Prevalence and determinants of hyperlactatemia among HIV-infected patients on combination anti-retroviral therapy in Ahero and Thika District Hospital

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DECLARATION

This thesis is my original work and has not been presented for a degree in any other University.

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DEDICATION

I dedicate this thesis to my beloved wife Susan Wanjiru and my daughter Victoria Wanjiru for the love and support they have given me throughout my study and particularly during the writing of this thesis. I must also thank my loving mother who sacrificed her all in order to ensure I get a decent education.

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF APPENDICES

LIST OF ABBREVIATIONS AND ACRONYMS

ABC	Abacavir
AIDS	Acquired Immunodeficiency Syndrome
ALT	Alanine Transaminase
ARV	Antiretroviral drugs
AZT	Azidothymidine (Zidovudine)
cART	Combination Antiretroviral Therapy
CCC	Comprehensive Care Clinic
CD	Compact disk used for storage of data
CD4 Count	Absolute count of CD4 T lymphocyte subset of white blood cells
CME	Continuous medical education
CPK	Creatinine Phosphokinase
D4T	Stavudine
DDI	Didanosine
DH	District Hospital
HAART	Highly Active Antiretroviral Therapy
HCO_3	Bicarbonate
HCW	Health care workers
HIV	Human Immunodeficiency Virus
JKUAT	Jomo Kenyatta University of Agriculture and Technology
KEMRI	Kenya Medical Research Institute
KES	Kenya shillings
MtDNA	Mitochondrial Deoxyribo-Nucleic Acid

NRTIs	Nucleoside reverse transcriptase inhibitors
PASW	Predictive Analytics Software (formerly called SPSS)
PI	Principal Investigator
PL/r	Boosted protease inhibitors e.g. Kaletra.
PLWH	People Living with HIV
SOP	Standard Operating Procedure
SPSS	Statistical Package for Social Scientists
TDF	Tenofovir
WHO	World Health Organization
µg	Micrograms

ABSTRACT

Nucleoside reverse transcriptase inhibitors (NRTIs) form the backbone of highly active antiretroviral therapy (HAART) in resource limited settings. Nucleoside reverse transcriptase inhibitors are preferred for use due to their low cost, ease of availability in fixed dose combination, ease of administration, and minimum interaction with food. However, they have been shown to cause mitochondrial toxicity resulting in drug toxicities including peripheral neuropathy, lipodystrophy and hyperlactatemia. The objective of this study was to determine the prevalence of hyperlactatemia and the associated risk factors among HIV-infected patients on combination antiretroviral therapy attending HIV clinics at Thika and Ahero District hospitals. This was a descriptive cross-sectional study and systematic sampling technique was used where all the eligible patients attending the two HIV clinics during the study period who consented to the study participated. A structured questionnaire was administered before collection of a blood sample to measure the lactate level. The overall prevalence of hyperlactatemia (lactate $\geq$2.5 mmol/l) in the population of HIV-infected patients on HAART was 41% of whom 3.1% had moderate (lactate $\geq$2.5-5 mmol/l) hyperlactatemia. Using multivariable logistic regression models the risk factors for hyperlactatemia was found to be stavudine- containing regimen, female gender, BMI of $\geq$ 24, a CD4 count of less than 200 at initiation of HAART and being on NRTIs for a period of more than 24 months. The study found that 80% of the patients with hyperlactatemia were symptomatic. The signs and symptoms included fatigue, muscle aches, abdominals symptoms, headache, and paraesthesia which were not significant. Because of the high prevalence of hyperlactatemia and the non-specificity of the clinical presentation, lactate meters should be availed to aid in accurate diagnosis of hyperlactatemia.

CHAPTER ONE

1.0 INTRODUCTION

1.1 BACKGROUND INFORMATION

Hyperlactatemia is defined as a persistent, mild to moderate increase in blood lactate concentration without metabolic acidosis, whereas lactic acidosis is characterized by persistently by increased blood lactate levels (usually >5mmol/l) in association with metabolic acidosis. Metabolic acidosis is defined as a state of decreased pH resulting from either a primary increase in hydrogen ions or a reduction in bicarbonate concentration.

Use of highly active antiretroviral therapy (HAART) since early 1990's, has been an important step in the fight against human immunodeficiency virus (HIV) infection worldwide. HAART refers to the use of at least three antiretroviral drugs (ARVs) to achieve maximal and durable viral suppression which results in immune reconstitution and a reduction in morbidity and mortality due to HIV infection. For maximal viral suppression, the three drugs should be selected from at least two classes of ARVs in order to inhibit the replication cycle at two different stages. HAART has provided extraordinary clinical benefit in HIV-infected patients by lowering morbidity and mortality (Palella *et al.*, 1998).

Three classes of antiretroviral drugs are widely used, namely nucleoside reverse transcriptase inhibitors (NRTIs), non-nucleoside reverse transcriptase inhibitors (NNRTIs),

and protease inhibitors (PIs). NRTIs inhibit reverse transcription by being incorporated into the newly synthesized viral DNA and preventing its further elongation (e.g. stavudine, zidovudine, abacavir, lamivudine, and emtricitabine). NNRTIs inhibit reverse transcriptase directly by binding to the reverse transcriptase enzyme and interfering with its function. They include nevirapine, efavirenz and delavirdine. Protease inhibitors (PIs) target viral assembly by inhibiting the activity of protease, an enzyme used by HIV to cleave new proteins for final assembly of new virions. They include saquinavir, indinavir, ritonavir, lopinavir, amprenavir, fosamprenavir, tipranavir, darunavir, atazanavir and nelfinavir. The first line regimen usually consists of two NRTIs in combination with one NNRTI with protease inhibitors usually being reserved for second line medication.

In Kenya, HAART was introduced in the public health facilities (including the study sites) in 2003 resulting in a dramatic change in the management of HIV infection from a fatal disease in the pre-HAART era to a chronic manageable condition. The management of HIV-infection requires a comprehensive care approach by a muiltidisplinary team of health care providers. These include counseling on adherence to care and on the use of ART, nutritional counseling, treatment of opportunistic infections, regular monitoring through clinical evaluation and laboratory investigations which include CD4 counts, biochemistry and hematology and provision of HAART for eligible patients. In most resource limited settings, including Kenya, NRTIs form the backbone of HAART (Kenya National ART guidelines, 2005 Edition). By the time the study was being conducted (March- May 2010), the first line antiretroviral regimen consisted of stavudine/ zidovudine + lamivudine + nevirapine/ efarvirenz. This regimen was recommended for use because of ease of administration, affordability and does not require refrigeration. These drugs are efficacious

but are also associated with numerous side effects. Use of stavudine for example has been associated with peripheral neuropathy, lipodystrophy and lactic acidosis and has been in use as the first line regimen in Kenya until July 2010 when the Ministry adapted the WHO 2009 recommendations on change of 1st line regimen (see appendix 2). This is not the case for developed countries whose drug regimen has been more tolerable and highly efficacious.

The common short term drug toxicities include nausea, vomiting, headache, peripheral neuropathy (stavudine), bone marrow suppression leading to anaemia (zidovudine), hallucination (efavirenz) and rash (nevirapine). Long term drug toxicity include lipodystrophy, lactic acidosis (stavudine and zidovudine), hepatotoxicity (nevirapine) and peripheral neuropathy (stavudine). Hyperlactatemia though thought to be rare is a life threatening condition that has been mainly associated with the long term use of NRTIs (Herman *et al.,* 2001; Holstein *et al.,* 2001). Nucleoside analogue reverse transcriptase inhibitors (NRTI) remain the cornerstone of antiretroviral therapy for HIV infection in resource-limited settings. These agents inhibit HIV replication through inhibition of reverse transcriptase enzyme, but have been shown to also inhibit human DNA polymerase and hence replication of mitochondrial DNA (mtDNA) leading to its depletion and resulting in toxicity including hyperlactatemia (Cote *et al.,* 2002). Severe lactic acidosis was first observed in HIV-infected persons when nucleoside analogue reverse-transcriptase inhibitor (NRTI) monotherapy was introduced prior to introduction of NRTI combination therapy (Forgang *et al.,* 1995; Brinkman *et al.,* 2000).

1.2 Problem statement

Hyperlactatemia is a common long term complication of use of NRTIs with a prevalence ranging from 15-35% in Western countries ((Boubaker *et al*., 2000) to 20-60% in developing countries (Wester *et al.,* 2007). Symptomatic hyperlactatemia has been shown to occur at a median period of six months after initiation HAART and the lactate level has been shown to be cumulative on use of NRTIs (Carr *et al*., 2000). Majority of HIV-infected patients in resource limited settings including Kenya, are on NRTI-based anti-retroviral drugs.

Severe NRTI-induced lactic acidosis has been showed to cause high mortality varying between 33-57% (Falco *et al.,* 2002). The mortality rate was higher with increase in lactate levels and was shown to be 80% with lactate levels of >10 mmol/l (Brinkman *et al.,* 1999; Falco *et al.,* 2002; McComsey *et al.,* 2002). Diagnosis of lactic acidosis is difficult due to the non-specific nature of clinical presentation (John *et al*., 2001) and the fact that most of the health facilities lack the required equipment to measure blood lactate levels.

1.3 Justification

In Kenya, combination anti-retroviral therapy became freely available in public hospitals in 2003. Since then the number of patients on combination ART has tremendously increased from less than 10,000 in 2003 to over 430,000 in 2010 (NASCOP, 2010). This study was conducted between March and May 2010 and by then the first line regimen in Kenya was stavudine/zidovudine + lamivudine + nevirapine/ effavirenz. The first line regimen was however changed to zidovudine/tenofovir + lamivudine + nevirapine/efavirenz at the time of data analysis through a circular (see appendix 2) from the Director of Medical Services

dated 15th July 2010 in order to adapt WHO rapid advice on use of anti-retroviral therapy released in November 2009.

Combination anti-retroviral therapy containing NRTIs have been shown to cause hyperlactatemia and lactic acidosis. Despite the wide use of these drugs in Kenya no study has been done to assess the magnitude of this problem. Mechanisms of diagnosis in the existing system include assessment of clinical presentations and laboratory procedures are nonexistent in most comprehensive care clinics (CCC). The signs and symptoms of hyperlactatemia has been shown to be non-specific (Falco et al., 2002) and majority of patients could be going undiagnosed therefore resulting in high morbidity and mortality. The prevalence of hyperlactatemia has been shown to be high in resource limited settings such as Botswana as compared to the developed countries and this study designed was to establish whether hyperlactatemia is a problem in our population. Despite the fact that NRTIs form the backbone of the first line therapy, no studies have been documented on the prevalence and correlates of hyperlactatemia in Kenya.

The study was designed to use both clinical and laboratory assessments in order to establish the role of laboratory diagnosis in management of hyperlactatemia at two clinics which fall between high and low HIV prevalence. A lactate meter manufactured by Roche was used to measure the lactate levels after clinical assessment which included both history taking and physical examination was conducted. The objective of this study was to establish the prevalence of hyperlactatemia and determine the factors associated with its development among HIV-infected patients on combination anti-retroviral therapy. The

information obtained will be useful in designing and modifying drug policy and change on the procedures for diagnosis which will result in reduced morbidity and mortality.

1.4 Research questions

1. What is the prevalence of hyperlactatemia among patients on treatment with NRTIs?
2. What factors are associated with hyperlactatemia among patients on NRTIs?

1.5 Objectives of the study

1.5.1 General Objective

To determine the prevalence and factors associated with development of hyperlactatemia among HIV-infected patients on treatment with Nucleoside reverse transcriptase inhibitors (NRTIs) in Thika and Ahero District Hospitals, Kenya.

1.5.2 Specific Objectives

1. To determine the prevalence of hyperlactatemia among patients on treatment with NRTI-based antiretroviral therapy
2. To determine the factors associated with development of hyperlactatemia among HIV-infected patients on treatment with NRTIs.

1.5.3 Assumptions

Hyperlactatemia due to nucleoside reverse transcriptase inhibitors is common among HIV-infected patients on antiretroviral therapy and often goes undetected.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Lactic acid

Lactic acid is a chemical compound that plays a role in various biochemical processes. Lactic acid is a carboxylic acid with the chemical formula $C_3H_6O_3$. It has a hydroxyl group adjacent to the carboxyl group, making it an alpha hydroxy acid (AHA). In solution, it can lose a proton from the acidic group and is known as lactate. In humans, L-lactate is constantly produced from pyruvate via the enzyme lactate dehydrogenase (LDH) in a process of fermentation during normal metabolism and exercise. The concentration of blood lactate is usually 1– 2 mmol/L at rest. It does not increase in concentration until the rate of lactate production exceeds the rate of lactate removal. Lactate, which is produced by the body all day long, is resynthesized by the liver (Cori Cycle) to form glucose. The glucose is released into the blood stream and used directly as a fuel by heart muscle, and by the liver to produce blood glucose and glycogen. In medicine, lactate is one of the main components of lactated Ringer's solution and Hartmann's solution used during resuscitation after blood loss due to trauma, surgery or burn injury.

2.1.1 Hyperlactatemia and Lactic Acidosis

HIV-associated disorders of lactate metabolism were first described in early 1990s, originally with the use of didanosine but they were later described with use of most of the NRTIs (Lai *et al.,* 1991; Bissuel *et al.,* 1994; Carr *et al.,* 2000). More recently, disorders such as myopathy, neuropathy, and pancreatitis have been shown to be part of a common

spectrum of conditions associated with mitochondrial toxicity (Bissuel *et al.,* 1994; Dalakas *et al*., 2000; Moore *et al*., 2000). Carr et al (2000) described the association between lactic acidemia and hepatic dysfunction with a fat redistribution syndrome dominated by lipoatrophy and weight loss.

Hyperlactatemia has been observed in HIV-infected patients receiving antiretroviral therapy. The spectrum of presentation ranges from mild to moderate symptomatic hyperlactatemia to fulminate and life threatening lactic acidosis. Hyperlactatemia is a long term adverse drug reaction that is mainly associated with the use of NRTIs, especially stavudine (Boubaker *et al.,* 2001).

Lactic acidosis is divided into two categories; those with evidence of a systemic impairment in tissue oxygenation (hypoxic) and those without impairment in tissue oxygenation (non-hypoxic). The causes of hypoxic lactic acidosis include hypoxemia, cardiac arrest, and tissue hypoperfusion due to shock, heart failure or sepsis while non-hypoxic lactic acidosis is caused by deregulation of cell metabolism as summarized in Table 1.1

Hyperlactatemia is defined as increased levels of lactate in blood and in this study was classified as mild (2.5-5 mmol/l), moderate (5-10mmol/l) and severe (>10mmol/l) (Guidelines for Anti-retroviral Drug Therapy in Kenya, 2005). Asymptomatic hyperlactatemia is a frequent finding among HIV-infected individuals receiving NRTI (Moyle *et al*., 2002). Symptomatic hyperlactatemia is characterized by nonspecific, predominantly gastrointestinal symptoms, reproducible elevated lactate levels, and hepatic

abnormalities which slowly resolve over weeks to months upon discontinuation of NRTI (Gerard *et al.,* 2000). Lactic acidosis represents the extreme form of NRTI-related hyperlactatemia syndromes and portends a poor prognosis (Falco *et al*., 2002). It is not clear whether severe hyperlactatemia is usually preceded by a period of asymptomatic hyperlactatemia and further longitudinal studies are required to establish this association.

Table 1.1: Causes of lactic acidosis

Classification of lactic acidosis	
Hypoxic	**Non-Hypoxic**
Ischemia	**Delayed clearance**
Shock, severe anaemia, cardiac arrest.	Renal or hepatic dysfunction
Global hypoxia	**Pyruvate dehydrogenase dysfunction**
Carbon monoxide poisoning	Sepsis, thiamine deficiency, alcohol, diabetic ketoacidosis, catecholamines excess
Respiratory failure	**Uncoupling of oxidative phosphorylation**
Severe asthma, cardic obstructive pulmonary disease (COPD), asphyxia	Cyanide, salicylates, methanol and ethylene glycol metabolite, anti-retrovirals, biguanides
Regional hypoperfusion	**Accelerated aerobic glycolysis**
Limb or mesenteric ischemia	Increased effect, sepsis, seizures, large fructose load and malignancies

Source: Kyle J Gunnerson M.D

2.2 Prevalence and risk factors for hyperlactatemia

Hyperlactatemia associated with mild or no symptoms has been reported in 8–21% of patients receiving at least one NRTI compared to 0–1% of patients not receiving antiretroviral therapy. Symptomatic hyperlactatemia is, however, less common and the prevalence has been shown to range between 1.5-2.5% (Carr *et al*., 2000). Other studies have shown the prevalence of elevated serum lactate levels among asymptomatic patients on NRTIs to range between 15-35% (Boubaker *et al*., 2000). In a randomized clinical trial

done in Botswana in a cohort of women, 20% to 60% of patients on NRTI therapy were shown to have elevated lactate levels with 2% developing lactic acidosis.(Wester *et al.*, 2007). In a prospective cohort study involving 1565 persons in a follow-up cohort by Alexander *et al.*, (2005), 662 (42.3%) persons had lactate levels of >2.4 mmol/L, and 49 (3.1%) persons had at least 1 episode of moderate hyperlactatemia (lactate levels ≥5 mmol/L). Another study has identified chronic mild hyperlactatemia with lactate concentrations of 1.5-3.5 mmol/l, as the most common form of hyperlactatemia with a prevalence of 65% (Mina *et al*., 2001).

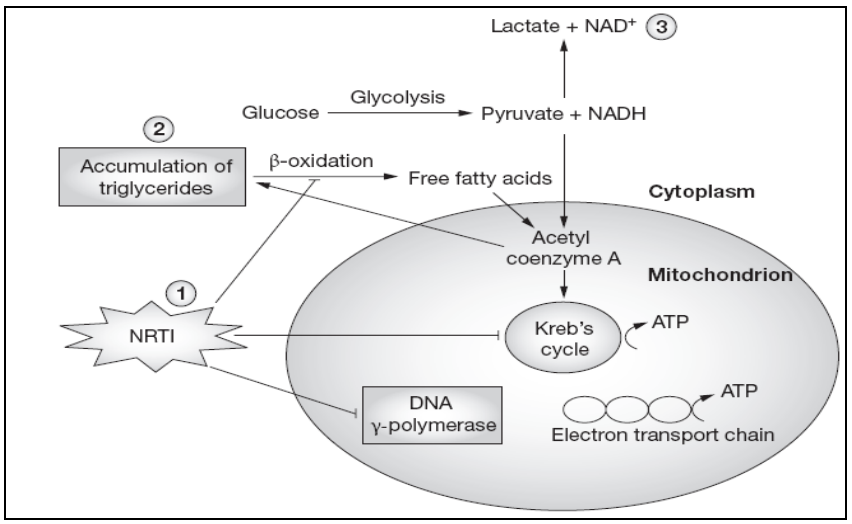
Risk factors for developing hyperlactatemia include increasing age >40 years, obesity (BMI> 30), gender (2.5 times more likely in females compared to males), use of NRTIs mostly stavudine or didanosine-based regimen, hepatitis C virus infection, pregnancy, low CD4 count at the start of therapy, renal insufficiency, intercurrent illness, increased waist-hip ratio, liver dysfunction and genetic predisposition (Fabrice *et al*., 2003, Alexander *et al.*, 2005). Stavudine and didanosine have been associated with increased risk of hyperlactatemia whereas zidovudine and lamivudine have been shown to have lower risk. An association between lipodystrophy, elevated urate levels, triglycerides, cholesterol, plasma glucose and hyperlactatemia has also been demonstrated (Boubaker et al., 2001).

2.3 Mechanisms of NRTI-induced hyperlactatemia

NRTI-induced hyperlactatemia is thought to be due to mitochondrial toxicity as a result of inhibition of DNA polymerase gamma, the enzyme responsible for mitochondrial DNA synthesis. Inhibition of DNA polymerase gamma leads to impaired pyruvate oxidation hence lactate production. Impaired β-oxidation results in conversion of fatty acids to triglycerides that accumulate in myocyte and hepatocyte cytosol, causing depletion in

Kreb's cycle substrates and decreased ATP production. Alternative energy sources through glycolysis are upregulated, which in addition to hepatic dysfunction, causes increased lactate levels (figure 1.2). Symptomatic hyperlactatemia has been associated with marked reductions in the ratio of mitochondrial to nuclear DNA, which averaged 43% lower than those of HIV-infected asymptomatic patients who have never been treated with ARVs. This ratio has been shown to increase significantly after the discontinuation of therapy (Lewis *et al.,* 1995; Hood *et al.,* 1998; Morris *et al.,* 1999; Cote *et al*., 2002; Claessens *et al.,* 2003; Montaner *et al.,* 2003; Walker *et al.,* 2004).

Mitochondrial dysfunction and cellular toxicity has been shown to be the common pathway for NRTI-related adverse effects on tissues (Brinkmann *et al*., 1998; Montaner *et al.,* 2003; Walker *et al.,* 2004) and for adverse effects which include hyperlactatemia, hepatic steatosis, myopathy, cardiomyopathy, peripheral neuropathy, pancreatitis, and lipodystrophy syndrome (Dalakas *et al*., 2001). As lactate is the product of anaerobic glycolysis, hyperlactatemia in normal aerobic conditions indicates mitochondrial dysfunction (John *et al*., 2001) which in turn leads to impaired synthesis of mitochondrial enzymes that generate adenosine triphosphate (ATP) by oxidative phosphorylation of glucose and fatty acids and hence accumulation of lactic acid (Lewis *et al*., 1995).



Source: Claessens et al., 2003[*]

Figure 1.1: Mechanisms of NRTI-induced Hyperlactatemia

2.4 Causes of hyperlactatemia

Hyperlactatemia without metabolic acidosis may result from increased lactate production or from diminished hepatic or renal clearance. This occurs in settings of adequate tissue perfusion, intact buffering systems and adequate tissue oxygenation. Hepatic dysfunction due to drug toxicities including NRTI-induced mitochondrial toxicity and due to chronic hepatitis (e.g. HCV infection), may lead to diminished hepatic clearance of plasma lactate (John *et al.*, 2002)

This has been observed under normal physiological circumstances or as a consequence of pathological conditions, such as hypoxia, circulatory insufficiency, mitochondrial enzyme defects, glycogen storage diseases, seizures, diabetes mellitus, ethanol poisoning, hepatic failure, malignancy, use of stavudine and didanosine and alcohol intoxication (Vrouenraets *et al.*, 2002; Moyle *et al.*,2002). The ability to cause mitochondrial dysfunction from the most to the least potent has been shown as zalcitabine > didanosine > stavudine >

[*] **1** NRTIs disrupt β-oxidation, the Kreb's cycle and transcription of essential enzymes needed for ATP production by inhibiting DNA γ-polymerase. **2** Increased triglycerides cause hepatic steatosis. **3** Lactic acid accumulates more rapidly than it can be excreted by the kidney causing hyperlactatemia.

zidovudine > abacavir = lamivudine = tenofovir (Birkus *et al*., 2002). Zalcitabine is no longer being used due to its association with lactic acidosis.

2.5 Diagnosis of hyperlactatemia

Diagnosis of hyperlactatemia is often difficult since most patients present with non-specific signs and symptoms and a high index of suspicion is required (Bonnet *et al*., 2003). The gold standard for diagnosis of NRTI-related mitochondrial toxicity is muscle or liver biopsy. Hepatic biopsy reveals macrovacuolar and microvacuolar steatosis, ballooned mitochondria and loss of cristae. Muscle biopsy shows red-ragged fibres, lipid droplets, muscle fibre atrophy, abnormal mitochondrial and decreased mitochondrial DNA (Claessens *et al.*, 2003). However, due to the invasiveness and impracticality of this procedure in clinical settings, diagnosis of NRTI-related hyperlactatemia requires demonstration of increased lactate levels in the absence of other known causes, such as dehydration, diabetes mellitus, malignancies, vigorous exercise, sepsis, hypoxemia, alcohol intoxication, renal failure, hyperthyroidism, and other drugs such as isoniazid and biguanides (Mizock *et al*., 1992).

Laboratory investigations include serum lactate, serum bicarbonate, transaminases, amylase, lipase, lactate dehydrogenase and creatinine phosphokinase. Serum lactate is the specific test for hyperlactatemia and the rest of the test are used to establish the cause of hyperlactatemia. Ultrasonography has been used to diagnose hepatomegally due to fat deposition in the hepatocytes but this could not be distinguished with other causes of hepatomegally (Bonnet *et al.,* 2003).

2.6. Clinical presentation of hyperlactatemia

Hyperlactatemia is defined as a persistent, mild to moderate increase in blood lactate concentration without metabolic acidosis, whereas lactic acidosis is characterized by persistent increase in blood lactate levels (usually more than 5 mmol/l) in association with metabolic acidosis. Metabolic acidosis is defined as a state of decreased pH resulting from either a primary increase in hydrogen ions or a reduction in bicarbonate concentration.

NRTI-related hyperlactatemia is associated with a spectrum of clinical presentations that include severe hyperlactatemia, less severe symptomatic hyperlactatemia and compensated chronic or intermittent asymptomatic hyperlactatemia (John *et al*., 2001). The primary clinical features of moderate to severe hyperlactatemia are non-specific and include fatigue, weight loss, myalgia, anorexia, nausea, vomiting, abdominal distension, abdominal pain, dyspnea, and cardiac dysrrhythmias with gastro-intestinal symptoms being the most common. In severe cases, features of hepatic dysfunction are common and include soft, tender hepatomegaly, peripheral edema, ascites, and encephalopathy. In rare cases, it may manifest with jaundice (Falco *et al.,* 2002; Cote *et al*., 2002; Bonnet *et al*., 2003). Onset of signs and symptoms can occur as early as four months from initiation of HAART (Falco *et al*., 2002).

2.7 Management of hyperlactatemia

The management of hyperlactatemia is dependent on the clinical presentation, associated risk factors and the lactate levels. Discontinuation of the causative NRTI is the mainstay of management. Severity of hyperlactatemia as well as presence of symptoms determines whether the patient should receive treatment as an inpatient or an outpatient. In severely ill

patients, haemodialysis has been used to correct metabolic acidosis (Brinkman *et al*., 2000; Bonnet *et al.,* 2003). Administration of co-factors including thiamine, riboflavin, L-carnitine, prostaglandin E and co-enzyme Q has been associated with lower mortality (Falco *et al.,* 2002). In a retrospective non-randomized study, supplementation with antioxidants (vitamin E (1 g/d), b-carotene (45 mg/d), acetylcysteine (800 mg/d), selenium (50 µg/d), *Gingko biloba* extract and several nutritional supplements containing vitamins B1, B2, B6, C, folates, zinc, flavonoids), resulted in statistically significant lower levels of lactic acidosis in the supplementation group (Lopez *et al*., 2003).

Other studies have shown that patients who stop HAART due to lactic acidosis are able to regain virologic suppression of similar magnitude or better after resuming HAART (Tyler *et al*., 2003). Current guidelines on ART in Kenya recommend that patients who have experienced symptomatic hyperlactatemia (lactate levels >2 mmol/l) should subsequently be treated with a NRTI-sparing antiretroviral regimen mainly Tenofovir after the lactate levels have normalize. For patients who continue to have high lactate levels despite being on an NRTI-sparing regimen, other risk factors which include liver failure, Hepatitis B or C infection, concurrent use of other drugs including acyclovir and metformin, and use of alcohol or cigarette smoking should be ruled out before NNRTIs and protease inhibitors are prescribed. In asymptomatic patients with lactate levels between 2 – 5 mmol/L, there is no evidence to suggest that any change in antiretroviral therapy is necessary, but there is no long-term safety data indicating whether or not adverse consequences may occur at levels within this range (Vrouenraets *et al*., 2001).

2.8 Mortality associated with hyperlactatemia

Severe NRTI-induced lactic acidosis is associated with a high mortality rate varying between 33-57% (Falco *et al.*, 2002). Higher lactate levels are associated with higher mortality rates (Brinkman *et al.*, 1999; Falco *et al.*, 2002; McComsey *et al.*, 2002). The overall mortality rate for patients with lactate levels >10 mmol/l has been shown to be 80% and the outcome is similar in both developed and developing countries. The mechanism of death is due to accumulation of lactic acid in the blood which halts the normal functioning of the cell which leads to multi-organ failure due to acid-base imbalance (Andrew *et al.*, 2001).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study Sites

The study was conducted in HIV clinics at Thika District Level-5 and Ahero District hospitals in Central and Nyanza Provinces respectively. The choice of the study sites was based on HIV prevalence since the study targeted a high and low prevalence region. HIV prevalence in Thika (Central Province) is 3.6% while that of Ahero (Nyanza Province) is 14.9% (KAIS, 2007 report). The study was therefore able to compare the outcomes of interest in the two regions and also was able to establish whether HIV prevalence affects prevalence of hyperlactatemia. Thika District Level-5 Hospital is located in Thika town about 50 kilometers to the North East of Nairobi, the capital city of Kenya. Ahero District Hospital is located along Nairobi-Kisumu highway about 30 kilometers from Kisumu city (Figure 3.1).

Both hospitals have well established HIV clinics with trained multidisciplinary teams of health care providers to deliver comprehensive HIV care and treatment services. At each hospital, the multidisciplinary team is composed of two clinicians, two nurse counselors, one pharmacist, one pharmaceutical technologist, three laboratory technologists, four peer counselors, one health records and information officer (HRIO). The HIV clinics at both hospitals have adequate rooms for service delivery including triaging, consultation, adherence counseling, pharmacy and chest clinic. At Thika District Level-5 Hospital, the HIV clinic has a laboratory where blood is drawn and taken to the main hospital laboratory for processing whereas at Ahero District Hospital all laboratory tests are done at the main

laboratory. The two hospitals were selected for this study because they have a large cohort of patients on ART, and have equipments including a lactometer, a FACS count machine for CD4 subset analysis and biochemistry analyzers that were required to conduct this study. Both public hospitals follow the National ART guidelines on the standard regimen to be used, when to initiate ARVs, how to monitor for drug toxicities and changing or substituting of therapy. As at March 2010, Thika District had enrolled a total of 6420 patients on HIV care with 2980 adults on ART while Ahero District Hospital had enrolled 6874 patients on HIV care with 3224 adults on ART (District Health records and Information Officers' report, March 2010). On average both hospitals attend to a total of 100-120 patients daily with about half of them being on ART. The study participants were recruited during their routine clinic days between March and May 2010.

Source: Source Oxford Cartographers

Figure 3.1: Map of Kenya showing Thika and Ahero study sites

3.2 Study Population

The study subjects were recruited during their routine clinic visits. A total of 255 HIV-infected adults (above 18 years) attending outpatient HIV clinics participated in the study at the two study sites.

3.2.1 Inclusion Criteria

All participants were HIV-infected adults who had been on combination antiretroviral therapy containing nucleoside reverse transcriptase inhibitors for at least 2 months. Patients who had changed their regimen due to drug toxicity but had been on the current regimen for more than two months were also included in the study.

3.2.2 Exclusion Criteria

The exclusion criteria included being less than 18 years in order to ensure that the participants were legally able to give consent. Sepsis is a known cause of hyperlactatemia and patients who were unwell with WHO defining clinical stage III or IV during the recruitment were excluded from the study. Patients with chronic diseases including renal failure, liver failure, diabetes mellitus and hypertension were also excluded from the study.

3.3 Ethical Consideration

Ethical approval for the study was obtained from KEMRI/ National Ethical Review Committee (See Appendix 9) and administrative clearance was obtained from the Provincial Directors of Medical Services in the two Provinces and from the medical

superintendents of the two hospitals. The patients were sensitized during the routine health talks which are held every morning before the clinic begins specifically on the objectives of the study, eligibility criteria and voluntary participation. For all eligible patients the questionnaire and the consent forms were inserted in their files. A written informed consent was obtained before collecting the demographic data and medical history via a structured questionnaire and medical records transcriptions. For those patients who were illiterate, their right thumb print was used and countersigned by a witness.

3.4 Study Design

This was a descriptive cross-sectional study, with a minimum sample size of 246 as illustrated below.

3.4.1 Sample size determination

To determine the minimum sample size, the prevalence of hyperlactatemia was assumed to be 20% based on studies done in similar set up in Botswana and the Fisher's formulae was used (Fisher et al., 1918). The figure of 20% was used since the prevalence in resource limited settings has been shown to range between 20- 60%. It was hypothesized that the prevalence of hyperlactatemia would be at least 20%. Statistical significance was determined at 95% confidence interval (CI) and a p value of < 0.05.

$$n = \frac{Z^2 pq}{d^2}$$

Where;

n = sample size

z = standard normal deviate set at 1.96 and corresponds to 95% confidence interval

p = prevalence (20%)

q = 1 – p (1-0.20)

d = degree of accuracy, P = 0.05

$$n=\frac{1.96x1.96x\ 0.20x\ 0.80}{0.05x0.05}$$

n = 246

Purposeful simple random sampling method was used to give an equal chance for the eligible patients attending the clinic to participate until the required sample size of 246 was obtained with at least 123 coming from each study site. The intention was to have equal number of participants from each of the two clinics but at the end of the recruitment period, the number of participants at Ahero DH was 124 while the number at Thika DH was 131. Both samples were higher than the minimum sample size. Since the HIV prevalence is 8.6% among females and 4.6% among males in the Kenya (KAIS, 2007 report), the number of females enrolled in HIV care and treatment is twice that of males. Due to this gender disparity, the sample size was obtained based on probability proportional to size (PPS) of the population in the clinic and hence 1/3 were men and 2/3 were females.

3.5 Clinical assessment

Recruitment was done during the routine clinic visits. As a standard of care, weight, height and temperature were taken at the triage desk by a nurse. One research assistant was placed at the triage desk in order to administer the consent form to all those who met eligibility criteria. For those who accepted to participate in the study, a questionnaire was put in their file. After the patient was seen by the clinician, they were referred to the study room.

In the study room, the second research assistant welcomed the client and abstracted data from the patient file in order to complete the sociodemographic section of the questionnaire. For any information that was incomplete, the research assistant sort clarification from the patient. For any missing information, that needed measurements, the study room had a thermometer, a weighing scale and height measure and the research assistant had been trained on how to take these measurements.

The patient would then proceed to the next desk where the principal investigator who was a medical doctor by training would take a detailed medical history on any chronic illnesses, weight loss, jaundice, alcohol intake, cigarette smoking, last menstrual periods (for women), fever, abdominal symptoms including nausea, vomiting, abdominal pain and diarrhea, fatigue, muscle aches, numbness, joint pains, difficulty in breathing, history of other medication or food supplements and history of long term drug toxicities including lipodystrophy and peripheral neuropathy. This was guided by a structured questionnaire (see Appendix 1). He also conducted chart abstraction to obtain information on date the client was enrolled into HIV clinic, WHO clinical staging at enrollment, anti-retroviral

drugs that the client was currently on, period on anti-retroviral therapy, CD4 count at enrollment and within the last six months and baseline alanine transaminase (ALT).

The patient was then requested to lie on couch and a physical examination was performed. The physical examination included a general examination with special emphasis on signs of yellowness of the conjunctiva or under the tongue, edema, pallor, signs of muscle wasting (especially facial and lower extremities), buffalo hump or abdominal enlargement which is pathognomonic of lipodystrophy on patients on stavudine based regimen. Systemic examination was then performed with emphasis on increased heart rate, increased respiratory rate which could be a sign of metabolic acidosis, abdominal distention which could be due to hepatomegaly and any abdominal tenderness. Central nervous system was also examined to establish any signs of peripheral neuropathy.

The patient was then allowed to rest for at least 15 minutes and the principal investigator measured the lactate levels using a lactometer (see full laboratory procedure below). The study team then thanked the patient for participating in the study. Any significant finding during the study was reported to the clinical team and action taken in line with the guidelines for Anti-retroviral Drug Therapy.

3.6 Lactate measurement

Accutrend® lactate meter by Roche diagnostics® was used to determine the blood lactate levels in this study (Fig. 3.1). The requirement for the test included: lactate meter, lancet, lactate strip, lactate controls, gloves and alcohol/spirit swabs. The patient was requested to

wash their hands with soap and allowed to rest for 15 minutes. When the hands were warm and dry, gentle massage of the index finger was done to encourage circulation of blood. Using a lancet, a prick was made on the side of the finger and a sufficiently large drop of blood obtained. The free hanging drop of blood was immediately directly applied on the test strip. The measurement chamber flap was closed and lactate measurement read after 60 seconds. Lactate levels were read from the meter and defined as normal if less than 2.5 mmol/l, those who had elevated lactate levels (hyperlactatemia) we classified as mild (2.5-5 mmol/l), moderate (5- 10 mmol/l) and severe (>10 mmol/l). Low values were denoted as (LO) and this meant that the amount of blood was inadequate. High values denoted as (HI) meant that the value was higher than 21.7 mmol/l and this could as a result of wrong blood collection techniques. For both results, a repeat test was done. Coding of the lactate meter was done for every new set of test strips and controls for both high and low levels were done after every 25 tests to ensure validity of the results. Management for those with hyperlactatemia was instituted in line with National algorithm on management of hyperlactatemia (see Appendix 6)

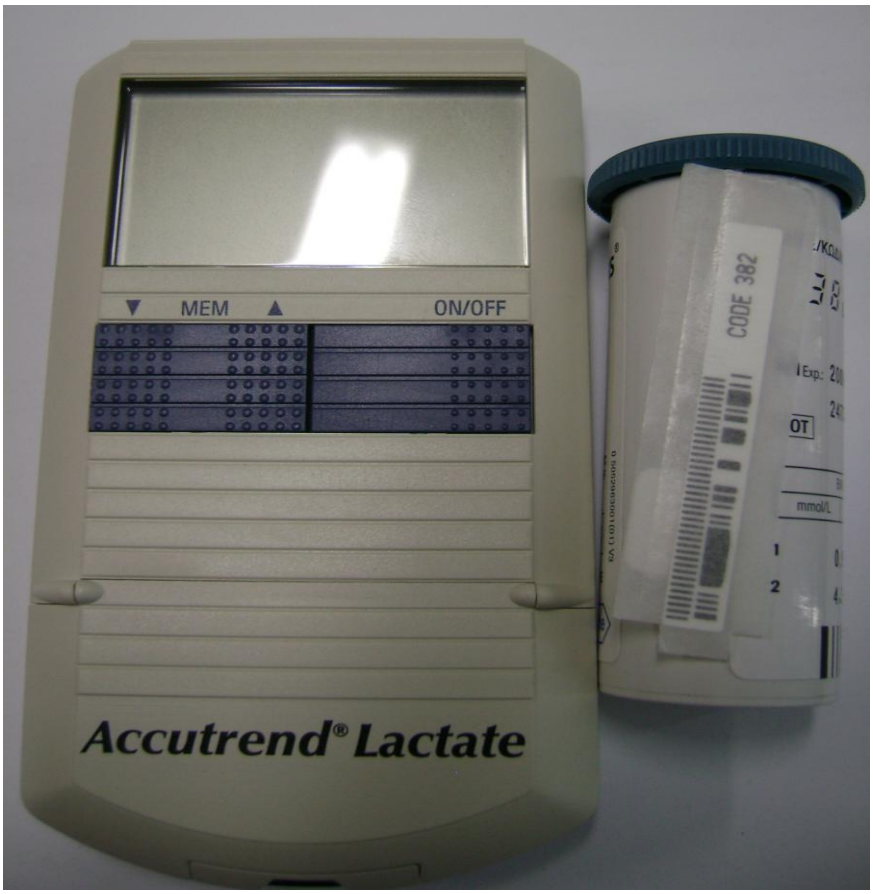


Figure 3.2: Accutrend® lactate meter and test strips used to measure lactate levels

3.7 Data analysis

Data collection was done using a structured questionnaire and abstraction from the patients files. Data was entered into a Microsoft Access® database before being imported for analysis using PASW® version 18†. Prevalence of hyperlactatemia was calculated using frequency tables by dividing the number of HIV-infected patients with lactate levels more than 2.5 mmol/l by the total number of study subjects (n=255). Since no patients had lactate levels more than 10 mmol/l, study subjects were put in three categories; normal, mild and moderate as explained above. Measures of central tendency i.e. the median and

† PASW (Predictive Analytics Software), formerly known as SPSS

the mean were used to compare the differences of characteristics of patients sampled as categorized by site.

Since the baseline characteristics were significantly different for body mass index, alcohol intake, period on ARVs, ART regimen, baseline CD4 count and WHO clinical staging between the two study sites, analysis of the two groups was done separately. Student t-test was used to determine any significant difference between the means in the two sites. Pearson's Chi square test was used to determine the association between hyperlactatemia and factors such as ART regimen, sex, age, BMI, clinical presentation, long term drug toxicities, period on ARVs and baseline CD4 count.

After associating hyperlactatemia and stavudine regimen, post hoc tests using multivariable logistic regression was used to determine the association between hyperlactatemia and a combination of the above risk factors. Odds ratio and P value at 95% confidence interval was used for interpretation of results. A P value of <0.05 was considered to be statistically significant.

CHAPTER FOUR

4.0 RESULTS

4.1 Sociodemographic characteristics of study participants

A total of 255 HIV-infected patients on anti-retroviral therapy for at least 2 months participated in the study (Ahero 124 and Thika 131). The table 4.1 shows the distribution of selected characteristics of the study population.

Table 4.1: Characteristics of the study population

Characteristic		Ahero DH Freq (%)	Thika DH Freq (%)	Overall Freq (%)	P value
Sex					
	Male	41 (33.0)	41 (31.3)	82 (32.2)	0.433
	Female	83 (67.0)	90 (68.7)	173 (67.8)	
Age					
	<40 years	203(80)	195(76.4)	143(56.1)	0.073
	≥ 40 years	52(20)	60(23.6)	112(0.439)	
Marital status					
	Married	65 (52.4)	61 (46.6)	126 (49.4)	**< 0.001**
	Widowed	50 (40.3)	26 (19.8)	76 (29.8)	
	Separated	2 (1.6)	31 (23.7)	33 (12.9)	
	Single	7 (5.6)	13 (9.9)	20 (7.8)	
Occupation					
	Unemployed	60 (48.4)	44 (33.6)	104 (40.8)	0.099
	Casual Laborer	24 (19.4)	35 (26.7)	59 (23.1)	
	Formal employment	6 (4.8)	12 (9.2)	18 (7.1)	
	Self employed	34 (27.4)	40 (30.5)	74 (29.0)	
BMI					
	BMI <24	71(57.3)	95 (72.5)	166(65.1)	**0.008**
	BMI ≥24	53 (42.7)	36 (27.5)	89(34.9)	
Alcohol intake and cigarette smoking					
	Alcohol intake	6 (4.8)	20 (15.3)	26 (10.2)	**0.005**
	Current smokers	1 (0.8)	5 (3.8)	6 (2.4)	0.120
Period on ARVs					
	< 24 months	36 (29.0)	24 (18.3)	60 (23.5)	**0.031**
	≥ 24 months	88 (71.0)	107 (81.7)	195 (76.5)	
ART Regimen					
	D4T+3TC+NVP/EFV	94 (75.8)	101 (77.1)	195 (75.5)	**0.004**
	AZT+3TC+ NVP/EFV	21 (16.9)	9 (6.9)	30 (11.8)	
	TDF+3TC+NVP/EFV	7 (5.6)	21 (16.0)	28 (11.0)	
	DDI+ABC+LP/r	2 (1.6)	0 (0)	2 (0.8)	
WHO clinical staging at enrollment					
	Stage 1 or 2	48 (39.3)	69 (60.0)	117 (49.4)	**0.001**
	Stage 3 or 4	74 (60.6)	46 (40.0)	120 (50.6)	

Key: D4T (Stavudine), 3TC (Lamivudine), NVP (Nevirapine), EFV (Efavirenz), AZT (Zidovudine), TDF (Tenofovir), LP/r (Kaletra), DDI (Didanosine), ABC (Abacavir) BMI (Body Mass Index)

4.1.1 Sex distribution

Two out of three participants were females. At Ahero the proportion of females was 67% which compared well with that of Thika (68.7%). There was no significant difference between the study sites (P=0.433) (Table 4.1).

4.1.2 Age distribution

The mean age was 41.3 years (range 20-81 years). Over half of the participants (56%) were above 40 years of age. A higher proportion of patients were aged 40 years and above at Ahero clinic (80%) as compared to Thika clinic (76%). The difference between the two sites was not significant (P=0.073) (Fig. 4.1).

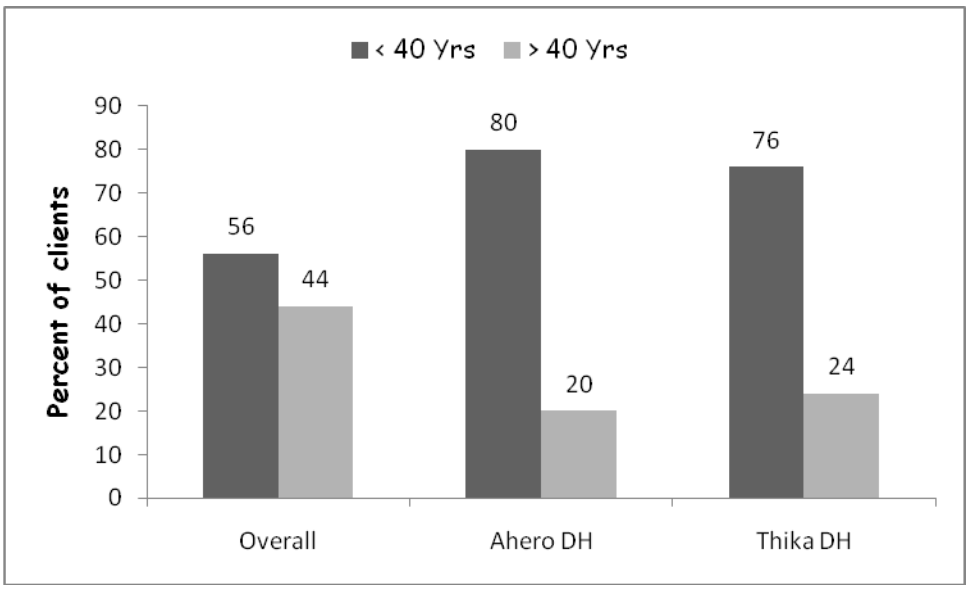


Figure 4.1: Age distribution among study participants

4.1.3 Marital Status

One out of two participants was married, thirty percent had lost a spouse (widow/ widower), thirteen percent had separated while eight percent were single. A higher proportion of patients had lost a spouse at Ahero clinic (40%) as compared to Thika clinic

(20%).Twenty four percent of patients had separated at Thika clinic compared to six percent in Ahero clinic. The difference between the two sites was significant P= < 0.001) (Table 4.1).

4.1.4 Occupation

Two out of three participants were unemployed. Twenty nine percent were self- employed, twenty three percent were casual labourers while seven percent were in formal employment. When comparing the two sites, slightly more participants were unemployed in Ahero (48%) than in Thika (34%), twice as many participants were in formal employment in Thika (10%) compared to Ahero (5%), twenty seven percent were casual labourers in Thika compared to \twenty percent in Ahero while thirty one percent were self-employed at Thika compared to twenty seven percent in Ahero. There was no significant difference between the two study sites (P=0.09) (Fig. 4.2).

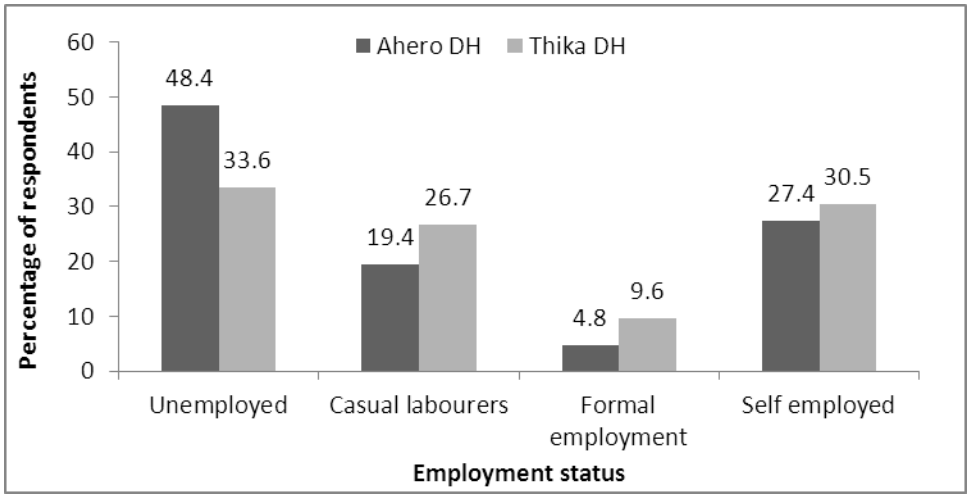


Figure 4.2: Occupation status of the participants

4.1.5 Body Mass Index (BMI)

Sixty five percent of patients had a BMI of <24 while thirty five percent had BMI of ≥ 24. The BMI at the two sites was significantly different (P=0.008) (table 4.1).

4.1.6 Alcohol intake

A total of 26 participants (10%) reported to be taking alcohol at the time of the interview. The number of participants taking alcohol were significantly higher at Thika DH than at Ahero DH (P=0.005). (Table 4.1)

4.1.7 Period on ARVs

Three out of four (77%) participants were on ARVs for a period more than 24 months. A higher proportion of patients in Thika clinic (82%) compared to Ahero clinic (71%) were on ARVs for a period of more than 24 months. The difference between the two study sites was significant (P=0.031) (Fig: 4.3).

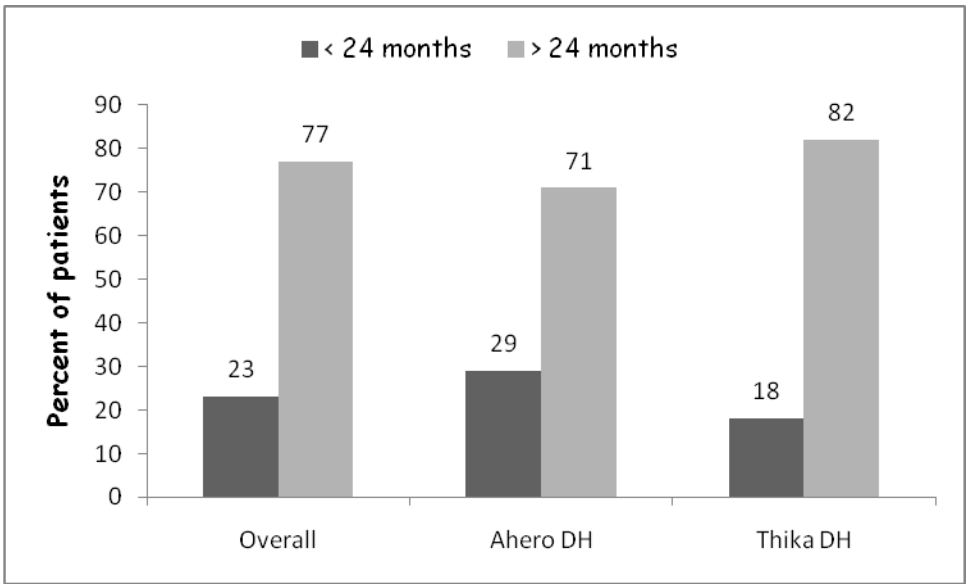


Figure 4.3: Period on ART regimen among patients

4.1.8 ART regimen

Of the 255 participants, 195 (75.5%) were on stavudine-based regimen, 30 (11.8%) were on zidovudine based regimen, 28 (11%) were on tenofovir based regimen and two (0.8%) were on second line (didanosine, abacavir and kaletra) (Table 4.1). The patients in the two clinics had a significant different treatment regimen with 17% in Ahero and 7% in Thika on zidovudine-based regimen while 16% (Thika) and 6% (Ahero) were on tenofovir-based regimen P=0.004 (Fig. 4.4).

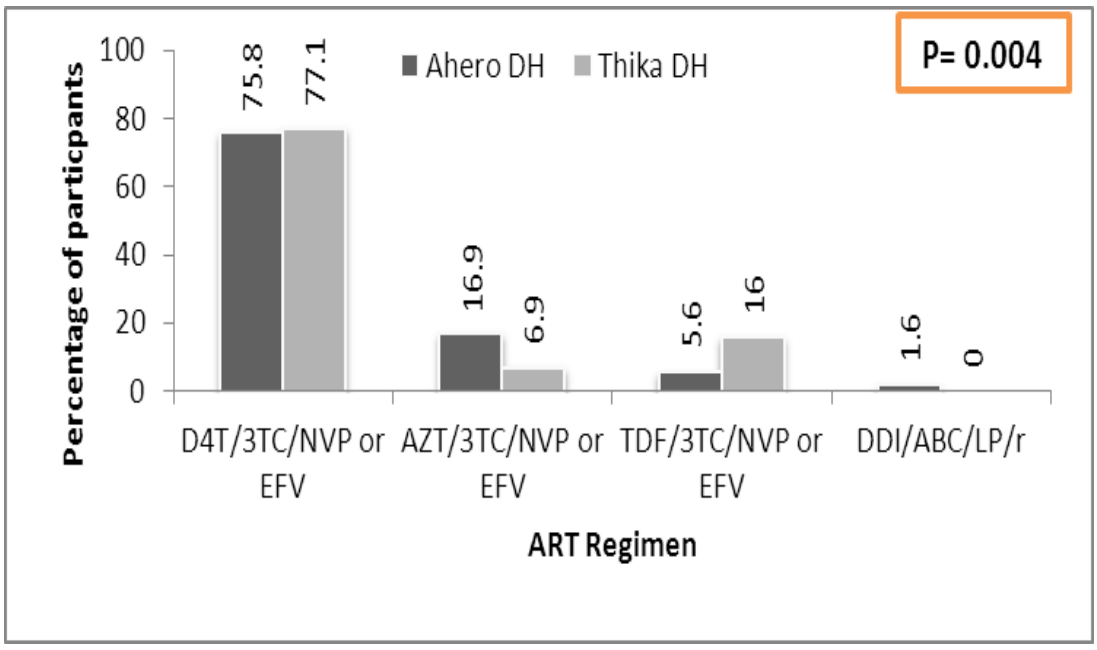


Figure 4.4: ART regimen among participants at Ahero and Thika clinics.

4.1.9 WHO defined clinical staging

Over half of the participants (51%) had WHO defined stage 3 or 4 at enrolment into the HIV clinic. A higher proportion of the patients in Ahero clinic were in WHO defined stage 3 or 4 (61%) compared to 40% in Thika clinic. This difference was statistically significant (P= 001) (Fig. 4.5).

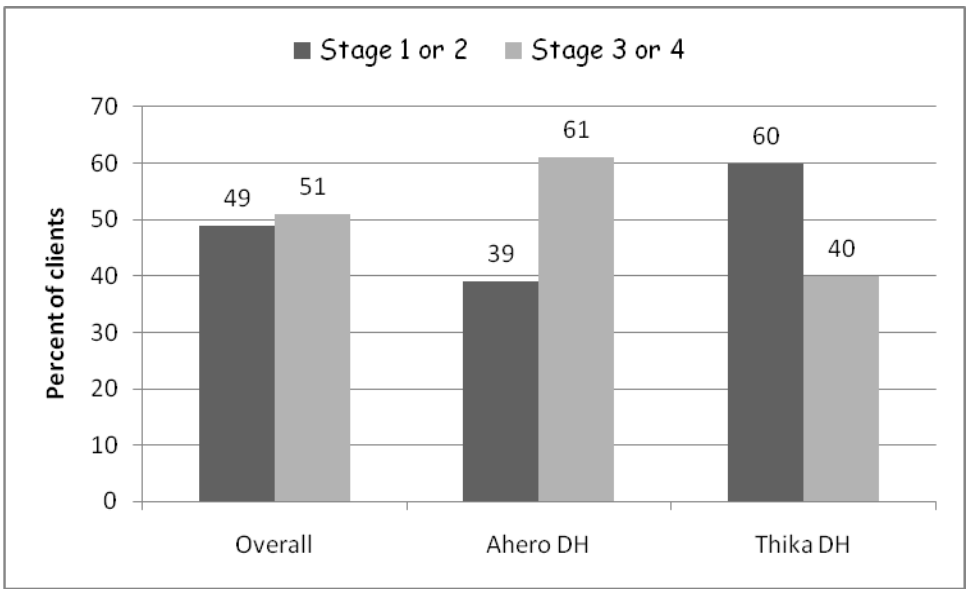


Figure 4.5: WHO defined clinical staging at enrollment

4.2 Prevalence of hyperlactatemia

The average lactate level among the study participants was 2.53 (median 2.4, IQR 1.8 – 3.0). The overall prevalence of hyperlactatemia (lactate ≥ 2.5 mmol/l) was 41%. Among those with elevated levels, 38% had mild hyperlactatemia (lactate ≥2.5-5 mmol/l) while 3% had moderate hyperlactatemia (lactate ≥5- 10 mmol/l) (Fig. 4.6). The study found that the prevalence of mild hyperlactatemia was slightly higher at Ahero (43%) than Thika (33%) while moderate hyperlactatemia was twice higher at Thika clinic (4%) than Ahero clinic (2%). There was no significant difference between the two study sites (P=0.242) (Fig. 4.7).

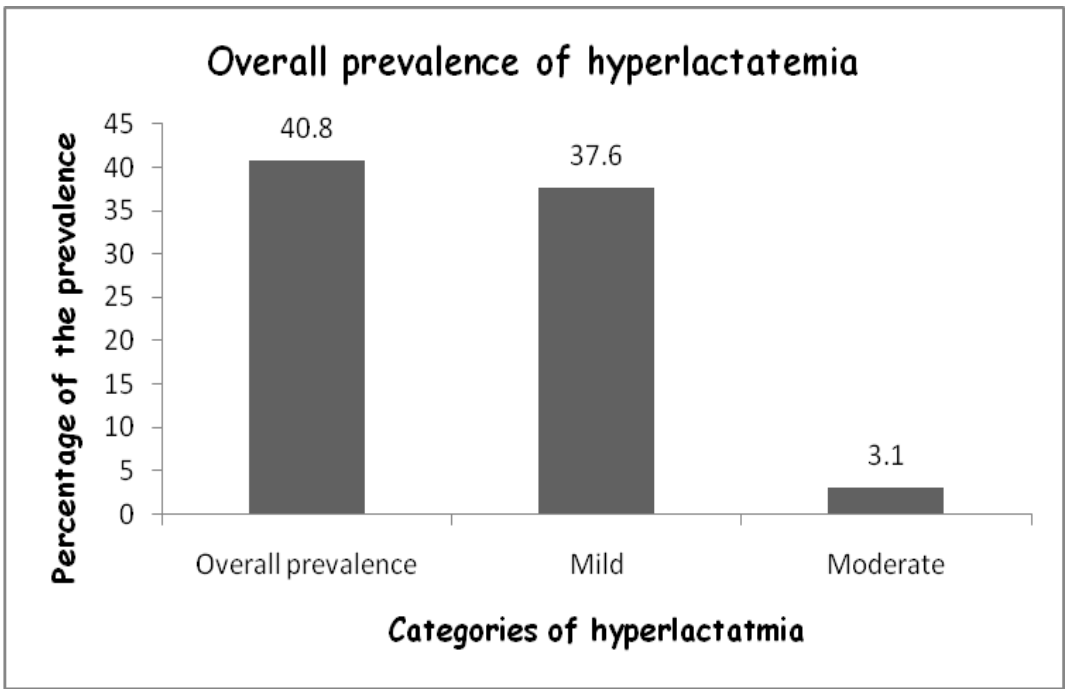


Figure 4.6: Overall prevalence of hyperlactatemia

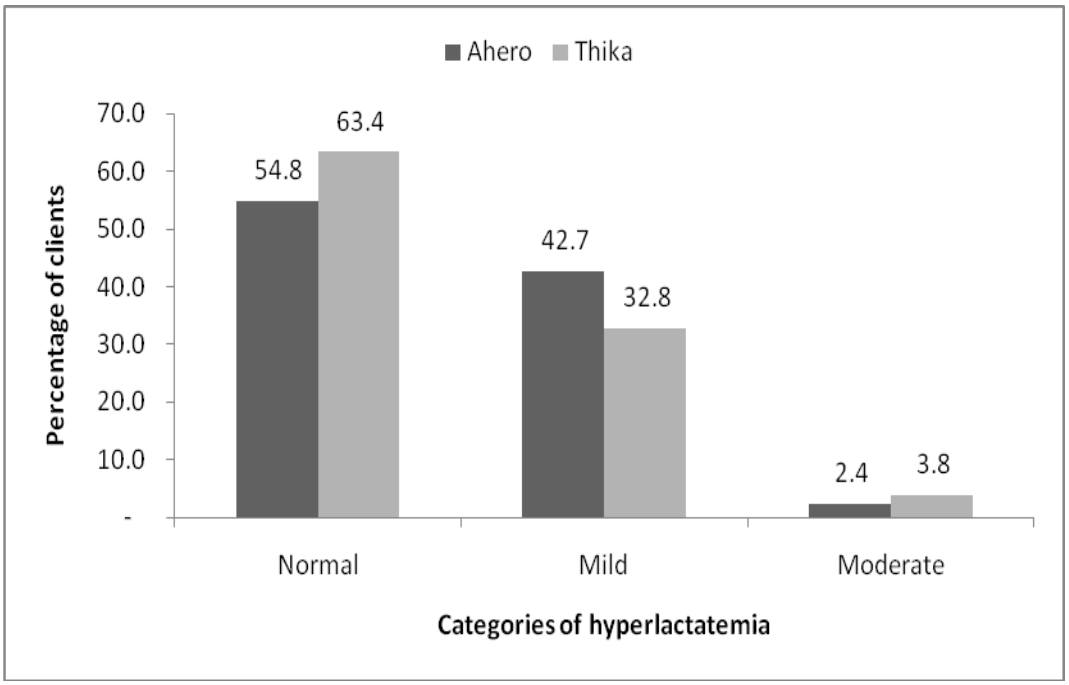


Figure 4.7: Prevalence of hyperlactatemia at Ahero and Thika DH

4.3 Risk factors for hyperlactatemia

Presence of hyperlactatemia was associated with use of stavudine based regimen (OR 1.87, 95% CI, 1.01- 3.47), female gender (OR, 2.42, 95% CI, 1.11-5.27), use of ARVS for more than 24 months (OR, 2.62, 95% CI, 1.97-7.04), BMI of ≥24 (OR 1.13(1.67-2.91) and baseline CD4 count of less than 200 cells/ul (OR 2.36, 95% CI, 1.01-5.50). All the above risk factors were significant at both study sites. This study did not show any association between age, renal insufficiency, hepatitis C co-infection, pregnancy and liver dysfunction as has been shown in other studies. In addition other risk factors evaluated in this study including cigarette smoking, alcohol intake, WHO defined clinical staging at enrollment and current CD4 but were all not significant (Table 4.2).

Table 4.2: Risk factors for development of hyperlactatemia

Risk factor	Overall OR (95% CI)	Ahero DH OR (95% CI)	Thika DH OR (95% CI)
Sex			
Male	1	1	1
Female	**2.42 (1.11 –5.27)**	**2.21(1.59 – 5.67)**	**2.17 (1.54 –5.54)**
Age			
<40 years	1	1	1
≥ 40 years	1.07 (0.70 - 2.70)	1.04 (0.46 -2.92)	0.88 (0.43 -2.79)
BMI			
BMI <24	1	1	1
BMI ≥24	**1.13 (1.67-2.91)**	**1.77 (1.38-2.58)**	**1.58(1.72-4.45)**
Alcohol intake (history)			
Yes	1.28(0.56-2.88)	1.23(0.24-6.33)	1.51(0.58-3.96)
No	1	1	1
ART Regimen			
D4T+3TC+NVP/EFV	**1.87 (1.01 – 3.47)**	**1.69(1.15-3.25)**	**1.27(1.18-3.43)**
AZT+3TC+ NVP/EFV	1.50 (0.48 – 4.71)	0.37(0.07-2.16)	3.0(0.47-19.04)
TDF+3TC+NVP/EFV	1	1	1
DDI+ABC+LP/r	OR < 0.001	OR < 0.001	n/a
WHO defined clinical staging at enrollment			
Stage 1&2	1	1	1
Stage 3&4	1.27(0.75-2.13)	1.19(0.57-2.48)	1.21(0.56-2.61)
Period on ARVs			
< 24 months	1	1	1
≥ 24 months	**2.62(1.97-7.04)**	**2.40(1.61-3.22)**	**2.89(1.65-5.48)**
Baseline CD4			
< 200 CD4 cells/ul	**2.36(1.01-5.50)**	**1.76(1.77-4.04)**	**2.22(1.46-3.25)**
≥ 200 CD4 cells/ul	1	1	1
Current CD4			
< 200 CD4 cells/ul	0.91(0.43-1.92)	0.77(0.29-2.04)	1.05(0.32-3.4)
≥ 200 CD4 cells/ul	1	1	1

4.4 Clinical presentation and hyperlactatemia

Of the 255 study participants, 48% presented with more than three signs and symptoms. A higher proportion of patients presented with more than 3 signs and symptoms at Ahero clinic (76%) as compared to Thika clinic (35%). The difference between the two sites was significant (P=0.017) (Fig. 4.8)

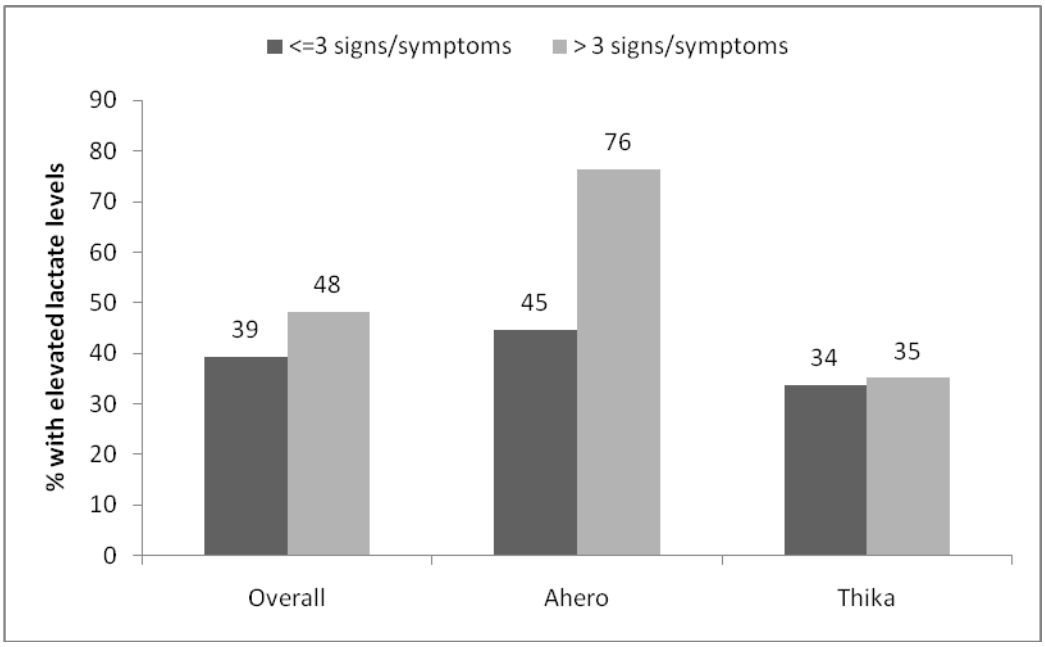


Figure 4.8: Occurrence of signs and symptoms among patients

Comparing the signs and symptoms between patients who had and those who did not hyperlactatemia revealed no significant difference between the two groups (P= 0.433) (Fig. 4.9).

Among the 104 patients diagnosed with hyperlactatemia, 80% were symptomatic (Thika 77% and Ahero 82%). The commonest signs and symptoms among patients with elevated lactic levels were fatigue, paraesthesia, lipodystrophy and peripheral neuropathy as shown in Figure 4.10. All clients with moderate hyperlactatemia were symptomatic. The mean number of signs and symptoms among those with mild hyperlactatemia was 3 compared to

6 among those with moderate hyperlactatemia. Three out of the eight patients (37.5%) with moderate hyperlactatemia presented with tender hepatomegaly.

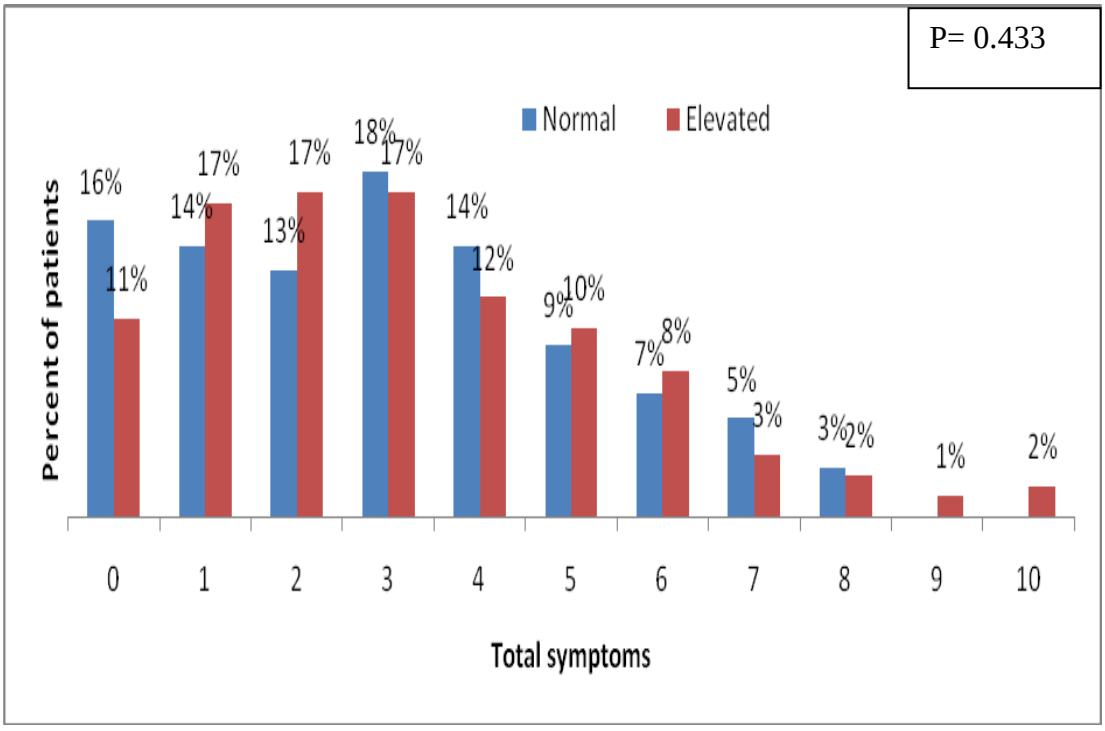


Figure 4.9: Presence of Signs and symptoms

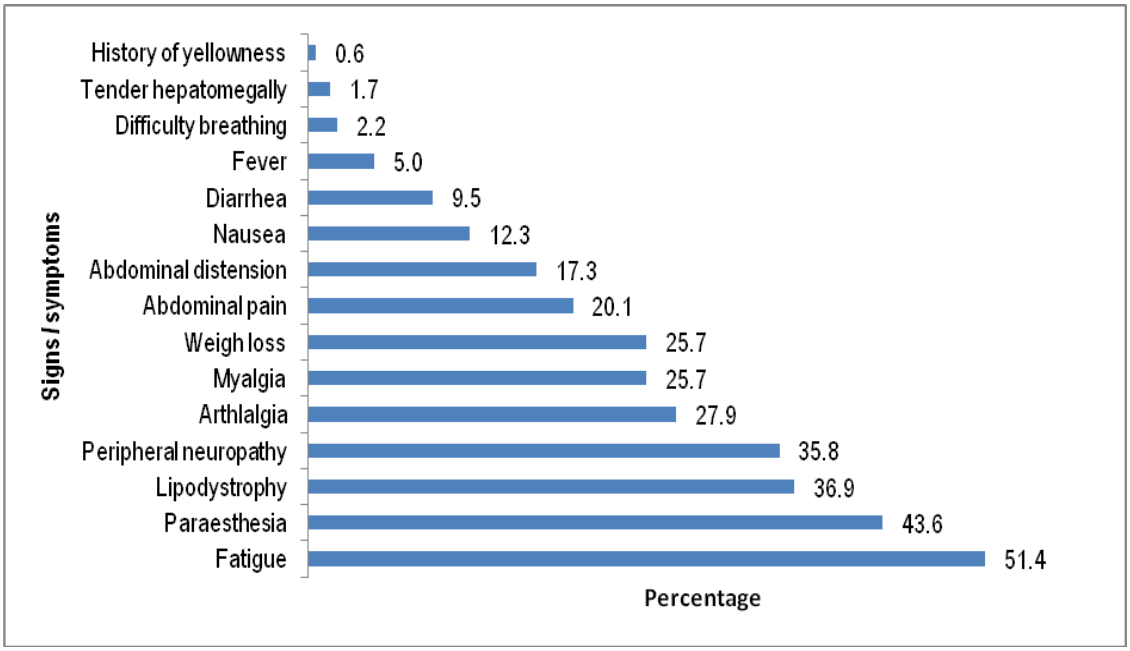


Figure 4.10: Signs and symptoms among patients with hyperlactatemia

4.5 Long term side effects and hyperlactatemia

The long term side effects that were assessed in this study included lipodystrophy and peripheral neuropathy. Overall, 34% of patients had lipodystrophy while 35% had peripheral neuropathy. Lipodystrophy (OR 1.16 95% CI 1.67-2.99) and peripheral neuropathy (OR 1.13 95% CI 1.67-2.94) were associated with occurrence of hyperlactatemia. The association for lipodystrophy was significant at both sites. However association of peripheral neuropathy at Thika DH was not significant (Table 4.3).

Table 4.3: Long term side effects and hyperlactatemia

Risk factor		Overall OR (95% CI)	Ahero DH OR (95% CI)	Thika DH OR (95% CI)
Lipodystrophy				
	Absent	1	1	1
	Present	**1.16 (1.67-2.99)**	**1.64 (1.59-5.60)**	**1.32 (1.64-3.74)**
Peripheral neuropathy				
	Absent	1	1	1
	Present	**1.13 (1.67-2.94)**	**1.39 (1.62-4.12)**	0.98 (0.47-2.04)

CHAPTER FIVE

5.0 DISCUSSION, CONCLUSIONS AND RECOMMENDATIONS

5.1 Discussion

Hyperlactatemia was defined as mild (lactate levels ≥ 2.5- <5 mmol/l), moderate (lactate levels ≥ 5-<10mmol/l) or severe (lactate levels ≥ 10mmol/l). The overall prevalence of hyperlactatemia (lactate ≥2.5 mmol/l) in the population of HIV-infected patients on HAART was 41% with 38% of participants having mild hyperlactatemia and 3% having moderate hyperlactatemia. These finding are similar to the study by Alexander (2005), which was a cohort follow-up study for 1566 HIV-infected patients of whom 1492(95%) were on combination anti-retroviral therapy containing stavudine-based regimen. The prevalence of hyperlactatemia (lactate ≥2.5 mmol/l) was 42.3% of whom 3.1% had moderate hyperlactatemia (lactate ≥5 mmol/l). A higher proportion of patients had hyperlactatemia at Ahero DH (45%) as compared to 37% in Thika DH of whom 2% and 4% had moderate hyperlactatemia respectively. The difference between the study sites was however not significant (P= 0.242).

The risk factors associated with hyperlactatemia in this study included, being on stavudine-based combination anti-retroviral treatment, female gender, being on ARVs for a period of more than 24 months, a CD4 count of less than 200 at initiation of therapy and having a BMI of 24 and above. Female gender was also associated with having moderate hyperlactatemia with seven out of eight patients (87.5%) being women. This study shows confirmation of stavudine, female gender, BMI, low baseline CD4 count as risk factors for hyperlactatemia but other risk factors identified by a study by Fabrice (2003) including age, renal insufficiency,

pregnancy, Hepatitis C infection, liver dysfunction, intercurrent illness and genetic predisposition were found not to be significant. However, presence of intercurrent illnesses, renal insufficiency, liver dysfunction were not assessed in this study because all patients with chronic illnesses including renal and liver diseases were excluded at recruitment. Genetic predisposition and Hepatitis C infection were not assessed in this study. This study showed new findings on the association of period on ARVs and hyperlactatemia. Patients on ARVs for more than 24 months were 2.62 times more likely to develop hyperlactatemia. The findings are important in patient management and also in policy formulation on who should be screened for hyperlactatemia.

Overall, 80% of patients with hyperlactatemia were symptomatic. The signs and symptoms included fatigue, paraesthesia, abdominal symptoms, arthralgia, muscles aches among others. These findings are similar with other studies (Falco et al., 2002; Cote et al., 2002; Bonnet et al., 2003). This study showed no significant difference between the signs and symptoms among patients presenting with hyperlactatemia and those with normal lactate levels. The current procedure in diagnosis of hyperlactatemia has been use of clinical signs and symptoms due to lack of lactate meters in majority of health facilities. Based on these findings this method of diagnosis is sub-optimal as it is not specific for hyperlactatemia and clinician end up missing many patients who require treatment resulting in high mortality. A model case study was identified in Thika DH during the course of the study. A 48 year old woman presented with nausea, vomiting, fatigue, abdominal pains, tender hepatomegaly, muscle aches and difficulty in breathing. The patient had presented a week earlier with similar symptoms and was admitted and discharged after two days. Lactate level estimation revealed that patient had moderate hyperlactatemia (lactate levels 6.5 mmol/l) and signs of metabolic acidosis. The patient was admitted, ARVs drugs stopped and management instituted in line with the National

guidelines. I believe that this is not an not an isolated case and many more such cases could be going undiagnosed due to lack of laboratory assessment.

As a follow up to this study a total of 83 patients (32%) who had hyperlactatemia and had signs and symptoms were managed in line with the National ART guidelines. This included stopping of the ARVs, weekly monitoring of the lactate levels and substitution with tenofovir based regimen. This group would have normally continued on treatment with the offending drugs but the use of lactometers assisted in diagnosing these cases hence improving the quality of patient care. These findings are similar to the study by Cote (2002) who showed that clinical presentation for hyperlactatemia to be non-specific and recommended that clinicians should have a high index of suspicion.

Three out of four patients (77%) were on stavudine based regimen. WHO (2009) recommended use of tenofovir or zidovudine based regimen as first line therapy. These recommendations were adapted in Kenya on 15th July 2010 (see Appendix 2) after this study. However the study found that 34% of patients had lipodystrophy and 35% had peripheral neuropathy but only 10% had been changed to tenofovir and 11% to zidovudine which is the standard management for lipodystrophy and peripheral neuropathy respectively. Other studies have shown association between lipodystrophy (45%) and hyperlactatemia (Saint-Marc *et al.*, 1999). Despite 117 (46%) of patients being on ARVs for more than twenty four months, only two (0.8%) of patients were on second line treatment. In a mature HIV program where patients have been on ARVs for more than 2 years, the proportion of patients on second line treatment range between 5- 15% (David et al., 2003). This shows that many patients with treatment failure are going undiagnosed which could compromise future anti-retroviral drug options due to resistance. This could be attributed to that fact that diagnosis

of treatment failure mainly relies on clinical and immunological criteria and viral load testing which is more sensitive is only reserved for those with suspected treatment failure.

The study found no significant difference in sex, age, occupation, and recent CD4 count between the two study sites. However there was a significant difference in factors such as body mass index, alcohol intake, period on ARVs, ART regimen, baseline CD4 count and WHO clinical staging at enrollment. It is hypothesized that the difference in these baseline characteristics can be attributed to the fact that due to the high prevalence of HIV in Nyanza Province, a lot of campaigns on stigma, HIV testing and treatment have been done hence patients are accessing treatment early at Ahero DH as compared to Thika DH. Low proportion of patients reported consuming alcohol (10%) or smoking cigarette (<1%) at the time of the study. However, many patients reported to have stopped taking alcohol and smoking after being enrolled into the HIV clinic after learning of the dangers associated with these practices.

This study had certain limitations. It was not possible to conduct an external validation of the lactometer used in order to ascertain the reliability of the results obtained. In addition, it was not possible to measure acidity of blood (pH) for the patients who had high lactate levels in order to confirm lactic acidosis. However, a study done to validate Accutrend® lactate meter as a screening tool for hyperlactatemia during antiretroviral therapy showed a sensitivity of 95.9% with a specificity of 63.8%. The study concluded that Accutrend® lactate meter is an appropriate device for screening of patients on HAART with suspected hyperlactatemia (Ernesto *et al.*, 2008). The study did not also investigate all the risk factors

associated with hyperlactatemia, however these findings are important and can provide information in designing future prospective studies in this subject.

5.2 Conclusion

The study found a 41% prevalence of hyperlactatemia (lactate $\geq$ 2.5 mmol/l) among patients on combination antiretroviral therapy containing a nucleoside reverse transcriptase inhibitor. Prevalence of moderate hyperlactatemia (lactate levels $\geq$5- 10 mmol/l) in this population was 3.1%. Although the 1st line regimen for anti-retroviral therapy in Kenya has changed from use of stavudine to tenofovir or zidovudine based regimen following WHO recommendations, NRTIs still remain the backbone of therapy for HIV-infected patients and these findings therefore are applicable even to the current situation.

The study found female gender, stavudine based anti-retroviral therapy, low CD4 count (<200cells/ul) at start of therapy, BMI of $\geq$24 and period on ARVs for more than 24 months as the risk factors for hyperlactatemia on patients on combination anti-retroviral therapy containing NRTIs.

Use of clinical signs and symptoms alone without lab-diagnosis is not sufficient and specific to make a diagnosis of hyperlactatemia among patients on combination anti-retroviral therapy. This is due to the non-specific nature of the signs and symptoms due to hyperlactatemia. Most HIV clinics lack laboratory capacity to assess hyperlactatemia.

No patient had severe hyperlactatemia and the prevalence of mild and moderate hyperlactatemia was varied between the two sites. This can be attributed to the differing baseline characteristics between the two sites.

Management of patients with long term drug toxicities including lipodystrophy and peripheral neuropathy remains a challenge as evidenced in this study.

5.3 Recommendations

The study found a high prevalence of hyperlactatemia among patients on stavudine based anti-retroviral regimen and health care workers should be sensitized on the importance of this side effect in order to improve the patient management.

Since being on ARVs for more 24 months was shown to be a risk factor for hyperlactatemia, special attention should be taken for this group of patients presenting with fatigue, abdominal symptoms, cuff muscle aches, paraesthesia, arthralgia, tender hepatomegaly, lipodystrophy, peripheral neuropathy and should undergo laboratory assessment by use of a lactate meter to improve diagnosis of hyperlactatemia. Patients on combination anti-retroviral therapy for more than 24 months should have a baseline lactate level done in order to rule out this condition.

Health facilities should be provided with a point of care lactate meters in order to be able to combine both clinical and laboratory assessment in diagnosis of hyperlactatemia. This will ensure timely diagnosis and substitution of the offending drug and improved quality of care. It will also reduce mortality since severe hyperlactatemia has been shown to cause high mortality.

Refresher training on long term side effects due to combination anti-retroviral therapy should be intensified in order to build the capacity of HCW to be able to diagnose and manage these side effects.

A database should be set up to document the side effects associated with use of combination antiretroviral therapy in order to inform the National Program for policy formulation.

A prospective cohort study among patients on the current combination anti-retroviral therapy is encouraged in order to establish the magnitude of this problem after change the of 1st line therapy.

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APPENDICES

Appendix 1: Study Questionnaire

Date of interview (*dd/mm/yyyy*): ____/____/_____

Part I: Social Demographic Data

1. Client's Name: _____ 2. Client's CCC Number:_____

3. a) Age: _____ b) Date of birth (dd/mm/yyyy)____/____/___

4. Sex: ☐ Male ☐ Female

5.Marital status Single ☐ Married ☐ Separated ☐ Widow ☐

6. Occupation Self Employed ☐ Casual Laborer ☐ Formal Employed ☐ Unemployed ☐

7.Weight: _____ (Kgs) 8. Height: _____ (cms) 9. BMI: _____

Part II: Clinical Assessment Information

a) ***Medical History (obtained from patient interview and chart abstraction)***

1. Date client enrolled into CCC (dd/mm/yyyy): ____/____/_____
2. WHO defined clinical staging at enrollment?

3. a) Have you lost weight in the last 3 months?

   b) If yes approximately how many kilograms: _____
4. Have you had any history of yellowness of eyes?
5. Do you take alcohol?
6. a) Do you smoke?

   b) If yes, how many cigarette sticks do you smoke per day? __
7. When was your last menstrual period? ----------

b) ***Signs and Symptoms***

8. Does the client have fever?

9. Are you experiencing any of the following:

   a) Nausea/ vomiting

   b) Abdominal pain

   c) Abdominal distension

   d) Diarrhoea

   e) Fatigue/ general malaise

   f) Muscle ache (especially cuff Muscles)

   g) Paraesthesia/ numbness

   h) Arthralgia (joint pain)

   i) Tender hepatomegally

   j) Difficulty in breathing

   k) Others: (Specify) _____

   l) None

c). Drug History

10. a) Are you taking other medications in addition to ARVs?

11. a) Are you on any food supplements? Yes ☐ NO ☐

    b). If yes, which type _____

11. Anti-retroviral drugs like any other drugs have side effects. Since you started taking the drugs, have you experienced any of the following side effects?

- Redistribution of fat leading to facial wasting and enlargement of the abdomen?

  Yes ☐ No ☐

- Tingling sensation of upper and lower extremities Yes ☐ No ☐

Others, (specify) _____

13. a) What combination antiretroviral therapy is the client currently on?

Stavudine + Lamivudine + NVP/EFV ☐ Zidovudine +Lamivudine + NVP/EFV ☐

Didanosine + Abacavir + Kaletra ☐ Tenofovir +Lamivudine + NVP/EFV ☐

b) Others, specify_____

14. For how long has the client being on ARVs _____ (months)

d). Laboratory Records Assessments

15. What was the CD4 counts

- At enrollment (Baseline) ……..
- Current (Within 6 months) ……..
- Date of last CD4 Count (dd/mm/yyyy)……/……../……..

16. What was the baseline ALT (Alanine transaminase)……..

17. Measure of lactate levels …….

Appendix 2: Circular on change of anti-retroviral therapy in Kenya.

MINISTRY OF PUBLIC HEALTH AND SANITATION

&

MINISTRY OF MEDICAL SERVICES

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Ref: NASCOP/CTS/17 **Date:** 15th July, 2010

- All Provincial Directors of Medical Services,
- All Provincial Directors of Public Health and Sanitation
- All Medical Officers of Health,
- All in-charges-Government, Mission Facilities and Private facilities,
- All Partners implementing HIV Programmes

Re: National Recommendations for Prevention of Mother To Child Transmission of HIV, Infant & Young Child Feeding and Antiretroviral therapy for children, adults and adolescents

The Government has come up with new recommendations based on the WHO guidance issued in 2010, program experience and other developments within the country. These recommendations cover the following areas:

1. Prevention of mother to child transmission of HIV (PMTCT)
2. Infant and young child feeding in the context of HIV (IYCF)
3. Antiretroviral Therapy (ART) for children , adolescents and adults

A. Prevention of Mother to child transmission of HIV (PMTCT)

i. All Pregnant women should be encouraged to start attending ANC early (as soon as they know they are pregnant, preferably in the first trimester).

16/7/2010

ii. All pregnant women should be counseled and tested for HIV during their **1st ANC visit** in line with the HIV testing and counseling guidelines and retesting should be done in the **3rd trimester for HIV negative pregnant** women.

iii. All pregnant Women who are not tested, opt-out or decline HIV testing during the first ANC visit should be offered testing in subsequent visit(s)

iv. All HIV positive pregnant women should be evaluated for eligibility for HAART during the 1st ANC visit using WHO staging and / or CD4 testing where available.

v. All HIV positive women with **CD4 count <350** or in **WHO stage 3 or 4** irrespective of CD4 count should be started on HAART as soon as possible regardless of gestational age. Those already on HAART before pregnancy should continue with their antiretroviral treatment **(refer to annex 1)**.

vi. All HIV positive pregnant women with CD4 count **>350** or in WHO **stage 1 or 2** should be given the following ARV prophylactic regimen:

- Zidovudine (AZT) should be started at **14 weeks** of pregnancy or at first contact thereafter, and
- Zidovudine (AZT) should be continued **in labour** and
- Single dose Nevirapine (sdNVP) should be started at the **onset of labour** and Lamivudine (3TC) should be started **in Labour** and
- Lamivudine (3TC) and Zidovudine (AZT) should be continued up to **one week** after delivery **(refer to annex 1 & 2).**

vii. HIV positive Women not eligible for HAART presenting for the 1st time after **38 weeks** should only be offered ARV prophylaxis during labour and up **to 1 week** after delivery as above (vi).

viii. All women presenting in the late antenatal , maternity or postnatal care clinic with unknown HIV status to be counseled and tested for HIV and managed accordingly

ix. All HIV exposed (children born to HIV positive mothers) breastfeeding infants should be started on Nevirapine prophylaxis from birth and continued throughout the duration of breastfeeding. **Nevirapine should be stopped one week after complete cessation of breastfeeding** (refer to annex 3).

x. All HIV exposed infants whose mothers are on HAART (with or without breastfeeding) should be started on Nevirapine prophylaxis from birth and continued up to **6 weeks of age.**

16-07-2010

xi. All HIV exposed infants who are on **not breastfed at all** should be started on Nevirapine prophylaxis from birth and continued up **to 6 weeks of age** (refer to annex 3).

xii. HIV exposed breastfeeding Infants whose mothers are not on HAART presenting for the 1st time in the post-partum period should be started on Nevirapine prophylaxis upon presentation to the Health facility. Diagnostic PCR testing should be conducted for these infants at **6 weeks** or at the earliest opportunity thereafter **(refer to EID algorithm - annex 4).**

xiii. Infants with PCR negative results should be continued on Nevirapine prophylaxis for the entire breastfeeding period until **one week after complete cessation of breastfeeding.**

xiv. Infants with positive PCR results should be started on HAART in line with peadiatric ART guidelines and Nevirapine prophylaxis **discontinued.**

B. Infant and young child feeding in the context of HIV

The government has reviewed available scientific evidence regarding infant and young child feeding and has come to a conclusion that breastfeeding with appropriate use of Anti retroviral drugs for mother and child is the best option for overall well being and survival of HIV exposed children.

Health service providers are therefore urged to ensure that all HIV positive pregnant women are evaluated for HAART eligibility and if not eligible are provided with ARV prophylaxis as per guidance in section A (refer to annex 1 & 2). **Nevirapine Prophylaxis for all HIV exposed children should be as per national guidelines.**

i. All mothers who are HIV negative or are of unknown HIV status should be encouraged and supported to exclusively breastfeed for the first 6 months and continue breastfeeding with appropriate complementary feeds introduced thereafter.

ii. All HIV positive mothers should be given information on available infant feeding options and counselled using recent scientific information on benefits and challenges of each options in order to help them make an informed choice **(refer to annex 5).**

iii. All HIV positive mothers who choose to breastfeed should be encouraged and supported to **exclusively breastfeed for the first 6 months** and continue breastfeeding with appropriate complementary feeds thereafter. Infants of these mothers should be provided with Nevirapine prophylaxis as recommended in point **ix** of section A of this circular (PMTCT).

4

16-07-2010

iv. HIV positive women who meet the AFASS criteria and chose not to breast-feed should be counselled and supported to do **exclusive replacement feeding for the first 6 months** and appropriate complementary feeds introduced thereafter. Infants of these mothers should be provided with Nevirapine prophylaxis as recommended in point **xi** of section A of this circular (PMTCT)

v. In special circumstances determined by clinicians involving infants who cannot breastfeed e.g. orphans or abandoned babies or where the mother has condition like mastitis preventing breastfeeding the infant should be provided with exclusive replacement feeding with appropriate complementary feeds introduced thereafter if determined by the clinician that not doing so will expose the infant to HIV infection **(refer to annex 6).**

C. Antiretroviral therapy for Children

HIV testing

i. All infants and young children whose HIV exposure status is not known at the time of the first visit to the health facility should have their exposure status established through:

- Testing and counselling the mother for HIV or
- Testing the infant using antibody test in situation where the mother is not available or not willing to be tested.

ii. All HIV exposed infants should be offered routine DNA PCR testing (Early Infant Diagnosis) at the 6 week immunization visit or at the earliest opportunity for infants seen after 6 weeks of age (refer to Annex 4)

iii. All HIV exposed infants should be offered Cotrimoxazole prophylaxis from the age of 6 weeks onwards till HIV status is confirmed.

iv. All HIV infected infants and children should be offered Cotrimoxazole prophylaxis from the age of 6 weeks onwards.

HAART Eligibility

v. All children aged **less than 18 months** confirmed HIV positive by PCR should be initiated immediately on HAART regardless of CD4 count or percentage, and their WHO clinical stage **(refer to Annex 7).**

vi. Initiation of HAART for children **older than 18 months** should be based on WHO clinical stage and / or CD4 recommendations as follows:

- 19-59 months : CD4 % of < 25% or count of <1000 or WHO stage 3 and 4 irrespective of CD4 levels

4

16-07-2010

- 5 – 12 years : CD4 % < 20% or count < 500 or WHO stage 3 and 4 irrespective of CD4 levels

ARVs to start with

vii. All infants and children already on HAART should continue with their current treatment regimens unless there are indications for change of regimes (**Refer to Annex 8**)

viii. All infants and children being newly initiated on HAART henceforth should be started on revised recommended national first line ART regimens for children (**Refer to Annex 8**)

ARVs to use in second line

ix. Recommendations on ARVs to use in second line have not changed from what is contained in current ART guidelines (**Refer to Annex 8**)

D. Antiretroviral therapy for adolescents and adults

When to start Anti retroviral treatment

i. All HIV positive patients should be started on HAART if CD4 count < 350 or WHO stage 3 and 4 irrespective of CD4 count **(Refer to Annex 9 & 10)**

What ARVs to start with

ii. **All new patients** initiating HAART should be started on:

- Zidovudine (AZT) + Lamivudine (3TC) + Nevirapine (NVP) or Efavirenz (EFV

OR

- Tenofovir (TDF) + Lamivudine (3TC) + Nevirapine(NVP) or Efavirenz (EFV) **(Refer to Annex 9)**

iii. **Patients currently on Stavudine (D4T) based 1st line regimens should be evaluated for side effects and where indicated changed appropriately** (Refer to Annex 9 & 11)

iv. 2nd line regimens include AZT/TDF/ D4T + 3TC + boosted Protease Inhibitor **(Refer to Annex 9)**

16-07-2010

ARVs to use in second line

i. Recommendations on ARVs to use in second line have not changed from what is contained in current ART guidelines (**Refer to Annex 8**)

The ministry has developed a plan for phased implementation of all recommendations contained in this circular and is currently revising all related national guidelines to reflect the changes

All health service providers should continue to provide PMTCT, IF, EID and ART services using existing guidelines as these recommendations are progressively phased in

All facilities are encouraged to integrate the provision of care for the mother and the child including PMTCT, EID, IYCF and treatment for children within the MCH service setting.

This circular supersedes all other previous circulars and recommendations.

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Director of Public Health & Sanitation

Dr. F. M. Kimani
Director of Medical Services
16/07/2010

Appendix 3: Consent Form (English)

This Informed Consent Form has two parts:

- Information Sheet (to share information about the research with you)
- Certificate of Consent (for signatures if you agree to take part)

PART I: INFORMATION SHEET

Purpose of the research

Antiretroviral drugs (ARVs) are used to reduce the multiplication of HIV virus in the body. The drugs can cause unwanted effects which include minor effects such as diarrhea, vomiting, body weakness headache and sometimes can cause major effects such as numbness of hands and feet, liver damage, kidney damage and elevated lactic acid levels which can cause death if not diagnosed early and treated appropriately. The study aims at establishing how common is the problem of elevated lactic acid among HIV-infected patients on treatment with ARVs. The information obtained will help the doctors to better take care of patients with HIV infection like you and possibly prolong their lives.

Participant selection

We are inviting all adults on treatment with ARVs for at least three months who attend clinic at this hospital to participate in the research

Voluntary Participation

Your participation in this research is entirely voluntary. It is your choice whether to participate or not. Whether you choose to participate or not, all the services you receive at this clinic will continue and nothing will change. You may also change your mind later and stop participating even if you agreed earlier and you will still continue receiving the routine care for HIV infected patients at this clinic.

What will be required of me if I decide to participate?

Upon enrollment into the study, the clinic staff will ask you some questions concerning yourself and your health during your routine monthly clinic visit. A small amount of blood, about two drops will be taken from you by pricking your index finger. We will also measure how tall you are and how much you weigh. The results of the test will be given to you at the same sitting by the attending health care worker and any necessary interventions done.

Duration

The research will take one day during the routine monthly clinic visits and therefore no extra cost in terms of transport will be required.

Risks

By participating in this research there is no risk that is anticipated but in case of any injury, immediate medical attention will be provided for free at the hospital. You will experience pain when being pricked to obtain blood but this will be done by qualified health care worker. Due to the time taken to ask you questions, the time taken during the clinic visit might be a little longer but the research team will do their best to ensure there is no unnecessary delay.

Benefits

If you participate in this research, you will have the following benefits: incase your lactic acid levels are elevated, you will receive immediate medical attention from the clinic staff, normal lactic acid levels will be an assurance to you and that you are tolerating the ARVs well. The information obtained will help the Government to make better decisions on the care of HIV-infected patients like you in future.

Reimbursements

You will not be given any money or gifts for taking part in this research.

Confidentiality

The information that we collect from this research project will be kept confidential. Information about you that will be collected during the research will be put away and no one but the researchers will be able to see it. Any information about you will have a number on it instead of your name. Only the researchers will know what your number is and we will lock that information up with a lock and key.

Sharing the Results

The knowledge that we get from doing this research will be shared with you through morning health talks at the clinic before it is made widely available to the public. Confidential information will not be shared. After these meetings, we will publish the results in order that other health care workers may learn from our research.

Who to Contact

If you have any questions you may ask them now or later, even after the study has started. If you wish to ask questions later, you may contact any of the following; Dr. Eliud Mwangi, Principal Investigator P.O. Box 25725, Nairobi. Tel: +254-057-2021252/ 0724688037 or Chairman, KEMRI/National Ethical Review Committee, P.O Box 54840, Nairobi. P.O Box 54840, Nairobi. You will be given a copy of this consent form

PART II: CERTIFICATE OF CONSENT

I have read the foregoing information, or it has been read to me. I have had the opportunity to ask questions about it and any questions that I have asked have been answered to my satisfaction. I consent voluntarily to participate as a participant in this research.

Print Name of Participant_____

Signature of Participant _____

Date _____

If illiterate

I have witnessed the accurate reading of the consent form to the potential participant, and the individual has had the opportunity to ask questions. I confirm that the individual has given consent freely.

Print name of witness_____ AND Thumb print of participant

Signature of witness _____

Date _____

Print name of researcher/clinician_____

(Person administering the questionnaire)

Signature_____ Date

_____

Appendix 4: Fomu ya Kukubalia

Fomu hii ina sehemu mbili:

Sehemu ya kwanza ni ya mafunzo na inaelezea kuhuzu utafiti

Sehemu ya pili ni kibali cha kukubali kushiriki kwenye utafiti

Sehemu ya kwanza: Mafunzo

Utangulizi: Tunakualika kushiriki kwenye utafiti huu kwa sababu wewe ni mtu anayeishi na virusi vya Ukimwi na umekuwa ukitumia madawa ya kupunguza makali ya ukimwi kwa muda unaozidi mwezi mitatu. Dawa hizi zinazopunguza makali ya virusi zinaitwa "antiretroviral drugs" au ARV. Kabla ukubali kushiriki utaelezewa zaidi juu ya utafiti wenyewe. Utapata pia fursa ya kujibiwa maswali yako yote. Utafiti huu unafanywa na watafiti kutoka Kenya Medical Research Institute (KEMRI) na Chuo Kikuu cha Jomo Kenyatta University of Agriculture and Technology (JKUAT).

Mathumuni ya Utafiti: ARVs zikitumiwa vema hupunguza makali ya virusi vya ukimwi. Pia ARVs zinamadhara yake kama vile kutapika, kuhara, kuwa na uchovu mwilini, kuumwa na kichwa ambayo hupungua baada ya kutumia haya madawa kwa muda. Madhara mengine ni mabaya na huenda yakasababisha kifo yazipotimbiwa haraka kama vile kufa ganzi kwa viungo vya mwili, kuharibiwa kwa maini na figo na kuongezeka kwa asidi kwa damu iitwayo "lactic acid". Kusudi la utafiti huu ni kubaini kiwango cha wale wanaopata hili dhara la kuongezeka kwa "lactic acid" mwilini baada ya kutumia madawa ya ARVs kwa muda unaozidi mwiezi mitatu. Ufahamu tutakaopata kwa utafiti huu utasaidia madaktari ili waweze kutibu watu walio na virusi kama wewe kwa ubora zaidi siku zijazo.

Watakao shiriki: Utifiti huu unafanyika katika kliniki maalum za utunzaji wagonjwa waliona virusi (CCCs) Hospitalini Thika na Ahero.Wote wanaokubali kushiriki kwenye

utafiti huu wamepokea dawa za kupunguza makali ya virusi vya ukimwi kwa muda wa miezi tatu au zaidi.

Lipi litahitajika kutoka kwangu nikiamua kushiriki katika utafiti huu?

Daktari wako atakuuliza maswali kuhusu hali yako ya mwili na vile unavyotumia madawa ya kupunguza virusi wakati wa kliniki yako ya mwezi. Damu kiwango cha matone mawili itatolewa kwa kudungwa kidole cha mkono, na tutapima uzito na urefu wako. Majibu yatatolewa kwa muda wa dakika chache na daktari wako atakuelezea juu yake.

Kuweka siri: Juhudi zote zitafanywa kudumisha siri juu ya hali yako ya afya. Utapewa nambari yako ya utafiti ya siri itakayokuwezesha kufikia habari juu ya afya yako kwenye utafiti. Habari zote kukuhusu katika utafiti zitawekwa salama salmini.

Manufaa: Manufaa utakayopata kwa kushiriki kwenye utafiti huu ni kama vile; kama kiwango cha “lactic acid” kiko juu utapata matibabu mapema kutoka kwa daktari wako, kama kiwango kiko sawa itakuwa ishara ya kuwa huna haya madhara na hii huenda ikakupa motisha ya kuendelea kutumia haya madawa. Ufahamu utakao tokana na utafiti unaweza kunufaisha watu wengine wanaoishi na virusi vya ukimwi siku za usoni.

Hatari: Kuna hatari ya maumivu au kuumia unapotolewa damu lakini madaktari waliohitimu ndio watakuhudumikia. Pia kwa vile utaulizwa maswali, huenda ukachukuwa muda zaidi ya kawaida kwenye kliniki lakini juhudi zote zitafanywa kupunguza muda utakao chukua kwenye kliniki. Kwa kukubali kushiriki kwenye utafiti unataambua na kukubali hatari zilizoko.Vilevile, una kubali kuwaondolea lawama zozote hospitali za Thika na Ahero, Wizara ya Afya, KEMRI, Chuo Kikuu cha JKUAT, iwapo kutatokea jambo au kudhurika, kufuatia kushiriki kwako.

Kushiriki kwa hiari: Kushiriki kwako kwenye utafiti huu ni kwa hiari bila shuruti. Unaweza kujiondoa wakati wowote unapoamua. Kutokubali kwako kushiriki katika utafiti hakutaadhuru huduma unazopokea za afya. Watafiti wanaweza kukatiza kushiriki kwako iwapo hautambatana na masharti ya utafiti.

Ruzuku: Hakuna ruzuku utakayaipokea kwa kushiriki kwenye utafiti huu.

Matokeo yenye maana: Matokeo yoyote ya maana yatakayopatikana baada ya utafiti huu kukamilika, yatawasilishwa kwako.

Maswali: Ukiwa na swali lo lote kuhusu utafiti huu tafadhali wasiliana na Dkt Eliud Mwangi SLP 25725-40100, Nairobi. Simu: 0724688037 au wasiliana na Mwenyekiti, KEMRI/National Ethics Review Committee, SLP 54840-00200, Nairobi. Simu: +254-020-272254114.

Utapewa nakala ya fomu hii

Sehemu ya pili: Ishara ya kukubali kushiriki

Mshiriki: Nimeelewa vema maelezo yalio hapo juu na nitashiriki katika utafiti.

_____ _____

Sahihi/kidole gumba Tarehe. Jina *(liandikwe)*

Shahidi: _____ _____.

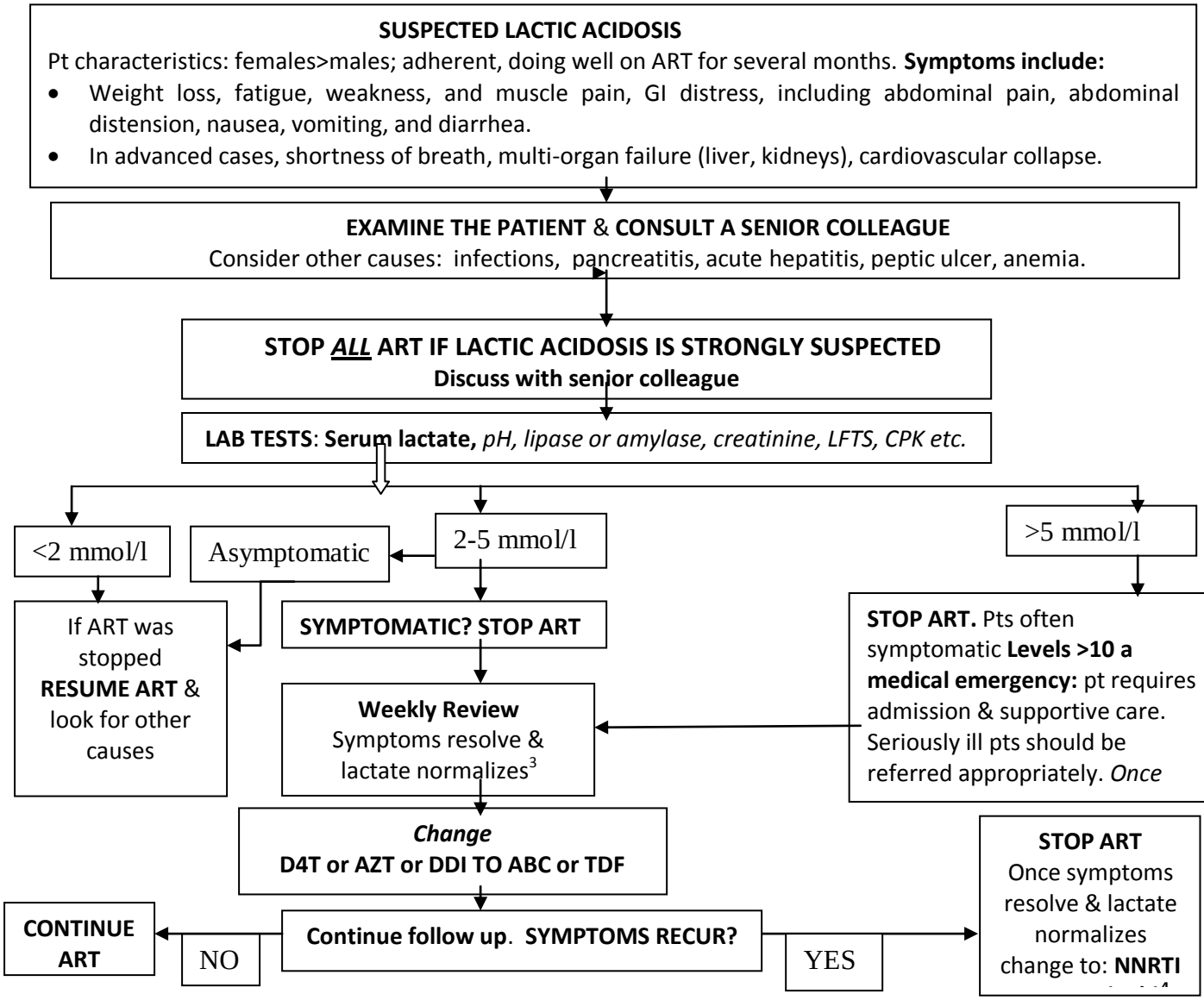
Sahihi Tarehe Jina *(liandikwe)*

Mchunguzi anayepokea kukubali:_____ _____

Sahihi na Tarehe *Jina(liandikwe*)

Appendix 5: Algorithm for the management of hyperlactatemia

(Adapted from the National ART clinical manual, 2nd edition page 43)



Normal	Mild (2-5mmol/l)	Moderate (5-10mmol/l)	Severe (> 10mmol/l)
Serum lactate < 2mmo/l	**Mild hyperlactatemia in asymptomatic pts requires no intervention. Symptomatic pts need NRTI modification**	STOP ART**. Provide supportive therapy. Substitute offending NRTI once pt recovers** **HIGH MORTALITY**	STOP ART. **Provide necessary supportive therapy. Substitute offending NRTI once pt recovers.** **VERY HIGH MORTALITY**

Appendix 6: SOP of measurement of blood lactate levels

Background

Accutrend® lactate meter by Roche diagnostics® will be used to determine the blood lactate levels in this study. A study done to validate Accutrend lactate meter for hyperlactatemia screening during antiretroviral therapy in resource limited settings gave a sensitivity of 95.9% with a specificity of 63.8%. The study concluded that Accutrend lactate meter is an appropriate device for screening of patients on HAART with suspected hyperlactatemia (Ernesto et al., 2008).

Standard Operating Procedure (SOP) for Lactate measurement by Accutrend® Lactate meter

1. Person responsible: Laboratory technologist, clinician or a nurse
2. Requirement for the test: Lactate meter, lancet, lactate strips, lactate controls, gloves, alcohol/spirit swabs, * floride oxalate tubes if transportation of blood will be required.
3. Sample to be used for the test: Venous blood or plasma
4. Precautions before and during sample collection:
    - Clients should have rested for at least 15 minutes
    - Do not apply tourniquet
    - Do not squeeze the finger as this will give false positive results
5. Sample collection and measurement of capillary blood
    - The client should wash their hands with soap and ensure they are warm and dry before lancing
    - Gently massage the client's finger before pricking on the side of the finger tip.

- Prick on the side of the finger and try to obtain a sufficiently large free hanging drop of blood without excessive pressing or squeezing.
- Apply the free hanging drop of blood directly from the finger to the yellow sample application area of the test strip. The application area **MUST** be completely covered with blood, otherwise wrong results will be obtained. If too little blood is applied, do not try to spread it or add a second drop, repeat the measurement with a new test strip. The drop must be applied immediately after pricking to avoid coagulation which will give incorrect results.
- Close the measurement chamber flap to start measurement. For lactate measurement it takes 60 seconds

NB: Venous blood can also be collected using floride oxalate tubes and lactate measurements done within 6 hours.

6. Results:

Low values (LO) - This denotes that the amount of blood was in adequate and you should repeat the test with a new test strip OR levels below 0.8 (blood values) and 0.7 (plasma values)

High values (HI) – This can be caused by wrong blood collection procedure, repeat the test with a new test strip OR levels above 21.7 (blood values) and 26 (plasma values).

Interpretation of results:

Normal levels (< 2.5 mmol/l); Mild hyperlactatemia (2.5-5 mmol/l); Moderate hyperlactatemia (5-10 mmol/l) and severe hyperlactatemia (5-10 mmol/l).

7. Management:

Refer to the algorithm on management of hyperlactatemia in the national guidelines

Appendix 7: Proposed lab request form for lactate measurement

(To be produced in duplicate)

LAB REQUEST FORM FOR LACTATE MEASUREMENT

Date of Test: _____/_____/_____ **Health facility:** ……

Unique patient ID. No (CCC No.): ……

Age: ……..

Sex: ☐ Male ☐ Female

Fever: ☐ Yes (>37.2) ☐ No (<37.2)

Weight (Kgs): ……..

Height (cms): …….

BMI: …….

Type of sample: ☐ Blood ☐ Plasma

Presenting signs and symptoms

Duration on current ART regimen (months): ……

ART Regimen: ……

Lactate levels: …….

Requested by: …….
(Name and signature)

Done by: ……
(Name and signature)

Appendix 8: Proposed register for recording diagnosis of patients with hyperlactatemia

Proposed register to record diagnosis and follow-up of patients with lactic acidosis due to ART

Serial No.	**Date**	**Age sex**	**Wght BMI**	**Unique pt No.**	**Presenting signs and symptoms**	**Duration on ART (months) ART Regimen**	**Lactate levels Date of test**				**Remarks**
							1st	2nd	3rd	4th	

Appendix 9: Ethical approval

KENYA MEDICAL RESEARCH INSTITUTE

P.O. Box 54840 - 00200 NAIROBI, Kenya
Tel: (254) (020) 2722541, 2713349, 0722-205901, 0733-400003; Fax: (254) (020) 2720030
E-mail: director@kemri.org info@kemri.org Website:www.kemri.org

KEMRI/RES/7/3/1 **March 23, 2010**

TO: **MR. ELIUD MWANGI (PRINCIPAL INVESTIGATOR)**
ITROMID STUDENT, TM310/0082/2007

THRO': **DR. YERI KOMBE,**
THE DIRECTOR, CPHR,
NAIROBI

Forwarded 24/3/10

RE: **SSC PROTOCOL NO. 1667 (*RE-SUBMISSION*): ASSESSING THE PREVALENCE AND CORRELATES OF HYPERLACTATEMIA AMONG HIV-INFECTED PATIENTS ON TREATMENT WITH NUCLEOSIDE REVERSE TRANSCRIPTASE INHIBITORS AT THIKA AND AHERO DISTRICT HOSPITALS, KENYA.**

Make reference to your letter dated March 11, 2010. Thank you for your response to the issues raised by the Committee. This is to inform you that the issues raised during the 175th meeting of KEMRI/National Ethical Review Committee held on 16th February 2010, have been adequately addressed.

Due consideration has been given to ethical issues and the study is hereby granted approval for implementation effective this **23rd day of March 2010,** for a period of twelve (12) months.

Please note that authorization to conduct this study will automatically expire on **22nd March 2011.** If you plan to continue with data collection or analysis beyond this date, please submit an application for continuing approval to the ERC Secretariat by **8th February 2011**.

You are required to submit any amendments to this protocol and other information pertinent to human participation in this study to the ERC prior to initiation. You may embark on the study.

Yours sincerely,

RCTKithinji

R. C. KITHINJI,
FOR: SECRETARY,
KEMRI/NATIONAL ETHICS REVIEW COMMITTEE