroine from as early as 10 years, with about 19% reporting to have started using heroine below20 years, with the mean duration on heroin use of was 15 ± 5 years.

The exposure time to MMT among participants varied. Fifty-two (52%) participants were on MMT for a period of 24 months or less whereas forty eight (48%) were on MMT for a period exceeding 24 months.

Table 3: Duration on heroin use and methadone treatment of study participants N=100

Duration	Frequency	Percent (%)
Duration on MMT (months)		
≤ 24	52	52
< 24	48	48
Mean duration on MMT 41± 38 Months		
Duration on heroin use (yearrs)		
≤ 10	40	40
11 -20	43	43
≥ 21	17	17
Mean duration on HU years 14±7.8 years		
Early age at fist heroin use (years)		
10-20	19	19
21-30	41	41
≥ 31	40	40
Mean AFHU 28± 9 years		

MMT= Methadone maintenance therapy, HU= heroin use, AFHU = age at first heroin use.

4.3 The effect of MMT on BMI

Whether MMT had any effect on BMI and if this was related to the duration on MMT exposure, age, duration on heroin use and early age at first heroin use were examined. The mean BMI before and after methadone treatment were calculated, and comparisons were made using paired t-test. As shown in figure 5, the mean (± SD) BMI after methadone treatment was significantly higher than before treatment ($p<0.0001$).

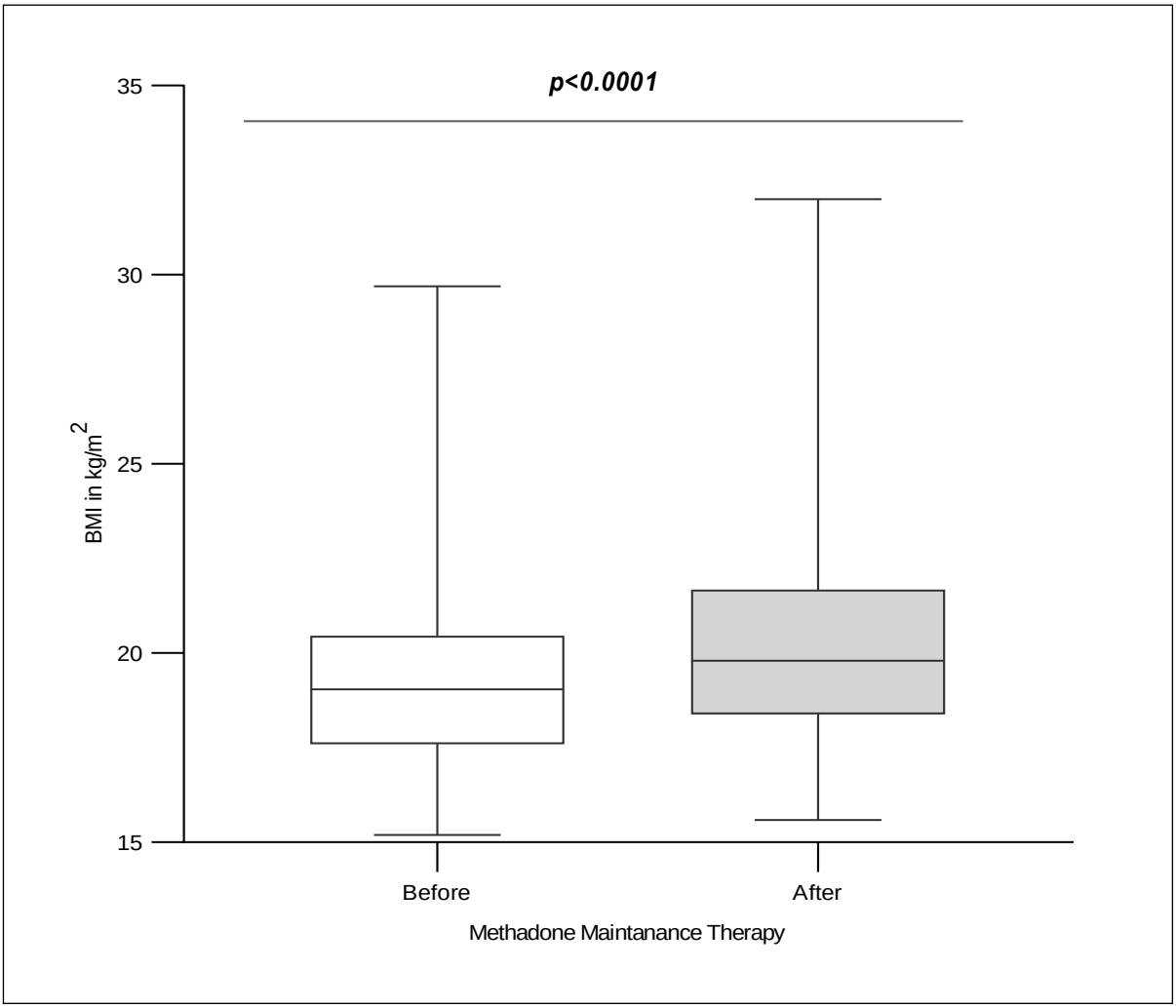


Figure 6: A box plot presenting BMI before and after methadone treatment. Before treatment 50% of the participants had BMI between 17.8 and 20.4 kg/m^2 (white box) while after treatment their BMIs were between 18.5 and 21.7kg/m^2.

4.3.1 Association between duration on MMT and BMI

We examined whether duration on MMT had any effect on BMI. As shown in table 3, fifty two study participants were on MMT for duration of 24 months or less and forty eight were on MMT for more than 24 months. Unpaired t-test statistics was performed to test if the differences in mean BMI between these two groups were significant. We noticed that at baseline (before treatment) there was no difference in the BMI between the two groups. However after methadone treatment, participants on MMT for more than 24 months had significantly higher mean BMI (21.28±2.5 kg/m^2) compared to those on MMT for less or equal to 24 months (20.05±2.5 kg/m^2),p=0.0480, t=2.00, two tailed, 95%CI =0.01075 to 2.443 kg/m^2. The mean (± SEM) increase in BMI was 1.227 ± 0.6128 kg/m^2, indicating that on average BMI is more likely to increase by 1.227kg/m^2after two years of methadone treatment.

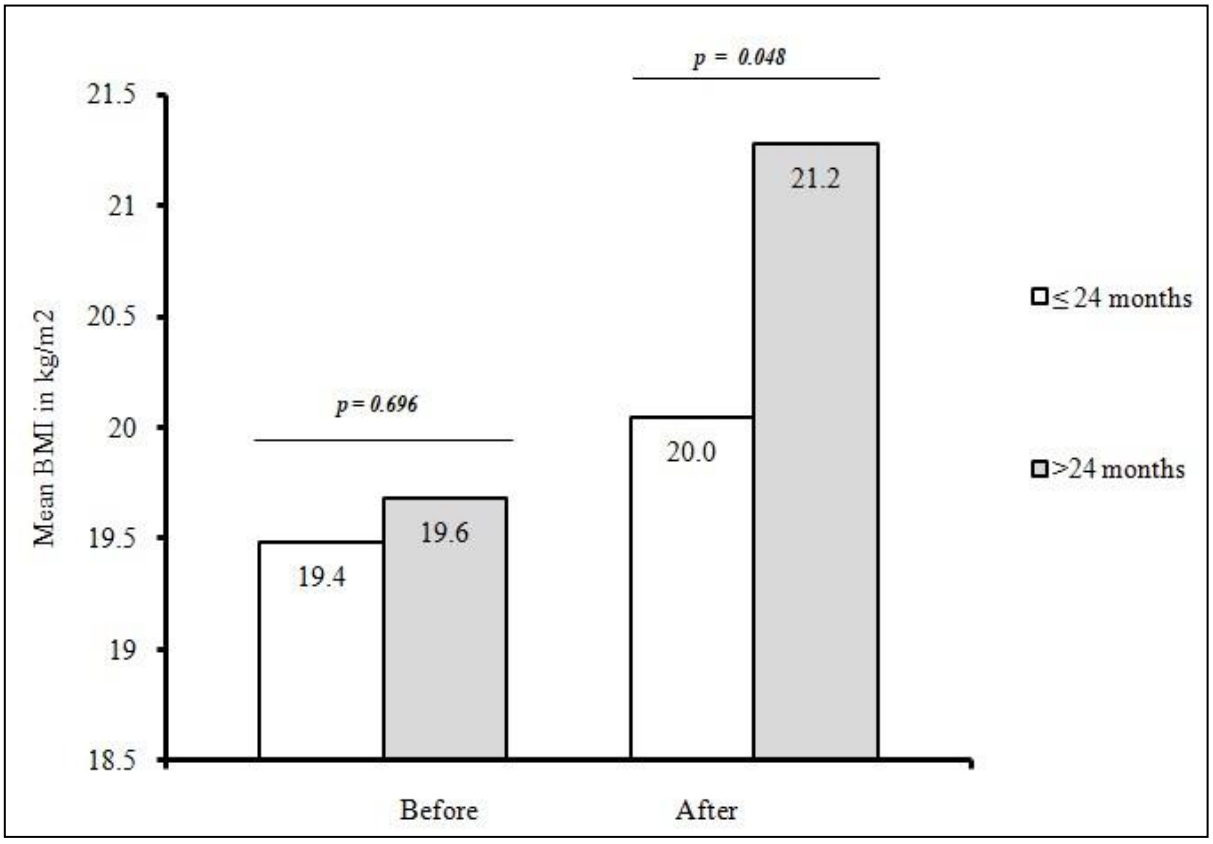


Figure 7: The relationship between BMI and duration on MMT. Before treatment the mean BMI of both groups were equal (n=52, p=0.696). After methadone treatment the mean BMI among groups on different duration on MMT were significantly different (n=48, p= 0.048).

4.3.2 Effect of age on BMI among participants on MMT

We examined whether age differences among MMT clients had any influence on BMI. We grouped participants into: aged ≤ 40 years as younger participants (n=42) and aged>40 years as older participants (n=58) and then their BMI were determined. The mean BMI of participants aged ≤ 40 years was 9.8 ± 2.7 kg/m^2 compared to 19.3 ± 2.3kg/m^2 of those aged > 40 (n=58) before MMT. After treatment, the mean BMI of participants aged ≤ 40 years remained higher and increased to 21.0 ± 3.4 kg/m^2 while those aged >40 increased to 20.3 ± 2.8 kg/m^2 . As shown in figure 8, Unpaired t test analysis revealed the difference in mean BMI between two age groups was not statistically significant after methadone use (p=0.319, t= 2.32, two tailed, 95%CI= -1.24 to 1.025 kg/m^2), suggesting that age was not a factor for changes in BMI among clients on MMT

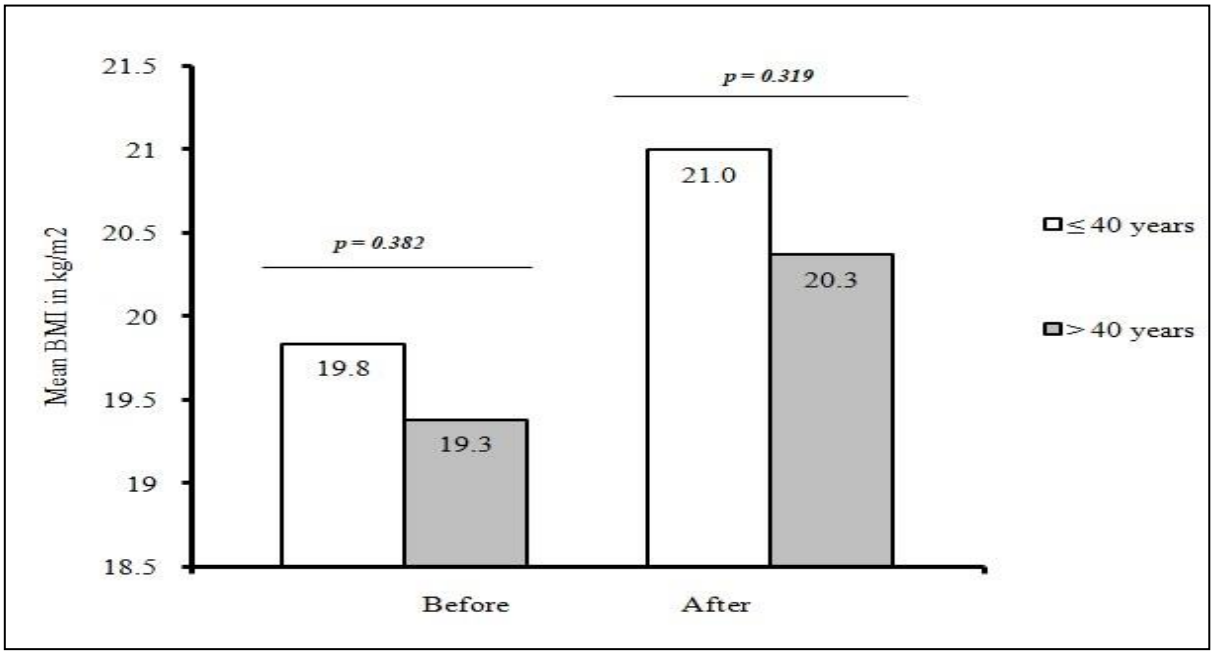


Figure 8: The association of BMI with participants' age. Open bars represent clients aged 40 years or below (n=42) and shaded bars represents clients aged above 40 (n=58). Those with young ages seemed to have higher mean values of BMI than older ones, however, it was noticed no significant difference in mean BMI between younger and older individuals on MMT (p>0.05).

4.3.3 Effect of age at first heroin use (AFHU) and duration on heroin use on BMI

(i) **Age at first heroin use (AFHU)**

We examined whether early age at first heroin use was associated with change in BMI among participants on MMT. We grouped the study participants based on the age at which they started using heroin into two groups: those started at age less than 30 years (n=58) and those who started at 30 years or above (n=42). The mean BMI of each group was calculated and analysed using unpaired t test. As shown in figure 8A, there was no significant difference in mean BMI between the two groups, p=0.360, t= 0.92, two tailed and 95% CI= -0.670 to 1.831 kg/m^2.

(ii) **Duration on heroin use (HU)**

Based on duration on previous heroin consumption, participants grouped into those on HU <15 years (n=45) and those on HU ≥ 15 years (n= 55). Using unpaired t test, we notice that the difference in mean BMI between groups after methadone treatment was statistically not significant, p=0.553, t= 0.595, two tailed, 95% CI = -1.62 to 0.87 kg/m^2. (See figure 8B)

Thus, AFHU and HU were not associated with change in BMI among participants on MMT.

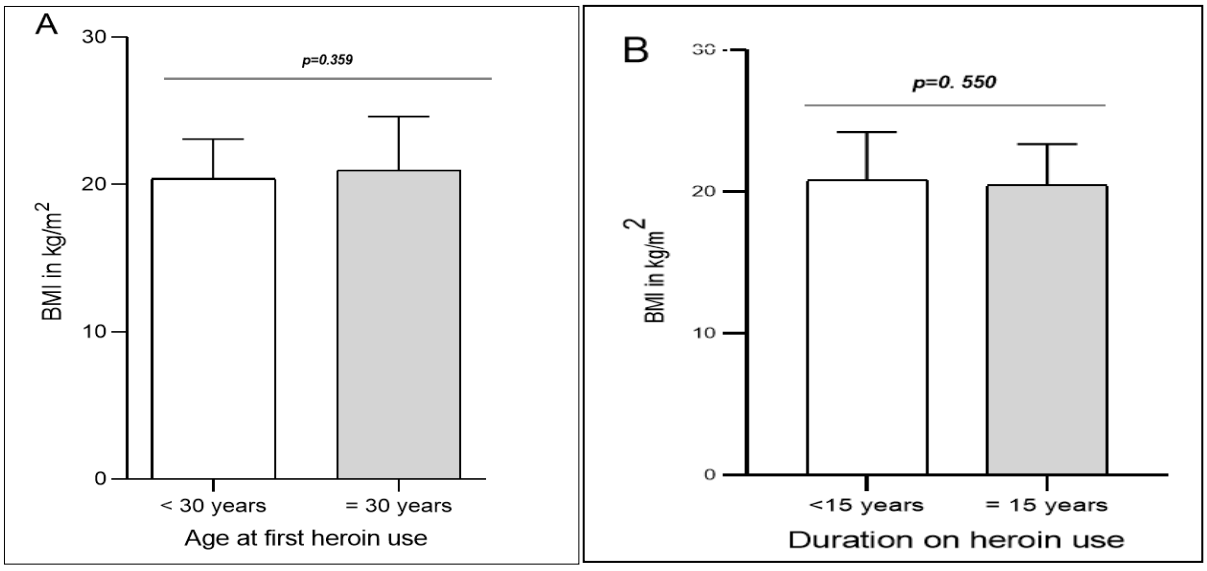


Figure 9: Association between age at first heroin use (A),and duration of heroine use(B)with BMI. Both age at first heroine use (p=0.359), and duration of heroine use(p=0.550) were not associated with changes in BMI following MMT.

4.4 Overall the use of Methadone increased clients weight

Clinically, BMI is categorized into underweight, normal and overweight/obese. To see whether MMT resulted in significant change in the clinical picture of individuals on MMT, a comparison of number of participants in these categories were made before and after MMT. Chi-square analysis showed that decrease in underweight and increase in overweight participants after methadone use was significant as shown in figure 9 ($p=0.010$, $X^2= 8.96$, $df= 2$). This indicates that clinically, methadone treatment improves the BMI of clients and thus overall health.

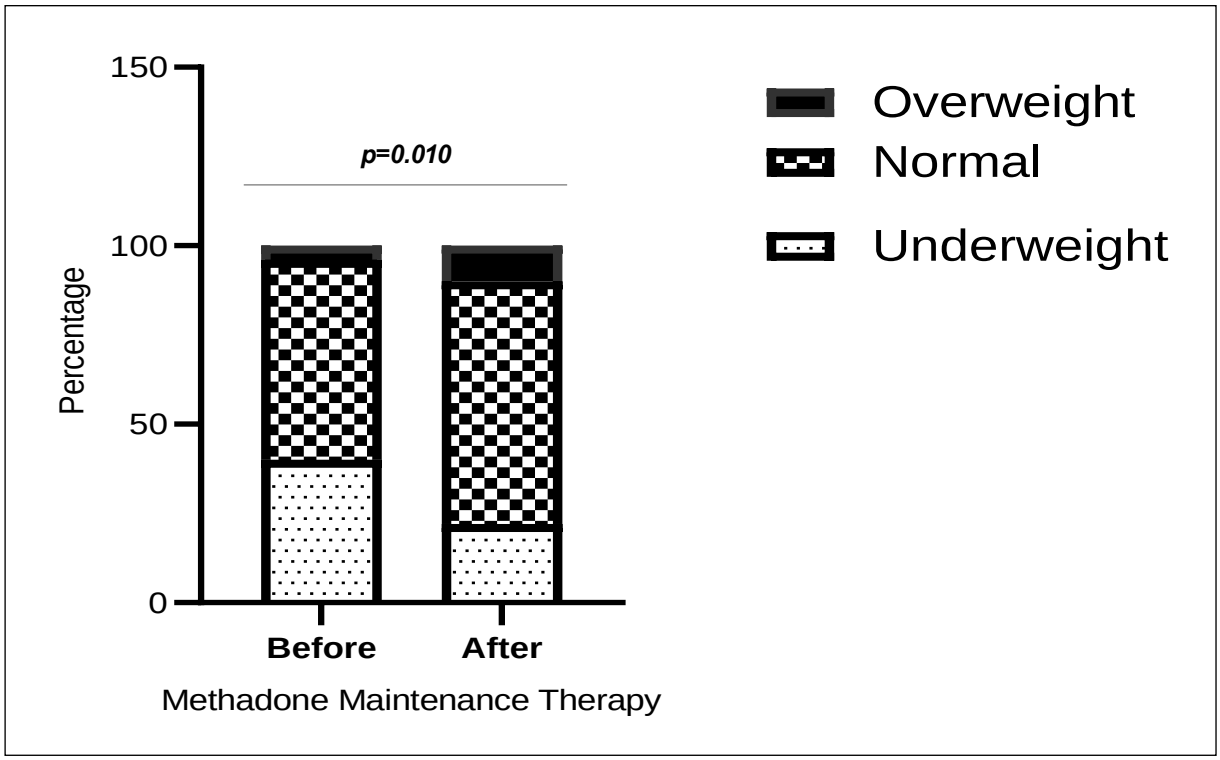


Figure 10: The proportions of participants BMI before and after MMT. Before methadone treatment, 40% of participants were underweight; and 56% participants were normal BMI. However, following methadone treatment the underweight decreased to 22%, and the proportional of normal individuals increased to 68%. Chi-square analysis of BMI before and after treatment were statistically significant with ($p<0.001$).

4.5 The effect of MMT on lipid profile

We determined lipid profile of 100 study participants on MMT and presented as proportions of abnormal levels of TC, LDL, HDL and TG (figure 10). Overall, 54% had one or more abnormal lipid levels. Decreased levels of HDL were most prevalent, rating 40% followed by elevated levels of TG, rating 21%. There was no baseline information on lipid levels that could have been used as bench mark for lipid analysis. However, we analyzed by comparing mean livid levels of various groups, to grasp the overall picture of their differences in lipid levels, and whether the difference is significant or not.

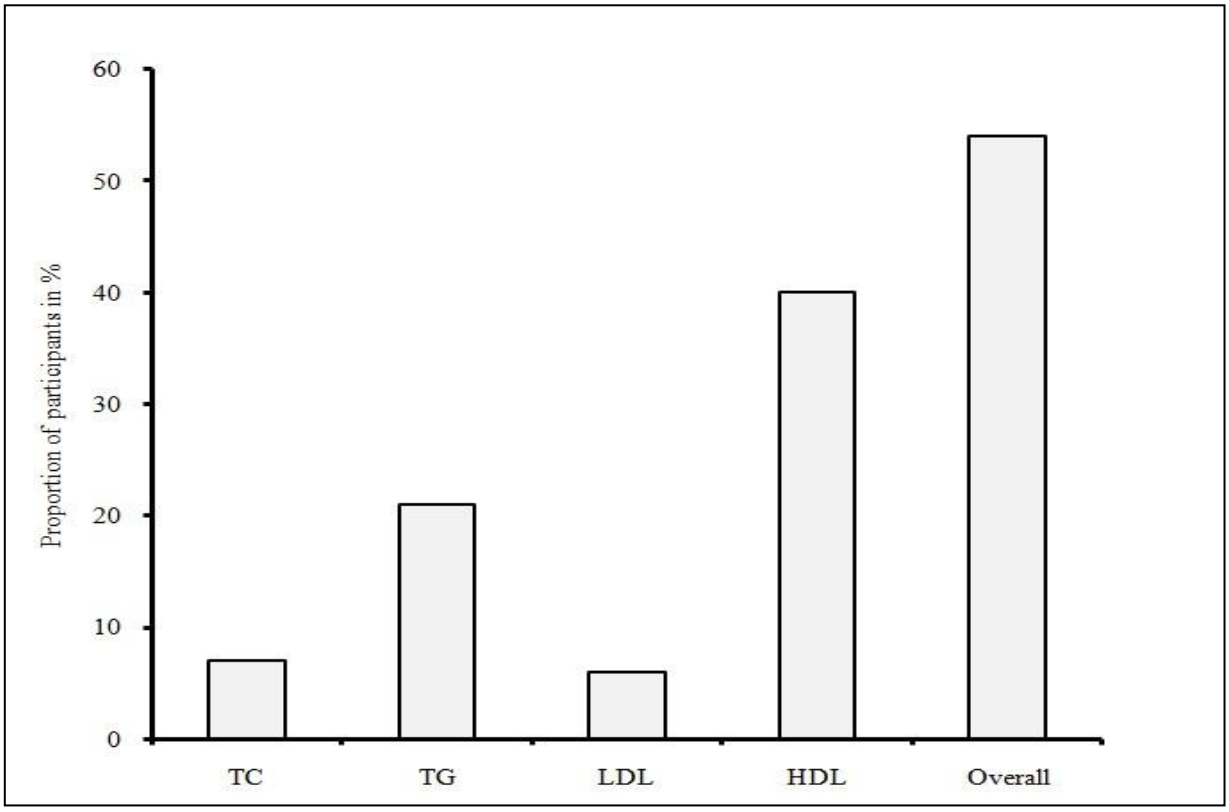


Figure 11: Illustrates the proportions of abnormal lipid levels among clients on MMT. Decreased level of HDL has the highest proportion followed by elevated levels of TG. The rates of proportions of high TC, high TG, high LDL and Low HDL are 7%, 21%, 6% and 40% respectively.

4.5.1 Association of lipid profile with MMT

The mean duration among 52%of study participants who were on MMT for 24 months or less was 12.6 ±7.9 months where as those of 48 participants who were on MMT for more than 24 months was 71.9± 33.3 months. As shown in figure 12, the mean lipid levels were high for individuals on MMT for more than 24 months except for TG. The magnitude of increase for TC, HDL and LDL were 30%, 20% and 10% respectively where as the mean TG level decreased by 10%. We observed significant difference in mean concentration of HDL (p=0.007, t =3.23) among individuals on different MMT duration. However, much cannot be concluded from these findings because the comparison is of different groups of participants.

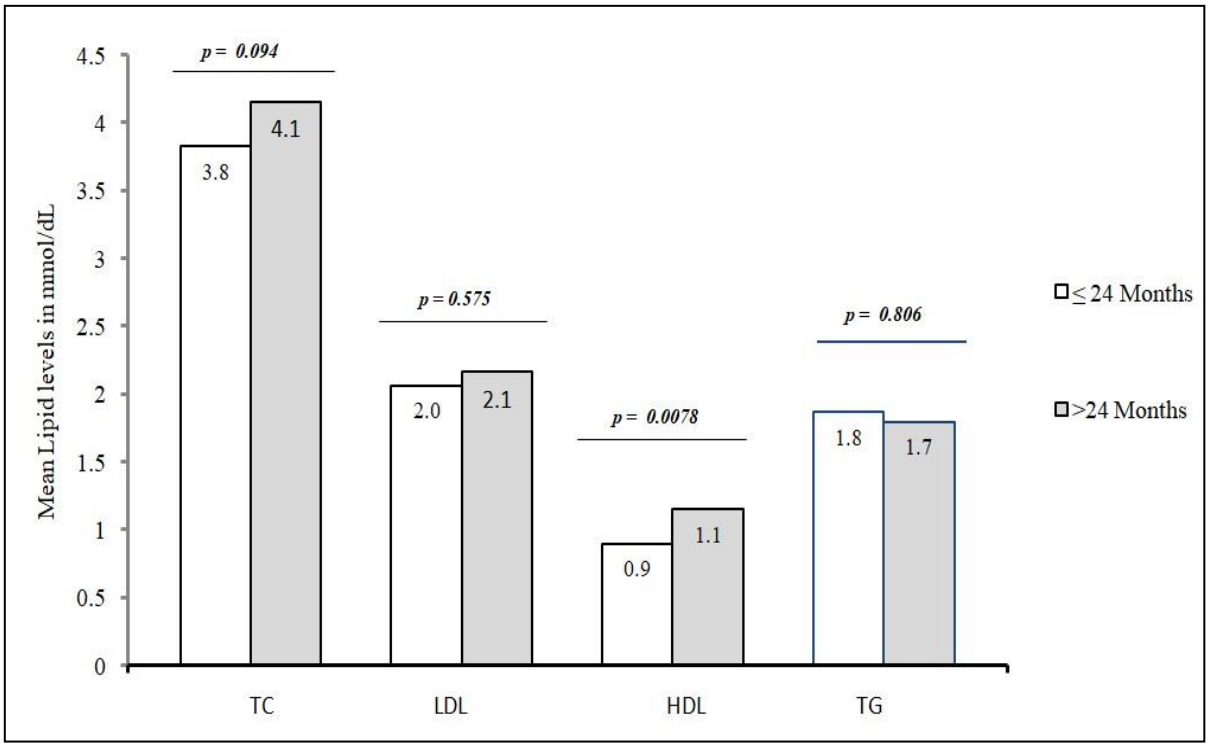


Figure 12: Illustrates the effects of duration on MMT in Lipid profile. The mean levels of TC, LDL, and HDL increase whereas TG decreases with increasing time on MMT. It was observed that the change was only significant in mean levels of HDL ($p<0.05$). Meaning individuals on MMT for more than 24 months are more likely to observe changes in the level of HDL than those with on MMT for less than 24 months.

4.5.2 The association of lipid profile and participant's sex

The mean lipid levels were different between females and males. As shown in figure 13, males had higher mean levels of TC, LDL and HDL while females had higher mean level of TG. The mean differences between females and males in mean serum concentration of LDL, HDL and TG was significant ($p<0.05$).

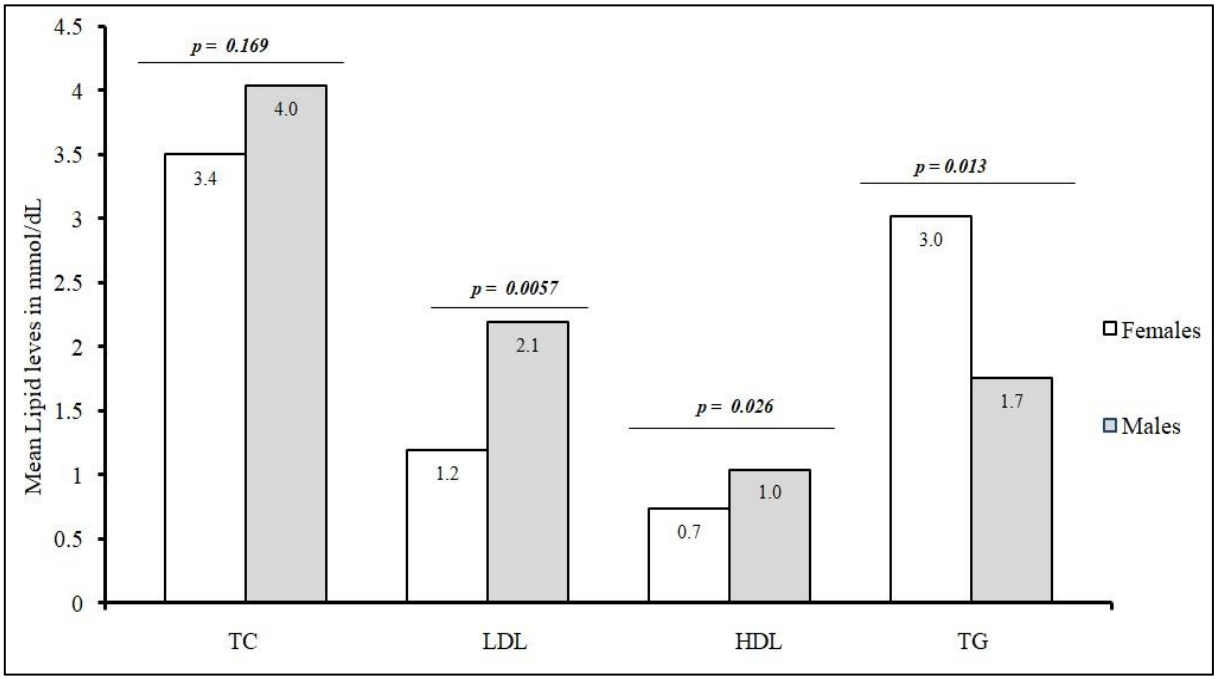


Figure 13: Illustrates the relationship between participants' sex and lipid Profile. Males (n=92) had higher mean levels of TC, LDL and HDL while females (n=8) had higher mean level of TG. The differences in mean levels of LDL, HDL and TG between males and females on MMT were statistically significant ($p<0.05$).

4.5.3 The association of lipid profile with others variables.

We examined whether age, duration on heroin use and age at first heroin use had an influence on levels of TC, LDL, HDL and TG. We observed insignificant increase in mean level of TG for individual aged above 40 years and insignificant decrease in mean level of TG among individuals who had used heroin for 15 years or more. However, age at first heroin use observed to have no significant association with the mean level of lipids

CHAPTER FIVE

DISCUSSION

This study examined the effect of MMT on body mass index and lipid profile of clients of previous heroine users, now on methadone maintenance therapy at Muhimbili National Hospital in Dar es Salaam Tanzania. Ninety two percent of the study participants were men, making up the majority of the sample. This implies a significant gender gap in Dar es Salaam's methadone treatment population. This observation aligns with findings from a study carried out in Dar es Salaam, Tanzania, to investigate the prevalence and determinants of Hepatitis C virus among a cohort of patients undergoing opiate therapy (79). The higher percentage of men is consistent with worldwide trends, since men are more likely than women to suffer from substance use problems, especially opioid addiction (80). The study participants had a mean age of 42 years, with a standard deviation of 9.3 years. The persons seeking methadone treatment ranged widely in age, from 19 to 62. The age distribution indicates that people of various ages with heroin disorder, use methadone treatment in Dar es Salaam. Seventy-eight percent of the study participants were under the age of fifty. This distribution indicates that the population in Dar es Salaam seeking methadone treatment is comparatively younger. Seventy-three percent of the study participants were not even in secondary school. Drug addiction, which affects youths, is a considerable factor contributing to school dropouts in Tanzania (81).

The study examined body mass index (BMI) of clients before (at baseline) and after MMT. The results of this study showed a significant increase in mean BMI among clients undergoing MMT. These findings suggest that MMT is associated with increase in BMI of clients investigated. The observed increase in BMI among clients on MMT aligns with existing literature that suggests methadone treatment can lead to weight gain. A substantial rise in BMI was found by Jennifer M et al, 2015 in United States ($p<0.001$) following methadone initiation (45). In the highlighted study the average body mass index rose significantly in 2 years of follow up (45).The improvement in BMI among clients on MMT may reflect better overall health and nutrition.

According to this study's findings, clients' BMIs changed significantly over the course of their MMT. The findings indicate that clients who were on MMT for more than 24 months experienced a significant increase in mean BMI. This implies that improvement in BMI is more closely linked to extended periods of methadone treatment. This finding is consistent with the results from a study by Sadek G et al 2016 on body composition changes associated with methadone treatment, which noted a statistically significant increase in BMI within one year from admission to treatment ($p < 0.05$) (46). However, we were not able to visualize significant change in mean BMI among individuals on MMT for 24 or less months (2years or less). Our sample had higher number of males.

The analysis reveals that age does not significantly affect BMI ($p=0.730$) among the participants on MMT. Despite small differences in mean BMI between the two age groups (the mean BMI for younger participants was 20.74 ± 0.62 kg/m², while that for older participants was 20.53 ± 0.62 kg/m²), the lack of statistical significance suggests that age alone may not be a critical factor in determining BMI in this population. Methadone's impact on BMI seems consistent across different age groups, indicating that the drug's metabolic effects (e.g., changes in appetite, lipid metabolism, and physical activity levels) may be similar regardless of age. This finding supports the idea that factors other than age, such as treatment duration (4,46), lifestyle (4,9,11,13), and baseline metabolic health (44,45,82), might play more significant roles in influencing BMI in MMT patients. While BMI tends to increase with age in the general population due to factors like reduced physical activity (44), changes in muscle mass (83), and hormonal shifts (76), the unique metabolic and lifestyle factors associated with MMT may override these typical age-related patterns.

The study investigated the effect of age at first heroin use (AFHU) and duration of heroin use (HU) on BMI among participants undergoing MMT. The findings suggest that neither early age at first heroin use nor longer duration of heroin use significantly influenced BMI in this population. These findings was consistent with prior studies in USA by Ji Li et al, 2017, heroine use reported to associate with weight loss, ultimately decreased BMI (84).

After MMT, 52.5% of participants who were underweight normalized their weight, while 10.7% of those with normal weight became overweight/obese, and only 2 participants regressed to being underweight. The changes in BMI categories were statistically significant ($p < 0.001$), indicating that MMT has a substantial impact on BMI. The observed trend aligns with findings from other studies, where MMT often leads to weight gain and a shift in BMI categories(46). The normalization of weight in previously underweight individuals and the increase in the proportion of overweight/obese patients are well-documented outcomes associated with methadone's effects on metabolism, appetite, and lifestyle(11,13,43,82).

The study investigated the effect of MMT on lipid profiles among 100 participants. The results revealed that 54% of participants had one or more abnormal lipid levels, with decreased HDL levels being the most prevalent (40%), followed by elevated triglycerides (21%). These findings are consistent with the results of prior study conducted in Spain by Gabriel G et al, 2018 for Caucasian population (2).Although the study did not provide a direct comparison with pre-treatment levels, it compared mean lipid levels across various groups to assess overall differences. This approach offers a snapshot of lipid abnormalities in the MMT population, reinforcing the need for regular lipid monitoring in clinical practice.

The significant increase in HDL levels in those on MMT for over 24 months is a positive outcome, as HDL helps remove excess cholesterol and lowers cardiovascular risk(28,85). The improvement may be linked to better lifestyle factors, such as enhanced nutrition and healthcare access during extended recovery. While increases in TC and LDL and a decrease in TG were observed with longer MMT duration, these changes were not statistically significant; indicating that other factors like diet(44), genetics(76), or comorbid (44) conditions may play a role in lipid levels.

CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

Heroine is one of the highly abused drugs and leads to severe debilitation of youth and loss of manpower. Introduction of methadone has resulted into positive changes on the wellbeing of these clients. The study revealed the following;

This study revealed that the use of methadone was associated with an increase in BMI, and that the longer the duration of therapy the higher the BMI.

Furthermore over50% of clients on MMT had abnormal lipid profile, particularly HDL (low) and TG (high**).**

6.2 Recommendations

Based on the benefits shown by the use of MMT in this study, we recommend expansion of MMT clinics to other urban centres in Tanzania and to consider determining lipid profile at enrolment, and during follow up. Furthermore a follow-up study comprising of larger sample size for a longer period to investigate the long term effects of methadone on lipid profile of clients attending MMT.

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APPENDICES

Appendix I: Consent Form (English Version)

No._____

Title of study

The effect of methadone maintenance treatment on body mass index and Lipid profile among clients attending MMT clinic at MNH, Dar es Salaam Tanzania.

Principal investigator

My name is Haji Ally Sangu, a postgraduate student at MUHAS for MSc. in Biochemistry. I am doing a research to examine lipid profile and body mass index among clients on methadone maintenance treatment, as part of my MSc. Study. I would like to invite you participate in this study because you meet the criteria for the study.

Use the following addresses to communicate with the researcher: E-mail sangukihunda@gmail.com. Phone number: +255 686 264 046.

Purpose of study

You are being asked to take part in a research study. Before you decide to participate in this study, it is important that you understand why the research is being done and what it will involve. Please read the following information carefully. Please ask the researcher if there is anything that is not clear or if you need more information. The purpose of this study is to examine lipid profile and body mass index of clients receiving methadone maintenance treatment.

Study procedures

I will ask you to tell me information like age, level of education, how long you are on MMT.
I will ask you to measure your height and weight when off shoes. Then you will donate about 5 mils of blood from vein of your left or right hand for laboratory lipid tests.

It will take only 5 to 10 minutes for you to complete all procedures. But results of this study can take three months or more depending on the availability of data and their analysis tools. All study procedures will take place at the department of psychiatric and mental health.

Risks

There is no risk that you will get from this study, except little pain you will feel during blood drawing due to syringe, but this is not something new to you. Blood drawing will be done by a phlebotomist.

Benefits

There will be no direct benefit to you for your participation in this study. However, we hope that the information obtained from this study may change the treatment modality you are getting now to include advices on dietary and activity**.**

Confidentiality

Your data provided in this study will be kept confidential**.** If results of this study are published or presented, individual names and other personally identifiable information will not be used. So, you will be assigned codenames/numbers that will be used in all research notes and documents.

Authorized persons such as my research supervisors may review the research data for the purpose identifying areas of improvement.

Voluntary participation

Your participation in this study is voluntary. It is up to you to decide whether or not to take part in this study. If you decide to take part in this study, you will be asked to sign a consent form. After you sign the consent form, you are still free to withdraw at any time and without giving a reason. Withdrawing from this study will not affect the relationship you have, if any, with the researcher. If you withdraw from the study before data collection is completed, your data will be returned to you or destroyed.

Consent

I have read and I understand the provided information and have had the opportunity to ask questions. I understand that my participation is voluntary and that I am free to withdraw at any time, without giving a reason and without cost. I voluntarily agree to take part in this study.

Participant's signature _____ Date _____

Investigator's signature _____ Date _____

Appendix II: Fomu Ya Ridhaa (Swahili Version)

Na._____

Athari za matumizi ya dawa ya methadone katika kuongeza uzito na profaili ya lipidi kwa wagonjwa wanaohudhuria kliniki ya methadone , MNH, Dar es Salaam Tanzania

Mtafititi mkuu

Naitwa Haji Ally Sangu, mwanafunzi wa shahada ya uzamili fani ya Biochemistry kutoka chuo cha MUHAS. Nafanya utafiti huu wa kuangalia profaili ya lipidi na indeksi ya uzito miongoni mwa watumiaji wa dawa ya methadone ikiwa ni sehemu ya masomo yangu. Ningependa nikuombe ushiriki katika utafiti huu kwa sababu unavyo vigezo vinavyohitajika kwenye utafiti. Mawasiliano yangu ni Email sangukihunda@gmail.com na simu namba 0686 26 40 46.

Lengo la utafiti

Unaombwa kushuriki katika utafiti huu , ni vizuri kabla hujafanya maamuzi ujue kwa nini utafiti huu unafanyika na nini kitahitajika. Lengo la utafiti ni kuangalia profaili ya lipidi na indeksi ya uzito miongoni mwa watumiaji wa dawa ya methadone.

Utafiti huu unafanyika ili kubaini uhusiano uliyopo kati ya dawa ya methadone na mpangilio wa aina za lipid mwilini pamoja na ongezeko la uzito kwa wanaopata matibabu. Ni muhimu kujua mienendo ya lipidi na ongezeko la uzito ili kujiepusha na magonjwa yasiyoambukizwa kama vile magongwa ya moyo na kisukari kwa kupata ushauri wa aina ya vyakula na mazoezi (kazi) ya kufanya.

Hatua za utafiti

Katika utafiti huu, nitakuuliza baadhi ya taarifa zako kama vile umri , elimu, muda uliotumia methadone . Nitakuomba pia kupima uzito pamoja na urefu na kisha nitakuomba utoe kiasi kidogo cha damu (mils 5) kwa ajili ya vipimo vya lipidi vitavyofanyika kwenye maabara.

Hatua zote zitafanyika katika idara ya afya ya akili. Na itachukua dakika tano hadi kumi kukamilisha hatua zote.

Madhara

Hakuna madhara utakayoyapata kwa kushiriki utafiti huu, isipokuwa utahisi maumivu kidogo wakati wa kutoa damu yatakayotokana na sindano. Ninaimani siyo jambo jipya kwako. Na kuna wataalam waliosomea kazi hii ya kutoa damu, hivyo unaweza pia usihisi kabisa maumivu.

Faida

Hakuna faida ya moja kwa moja utakayopata kwa kushiriki kwako kwenye utafiti huu. Hata hivyo ushiriki wako ni muhimu zaidi kwa sababu taarifa zitakazopatika zinaweza kutumika kuboresha huduma unayoipata sasa ikiwemo kupata ushauri wa aina ya vyakula vya kutumia na aina ya kazi za kufanya ukiwa unatumia hii dawa.

Usiri

Taarifa zako zote zitakuwa ni za siri kati yangu na yako. Na ikitokea matokeo ya utafiti yanatakiwa yatangazwe, jina lako na kitu chochote kinakutambulisha havitakwepo.

Watu wanaoruhiswa kuangalia matokeo ya utafiti huu ni msimamzi wangu , kwa lengo la kushauri sehemu za kurekebisha na kuboresha.

Ushiriki wa hiyari

Ushiriki wako katika utafiti huu ni wa hiyari. Unaruhusiwa kuamua au kutoamua kushiriki.
Kama utaamua kushiriki, utaombwa kusaini fomu ya ridhaa. Unaweza kuamua kutoendelea na utafiti muda wowote hata baada ya kusaini fomu ya ridhaa bila kutoa sababu yeyote na kujitoa kwako hakutoathiri mahusiano yetu tuliyokuwa nayo.

Nimesoma na kuelewa taarifa zote zilizotolewa na nimepata nafasi ya kuuliza maswali. Na kwamba nimeelewa ushiriki wangu ni wa hiyari na niko huru kujitoa muda wowote bila kutoa sababu yoyote.

Kwa hiyari yangu nimekubali kushiriki katika utafiti huu.
Saini ya mshiriki _____ Tarehe _____
Saini ya mtafiti _____ __Tarehe _____

Appendix III: Case Report Form (CRF)

Study Number …….

Hospital Reg No……

Questionnaire for participants

"THE EFFECT OF METHADONE MAINTENANCE TREATMENT ON BODY MASS INDEX AND LIPID PROFILE AMONG CLIENTS ATTENDING MMT CLINIC AT MUHIMBILI NATIONAL HOSPITAL IN DAR ES SALAM, TANZANIA"

DEMOGRAPHIC INFORMATION

1. Sex ……..(M for male, F for female)

2. Age…….

3. Educational level ……

DURATION ON METHADONE USE

4. How long have you been using methadone? .....

5. Do you use other drugs apart from methadone? Yes ( ) No ( )

If yes, what is/are the name (s) of the drug? ……..

6. How long have you been using illicit drugs?……..

HEIGHT AND WEIGHT

	Height (cm)	Weight (kg)	BMI (kg/m^2)
Baseline			
At treatment			

7. Baseline Height……..(cm). Baseline weight…….(kg) Baseline BMI…… kg/m^2

8. Height …….cm. Weight at study …….kg . BMI at a study ……....kg/m^2

LIPID PROFILE (For laboratory use)

9. Will you donate about 5mls of blood from the anterior vein for the following laboratory tests? Yes ( ) No ( ).

Lipid profile	TC	TG	LDL	HDL
Results				

Thank you for your participation.

Appendix IV: Approval for ethical clearance

UNITED REPUBLIC OF TANZANIA

MINISTRY OF EDUCATION, SCIENCE AND TECHNOLOGY

MUHIMBILI UNIVERSITY OF HEALTH AND ALLIED SCIENCES

OFFICE OF THE DIRECTOR – RESEARCH, PUBLICATIONS AND INNOVATION

Ref. No.DA.282/298/01.C/2039 Date: 07/02/2024

MUHAS-REC-02-2024-2039
Haji Ally Sangu
College of Medicine,
MUHAS

RE: APPROVAL FOR ETHICAL CLEARANCE FOR A STUDY TITLED: LIPID PROFILE AND BODY MASS INDEX AMONG CLIENTS ON METHADONE MAINTENANCE TREATMENT AT MUHIMBILI NATIONAL HOSPITAL IN DAR ES SALAM, TANZANIA

Reference is made to the above heading,

I am pleased to inform you that the Chairman has on behalf of the University Senate, approved ethical clearance of the above-mentioned study, on recommendations of the Senate Research and Publications Committee meeting accordance with MUHAS research policy and Tanzania regulations governing human and animal subjects research.

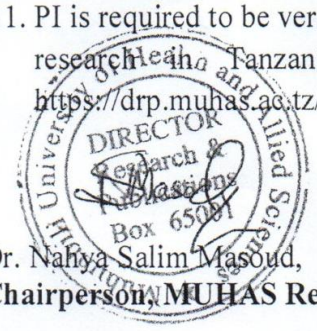
APPROVAL DATE: 07/02/2024
EXPIRATION DATE OF APPROVAL: 07/02/2025

STUDY DESCRIPTION:
Purpose:
The purpose of this case study is To examine the lipid profile and body mass index among clients on Methadone maintenance treatment at Muhimbili National Hospital in Dar es salam, Tanzania.

The approved protocol and procedures for this study is attached and stamped with this letter, and can be found in the link provided:
https://irb.muhas.ac.tz/storage/Certificates/Certificate%20-%2015143.pdf and in the MUHAS archives

The PI is required to:

1. Submit bi-annual progress reports and final report upon completion of the study.
2. Report to the IRB any unanticipated problem involving risks to subjects or others including adverse events where applicable.
3. Apply for renewal of approval of ethical clearance one (1) month prior its expiration if the study is not completed at the end of this ethical approval. You may not continue with any research activity beyond the expiration date without the approval of the IRB. Failure to receive approval for continuation before the expiration date will result in automatic termination of the approval for this study on the expiration date.
4. Obtain IRB amendment (s) approval for any changes to any aspect of this study before they can be implemented.
5. Data security is ultimately the responsibility of the investigator.
6. Apply for and obtain Data Transfer Agreement (DTA) from NIMR if data will be transferred to a foreign country.
7. Apply for and obtain Material Transfer Agreement (MTA) from NIMR, if research materials (samples) will be shipped to a foreign country.
8. Apply for and obtain permission to publish the results from National Institute for Medical Research. The application should be before submitting to a publisher and not concurrently through the email publications@nimr.or.tz.
9. Any researcher, who contravenes or fail to comply with these conditions, shall be guilty of an offence and shall be liable on conviction to a fine as per NIMR Act No. 23 of 1979, PART III section 10 (2)
10. The PI is required to ensure that the findings of the study are disseminated to relevant stake holders.
11. PI is required to be versed with necessary laws and regulatory policies that govern research in Tanzania. Some guidance is available on our website https://drp.muhas.ac.tz/.



Dr. Nahya Salim Masoud,
Chairperson, MUHAS Research and Ethics Committee

Appendix V: Permission to collect data at MNH

THE UNITED REPUBLIC OF TANZANIA

MINISTRY OF HEALTH

MUHIMBILI NATIONAL HOSPITAL

In reply please quote:

Ref. No.: MNH/CRTCU/Perm/2024/081 Date: 12th February, 2024

Head of Department,
Psychiatry and Mental Health
Muhimbili National Hospital

RE: PERMISSION TO COLLECT DATA AT MNH

Name of Student	Haji Ally Sangu
Title	**"Lipid Profile and Body Mass Index Among Clients on Methadone Maintenance Treatment at MNH Dar es Salaam Tanzania."**
Institution	Muhimbili University of Health and Allied Sciences
Supervisor	Dr. T. Mselle (MUHAS)
Co – supervisor	Dr. I. Haruna (MNH)
Period	12th February 2024 to 30th March, 2024

Approval has been granted to the above principal investigators to collect data at MNH. Dissemination of study findings at MNH shall be overseen by **Dr. I Haruna** (MNH) supervisor.

Kindly ensure the named principal investigator abide to the ethical principles and other conditions of the research approval.

Sincerely,

Dr. Charles K. Pallangyo
For: Head of Clinical Research, Training and Consultancy Unit

c.c DMS
c.c Dr. I. Haruna
c.c Haji Ally Sangu

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