FIRST LINE ANTI- TB DRUG RESISTANCE AMONG HIV INFECTED PATIENTS ATTENDING COMPREHENSIVE HEALTH CARE CENTRE, NAIROBI, KENYA

LUCY OBONYO NYANG'AU

MASTER OF SCIENCE

(Medical Microbiology)

JOMO KENYATTA UNIVERSITY OF

AGRICULTURE AND TECHNOLOGY

2016

First line anti- TB drug Resistance among HIV infected Patients attending Comprehensive health care centre, Nairobi, Kenya

Lucy Obonyo Nyang'au

A thesis Submitted In Partial Fulfillment of the requirements for the degree of Master of Science in Medical Microbiology, Jomo Kenyatta University of Agriculture and Technology.

2016

DECLARATION

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

Signature..... Date…….

Lucy Obonyo Nyang'au

This thesis has been submitted for examination with our approval as the university supervisors

Signature..... Date…….

Prof. Zipporah Ng'ang'a

JKUAT, KENYA

Signature..... Date……

Dr. Evans Amukoye

CRDR, KENYA

DEDICATION

I dedicate this thesis to my immediate families, who have tirelessly supported me all along. Their prayers and moral support have been the driving force that has enabled me to achieve the targeted goal in my studies.

ACKNOWLEDGEMENTS

I owe special thanks to my supervisors Prof. Zipporah Ng'ang'a of JKUAT, Dr. Evans Amukoye of KEMRI for their guidance, encouragement, and support throughout the project work. Thanks and appreciation to all KEMRI ITROMID and JKUAT staff, for playing a role either directly or indirectly in the success of this project. May God bless you.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF APPENDICES

ACRONYMS/ABBREVIATIONS

AFB	Acid Fast Bacilli
AIDS	Acquired Immune Deficiency Syndrome
ART	Anti-retroviral Therapy
ATCC	American Type Culture Collection
CCC	Comprehensive Care Centre
CD4	Cluster of Differentiation
DOTS	Directly Observed Treatment Short Course
DR	Drug Resistance
DRTB	Drug Resistant Tuberculosis
DST	Drug Susceptibility Testing
ERC	Ethical Review Committee
FDC	Fixed Dose Combination
GTP	Global Tuberculosis Program
HCL	Hydrochloric Acid
HIV	Human Immune Deficiency Virus
IPT	Isoniazid preventive therapy
IUALTD	International Union Against, Tuberculosis and Lung Diseases
JKUAT-BPS	Jomo Kenyatta University of Agriculture and Technology, Board of Postgraduate Studies
KEMRI	Kenya Medical Research Institute
LAB	Laboratory
LJ	Lowenstein Jensen
MDR-TB	Multi Drug Resistant Tuberculosis
MGIT	Mycobacterium Growth Indicator Tube
Min	Minutes
MLS	Millilitres
MTB	*Mycobacterium Tuberculosis*
NALC	N-Acetyl L-Cysteine
NAOH	Sodium Hydroxide

NASCOP	National Aids and Sexually Transmitted Diseases Control Program
NLTP	National Leprosy and Tuberculosis Control Program
PC	Personal Computer
SPSS	Statistical Packages for Social Scientists
SSC	Scientific Steering Committee
TB	Tuberculosis
WHO	World Health Organization
PANTA	Polymyxin B, Ampotericin B, Nalidixic Acid, Azlocillin

DEFINITION OF TERMS USED

Drug Resistance	This is a state when *Mycobacterium tuberculosis* organisms are resistant to antimicrobial agents at the levels attainable in blood and tissue.
Resistance Patterns	These are drug resistance sequences.
Mono-Resistance	Resistance to only one drug
Poly-Resistance	Resistance to more than one drug, except to both H and R
Multi-drug Resistance	Resistance to at least both H and R with or without resistance to any other TB drugs
Re-treatment TB Cases	These are people with relapse, failure in TB treatment **or** have treatment after defaulting.
New TB Cases	These are cases having TB disease for the first time and thus never been treated for it.
Spot Sample	Sample taken as soon as patient presents to the health facility.
Participants	These are human subjects involved in the research process

ABSTRACT

Drug resistant tuberculosis occurs when *Mycobacterium tuberculosis (MTB)* organisms become resistant to antimicrobial agents at the levels attainable in blood and tissue. This is a major challenge in tuberculosis care and control. Scarce data exists in areas with high rates of tuberculosis and Human Immunodeficiency Virus co- infection. The study aimed at determining first line anti-TB drug resistance. A cross-sectional study was conducted among new and re-treatment HIV infected pulmonary tuberculosis patients fifteen years and older in Nairobi, Kenya in 2013. A total of 215 patients were enrolled for the study. One hundred and thirty eight results were analyzed, 79 (57.2%) were male and 59 (42.8%) female. New cases were 34% while 66% were retreatment cases. Sputa with bacteriologically confirmed pulmonary tuberculosis were cultured on Mycobacterium Growth Indicator Tube media. Strains of MTB complex were subjected to drug susceptibility testing for isoniazid, rifampicin, streptomycin, and ethambutol using the proportional method on (MGIT). Forty three (31.2%) isolates showed resistance to at least any one drug tested, while 112 (81.2%) were susceptible. Resistance to any one drug tested regarding treatment status, showed high resistance to isoniazid in re-treatments and none to new cases (17.6% and none) respectively. Six (6.6%) and none were multi drug resistant among retreatment and new cases respectively. Analysis of CD4 count revealed a median CD4 count of 286 cells /µl, indicating advanced H.I.V disease. Resistance pattern among patients with CD4 count of < 200 cells /µl and CD4 $\geq$200 cells /µl, showed high resistance to isoniazid 6 (11.8%) and 10 (11.5%) respectively. Three (5.9%), and 3(3.4%) isolates, had multidrug resistance against isoniazid and rifampicin. There were no significant associations between the various resistant patterns and levels of CD4. The study revealed high levels of drug resistance among those previously treated. This implies that the drug resistant strains are not passed to the new cases.

CHAPTER ONE
INTRODUCTION

1.1 Background Information

Tuberculosis is a serious public health problem, a third of the world population are infected with TB, 9 million develop TB disease and close to 1.4 million die annually, 610,000 develop MDR-TB while 8% of TB cases are co-infected with HIV (WHO, 2013). Ninety five percent of these TB cases occur in developing countries, where 1 in 14 new cases occur in individuals who are infected with HIV (WHO, 2013). The incidence of TB associated with HIV is believed to have peaked at 1.39 million in 2010 and is now decreasing (Zwang *et al.,* 2012). However TB remains the most common cause of death among patients with AIDS, killing one in three patients (WHO, 2013). According to the WHO global report of 2013, 1.0-1.2 million (11-14%) TB cases were among people living with HIV. The proportion of TB cases co-infected with HIV was highest in African region countries. Overall, 34% of TB cases were estimated to be coinfected with HIV in this region, which accounted for 78% of TB cases among people living with HIV worldwide. In parts of Southern Africa, more than 50% of TB cases were coinfected with HIV.

Africa remains the part of the world where drug resistance surveillance data are most lacking, largely as a result of weak laboratory infrastructure. In the African and European regions in 2013, the numbers of notified MDR-TB cases were equivalent to 74% and 61% respectively, of the estimated MDR-TB cases among notified pulmonary TB patients. Highest rates of TB are reported in the countries of Eastern Europe, where weakened economies and public health efforts are the main causes of its resurgence, and where internationally recommended control strategies need further expansion and strengthening (WHO, 2011a). In Western Europe, there are pockets of increasing incidence, particularly in major cities with socially marginalized immigrants from high burden TB countries (WHO, 2012a; Chan *et al.,* 2011*)*.

Studies on drug resistance in various countries in the 1960's showed a much higher incidence of drug resistance in developing than developed countries (Bonnet *et al.,* 2010). Resistance to isoniazid and streptomycin was more than resistance to rifampicin and ethambutol and the rate of primary drug resistance to isoniazid as a single agent ranged from 0 to 16.9% among HIV infected individuals (Taha *et al.,* 2013). Isoniazid forms the core of anti-tuberculosis drugs, and its use in TB preventive therapy has been known to reduce incidence in high risk individuals for more than 40 years (Ferebee *et al.,* 2012). Despite the confirmed efficacy of preventive therapy, concerns about drug resistance, have limited its uptake (Ferebee *et al.,* 2012). HIV infection by impairing the cell mediated immunity is the most potent known risk factor for the reactivation of latent TB infection and rapid progression to active disease (Mcshane, 2010).

Overall an estimated 8% of new cases are attributable to HIV co- infection (WHO /GTP, 2012). An estimated 13% of the 1.5 million TB deaths in 2010 were attributed to HIV infection, but in the African region this proportion has been much higher because of the high HIV prevalence (Corbett *et al.,* 2010). Some groups of people are at higher risk for TB. Risk factors for TB infection include; poor housing and crowding, large pool of untreated persons. Risk of developing disease after infection is increased by low immunity, this includes: HIV, diabetes, cancer, old age (Squire *et al.,* 2011). Globally, people living with HIV are 29 times more likely to develop TB disease than those, who are HIV negative (WHO/GTP, 2012). Control of drug resistance involves; identifying and treating TB drug resistant cases, treating all TB cases, treating latent TB, improving cure rate, active case finding, and reducing development of secondary drug resistance by improving adherence (DLTLD, 2012).

Introduction of the first anti-tuberculosis drugs, isoniazid, streptomycin, para-amino salicylic acid, was slowly followed by resistance, which was observed in clinical isolates of *MTB* (Pozniak *et al.,* 2011). Over 60% of new cases of pulmonary TB in most developing countries are now co-infected with HIV (Squire *et al.,* 2011). The WHO approach (identifying of TB bacilli microscopically) to TB diagnosis is failing

in a number of HIV infected patients, as smear negative TB has been linked to poor treatment outcomes, including death (Blomberg *et al.,* 2011). According to the national MDR-TB surveillance data, approximately 150 HIV positive individuals were diagnosed with MDR-TB and three cases confirmed with XDR -TB in Kenya (WHO, 2013). These figures have been on the rise because of inadequate and insensitive diagnostic methods, leading to increased mortality and morbidity among those infected (DLTLD, 2012).

The emergence of *Mycobacterium tuberculosis* strains that are resistant to antimicrobial agents, has received much attention owing largely to the dramatic outbreaks of multidrug resistant tuberculosis in HIV infected patients in the United States. These outbreaks have been characterized by delayed diagnosis, inadequate treatment regimens, high mortality and significant rates of nosocomial transmission (Deun *et al.,* 2012). Resistance of *M. tuberculosis* to antimicrobial agents is a world wide problem in both immunocompetent and HIV infected populations (Cole, 2010). The true global magnitude of drug resistance is not well described (Raviglione *et al.,* 2011). There are several limitations to adequate assessment of this problem, especially in developing countries. In many areas, there are few or no facilities for culture of *M. tuberculosis*, and where they exist, antimicrobial susceptibility testing is often not performed. In many surveys, small or non representative populations have been sampled or and there have been few longitudinal data bases. This makes it difficult to monitor trends (Deus *et al.,* 2012).

World wide an estimated 3.5% (95% C.I: 2.2-4.7 %) of new cases and 20.5% (95% C.I 13.6-27.5%) of previously treated cases have MDR-TB. Extensively drug resistant TB (XDR-TB) has been reported by 100 countries. On average, an estimated 9.0% (95 % C.I: 6.5-11.5 %) of people with MDR-TB have XDR-TB. Despite the progress in the detection of MDR-TB cases a major diagnostic gap remains. Fifty five percent of reported TB patients estimated to have MDR-TB were not detected in 2013. The detection figures were lowest in the Eastern Mediterranean region (22%) and the Western Pacific region (16%). Innovative approaches and more

funding to increase the uptake of programmatic management of DRTB globally are urgently required to detect and enroll more patients on MDRTB treatment, and to improve outcomes.

1.2 Statement of the problem

Tuberculosis, HIV, anti-TB drug resistance and poverty are emerging in third world countries as significant threats to public health, resulting in high mortality and morbidity; especially in people with HIV (Saadoun *et al.*, 2011). Antimicrobial resistance is a global concern because infections caused by resistant strains often fail to respond to the standard treatment resulting in prolonged illness and greater risk of death (Nunes *et al.*, 2011). The exact burden of drug resistant TB in Kenya is currently not known, as no recent drug resistance survey has been done. This poses a challenge because it is hard to make informed decisions or to rate progress made in drug resistant TB control, without an accurate reference (Abate and Miorner 2011). The threat of drug resistant TB has been heightened by the displacement of an estimated 300,000 people, of which settlement has taken along time in Kenya's 2007 political crisis,

1.3 Justification

Gaps in knowledge on diagnostics and detection of TB, and DRTB exist in areas with high HIV prevalence, especially developing countries (Taha *et al.*, 2013). Diagnosis of TB in HIV patients is difficult because transfer of bacilli into respiratory secretions is markedly reduced; leading to increased smear negatives (Kawai, 2010). Lack of accurate reference poses a challenge in making informed decisions on tuberculosis control, (Getahun *et al.*, 2012). In Kenya tuberculosis in HIV causes an average mortality of 30% (NASCOP, 2012). This Mortality is attributed to undiagnosed DRTB cases, because of inaccessibility to sensitive techniques (NASCOP, 2012). Therefore additional specialized services are needed to ensure that patients with HIV and TB have better treatment outcomes. The increased risk of TB drug resistance in HIV needs heightened attention (Getahun *et al.*, 2012). Currently

drug susceptibility testing is carried out only in re-treatment cases and those found to be failing, this may be too late for HIV patients (Kawai, 2010). WHO recommends continued studies of drug resistance especially isoniazid in the context of isoniazid preventive therapy (IPT) therapy, which is increasingly being promoted but raises concern about risk for isoniazid resistant TB development.

1.4 Research questions

- What is the prevalence to first line TB drugs among HIV infected TB (new and retreatment cases) attending comprehensive care centre in Nairobi Kenya?
- What are the patterns of resistance to first line tuberculosis drugs among HIV infected TB attending comprehensive care centre in Nairobi Kenya?
- What are the patterns of TB drug resistance with respect to CD4 cell count, among HIV infected TB patients attending comprehensive care centre in Nairobi Kenya?

1.5 Objectives

1.5.1 General objective

- To determine first line anti-TB drug resistance and CD4 count among HIV infected patients attending a comprehensive care centre in Nairobi Kenya.

1.5.2 Specific objectives

1. To establish the prevalence of drug resistance to first line TB drugs among new and re-treatment TB cases.
2. To determine drug resistance patterns to first-line TB drugs with respect to CD4 count.
3. To determine drug resistance patterns to first-line TB drugs among new and retreatment cases.

CHAPTER TWO
LITERATURE REVIEW

2.1 Mycobacterium tuberculosis

Mycobacterium tuberculosis is a pathogenic bacterium that belongs to the class actinomycetes, order actinomycetales and family Mycobacteriaceae (Chan *et al.,* 2011). The genus *Mycobacterium* includes obligate parasites, saprophytes and intermediate forms (Chan *et al.,* 2011). The bacterium is typically slender, straight or slightly curved and rod in shape. The size of the bacillus ranges from 0.3um to 0.6um in width and from 0.5um to 0.4um in length. This bacterium is slow growing, non-capsulated, non-spore forming, non motile and lipid rich organism (Lawn and Nicol, 2011). The cell wall of *M.tuberculosis* is similar to that of gram positive organisms except that it has higher lipid content. The lipids are long chain fatty acids that present difficulties during staining, therefore heat or increased concentration of stain is required to achieve staining (Lawn and Nicol, 2011). This unique cell wall of Mycobacteria is also responsible for its resistance to the lethal effects of acids, alkalis, and detergents. This characteristic is fully exploited for isolation of the organism from other bacteria for culturing (Pozniak *et al.,* 2011). *M.tuberculosis* optimally grows at temperature range of 35°C- 37°C (Cole, 2010).

2.2 Transmission of Mycobacterium tuberculosis

Tuberculosis is spread from person to person by aerogenic transmission. The source of infection is a patient with pulmonary TB who coughs and spreads tiny droplets. A single cough may produce up to 3000 droplets, where each one contains one or more tubercle bacilli (Dye *et al.,* 2011). Under normal circumstances small proportions (about 10%) of all individuals who are infected by the tubercle bacilli develop the disease in their lifetime. Vast majority (90%) of people exposed to the bacteria except those with HIV infection do not develop the disease (Uplekar *et al.,* 2010).

2.3 Pathology of Mycobacterium tuberculosis

Healthy people exposed to relatively low numbers of bacteria generally clear them before appreciable damage to the lungs occurs. But if the phagocytic cells do not clear the infection, T cells, polymophonuclear cells, and macrophages continue to be attracted to the area where bacteria are growing (Cole, 2010). In some cases the phagocytes fail to kill the bacteria and the T cells and macrophages protect the growing lesion with a thick fibrin coat. The walled off lesion is called tubercle. Tubercles eventually calcify, giving rise to hard edged lesions visible in chest X-rays. Phagocytes unsuccessfully trying to kill the bacteria cause considerable damage to the lung tissue by releasing lysosomal enzymes and tumor necrosis factor (TNF). Tumor necrosis factor (TNF) causes tissue damage and is probably responsible for the weight loss that occurs in people with tuberculosis. Initially the areas where bacteria are growing have a thick cheese like appearance. As bacteria continue growing and phagocytes continue to enter the area, the necrotic region becomes much more liquid (Boehme *et al.*, 2011).

2.4 Clinical manifestation of *Mycobacterium tuberculosis*

Mycobacterium tuberculosis has many manifestations affecting bone, the central nervous system and many other organ systems, but it is primarily a pulmonary disease (Drobniewski *et al.*, 2011). Pulmonary tuberculosis patients usually have weight loss and productive cough for more than three weeks. Symptoms like haemoptysis, chest pain, dyspnea, fever, night sweats and anorexia have also been shown to be common among TB patients (Deus *et al.*, 2012; Goyal *et al.*, 2012). Cough is the commonest presentation, initially it may be non productive, but as inflammation and tissue necrosis ensue, sputum is produced (Suzuki *et al.*, 2010). Elderly individuals with TB may not display typical signs and symptoms of TB infection, because they may not mount a good immune response (WHO, 2011b). Active TB infection in this age group may manifest as non resolving pneumonitis (Gillespie, 2011; Yoon *et al.*, 2012)

2.5 TB/HIV Co-infection

Tuberculosis and HIV nine million new cases have been closely linked since the emergence of AIDS. HIV infection has contributed to significant increase in the world wide incidence of TB (Dye *et al.,* 2011). By producing a progressive decline in cell mediated immunity; HIV alters the pathogenesis of TB, leading to more frequent extra-pulmonary involvement and atypical radiographic manifestations (Espinal *et al.,* 2012). Globally, TB is the most common opportunistic infection affecting HIV infected individuals and the most common cause of death among them (Raviglione *et al.,* 2011). WHO estimates indicate that one third of the world's population is infected with MTB, resulting in an estimated of TB and nearly 1.4 million deaths yearly (WHO, 2012a). Approximately ten million individuals are estimated to be co-infected with MTB and HIV, and over 90% of these dually infected individuals reside in developing nations. In some areas of sub- Saharan Africa, the rates of co-infection exceed 1000 per 100,000 (Saadoun *et al.,* 2011; WHO, 2012b). The global distribution pattern of TB has changed considerably since the late 1990's, when most of the sub Saharan African countries were categorized together in the highest infection rate class. This change is due to the emergence of HIV/AIDS in Africa. The rate of TB infection rises sharply with an increased prevalence of HIV, as TB is an opportunistic disease in those with weakened immune system (Suzanne *et al.,* 2011).

2.6 Global epidemiology of TB drug resistance

Drug resistant tuberculosis occurs when Mycobacterium tuberculosis (MTB) organisms are resistant to antimicrobial agents at levels attainable in blood. The causes of drug resistance include, host factors: prior exposure to TB drugs, poor adherence to treatment by the patient, mal-absorption of the drugs and genetic susceptibility of the host (Gillespie, 2011). Environmental factors: inefficiency in the TB program by the care providers and partners and prescription errors by the clinician. Agent factors: genetic changes of the Mycobacterium genotype (Sharma & Mohan 2010; Gillespie, 2011). Drug resistant tuberculosis (DRTB) is a global threat

to TB control programs, with half a million new cases being notified to the WHO in year 2012 (WHO, 2012a).The global distribution of DRTB was poorly defined until 1994, when the international union Against TB and lung Disease and World Health Organization (WHO/IUATLD) launched the global project on anti-tuberculosis drug resistance surveillance. The first report was based on data collected from 35 countries in 5 continents, representing approximately 50,000 patients worldwide. Resistance to anti-TB drugs such as isoniazid and streptomycin was found in all 35 countries and regions surveyed. Overall, 9.9% of patients with tuberculosis had drug resistance, and multidrug resistant tuberculosis ranged from none to 14% in new cases and none to 54% in re-treatment cases (WHO, 2011b).

In new TB cases, the prevalence of resistance to more than one drug ranged from 1.7% in Uruguay to 36.9% in Estonia, with a median of 10.7%. MDR-TB ranged from 0-16% for new cases and 0- 48% re-treatment cases (WHO, 2012a). A number of regions with a high prevalence of drug resistance among new cases were identified, including Ivanovo blast in the Russian Federation (32.4%), Latvia (19.9%) and Henan province, China (35%). Particular areas of concern with a high prevalence of MDR-TB in new TB cases were Estonia (14%), Latvia (9%), Ivanovo (9%), and Tomsk Oblasts (6.5%), in the Russian Federation, and Henan province (11%) in China. The high rates of MDR-TB found in the two most populous countries in the world, China and India is of significance because these countries account for 40 % of all TB cases worldwide. In Henan province, the most populous province in China, 11 % of new cases had MDR-TB. In Tamil Nadu, India, 3.4% of new cases had MDR-TB, the prevalence of isoniazid resistance was 15% and rifampicin resistance was found in 4.4%.

In a global survey conducted by WHO/GTP, (2013) resistance to isoniazid and streptomycin was more than resistance to ethambutol and rifampicin among HIV infected patients. The rate of primary drug resistance to isoniazid as a single agent ranged from 0 to 16.9%. There were several reports of high rates of primary resistance to isoniazid from Africa, Asia, and North America (WHO, 2011a). The rate

of resistance to isoniazid was 10.2% in Kenya, 16.9% in Karnataka State, India, and 12.2 % in Haiti. In contrast most regions in Europe, Oceania and South America reported low rates of isoniazid resistance. The rates of primary resistance to streptomycin were similar to the rates of primary resistance to isoniazid; these rates ranged from 0.1% to 23.5% and 0 to 16% respectively. Ethambutol had a rate ranging from 0 to 4.2 %. Resistance to multiple drugs varied by region and was more common when resistance was acquired rather than primary (WHO/IUATLD, 2013). According to the recent international union Against TB and lung Disease and World Health Organization survey, resistance is not yet a problem in most sub-Saharan African countries. This may be because rifampicin based regimens have been introduced only recently. In addition, rifampicin sparing regimens are often used in the continuation phase, and a growing number of countries use directly observed therapy. However countries such as Mozambique, Cote D'Ivoire, Cameroon, Argentina, the Dominican Republic, and Mexico are a concern because the prevalence of MDR-TB in new cases is approximately 3%.

The full magnitude of TB drug resistance is still unknown in a number of countries with high TB incidence, such as Democratic Republic of the Congo, Ethiopia, Kenya, Nigeria, Indonesia, Bangladesh and Pakistan.World Health Organization/International Union Against, Tuberculosis and Lung Diseases IUATLD reports show that a well functioning TB control program is associated with low prevalence of TB DR and that previous anti-TB therapy is a strong predictor of drug resistance. Although HIV infection has been linked to primary MDR-TB in institutional outbreaks in the developed world, this association was not found in the geographic hot spots of resistance (Drobniewski *et al.,* 2011). There is no evidence that HIV infection is associated with the development of MDR-TB. In settings where MDR-TB is being transmitted, HIV infected people who acquire infection with a resistant organism are at a greatly increased risk of progressing to clinical illness with the resistant strain (Shah *et al.,* 2010). In addition, because HIV infected people are more likely than healthy individuals to visit hospitals and other health facilities where MDR-TB is transmitted, they have an added risk for infection (Kassu

et al., 2011).

2.6.1 TB drug resistance in Kenya

Kenya participated in the global anti-TB DR survey WHO/GTP (2012), and reported no MDRTB. The isoniazid mono-resistance was however reported to be 5% and 10% for primary and combined resistance respectively (WHO, 2013). For streptomycin a combined resistance of 2% was reported, no resistance to rifampicin was reported. A study conducted by Keter *et al.* (2014), in North Eastern Kenya to estimate and compare patterns of drug resistance among refugees from Somalia and non refugee population, showed that drug resistance was significantly higher in patients from the refugee population (10.3 %) compared to patients from the non refugee population (4.7%). In addition, 2.7% of the cases of DRTB among the refugee population were MDRTB while the non refugee population had no MDRTB. However the real burden and patterns of DRTB in Kenya is not well known and needs further assessment. The control of drug resistant TB is technically demanding and may pose a threat to the successes gained in TB control over years (WHO, 2013b). WHO estimates that there were around 2000 cases of MDRTB in Kenya in 2007, although only 4.1 % of these cases were diagnosed and notified (WHO, 2012b). In Kenya, the threat of MDR-TB has been heightened by the displacement of an estimated 300,000 people in Kenya's recent political crisis (WHO/GTP, 2012). During the crisis, many people were displaced, which meant disruption from drugs.

2.7 Diagnosis of drug resistant tuberculosis

Conventional methods that use solid media (Lowenstein Jensen media or 7H10 middlebrook agar) take 6 to 9 weeks to obtain results. The more rapid methods such as Bactec are very expensive for routine use in high TB endemic countries. Therefore simple, rapid and relatively inexpensive methods are desired particularly for low income countries (Nunes *et al.,* 2011).

2.7.1 Proportion method

This method is based on the principle of calculating the proportion of resistant bacilli present in a strain. High and low dilutions of bacteria are inoculated on drug free and drug containing media, in order to provide numerable colonies. The ratio of the number of colonies between the the two media indicates the proportion of resistant bacilli present in the strain. Resistance is defined as growth above (1%) in drug containing media as compared to drug free control. This method is preferred to others because it does not require standardization of inoculum size (Lonroth *et al.,* 2011).

2.7.2 Colorimetric assay: 3-(4, 5-dimethylthiazol-2-yl-2, 5-diphenyl tetrazolium bromide assay.

This assay is a colorimetric method that uses a yellow tetrazolium salt, 3-(4, 5-dimethylthiazol-2-yl-2, 5-diphenyl tetrazolium bromide (MTT) that is converted by dehydrogenase enzyme in living cells to produce insoluble blue formazan crystals. The formazan can then be measured by a spectrophotometer. The amount of formazan produced is directly proportional to the number of living cells (Standford *et al.,* 2012). The same principle can be used to detect the viability of *M. tuberculosis* after exposure to rifampicin. The presence of rifampicin resistance is confirmed when at least 1% of the bacterial populations are composed of drug resistant strains. The assay is cheap, requires less than 4 to 5 weeks and is visually readable (Kent & Kubica, 2010).

2.8 Treatment of tuberculosis

2.8.1 Conventional treatment

The discovery of streptomycin in 1944 marked the beginning of the era of effective chemotherapy for mycobacterial disease, and the initial clinical trials of isoniazid in 1952 increased the effectiveness of the chemotherapy (Uplekar *et al.,* 2011). Currently tuberculosis is effectively treated using a combination of the firstline drugs, that include; isoniazid, streptomycin, ethambutol, rifapmicin and

pyrazinamide. Resistance to all of these drugs occurs at high frequency when used alone.That is why the drugs are given in combinations (Chonde *et al.*, 2010). These drugs are given to TB patients in two phases for at least 6 months. During the initial intensive phase of treatment, which lasts two months, the patients are treated with four drugs (isoniazid, rifapmicin, pyrazinamide, ethambutol and streptomycin) to ensure that mutants that are resistant to a single drug cannot emerge. In the following four or six months, the continuation phase, two drugs (isoniazid, rifapmicin or isoniazid, ethambutol) are given to all any persisting organisms (Espinal *et al.*, 2012).

In Kenya, all the five drugs (isoniazid, rifapmicin, pyrazinamide, ethambutol and streptomycin) are used as anti-TB drugs. These drugs are available in loose and fixed dose combinations (FDC). The fixed dose combination drugs exist in two forms 2FDC and 3FDC formulations. Ethambutol and isoniazid (RH) exist in 2FDC while rifampicin, isoniazid and pyrazinamide (RHZ) are available as 3FDC.

2.8.2 Fixed dose combination therapy

Drug resistance in most tuberculosis patients predominantly arises from multiple interruptions of treatment (WHO/IUATLD, 2012).When using loose drug formulations; patients are more prone to interrupt their treatment on some drugs while not on others, thereby creating arisk of monotherapy and selection of drug resistant mutants. FDC is an anti-TB drug formulation where two or more anti-tuberculosis drugs are prepared in fixed proportions in the same formulation. This is a strategy adopted to avoid problems of drug resistance. Therefore WHO and IUATLD recommend the use of FDC; this will reduce the emergence of drug resistant tuberculosis, simplify treatment by minimizing prescription error, increase patient compliance, and reduce the risk of misuse of rifampicin for conditions other than tuberculosis (WHO/GTP 2013).

CHAPTER THREE
MATERIALS AND METHODS

3.1 Study area

The study site was a comprehensive care centre within the Mathare slum, in Maryland comprehensive care health centre. This centre offers comprehensive services for T.B and H.I.V patients, in terms of treatment and diagnosis. Mathare is one of the oldest slums in Nairobi, Kenya. The area is one of the most impoverished in Kenya with (70-80%) of people living below the poverty line (KBS, 2012). The HIV prevalence in the adult population of Mathare slum, Nairobi was 15% with 50% of patients with TB being co-infected with HIV (Bonnet et *al.*, 2010). Casual employment and informal businesses constitute the principal economic activities. The area has a population of nearly 150,000 and culturally heterogenous (Fig.3.1).

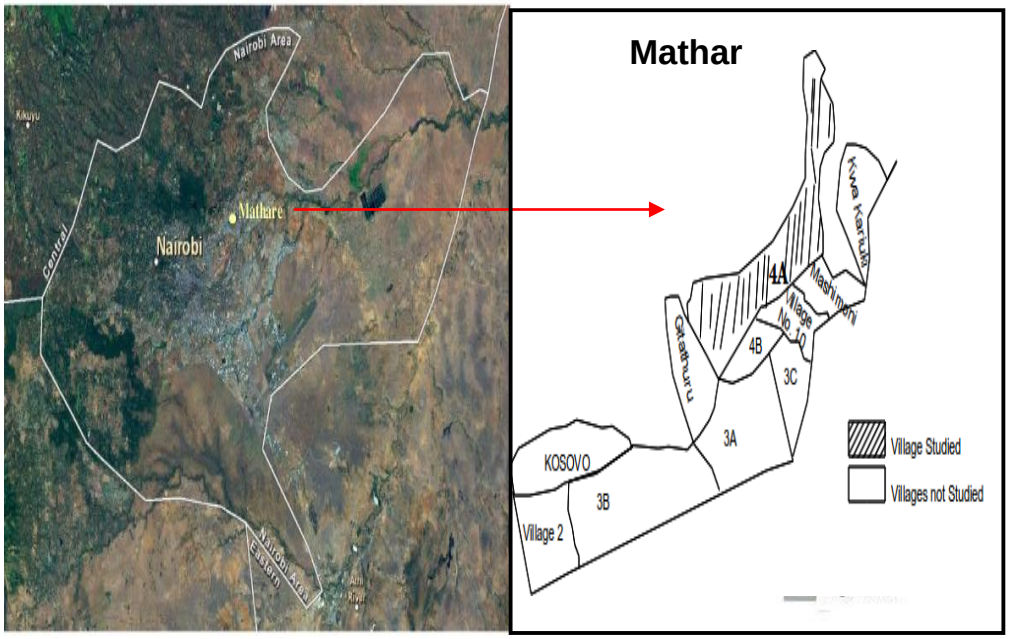

Figure. 3.1 Map of Nairobi locating Mathare

Source: MuST, UoN-UIP & UCB GeoEye Image (2010).This information is a product of a participtory Mapping process

3.2 Study design

This was a cross sectional descriptive study assessing the prevalence of resistance to first line anti-TB drugs and the resistance patterns among HIV/TB coinfected patients, whereby new and re-treatment cases were captured.

3.3 Study population

The study population of nearly 150,000 comprised of new and re-treatment TB cases, of different ethinic backgrounds.

3.3.1 Inclusion criteria

Positive status for HIV, 15 and above years of age, acceptance to sign the informed consent form by self if above 18 years, or by guardian if below 18 years, TB bacteriologically positive by geneXpert

3.3.2 Exclusion Criteria

Negative HIV status, below 15 years of age, inability to produce sputum, unable to consent or withdrawal from consent

3.4. Sample size determination

In a cross sectional study in Nairobi on T.B drug resistance, among pulmonary T.B cases by Ogaro *et al.,* 2012, a drug resistance prevalence of 10% was established among reatreatment T.B cases. Using Fishers *et al.*(1998), formula and 10% as the working prevalence rate (P) for drug resistance and assuming a standard error (Z) from the mean of 1.96 and in absolute precision (D) of 5% sample size (N) was calculated as follows. It was adjusted for attrition/refusals which were estimated for 5%.

$$n = \frac{Z^2 \propto/_2 P(1- P)}{D^2}$$

$$= \frac{1.96^2 \times 0.1(0.9)}{0.05^2} = 138 \text{ Subjects}$$

Where n = minimum sample size

Z = 1.96 Value corresponding to the 95% confidence interval

P = the prevalence of Multi drug resistant tuberculosis

D = 0.05 (The allowable error margin)

The estimated sample size was 138; it was adjusted to allow for attrition/refusals which were estimated for 5% thus:

$$n = \frac{138}{(1-0.05)} = \frac{138}{0.95} = 145 \text{ Subjects}$$

3.5 Sputum collection, transportation and storage

Two sputum samples (Spot and morning) were collected in clean wide mouthed sterile containers, from two hundred and fifteen study participants above 15 years. Genexpert was performed on one of the samples from each of the participants, in Maryland comprehensive care centre laboratory. Samples which were positive for MTB, were then transported by Smithline courier sevices to the central reference TB laboratory, in Kenyatta hospital grounds.They were stored at 4°C prior to processing.

3.6. Identification of *M. tuberculosis*

The technique used for detection of *Mycobacterium tuberculosis* from sputum samples was Genexpert adopted from WHO (2011a). Two ml of the sample buffer was added to one ml sputum, mixed well and left to stand for 10 minutes. The

mixture was shaken and left to stand for another 5 minutes. Two milliters of the supernatant was fed into each cartridge and loaded into the Genexpert machine (Hernandex *et al.*, 2011).

3.6.1 Isolation of *M. tuberculosis*

3.6.2 Decontamination of samples

The sputum was transferred into a 50 ml falcon tube with a cap. Sodium hydroxide, N-acetyl-L-cystein (NAOH- NALC solution was added in equal volume to the sputum and cap tightened (Hernandex *et al.*, 2011). The contents were lightly vortexed for 30 seconds. The tubes were inverted so that the whole tube was exposed to the Sodium hydroxide, N-acetyl-L-cystein (NAOH-NALC) solution.The tubes were let to stand for 20 min and vortexed lightly every 5-10 minutes. Phosphate buffer was added (PH 6.8) up to the top ring of the centrifuge tube and lightly vortexed.The specimen was centrifuged at 3000g for 20 minutes.

The tubes were allowed to stand for 5 minutes to allow aerosols to settle, then the supernatant was decanted into a bottle containing mycobactericidal disinfectant and 1ml of phosphate buffer added to re -suspend the sediment. The sediment was used for inoculating MGIT tubes.

3.6.3 Culture of *Mycobacterium tuberculosis* using MGIT

Mycobacterium growth indicator tube (MGIT), PANTA (Polymyxin B, Amphotericin B, Nalidixic Acid, and Azlocillin) was reconstituted with 15 ml MGIT growth supplement, and 0.8 ml of this enrichment was added to each MGIT tube in use (Kent & Kubica, 2010). MGIT tubes were labelled with respective specimen numbers. The cap was unscrewed and aseptically 0.8 ml of MGIT growth supplement/PANTA was added to each MGIT tube. While working under biologic safety cabinet, 0.5 ml of well mixed processed specimen was added to the appropriately labeled MGIT tubes. The tubes were recapped tightly and mixed by inverting several times, caps were then wiped with a mycobactericidal disinfectant

and the inoculated tubes left at room temperature for 30 minutes.

All inoculated MGIT tubes were entered in the BACTEC MGIT 960 instrument after scanning each tube. The MGIT tubes were incubated until the instrument flaged them positive. After a maximum of six weeks, the instrument flaged the tubes negative if there was no growth at 37°C (Kent & Kubica, 2010). Negative and positive controls were included along the clinical specimens. Phosphate buffer and MTB H37RvATCC27294 were used respectively. To facilitate confirmative identification of *MTB*, positive tubes were subcultured onto LJ slants by inoculating 0.1 ml of well mixed positive MGIT broth onto LJ medium. The subcultures were incubated at 37°C for 3 weeks.

3.7. Confirmative identification of *M. tuberculosis* Niacin test

One ml of sterile water was added to the culture slant. The tubes were placed horizontally for 30 minutes for extraction of niacin (Suzuki *et al.*, 2010). 0.5 ml of the fluid mixture was transffered to a clean screw cap tube, then 0.5 ml of the 4% aniline solution and 0.5 ml of 10% cyanogen bromide was added sequentially. The tubes were closed and observed for formation of color, with no color indicated negative result, while yellow color indicated positive result (Sharma *et al.*, 2011). Extract from an un-inoculated tube of medium and from culture of *M. Tuberculosis* H37Rv were used as negative and positive controls respectively.

3.8 Drug susceptibility testing using MGIT

Five MGIT tubes were labelled for each test culture, one was labelled GC (growth control without drug) one for streptomycin, one for isoniazid, one for rifampicin, and one for ethambutol. 0.8 ml of BACTEC 960 streptomycin, isoniazid, rifampicin, ethambutol supplement was added aseptically to each of the MGIT tubes. 0.1 ml of streptomycin was added aseptically in the streptomycin labeled tube; and similarly to isoniazid, rifampicin, and ethambutol labeled tubes using separate micropipette tip for each drug. No drug was added to the growth control tube. Zero point five

millilitres of the well mixed culture suspension (inoculum) was added into each of the drug containing tubes using a pipette, but not to the control. For the control the test culture suspension was diluted 1:100 by adding 0.1 ml of the test culture suspension to 10.0 ml of sterile saline. Zero point five millilitres of the diluted suspension was added into the growth control tube. The caps were tightened and the inoculated broth well mixed by gently inverting the tube several times.

Labelled tubes in the correct sequence were placed in the set carrier (growth control, streptomycin, isoniazid, rifampicin, and ethambutol). The Susceptibility set carrier was entered into the BACTEC MGIT 960 instrument using the susceptibility test set entry feature. The order of the tubes was growth control, streptomycin, isoniazid, rifampicin, and ethambutol for the standard testing.The instrument monitored the entered susceptibility test set. Once the test was complete (within 4-21 days) the instrument indicated that the results were ready. The susceptibility set carrier was scaned and report printed. The instrument print out indicated susceptibility results for each drug. Results were qualitatively reported as either susceptible (S) resistant (R) or indeterminate (X). Susceptible (the growth units of the drug tube is less than 100), resistant (the growth unit of the drug is 100 or more), Error- Indeterminate results when certain condition occur which may affect the test, such as growth units of the control reaches >400 in less than 4 days.

3.9 CD4 count

The CD4 counts were obtained from the Maryland comprehensive care centre laboratory registers. This is because patients at the comprehensive care centre had CD4 determined at baseline, and every 3-6 months. The latest CD4 count was used because culture and DST results took 6-8 weeks to access. The Becton Dickinson Facscount technique was used and quality controls both internal and external were done according to an established protocol (Standford *et al.,* 2012).

3.10 Data management and analysis

Quantitative data from the laboratory results was double entered into a computer database designed using MS-Access. Data cleaning and validation was performed in order to achieve a clean dataset that was then exported into a Statistical Package format (IBM SPSS) for analysis. File back-up was regularly done to avoid any loss or tampering. All the questionnaires (Appendix IV) which were administered upon patient consent, to capture demographic information were stored in a lockable drawer for confidentiality.

Data analysis was conducted using IBM SPSS version 21.0 statistical software. Descriptive statistics such as proportions were used to summarize categorical variables while measures of central tendency such as mean, standard deviation, and range were used to summarize continuous variables. Relationship between resistance to any of the four TB drugs (isoniazid, rifampicin, ethambutol and streptomycin) by age, and gender was tested using Pearson's Chi-square test. Similarly, Pearson's Chi-square test was used to test the association between different patterns of resistance to any of the four TB drugs (isoniazid, rifampicin, ethambutol, and streptomycin) and; treatment status as well as CD4 count level. Odds Ratio (OR) and 95% Confidence Interval (CI) was used to estimate the strength of association between different resistance patterns and treatment status, as well as CD4 count level. A p-value of < 0.05 was considered statistically significant.

3.11 Ethical considerations

Ethical clearance for the study was obtained from KEMRI Scientific Steering Committee (Appendix VI) and Ethical Review Committee (Appendix V). Consent was obtained from the study participants, and for those below 18 years consent was obtained from parents/ guardians.

CHAPTER FOUR

RESULTS

4.1 Demographic characteristics of the study population

In this study a total of 215 patients were enrolled, Out of these, 145 became sputum genexpert positive, with 70 becoming negative (Figure 4.1). Of the study participants, 79 (57.2 %) were male 59 (42.8%) female (Figure 4.2). Most study participants (52.2%) were aged 35 years and above, with (47.8%) being 35years and below (Figure 4.3). Forty seven (34.1%) of these patients were new while 91 (65.9%) were re-treatment cases (Table 4.1).

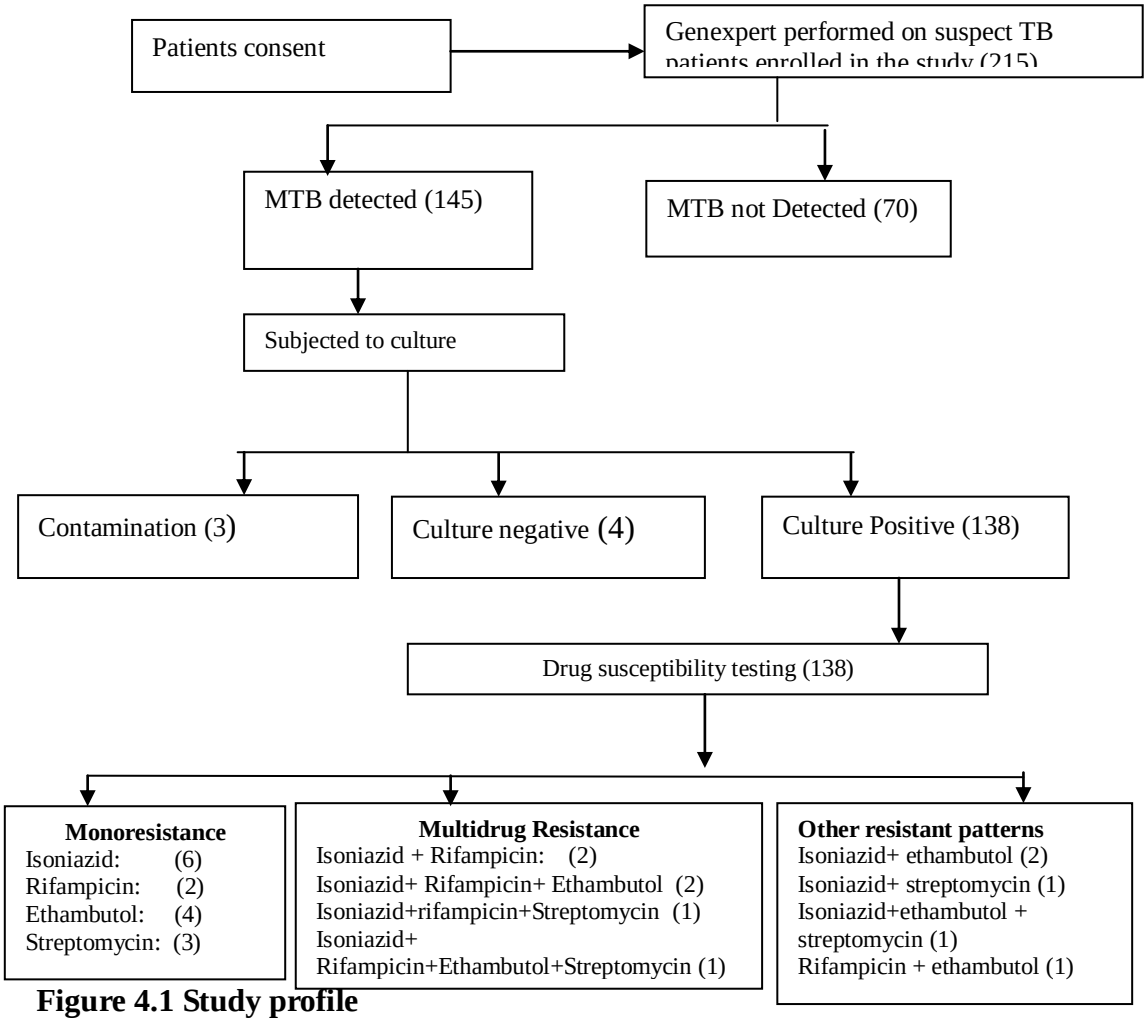

Figure 4.1 Study profile

Gender distribution was analyzed as shown in Figure 4. 2. Majority of the patients were males accounting for 79 (57.2 %) while females were 59 (42.8 %) (Figure 4.2)

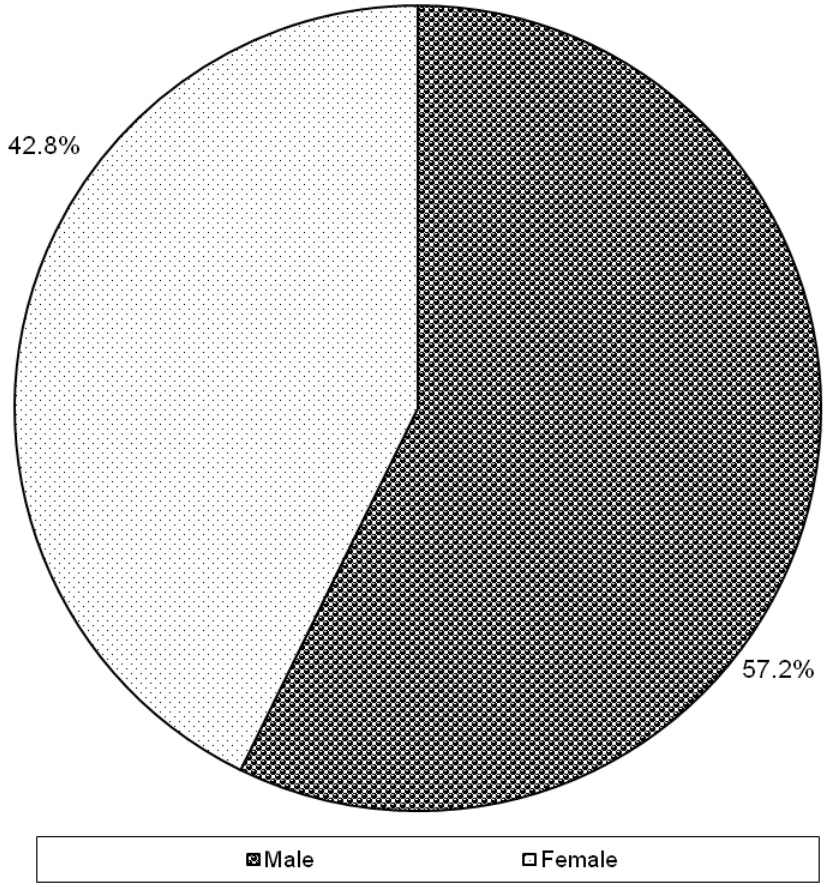

Figure 4.2: Gender distribution among the study patients

Age distribution among the participants was normally distributed with a mean age of 35.0 (±10.0 SD), and an Inter quartile range (IQR) of 12 ranging between 29 and 41 years. Most of the patients (52.2 %) were aged 35years and over, while (47.8 %) were 35years and below (Figure 4.3)

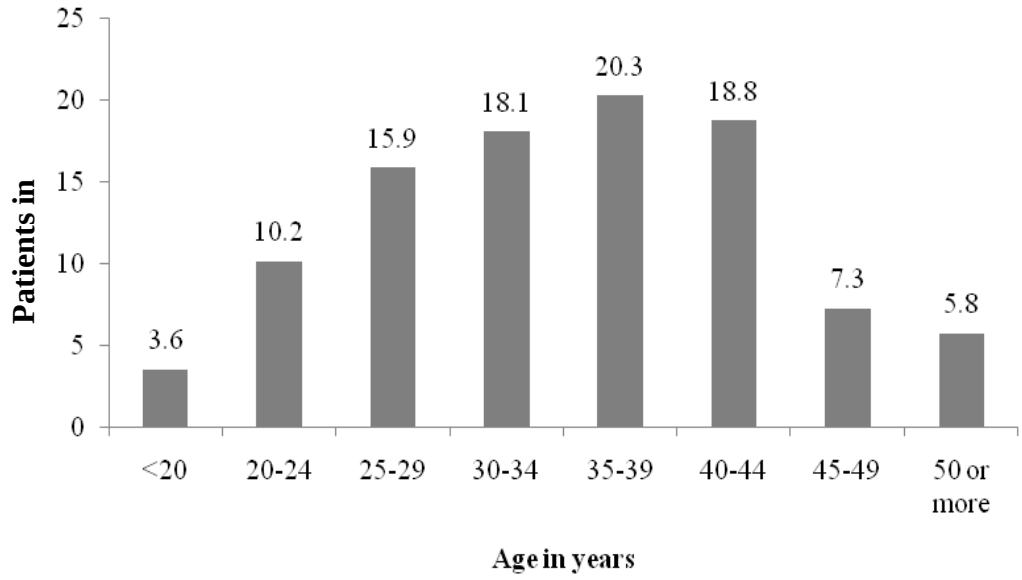

Figure 4.3: Age distribution among study participants

4.2 Mycobacterium tuberculosis confirmatory testing

Twenty (14.5%) of the sample (N) isolates subjected to Niacin test were all positive for *mycobacterium tuberculosis.*These isolates were randomly selected from the 138. This showed that those diagnosed with mycobacterium tuberculosis were highly likely to have *mycobacterium tuberculosis*

4.3 Antibiotic susceptibility testing of *Mycobacterium tuberculosis* isolates to treatment status

Antibiotic susceptibility testing was performed on 138 isolates. 112 (81.2%) isolates were sensitive to all drugs tested. The proportion of isolates that were sensitive to all drugs among retreatment and new cases was 66 (72.5%) and 46 (97.9%) respectively,

(OR=0.06 [95% CI=0.01 – 0.44]; P<0.001). Resistance to any of the drug tested among new patients was as follows; ethambutol, 1(2.1%), isoniazid, none, streptomycin, none and rifampicin, none. Among previously treated patients the resistance pattern was; isoniazid, 16 (17.6%), ethambutol, 10 (11.0%), rifampicin, 9 (9.9%), and streptomycin, 7 (7.7%). None and 6 (6.6%) isolates from new and previously treated patients were multidrug resistant defined as resistance to at least both isoniazid and rifampicin.The six isolates were not characterized further to show if they were new strains. Resistance to isoniazid and rifampicin was significantly associated with TB retreatment (P=0.001, and P=0.028 respectively).

Table 4.1: Drug susceptibility testing of first-line anti-tuberculosis drugs to treatment status

Antibiotic	Total (N=138)		RT (n=91)		New (n=47)		OR	95% CI		p value
	n	%	n	%	N	%		Lower	Upper	
Sensitivity to all	112	81.2	66	72.5	46	97.9	0.06	0.01	0.44	**<0.001**
Any resistance pattern										
Isoniazid (H)	16	11.6	16	17.6	0	0.0	UD	UD	UD	**0.001**
Rifampicin (R)	9	6.5	9	9.9	0	0.0	UD	UD	UD	**0.028**
Ethambutol (E)	11	8.0	10	11.0	1	2.1	5.68	0.70	45.79	0.098
Streptomycin (S)	7	5.1	7	7.7	0	0.0	UD	UD	UD	0.095
Total resistance to any drug tested	43	31.2	42	46.2	1	2.1				
Monoresistance TB										
Isoniazid (H)	6	4.3	6	6.6	0	0.0	UD	UD	UD	0.095
Rifampicin (R)	2	1.4	2	2.2	0	0.0	UD	UD	UD	0.548
Ethambutol (E)	4	2.9	3	3.3	1	2.1	1.57	0.16	15.50	1.000
Streptomycin (S)	3	2.2	3	3.3	0	0.0	UD	UD	UD	0.551
Multi drug resistance TB (MDR TB)										
Isoniazid+Rifampicin	2	1.4	2	2.2	0	0.0	UD	UD	UD	0.548
Isoniazid+Rifampicin+Ethambutol	2	1.4	2	2.2	0	0.0	UD	UD	UD	0.548
Isoniazid+Rifampicin+Streptomycin	1	0.7	1	1.1	0	0.0	UD	UD	UD	1.000
Isoniazid+Rifampicin+Ethambutol+Streptomycin	1	0.7	1	1.1	0	0.0	UD	UD	UD	1.000
Total MDR TB	6	4.3	6	6.6	0	0.0	UD	UD	UD	0.095
Other resistant Patterns										
Isoniazid+Ethambutol	2	1.4	2	2.2	0	0.0	UD	UD	UD	0.548
Isoniazid+Streptomycin	1	0.7	1	1.1	0	0.0	UD	UD	UD	1.000

Isoniazid+Ethambutol+Streptomycin	1	0.7	1	1.1	0	0.0	UD	UD	UD	1.000
Rifampicin+Ethambutol	1	0.7	1	1.1	0	0.0	UD	UD	UD	1.000

RT-Retreatment, OR-Odds Ratio, CI- Confidence Interval, P-Value- Level of Significance

4.4 Antibiotic susceptibility testing of *mycobacterium tuberculosis* isolates to CD4

Analysis of CD4 count among the patients revealed that median CD4 count was 286 cells/µl ranging between 2 and 859 cells/µl. A classification of CD4 was done using a cut-off of 200 cells /µl. A total of 51 (37.0%) patients had CD4 count (<200) cells/µl while 87 (63.0%) patients had CD4 count ≥200 cells/µl. Forty two (82.4%) of patients with CD4 count <200 cells/µl and 70 (80.5%) of patients with CD4 count ≥200 cells/µl were sensitive to all anti-tuberculosis drugs (Table 4.2).

Resistance to any of the drugs tested among patients with CD4 count of<200 cells/µl was as follows; isoniazid 6 (11.8%), rifampicin 5 (9.8 %), ethambutol 4 (7.8 %), streptomycin 3 (5.9%). Among patients with CD4 count ≥200 cells/µl the resistance pattern was isoniazid 10 (11.5%), ethambutol 7 (8.0 %), rifampicin 4 (4.6 %), and streptomycin 4 (4.6%). Three (5.9 %), and 3 (3.4%) isolates from patients with CD4 count <200 cells/µl, and those with CD4 count ≥200 cells/µl respectively, were multidrug resistant TB (MDR TB) defined as resistant to at least both isoniazid and rifampicin. There was no statistical significant difference between CD4 count and TB drug resistance patterns with P=0.670 (Table 4.2).

Table 4.2 Drug susceptibility testing of first-line anti-tuberculosis drugs to CD4 counts

Antibiotic	Total (N=138)		CD4<200 cells/µl (n=51)		CD4>=200 cells/µl (n=87)		OR	95% CI		p value
	N	%	n	%	n	%		Lower	Upper	
Sensitivity to all	112	81.2	42	82.4.	70	80.5	1.13	0.46	2.77	0.784
Any resistance										
Isoniazid (H)	16	11.6	6	11.8	10	11.5	1.03	0.35	3.01	0.962
Rifampicin (R)	9	6.5	5	9.8	4	4.6	2.26	0.58	8.82	0.290
Ethambutol (E)	11	8.0	4	7.8	7	8.0	0.97	0.27	3.50	1.000
Streptomycin(S)	7	5.1	3	5.9	4	4.6	1.30	0.28	6.04	0.709
Total resistance for any drug tested	43	31.2	18	35.3	25	28.7				
MonoresistanceTB										
Isoniazid (H)	6	4.3	1	2.0	5	5.7	0.33	0.04	2.89	0.413
Rifampicin (R)	2	1.4	2	3.9	0	0.0	UD	UD	UD	0.135
Ethambutol (E)	4	2.9	0	0.0	4	4.6	UD	UD	UD	0.296
Streptomycin(S)	3	2.2	1	2.0	2	2.3	0.85	0.08	9.61	1.000
Multi drug resistance TB(MDRTB)										
Isoniazid+Rifampicin	2	1.4	1	2.0	1	1.1	1.72	0.11	28.11	1.000
Isoniazid+Rifampicin+Ethambutol	2	1.4	1	2.0	1	1.1	1.72	0.11	28.11	1.000
Isoniazid+Rifampicin+Streptomycin	1	0.7	0	0.0	1	1.1	UD	UD	UD	1.000
	1	0.7	1	2.0	0	0.0	UD	UD	UD	0.370
Total MDR TB	6	4.3	3	5.9	3	3.4	1.75	0.34	9.01	**0.670**
Other resistant Patterns										
Isoniazid+Ethambutol	2	1.4	1	2.0	1	1.1	1.72	0.11	28.11	1.000
Isoniazid+Streptomycin	1	0.7	0	0.0	1	1.1	UD	UD	UD	1.000
Isoniazid+Ethambutol+Streptomycin	1	0.7	1	2.0	0	0.0	UD	UD	UD	0.370
Rifampicin+Ethambutol	1	0.7	0	0.0	1	1.1	UD	UD	UD	1.000

RT-Retreatment, CD4 Cluster of differentiation , OR-Odds Ratio, CI- Confidence Interval, P-Value- Level of Significance

CHAPTER FIVE

DISCUSSION, CONCLUSIONS & RECOMMENDATIONS

5.1 Resistance patterns of first line TB drugs

The overall resistance to all the drugs tested (18.8 %) was lower than that reported in earlier studies in Kenya, where 30.1 % of the isolates were resistant to at least one drug (Ndungu *et al.,* 2012). The results of this study differ with that conducted in Central Asia where resistance was 30.5 % (Suzanne *et al.,* 2011). Resistance rates in the present study were higher than rates observed in studies in Tanzania where only 14 out of 280 (5.83 %) isolates were resistant to at least one drug (Uplekar *et al.,* 2011), while in South Africa and Korea total resistance to the drugs tested was 7.4 % (Magana *et al.,* 2010) and 18.7 % (Lee &Chang, 2011) respectively. These differences would be attributed to the accelerated laboratory strengthening for the diagnosis of drug resistant TB and pursued high quality DOTS expansion and enhancement. This would lead to a decrease in the number of drug resistant cases as most of the patient's access diagnosis and treatment (Hom *et al.,* 2012).

Results of this study showed that resistance to any one drug tested was 2.1% and 46.2% among new and retreatment cases respectively, while MDR-TB prevalence was (none and 6.6%) among new and retreatment cases respectively (Table 4.1). This contrasts with studies conducted in Uganda, where resistance to any of the anti-TB drugs was 8.3% and 30.9%, among new and retreatment cases respectively; with an MDR-TB prevalence of 1.4 % and 12.1% among new and retreatment cases respectively (Deus *et al.,* 2012). In the Republic of Tanzania, there were 8.3% and of 20% resistance cases among new and retreatment cases respectively (Chonde *et al.,* 2010). While in Kenya there were 20% and 45% resistance cases among new and retreatment cases respectively (Ogaro *et al.,* 2012). Resistance to any one drug of 46.2%, in this study, 30.9%, 20%, 45% among retreatment cases in studies conducted in Uganda, Tanzania, and Kenya respectively raises concerns about the quality of directly observed therapy and adherence to treatment (Chonde *et al.,* 2010).

Resistance to any one drug of 2.1 % among new patients in this study implies no ongoing transmission of drug resistant strains in the community. This may suggest strengthened infection control measures which should therefore be further strengthened through dissemination of TB infection control guidelines by the National Leprosy and Tuberculosis Program (NLTP). All these were one-time studies performed in single facilities in the respective countries, similarly the present investigation was a one-time study performed in a single facility.

Globally in 2012, data from drug resistance surveys and continuous surveillance among TB cases suggested that 3.6% new diagnosed and 20% previously treated TB cases had MDR- TB and were found in Eastern Europe and Central Asia, while in some countries more than 20% of new TB cases and more than 50% previously treated TB cases had MDR-TB (Uplekar *et al.*, 2011). These previous studies compare with this study, where there is high percentage of retreatment cases being prone to drug resistance compared to new cases. The fact that most of these patients are previously treated for TB could be a possible risk factor for the development of resistance. A study carried out by Ogaro *et al.* (2012) in Kenya, showed that among previously treated patients 153 (76.9%) and new 312 (84.5%) patients were fully sensitive to all anti-tuberculosis drugs tested. Any resistance pattern among new patients was as follows; isoniazid 38 (10.3%), ethambutol 19 (5.1%), streptomycin 16 (4.3%), and rifampicin 3(0.81%) (Ogaro *et al.*, 2012). Among previously treated patients the resistance pattern was; isoniazid 34(18.1%), streptomycin 21(10.5%), ethambutol 14 (7.02%), and rifampicin 18 (9.04%). Two (0.54%) and 17 (8.54%) isolates from new and previously treated patients respectively were multi-drug resistant. This compares with this study; there is high percentage of retreatment cases being prone to multi- drug resistance compared to new cases.

Any resistance to isoniazid in this study was 11.6 % which was high compared to results obtained in earlier studies in Ethiopia, where one isolate was resistant to isoniazid (Kassu *et al.*,2011), with Bangladesh at 5.4 % (Deun *et al.*, 2012) and Sri-lanka at 12.2% (Magana *et al.*,2010). According to WHO, isoniazid resistance rates

higher than 10% can predict the development of multidrug resistant tuberculosis. This high resistance may be caused by both its wide use in the treatment of TB as a first-line drug or poor compliance by patients (Kassu *et al.,* 2011). In this study, any resistance to rifampicin was 6.5% which is higher than that observed in earlier studies in Tanzania, where resistance was 1.3% (Chonde *et al.,* 2010). This rate is also higher than reports from studies in Bangladesh where resistance was 0.5% (Magana *et al.,* 2010) and Ethiopia where resistance to rifampicin ranged from none to 1.8% (Uplekar *et al.,* 2011; Lee & Chang 2011). These differences may be attributed to the accelerated laboratory strengthening for the diagnosis of drug resistant TB and pursued high quality DOTS expansion and enhancement. This may lead to a decrease in the number of drug resistant cases as most of the patient's access diagnosis and treatment (Gandhi *et al.,* 2010). Rifampicin has several adverse effects such as nausea, vomiting, rashes, hepatitis, GIT upset, fever, and jaundice, which could result in non adherence and hence may lead to the selection of resistant strains.

Any resistance to streptomycin in this study was 5.1 % which was comparable to the resistance of 5.2 % (Ndungu *et al.,* 2012) reported in another study in Kenya but lower than that reported in Ethiopia 26 % (Kassu *et al.,* 2008) and Sri-Lanka 9.9 % (Nunes *et al.,* 2011). Any resistance to ethambutol in this study was 8.0 % which was higher than rates in Ethiopia 2.7 % (Kassu *et al.,* 2008). It was however; lower than that reported in Sri-lanka where 14.5% resistance was reported (Nunes *et al.,* 2011). Ethambutol enhances the effect of many drugs including beta lactams to different *Mycobacterium* species and can be used to develop a regimen for MDR-TB (Abate & Miorner, 2011).

In this study, a high number of patients with TB showed isoniazid resistance but significant susceptibility to rifampicin. It is therefore possible for these patients to recover fully if WHO guidelines for re-treatment are followed under strict supervision to prevent them from developing MDR-TB. However, the high rate of isoniazid resistance is significant since it is a first line drug which is used throughout

the course of treatment. This indicates a high probability for developing MDR-TB in the future since it has been observed that MDR often develops from initial isoniazid mono-resistant strains. Isoniazid is also the drug of choice for chemoprophylaxis of TB and is used in developed countries for treating latent TB (Hom *et al.*, 2012). The high level of isoniazid resistance among the study population is an indicator that this drug will not be useful for chemoprophylaxis in future. Sensitivity to all drugs tested was 46 (97.9 %) among new cases in the present study (Table 4.1). These findings imply no ongoing transmission of drug resistant strains in the community.

5.2 CD4 count and TB drug resistance

Worldwide incidence of TB is increasing, particularly in areas where HIV is prevalent (WHO/GTP 2012). The effect of CD4 count on TB drug resistance is varied in various studies and it is often difficult to compare data because of relatively small patient numbers in previous studies and few documented data (Mcshane, 2010). In this study, three (5.9%), and 3 (3.4%) isolates from patients with CD4 count <200 cells/µl, and those with CD4 count ≥200 cells/µl respectively, had multidrug resistant TB (MDR TB) defined as resistant to at least both isoniazid and rifampicin. The median CD4 count was 286 cells/µl, ranging between 2 and 859 cells /µl. This contrasts with a study carried out by Shah *et al.*, (2010) in South Africa's Tugela Ferry from 2009 to 2010 which found that of the 272 MDR-TB and 382 XDR-TB cases, 90% and 98% were co-infected with HIV with median CD4 counts of 41 cells/µl and 36 cells/µl (Gandhi *et al.*, 2010).

This study also contrasts with another, carried out by Gandhi *et al.*(2010) in Kwazulu Natal, South Africa, of the 1,539 patients tested, 542 (35 %) had culture-positive TB, with MDR-TB in 221 (41 %) of those with culture-positive TB. Of the MDR-TB cases, 53 (24 %) had XDR-TB, of which all of the 44 patients who were tested for HIV were infected with HIV. They had a median CD4 count of 63 cells/µl. In another study conducted by Hom *et al.* (2012) in Durban South Africa, of the 1035 patients who had complete culture results, the estimated prevalence of resistance to

any of the antituberculous drugs was 7.4% (95% C.I 4-0-12.4) with a median CD4 count of 92 cells/µl (IQR 42-150cells/µl). These differences would be attributed to the extent of H.I.V disease as was observed by Hom *et al.*, (2012). Based on this study, the CD4 count does not predict the occurrence of drug resistant TB, because there were no significant associations between the various resistant patterns and levels of CD4. Some studies show that CD4 count does not have significant effects on MDR-TB development based on there being no difference found in sputum cultures of HIV positive and negative individuals with MDR TB (Kamerbeek & Kolk 2013). These studies propose that MDR-TB may be impacted by previous antibiotic treatment, with individuals who have had previous T.B treatment being five times more likely to develop T.B drug resistance as was also observed by Sharma and Mohan (2010).

5.3 Conclusions

- There is a decline in TB drug resistance both for new and retreatment cases (None and 6.6% respectively).
- CD4 count does not predict the occurrence of drug resistant tuberculosis among the HIV patients with CD4 count of > or < 200 cells/ µl/ (P-value 0.670).
- Resistance to any one of the anti TB drugs was high with isoniazid (11.6%).

5.4 Recommendations

- All TB patients new or re-treatment should have culture and drug susceptibility tests, before commencement of TB treatment.
- All patients with resistance to either of the TB drugs should be commenced on the recommended treatment regimen to prevent further spread of the drug

resistant strains.

- There is need to implement molecular techniques in diagnosis of T.B in H.I.V individuals, for example the genexpert. This will ensure faster detection of drug resistant T.B especially in HIV individuals.
- There is need for characterization of the drug resistant isolates to determine whether they are new strains emerging due to drug interactions
- Nation wide tuberculosis drug resistance surveillance is needed to monitor drug resistance in the country
- Strengthening drug susceptibility testing capacity is a high priority for national TB programmes, especially in Kenya.

REFERENCES

Abate, G. & Miorner, H. (2011). Susceptibility of Multidrug resistant strains of *Mycobacterium tuberculosis* to amoxicillin in combination with clavullanic acid and ethambutol. *Journal on Antimicrobial Chemotherapy 42*(6),735-740.

Blomberg, B., Spinaci, S., Fouri, B. & Laing, R. (2011).The rationale of recommending fixed dose combination tablets for treatment of tuberculosis. *Bulletin of World Health Organisation.79,*61-68

Boehme, C.C., Nicol, M.P., Nabeta, P., Michael, J.S. & Gotuzzo, E. (2011). Feasibility diagnostic accuracy and effectiveness of decentralized use of the Xpert MTB/RIF test for diagnosis of tuberculosis and multi drug resistance: a multicentre implementation study. *Lancet 377,*1495-1505

Bonnet, M., Ramsay, A., Githui, W., Gaqnidze, L., Varaine, F., & Guerin, P. (2010). An opportunity to optimize smear microscopy for Tuberculosis diagnosis in settings of high prevalence of HIV. *Clinical infectious Diseases 15,* 98-120

Chan, E.D., Chan, J.F., Globe, M. & Iseman, M.D. (2011). Epidemiology and control of tuberculosis in Western European cities. *International Journal of Tuberculosis and Lung Diseases 7,*751-757

Chonde, T.M., Basra, D., Mfinanga, S.G., Range, N. & Lwilla, F. (2010). National anti-tuberculosis drug resistance study in Tanzania. *International Journal Tuberculosis and Lung Diseases 14(*8), 967-972

Cole, S.T. (2010). *Mycobacterium tuberculosis*: drug resistance mechanisms. *Trends in microbiology, 2,*411-415

Corbett, E.L., Watt, C.J., Walker, W., Maher, D. & William, B.G. (2010). The growing burden of tuberculosis in HIV infected patients: global trends and

interactions with the HIV epidemic. *International Journal of Tuberculosis and Lung Diseases 163*, 1009-1021

Deun, V. A., Aung K.J., Chowdhury, S., Saba, S., Paukay, A., Ashraf, A., Rigout, L., Fissette, K. & Portael, F. (2012). Drug susceptibility of *Mycobacterium tuberculosis* in rural area of Bangladesh and its relevance to the National treatment regimens. *International Journal on Tuberculosis and Lung Disease 3*,143-148

Deus, L., Francis, A., Kenneth, M., George, W. K., Willy, W., Rosemary O., Julius, N.K., Ann, A., Anand, D. & Moses, L. J.(2012). Anti-tuberculosis drug resistance among new and previously treated sputum smear positive tuberculosis patients in Uganda. *Tropical Medicine and International Health 19(7)*, 1016-1020

Division of Leprosy, Tuberculosis and Lung Disease (2012). Guidelines for the management of multidrug resistant TB in Kenya, Division of Leprosy, Tuberculosis and Lung Disease, Nairobi.Government Press: Kenya

Drobniewski, F.A., Nikolayevskyy, V., & Maxeiner, H. (2011). Tuberculosis, HIV sero-prevalence and intravenous drug abuse in prisoners. *The England Journal of Medicine 26*(2), 294-304

Dye, C., Sheele, S., Dolin, P. & Pathania, V. (2011) Global burden of TB. Estimated incidence, prevalence and mortality by country. *British Medical Journal 282*,677-686

Espinal, M. A., Laszlo, A., Simonsen, L., Boulahbal, F., Kim, S. J. & Reneiro, A. (2012). Global trends in resistance to anti-tuberculosis drugs. World Health Organization- International Union Against Tuberculosis and Lung Disease. *British Medical Journal 344,*1294-303

Fisher, M., Kasiulevicius, V., Sapoka, V., & Filipaviciute, R. (1998). Statistical methods for rates and proportions. 2nd edition. Oxford: Oxford University Press

Ferebee S.H., Comstock, G.W. & Hammes, L.M. (2012). A controlled trial of community wide Isoniazid prophylaxis in Alaska. *Review on Respiratory Diseases 95*,935-943

Getahun, H., Havlir, D. &Granich, R. (2012). Paradigm shift to address drug resistant tuberculosis in people living with HIV needed and needed now. *Tropical Medicine and International Health 14* (4), 376-378

Goyal, M., Saunders, N.A., & Embden, J.D.A. (2012). Differentiation of *mycobacterium tuberculosis* isolates by spoligotyping and IS6110 restriction fragment length polymorphism. *Journal on clinical microbiology 35*,647-651

Gillespie, S.H. (2011). "Evolution of drug resistance in *Mycobacterium tuberculosis*, clinical and molecular perspective" *Antimicrobial Agents Chemotherapy 46*, (2) 267-274

Gandhi, N.R., Nunn, P., Dheda, K., Schaaf, H.S., Zignol, M., Sooligen, Van. D. (2010). Multi drug resistant and Extensively drug resistant tuberculosis: a threat to global control of tuberculosis. Lancet.375: 1830-43 10.1016/S0140-6736(10) 60410-2[PubMed] [Cross Ref]

Geneva, WHO. (Global tuberculosis control report. Retrieved from http://www.who.int/tbpublication/global report/2012/en/ind.html

Hernandex, G., Cook, E., Kunimoto, D., Black, W.A. & Elwood, R.K. (2011). Transmission of TB from smear negative patients: a molecular Epidemiology study. *Thorax 59*, 286-290

Hom, K.J., Bingxia, W., Senica, C., Giddy, J., G., Mazibuko, M., Rochelle, P., Walensky, E., Losina, A., Freedberg, I. & Bassett, V. (2012). Drug resistant tuberculosis among HIV infected patients starting antiretroviral therapy in Durban South Africa .*PLos ONE/www.plosone.org volume 7/issue8/e43281*

Keter, K.L., Cherogony, K.S., Korir, K.R., Mutai, C. (2014). Surveillance of drug resistant tuberculosis in refugee and non refugee populations in North Eastern Kenya. *International Journal on Tuberculosis and Lung Disease 3,* 45-52

Kamerbeek, J.K., & Kolk, M. (2013). Simultaneous detection and strain differentiation of mycobacterium tuberculosis for diagnosis and epidemiology. *Journal on clinical microbiology 38,* 907-914

Kenya Bureau of Statistics (2012). *Kenya demographic and Health Survey :Basic report* .3, 8-9) , Nairobi*:* Central bureau of statistics

Kassu, D., Daniel, A., Eshatu, L., Mekdes, G.M. & Benium, F. (2011). Drug susceptibility of *Mycobacterium tuberculosis* isolates from smear negative pulmonary tuberculosis patients, Addis Ababa, Ethiopia. *Ethiopian Journal on Health Development 22*(2), 212-5

Kawai, V., Soto, G., Gilman, R. H. & Evans, C.A. (2010). "Tuberculosis, mortality, drug resistance, and infectiousness in patients with HIV infection in Kenya" *Journal on Tropical Medicine Hygiene 75* (6), 1027-1033

Kent, P.T. & Kubica, G.P. (2010). Public health Mycobacteriology. A guide for level III laboratory centers of Disease control, Atlanta. *Publication (86), 345-456*

Lawn, S.D. & Nicol, M.P. (2011). Xpert-MTB/RIF assay; development, evaluation, and implementation of a new rapid molecular diagnostic for tuberculosis and rifampicin resistance. *Future of Microbiology 6*(9),1067-82.*doi:10.2217/fmb.11.84*

Lee, J.H. & Chang, J.H. (2011). Drug resistant tuberculosis in tertiary referral teaching hospital in Korea. *Korean Journal on International Medicine 16,* 173-179

Lonroth, K., Juramillo, E., Williams, B., Dye, C. & Raviglione, M. (2011). Drivers of tuberculosis epidemics. The role of risk factors and social determinants.*Journal on Social sciences and Medicine.68* (12), 2240-2241

Magana, A.D.N., Pirera, A.J., Senarathe, V. &Chandrasekharan, N.V. (2010). Patterns of drug resistance and RFLP analysis of *Mycobacterium tuberculosis* strains isolated from recurrent tuberculosis patients in Sri- Lanka. South- East - Asian. *Journal on Tropical Medicine and Public Health 41*,583-589.

Mcshane, H. (2010). Co-infection with HIV and TB: Double trouble. *International British Medical Journal 16*(2), 95-100

National Aids and sexually transmitted infections control Program (2012). Annual report. Retieved from: http://www.nascop.co.ke/docs/annual report 2012.

Ndungu, P.W., Kariuki, S., Ng'ang'a, Z., & Revathi, G., (2012). Resistance patterns of *Mycobacterium tuberculosis* isolates from pulmonary tuberculosis patients in Nairobi.*Journal on Tuberculosis and Lung disease Lancet 6(01),33-9.*

Nunes, E.A., Decapitani, E.M., Coelho, E., Panunto, A.C., Joaquim, O.A. & Ramos, M.C. (2011). *Mycobacterium tuberculosis* and non tuberculosis *Mycobacterium* isolates among patients with recent HIV infection in Mozambique. *Journal on Tuberculosis and Lung disease 4*,571-575

Ogaro, T.D., Githui, W., Kikuvi, G., Okari, J., Wangui, E. & Asiko, V. (2012). Anti-tuberculosis drug resistance in Nairobi, Kenya. *African Medical Journal of Health Sciences 2,1-2*

Pozniak, A.L., Miller, R.F., Lipman, M.C.I., Freedman, A.R. & Ormerod, L.P.

(2011). Treatment guidelines for tuberculosis and HIV infection. *Lancet 6*,62-83

Raviglione, M.D., Snider, D.E. & Kochi, A. (2011). Global epidemiology of TB: Morbidity and Mortality of Worldwide epidemic. *Lancet 273, 220-226*

Standford, J. L., Grange, J.M. & Pozniak, A. (2012). The promise of Immunotherapy for tuberculosis. *Respiratory Medicine 88* (1), 3-7.

Suzanne, H., Orozco, J. D., Male, R., Gerdes, S., Falzon, D., Darebay, D., Yared, K. & Mohammed, A. (2011). Multidrug resistance in Central Asia. *Emergency Infectious Diseases 10*, 1210-1215

Saadoun, D., Calatroni, I., Launay, D., Memain, N. Marchal, G., Dupont, B., & Bouchaud, D. (2011). Tuberculosis in HIV infected patients: a comprehensive review. *Clinical Microbiology Infections.10*,388-198.

Squire, S.B., Harries A.D. & Whitty C.J. (2011). Smear negative pulmonary tuberculosis in a DOTS program: Poor outcome in an area of high HIV sero-prevalence. *International Journal of Tuberculosis and Lung Diseases 3*,521-543

Suzuki, K., Tsuyuguchi, K., Matsumoto, H., Tamaru, A., Makino, M., Mizuguchi, Y. & Taniguchi, H. (2010). Evaluation of Mycobacteria growth indicator tube for drug susceptibility testing of *Mycobacterium tuberculosis* isolates (4^{th}ed).Oxford: Oxford University press.

Shah, N., Wright, A., Bai, G., Barrera, L., Boulahbal, F. & Martan, N. (2010). Worldwide emergence of extensively drug resistant tuberculosis. *Emergency of Infectious Diseases 13*,380-387

Sharma S.K. & Mohan A. (2010). "Multidrug resistant tuberculosis" *Indian Medical Journal 120,* (4) 354-376

Taha, N., Hamed, A., Qurechi, J., Ahmad, B. & Abraham, S. (2013). Rifampicin resistance profile of *mycobacterium tuberculosis* isolated from HIV patients. *British Medical Journal 326,*1514-1521.

Uplekar, M.W., Rangan, S., Weiss, M.G., Ogden, J. & Burgdorff, M.W. (2011). Attention to gender issues in tuberculosis control. *International Journal on Tuberculosis and Lung Disease 5,*220-224.

WHO/GTP (2013). Multi drug and extensively drug resistant TB: Global report on surveillance and response. Report no. WHO/GTP/ TB/2013.3.Geneva: The Organizations; 2013 Facilities, congregate settings and households, WHO: Geneva

World Health Organization (2013). Global tuberculosis control: Epidemiology, strategy, financing and planning. WHO: Geneva

World Health Organization, International union Against Tuberculosis and Lung disease (2013). Interim recommendations for the surveillance of drug resistance in tuberculosis.WHO: Geneva

WHO/GTP (2012) Global Tuberculosis Control program. WHO/GTP/TB/2012.2.Geneva.

World Health Organization, International union Against Tuberculosis and Lung disease (2012). Guidelines for surveillance of drug resistance in tuberculosis, World Health Organization Document WHO/TB/96.216, 1-35

World Health Organization (2012a). Global tuberculosis control- epidemiology, strategy, financing: WHO report 2012a, World Health Organization, Geneva, Switzerland.WHO: Geneva

World Health Organization (2012b). WHO policy on tuberculosis infection control in health care.WHO: Geneva

World Health Organization (2011a). Anti-tuberculosis drug resistance in the world 4th global report.WHO: Geneva

World Health Organization (2011b). Guidelines for the programmatic management of drug resistant tuberculosis: Emergency update 2008, WHO: Geneva Retrieved from http://www.who.int/tb/publications/global-report/2011b

Yoon, C., Cattamanchi, A., Davis, J.L., Worodria, W. & Den, B. S. (2012). Impact of Xpert MTB/RIF Testing on Tuberculosis Management and outcome in Hospitalized Patients in Uganda. *PLos ONE 7(11)*.

Zwang, S.S., Kim, H.R., Kim, H.J., Kim, M.J., Lee, S.M., Yoo, C.G. & Kim, Y.W. (2012). Impact of resistance to first line and injectable drugs on treatment out comes in MDR-TB. *European Respiratory Journal 33,581-585*

APPENDIXES

Appendix I: Patient Written Consent form

Informed Consent For The Participants (Filled by participant /Guardian incase of illiterate study subject)

Title of Research: First Line Tb Drug Resistance among HIV Infected Patients Attending Maryland Comprehensive Care Centre, Mathare 4a, Nairobi, Kenya

Investigators: Lucy Nyanga'u, Jomo Kenyatta University of Agriculture and Technology, Dr. Amukoye: KEMRI-CRDR, Prof. Zipporah: JKUAT

Purpose of the Study

The aim of this study is to help understand the risk that drug resistant TB poses to the health of people infected with HIV, attending Maryland health centre. This study will not follow up participants; participants will give two sputum samples. This study will take six months to complete.

Procedures

The study will involve obtaining sputum samples, from the participants. The participants will be given well labelled sputum containers and explained by the principal investigator, how and where at home to void sputum early in the morning. The sputum will then be brought by the participant to Maryland TB Lab. Spot samples will be taken in the facility, for those participants who reach early in the morning (8.00-8.30a.m) and would have not taken breakfast. The sputum will be voided in a secluded place (sputum booth). Explanation on how to void sputum will be done by the principal investigator. Two samples (spot and morning) are expected from each participant. One will be processed for geneXpert in Maryland lab; the other will be transported to the National central reference TB lab by the smith line courier services for culture and DST. The study will also involve accessing the

participant's lab reports on the number of cells which help fight diseases in the body. The study will focus on first line anti- TB drug resistance among HIV patients in Maryland facility.

Benefits

Free laboratory expenses incurred during specimen processing. Free treatment for those found with drug resistant TB, in relevant public health facilities. And Maryland health centre for those with drug susceptible TB.

Risks and Discomfort

There is no risk involved in obtaining sputum sample. Treatment will not be delayed upon diagnosis.

Withdrawing Participation and Participants Data

If you feel you cannot carry on with the study you are free to stop participation. You have a choice to do so and we will still appreciate your willingness to participate in the study. If the participant withdrawals consent, the data is not usable and cannot be used for data analysis. Withdrawal on death or missing participant, the data can be considered for analysis. But appropriate statistical methods will be used to adjust for analysis of the data.

Confidentiality

The information we get from you is purposely for research and will not be revealed to anybody. Your information will be confidential. This will be achieved by use of serial numbers to code participants Names. Names will not be used as identities.

Contact Information

If you have any questions regarding this study, please contact Lucy Obonyo cell:

0722816265, P.O BOX 4899-00200 Nairobi.

Questions about your participation rights should be directed to the secretary KEMRI National Ethical Review Committee, P.O BOX 54840, 00200, Nairobi, Telephone Numbers: 020-2722541, 0722205901, 0733400003, Email address:erc@kemri.org

Declaration

Having read and understood the purpose of the study, I willingly accept to take

Part in it

Signature…… Date……

Note: By putting your signature, you are agreeing that: You have read this consent form and have been given the opportunity to ask questions.

You have known the risks and they have been explained to your satisfaction

You understand Jomo Kenyatta University and KEMRI has no policy or plan to pay for any injuries you might receive as a result of participation in this research

Your participation in this research is given at your free will

Witness

Signature…….. …..
Date…….

The participant received a copy ☐

Appendix II: Patient Written Parental Consent form

Informed Consent for the Participants (Filled by Guardian in case of Minors)

Title of Research: First Line Tb Drug Resistance among HIV Infected Patients Attending Maryland Comprehensive Care Centre, Mathare 4a, Nairobi, Kenya

Investigators: Lucy Nyanga'u, Jomo Kenyatta University of Agriculture and Technology, Dr. Amukoye: KEMRI-CRDR, Prof.Zipporah: JKUAT

Purpose of the Study

The study aims to help understand the risk that drug resistant TB poses to the health of people infected with HIV, attending Maryland health centre. This study will not follow up participants; participants will give two sputum samples. This study will take six months to complete.

Procedures

The study will involve obtaining sputum samples, from the participants. The participants will be given well labelled sputum containers and explained by the principal investigator, how and where at home to void sputum early in the morning. The sputum will then be brought by the participant/guardian to the Maryland Lab. Spot samples will be taken for those participants who reach early in the morning (8.00-8.30a.m) in the facility and would have not taken breakfast. The sputum will be voided in a secluded place in the facility (sputum booth). Explanation on how to void sputum will be done by the principal investigator. Two samples (spot and morning) are expected from each participant. One will be processed for geneXpert in Maryland lab. The other will be transported to the National central reference TB lab, by the smith line courier services, where culture and DST will be done. The study will also involve accessing the participant's lab reports on the number of cells which help fight diseases in the body (CD4). The study will focus on first line anti- TB drug

resistance among HIV patients in Maryland facility.

Benefits

There will be no payment for any lab procedures done. Free treatment will be given for those found with drug resistant TB in relevant public health facilities, and in Maryland health centre for those with drug susceptible TB.

Risks and Discomfort

There is no risk/ danger involved in obtaining sputum sample. Treatment will not be delayed upon diagnosis.

Withdrawing Participation and Participants Data

If you feel the participant cannot carry on with the study you are free to stop his or her participation. You have a choice to do so and we will still appreciate the willingness to participate in the study. Your decision will involve no penalty or loss of benefits normally available for you or your child. If the participant withdrawals consent, the data is not usable and cannot be used for data analysis. Withdrawal on death or missing participant, the data can be considered for analysis. But appropriate statistical methods will be used to adjust for analysis of the data.

Confidentiality

The information we get from you is purposely for research and will not be revealed to anybody. Your information will be confidential .This will be achieved by use of serial numbers to code participants Names. Names will not be used as identities.

The Contact Information

If you have any questions regarding this study, please contact Lucy Obonyo cell: 0722816265, P.O BOX 4899-00200 Nairobi.

Questions about his/her participation rights should be directed to the secretary KEMRI National Ethical Review Committee, P.O BOX 54840, 00200, Nairobi, Telephone Numbers: 020-2722541, 0722205901, 0733400003, Email address:erc@kemri.org

Declaration

Having read and understood the purpose of the study, I willingly accept my Child to take Part in it

Printed Name of Child..... Date.....

Printed Name and Signature of Parent..... Date.....

Signature of Principal Investigator …..... Date.....

Note: By putting your signature, you are agreeing that: You have read this consent form and have been given the opportunity to ask questions.

You have known the risks and they have been explained to your satisfaction

You understand Jomo Kenyatta University and KEMRI has no policy or plan to pay for any injuries you might receive as a result of participation in this research

Participation of your child in this research is given at your free will

Witness

Signature…….. …..

Date…….

The participant received copy ☐

Appendix III: Adolescent Participant Assent Form

You are asked to help us in the project described below. Your parent or guardian has given his okay, but you get to decide if you want to be in this study or not. You may stop or quit the study at any time by telling us and it is okay. If you want to know more about the study, it is okay to ask questions.

Title of Research: First Line Tb Drug Resistance among HIV Infected Patients Attending Maryland Comprehensive Care Centre, Mathare 4a, Nairobi, Kenya

Investigators: Lucy Nyanga'u, Jomo Kenyatta University of Agriculture and Technology, Dr. Amukoye: KEMRI-CRDR, Prof. Zipporah: JKUAT

Assent Form

Purpose of Study

This study is to help understand the dangers drug resistant TB brings to people living with HIV and attending Maryland health centre. The people who will agree to participate will give two sputum samples. This study will take six months to complete.

Procedures

This study will involve obtaining sputum samples from you. You will be given well labelled sputum containers by the principal investigator and explained how and where at home to produce the sputum early in the morning. You/guardian will then bring the sputum to the facility. A spot sample will be obtained from you, if you reach early in the morning (8.00-8.30am), this will be done in a sputum booth away

from other people within the facility. Two samples spot and morning are expected from you. One will be processed here for identification of TB (Maryland lab) and the other will be transported to the National central reference TB lab by smith-line courier services. In the reference lab the TB micro-organism will be grown in certain specific foods and then checked for which drugs can/ cannot kill it. The study will also access reports on the number of soldiers in your blood which help fight disease in your body.

Risks/Discomfort

There is no danger involved in obtaining your sputum sample. Treatment will not be delayed upon diagnosis.

We will do everything to make sure you do not get hurt in any way. We will be the only people who know what you say and do.

If you understand what you are being asked to do and you decide to help. You are asked to sign your name below.

Printed Name and Signature of
Adolescent.....Date.....

Researcher's
Signature.....Date.....

The participant received copy ☐

Appendix IV: Partcipant Questionnaire Form

Questionnaire for the Participants: (Filled by investigators for those who have already consented)

You are asked to help us in the project already described to you. You will be asked some questions as pertains to your health; you are not obliged to answer if you wish not to. It is okay to ask questions as well, where need be.

Title of Research: First Line Tb Drug Resistance among HIV Infected Patients Attending Maryland Comprehensive Care Centre, Mathare 4a, Nairobi, Kenya

Investigators: Lucy Nyanga'u, Jomo Kenyatta University of Agriculture and Technology, Dr. Amukoye: KEMRI-CRDR, Prof. Zipporah: JKUAT

1. How old are you? ☐

2. Do you cough? Yes ☐ No ☐

If yes how long have you coughed

More than two weeks ☐ Less than two weeks ☐

3. When is the cough worse?

Day ☐ Night ☐ All the time ☐

4. Does the cough produce sputum?

Yes ☐ No ☐

If yes how is the sputum?

Mucopurulent ☐ Salivary ☐ Bloody ☐

5. Do you have night sweats?

Yes ☐ No ☐

6. Do you have a history of contact with someone who had TB?

Yes ☐ No ☐

7. Have you been treated for TB before?

Yes ☐ No ☐

8. Do you have chest pain?

Yes ☐ No ☐

9. Gender of participant? M ☐ F ☐

Declaration

Having actively participated in the filling of this questionnaire and confirmed that the same is true on paper, i willingly sign / guardians sign in place of minors and illiterate participants.

Printed Name of participant/ Child..... Date.....

Printed Name and Signature of Guardian..... Date.....

Signature of Principal Investigator …..... ……..Date.....

Note: By putting your signature, you are agreeing that: the questionnaire as been

filled with you actively participating and have been given the opportunity to ask questions.

Witness

Signature…….. …..

Date…….

The participant received copy ☐

APPENDIX V: Ethical Review Committee Approval Form

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 **April 19, 2013**

TO: **LUCY NYANG'AU,**
PRINCIPAL INVESTIGATOR

THRO': **DR. EVANS AMUKOYE**
THE DIRECTOR, CRDR,
NAIROBI

Dear Madam,

RE: **SSC PROTOCOL NO.2475 (INITIAL SUBMISSION): FIRST LINE ANTI-TB DRUG RESSISTANT AMONG HIV INFECTED PATIENTS ATTENDING MARYLAND COMPREHENSIVE CARE CENTRE, MATHARE 4A, NAIROBI, KENYA.**

This is to inform you that during the 214th meeting of the KEMRI/ERC held on 23rd April 2013, the above referenced study was reviewed.

The Committee notes that the above referenced study aims to determine first line anti-TB Drug resistance among HIV infected patients attending Maryland comprehensive care Centre, in Mathare 4A, Nairobi.

The committee determines that the issues that were raised during the 212th meeting of the KEMRI/ERC held on 26TH February 2013 have been adequately addresses. The study is therefore granted approval for implementation effective this **23rd April 2013** for a period of one year. Please note that authorization to conduct this study will automatically expire on **April 22, 2014**.

If you plan to continue data collection or analysis beyond this date, please submit an application for continuation approval to the ERC Secretariat by **March 11, 2014**. The regulations require continuing review even though the research activity may not have begun until sometime after the ERC approval.

You are required to submit any proposed changes to this study to the SSC and ERC for review and the changes should not be initiated until written approval from the ERC is received. Please note that any unanticipated problems resulting from the implementation of this study should be brought to the attention of the ERC and you should advise the ERC when the study is completed or discontinued.

APPENDIX VI: Scientific Steering Committee Approval Form

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

ESACIPAC/SSC/101299 8th January, 2013

Lucy Nyang'au

Thro'
Director, CRDR
NAIROBI

REF: SSC No. 2475 (Revised) – First Line Anti-TB drug resistance among HIV infected patients attending Maryland Comprehensive Care Centre, Mathare 4A.

I am pleased to inform you that the above mentioned proposal, in which you are the PI, was discussed by the KEMRI Scientific Steering Committee (SSC), during its 197th meeting held on 4th, December, 2012 and has since been approved for implementation by the SSC.

Kindly submit 4 copies of the revised protocol to SSC within 2 weeks from the date of this letter i.e., 22nd January, 2013.

We advise that work on this project can only start when ERC approval is received.

Sammy Njenga, PhD
SECRETARY, SSC

Kiambatisho cha VII: Fomu ya Kibali/Idhini ya Mshiriki

Ufahamu wa Idhini hii kwa Washiriki: Ijazwe na mshiriki mwenyewe /mdhamini iwapo anayehusishwa bado hafahamu kusoma na kuandika.

Mada ya Utafiti: Viwango vya juu vya kifua kikuu kisichosikia dawa miongoni mwa walioambukizwa Virusi vya Ukimwi wanaopokea huduma katika kituo cha afya cha Maryland, Mathare 4A. Nairobi nchini Kenya.

Watafiti: Lucy Nyanga'u: Chuo Kikuu cha Kilimo Na Teknolojia Jomo Kenyatta, Dkt. Amukoye: KEMRI-CRDR, Prof. Zipporah: JKUAT.

Lengo la Utafiti huu

Lengo kuu la utafiti huu ni kusaidia katika kuelewa hatari inayoletwa na kifua kikuu kisichosikia dawa kwa afya ya watu walioambukizwa Virusi vya Ukimwi, wanaopokea huduma katika kituo cha Afya cha Maryland, kilicho eneo la Mathare 4a.Utafiti huu hautawafuatilia wanaoshiriki; wanaoshiriki watatoa kikohozi mara mbili. Utafiti huu utachukua miezi sita kukamilika

Utaratibu

Utafiti huu utahusika na kupata sampuli mbili tu za kikohozi cha anayeshiriki. Washiriki watapewa vichupa vya kuwekea kikohozi vilivyoandikwa vizuri na kuelezewa na mtafiti mkuu, jinsi na ni wapi wanafaa kutolea kikohozi chao nyumbani kwao, asubuhi na mapema.

Kikohozi hicho kitaletwa na mhusika hadi katika kituo cha afya cha Maryland katika maabara ya TB.

Kikohozi cha kwanza kitatolewa hospitalini punde tu mshiriki afikapo katika kituo hicho, kwa wale washiriki watakaowasili hospitalini asubuhi mapema baina ya saa mbili na saa mbili unusu na hawatakuwa wamepata chemsha kinywa au chai ya

asubuhi.

Kikohozi chao kitatolewa katika mahali maalum palipotengwa (chumba cha kikohozi) mbali na watu walioko hospitalini. Maelezo jinsi ya kutoa kikohozi hicho yatatolewa na mtafiti mkuu. Kwa hivyo sampuli mbili za kikohozi zitatolewa kutoka kwa kila mhusika (cha kwanza ufikapo hospitalini, na ya pili asubuhi mapema nyumbani)

Kikohozi kimoja kitapimwa kwa sababu ya kutambua uwezo wa uwepo wa kifua kikuu katika maabara ya Maryland; na kingine kitasafirishwa hadi Maaabara kuu ya Kitaifa ya kurejelea masuala ya Kifua Kikuu kupitia kwa huduma ya uchukuzi ya Smith-line ili kichunguzwe na kipimwe uwezo wa kuhimili dawa. Pia utafiti huu utahusika na kutathimini ripoti za maabara za mshiriki huyo kuhusu idadi ya chembechembe zinazosaidia katika kupigana na magonjwa mwilini. Utafiti utamakinika kwanza na viwango vya juu vya Kifua kikuu kisichosikia dawa miongoni mwa walioambukizwa Virusi vya Ukimwi katika hospitali ya Maryland.

Faida za Utafiti huu

Gharama ya kupima kikohozi na mengineyo maabarani haitalipiwa. Pia watakaopatikana na hali ya kuwa na Kifua Kikuu kisichosikia dawa watapata matibabu kutoka kwa vituo vya afya vya umma na waliona kifua kikuu kinachotibika kutoka kituo cha afya cha Maryland. Matibabu yatakuwa bila malipo.

Hatari na Usumbufu

Hakuna hatari kwa kutoa kikohozi. Matibabu hayawezi kucheleweshwa hata kidogo wakati wa kusubiri uchunguzi ufanywe maabarani.

Kujiondoa katika kushiriki na habari za mhusika

Ukihisi ya kuwa hutaki kuendelea na utafiti huu uko huru kuacha kushiriki. Unaweza kufanya hivyo na bado tutashukuru kwa jinsi ulivyokubali kushiriki katika utafiti

huu.

Iwapo mshiriki ataondoa idhini yake, habari yake haitumiki tena na haiweza kutumiwa katika uchambuzi wa data zetu. Iwapo mshiriki anatoka katika utafiti huu kupitia kwa kifo au kupotea, basi data hiyo inaweza kuangaliwa kama itakayotumika katika uchunguzi. Lakini njia mwafaka za kitarakimu zitatumika kurekebisha uchunguzi wa data hiyo ili iwe sahihi

Usiri

Habari yote tunayopata kutoka kwako inalenga kutumiwa katika utafiti tu na haitafichuliwa kwa mtu mwingine yeyote. Habari yako yote itakuwa siri. Hili litafanikiwa kwa kutumia nambari za siri za kuratibia ili kuficha wahusika wasitambulike.

Majina: Majina hayatatumika kama kitambulisho cha mshiriki.

Anwani za kutufikia

Iwapo una maswali yoyote kuhusiana na utafiti huu, tafadhali wasiliana na Lucy Obonyo Nambari ya Simu ya mkono: 0722816265, Sanduku la Posta 4899-00200 Nairobi.

Maswali kuhusu haki za kushiriki kwako yanafaa kuelekezwa kwa Mhazili wa Kamati ya Kitaifa ya Kuangalia Maadili, KEMRI Sanduku la Posta 54840, 00200, Nairobi, Nambari za simu: 020-2722541, 0722205901, 0733400003, Barua pepe: erc@kemri.org

Msimamo wa mshiriki

Baada ya kusoma na kuelewa lengo la utafiti huu, mimi nimekubali kushiriki kwa hiari yangu

Sahihi…… Tarehe……

Tazama: Kwa kutia sahihi yako hapa, umekubali kuwa, umesoma na kuielewa fomu hii na umepewa nafasi ya kuuliza maswali.

Umefahamu hatari zote na zimeelezwa kwako hadi ukaridhika.

Kuwa Chuo Kikuu cha Kilimo Na Teknolojia cha Jomo Kenyatta na Taasisi ya KEMRI havina sera au mpango wa kukulipa iwapo utapata maumivu yoyote kutokana na kushiriki katika utafiti huu.

Kushiriki katika utafiti huu ni mapenzi yako na hiari yako mwenyewe bila kulazimishwa.

Shahidi

Sahihi…… Tarehe ……

Mshiriki alipata nakala hii ☐

Kiambatisho Cha VIII: Fomu ya Kibali/Idhini ya Mshiriki (Kwa Mlezi)

Ufahamu wa Idhini kwa Washiriki: Ijazwe na mdhamini iwapo anayehusishwa bado ni mtoto au mdogo wa umri.)

Mada ya Utafiti: Viwango vya juu vya kifua kikuu kisichosikia dawa miongoni mwa wagonjwa walioambukizwa Virusi vya Ukimwi wanaopokea huduma katika kituo cha afya cha Maryland, Mathare 4A. Nairobi nchini Kenya.

Watafiti: Lucy Nyanga'u: Chuo Kikuu cha Kilimo Na Teknolojia Jomo Kenyatta, Dkt. Amukoye: KEMRI-CRDR, Prof. Zipporah: JKUAT

Lengo la Utafiti huu

Lengo kuu la utafiti huu ni kusaidia katika kuelewa hatari inayoletwa na kifua kikuu kisichosikia dawa kwa afya ya watu walioambukizwa Virusi Vya Ukimwi, wanaopokea huduma katika kituo cha Afya cha Maryland, kilicho eneo la Mathare 4a.Utafiti huu hautawafuatilia wanaoshiriki; wanaoshiriki watatoa sampuli mbili za kikohozi. Utafiti huu utachukua miezi sita kukamilika

Utaratibu

Utafiti huu utahusika na kupata sampuli mbili za kikohozi cha anayeshiriki. Washiriki watapewa vichupa vya kuwekea kikohozi vilivyoandikwa vizuri na kuelezewa na mtafiti mkuu, jinsi na ni wapi wanafaa kutolea kikohozi nyumbani kwao, asubuhi na mapema.

Kikohozi hicho kitaletwa na mshiriki/mdhamini wake hadi katika kituo cha afya cha Maryland katika maabara ya TB.

Kikohozi cha kwanza kitatolewa hospitalini, punde tu mhusika afikapo katika kituo hicho, kwa wale washiriki watakaowasili hospitalini asubuhi mapema baina ya saa mbili na saa mbili unusu na hawatakuwa wamepata chemsha kinywa au chai ya

asubuhi.

Kikohozi chao kitatolewa katika mahali maalum palipotengwa (chumba cha kikohozi). Maelezo jinsi ya kutoa kikohozi hicho yatatolewa na mtafiti mkuu. Kwa hivyo sampuli mbili za kikohozi zitatolewa kutoka kwa kila mshiriki (cha kwanza ufikapo hospitalini, cha asubuhi mapema nyumbani)

Kikohozi kimoja kitapimwa kwa sababu ya kutambua uwezo wa uwepo wa kifua kikuu katika maabara ya Maryland; na kingine kitasafirishwa hadi Maaabara kuu ya Kitaifa ya kurejelea masuala ya Kifua Kikuu kupitia kwa huduma ya uchukuzi ya Smith-line ili kichunguzwe na kipimwe uwezo wa kuhimili dawa. Pia utafiti huu utahusika na kutathimini ripoti za maabara za mshiriki huyo kuhusu idadi ya chembechembe zinazosaidia katika kupigana na magonjwa mwilini yaani CD4. Utafiti utamakinika kwanza na Viwango vya juu vya Kifua kikuu kisichosikia dawa miongoni mwa walioambukizwa Virusi vya Ukimwi katika hopitali ya Maryland.

Faida za Utafiti huu

Gharama ya kupima kikohozi na mengineyo maabarani haitalipiwa. Pia watakaopatikana na hali ya kuwa na Kifua Kikuu kisichosikia dawa watapata matibabu kutoka kwa vituo vya afya vya umma na waliona kifua kikuu kinachotibika kutoka kituo cha afya cha Maryland. Watatibiwa bila malipo.

Hatari na Usumbufu

Hakuna hatari yoyote ya kutoa kikohozi. Matibabu hayawezi kucheleweshwa hata kidogo wakati wa kusubiri uchunguzi ufanywe maabarani.

Kujiondoa katika kushiriki na habari za mhusika

Ukihisi ya kuwa hutaki kuendelea na utafiti huu uko huru kuacha kushiriki. Unaweza kufanya hivyo na bado tutashukuru kwa jinsi ulivyokubali kushiriki katika utafiti huu.

Iwapo kama mshiriki utaondoa idhini yako, basi habari yako haitumiki tena na haiweza kutumika katika uchambuzi wa data zetu. Iwapo mshiriki anatoka katika utafiti huu kupitia kwa kifo au kupotea, basi data hiyo inaweza kuangaliwa kama itatumika katika uchunguzi. Lakini njia mwafaka za kitarakimu zitatumika kurekebisha uchunguzi wa data hiyo ili iwe sahihi.

Usiri

Habari yote tunayopata kutoka kwako inalenga kutumiwa katika utafiti tu na haitafichuliwa kwa mtu mwingine yeyote. Habari yako yote itakuwa na kubakia siri. Hili litafanikiwa kwa kutumia nambari za siri kuratibia ili kuficha wahusika wasitambulike.

Majina: Majina hayatatumika kama kitambulisho cha mshiriki.

Anwani za kutufikia

Iwapo una maswali yoyote kuhusiana na utafiti huu, tafadhali wasilina na Lucy Obonyo Nambari ya Simu ya mkono: 0722816265, Sanduku la Posta 4899-00200 Nairobi.

Maswali kuhusu haki za kushiriki kwako yanafaa kuelekezwa kwa Mhazili wa Kamati ya Kitaifa ya Kuangalia Maadili, KEMRI. Sanduku la Posta 54840, 00200, Nairobi, Nambari za simu: 020-2722541, 0722205901, 0733400003, Barua pepe: erc@kemri.org

Msimamo wa mshiriki

Baada ya kusoma na kuelewa lengo na madhumuni ya utafiti huu, mimi nimekubali mtoto Wangu ashiriki katika utafiti huu.

Jina la Mtoto..... Tarehe.....

Jina na sahihi ya mzazi..... Tarehe.....

Jina na sahihi ya Mtafiti Mkuu..... Tarehe.....

Tazama: Kwa kutia sahihi yako hapa, unakubali kuwa; Umesoma na kuelewa fomu hii ya idhini na umepewa nafasi ya kuuliza maswali.

Umefahamu hatari zilizopo na zimeelezwa kwako kwa njia ya kukuridhisha.

Unaelewa kuwa Chuo Kikuu cha Jomo Kenyatta na Taasisi ya KEMRI, havina sera au mpango wa kukulipa iwapo utapata maumivu yoyote kutokana na kushiriki katika utafiti huu.

Kushiriki kwa mtoto wako katika utafiti huu ni kwa hiari na mapenzi yako mwenyewe bila kulazimishwa.

Shahidi:

Sahihi……. …..
Tarehe…….

Mshiriki alipata nakala hii ☐

Kiamabatisho Cha IX: Fomu ya Kibali/Idhini ya Kushiriki Kwa Kijana (Mvulana au Msichana)

Unaulizwa utusaidie katika mradi wa utafiti ulioelezewa hapa chini. Mzazi wako au mdhamini wako amekubali, lakini ni lazima uamue iwapo ungependa kushiriki katika utafiti huu au la. Unaweza kusimamisha na hata kujiondoa katika utafiti huu wakati wowote kwa kutufahamisha tu na itakuwa sawa.

Ukitaka kufahamu zaidi kuhusu utafiti huu, uko huru kuuliza maswali yoyote

Mada ya Utafiti: Viwango vya juu vya kifua kikuu kisichosikia dawa miongoni mwa wagonjwa walioambukizwa Virusi Vya Ukimwi wanaopokea huduma katika kituo cha afya cha Maryland, Mathare 4A. Nairobi nchini Kenya.

Watafiti: Lucy Nyanga'u: Chuo Kikuu cha Kilimo Na Teknolojia Jomo Kenyatta, Dkt. Amukoye: KEMRI-CRDR, Prof. Zipporah: JKUAT

Fomu ya kibali/kukubali

Lengo la Utafiti huu

Lengo kuu la utafiti huu ni kusaidia katika kuelewa hatari inayoletwa na kifua kikuu kisichosikia dawa kwa afya ya watu walioambukizwa Virusi vya Ukimwi, wanaopokea huduma katika kituo cha Afya cha Maryland, kilicho eneo la Mathare 4a.Utafiti huu hautawafuatilia wanaoshiriki; wanaoshiriki watatoa sampuli mbili za kikohozi. Utafiti huu utachukua miezi sita kukamilika.

Utaratibu

Utafiti huu utahusika na kupata sampuli mbili za kikohozi cha anayeshiriki. Wewe utapewa vichupa vya kuwekea kikohozi vilivyoandikwa vizuri na kuelezewa na

mtafiti mkuu, jinsi na ni wapi panafaa kutolea kikohozi nyumbani kwenu asubuhi na mapema.

Kikohozi hicho kitaletwa na mzazi au mdhamini wako hadi katika kituo cha afya cha Maryland.

Kikohozi cha kwanza kitatolewa hospitalini punde tu mhusika afikapo katika kituo hicho, kwa wale wahusika watakaowasili hospitalini asubuhi mapema baina ya saa mbili na saa mbili unusu na hawatakuwa wamepata chemsha kinywa au chai ya asubuhi.

Kikohozi chako kitatolewa katika mahali maalum palipotengwa (chumba cha kikohozi) mbali na watu wengine wanaokuja hospitalini. Kwa hivyo sampuli mbili za kikohozi zitatolewa kutoka kwako (cha asubuhi mapema nyumbani na cha kwanza ufikapo hospitalini).

Kikohozi cha kwanza kitapimwa hapa katika maabara ya kituo cha Maryland ili kutambua kama kina TB; na kingine kitasafirishwa hadi katika maabara kuu ya Kitaifa ya kurejelea masuala ya Kifua Kikuu kupitia kwa huduma ya uchukuzi ya Smith-line. Katika maabara hiyo ya Kitaifa kurejelea viini vya Kifua Kikuu vitakuzwa katika vyakula mahsusi na kuchunguzwa ni dawa zipi zinaweza kuviua au ni zipi haziwezi kuviua. Pia utafiti huu utahusika na kuchunguza ripoti za maabara kuonyesha idadi ya askari au chembechembe za damu zilizo katika damu yako zinazosaidia katika kupigana na magonjwa mwilini mwako.

Hatari/Usumbufu

Hakuna hatari yoyote kwa kutoa kikohozi. Matibabu hayawezi kucheleweshwa hata kidogo wakati wa kusubiri uchunguzi ufanywe maabarani.

Tutafanya kila linalowezekana kuhakikisha ya kuwa hutaumia kwa njia yoyote. Ni sisi tu tutakuwa tunafahamu yale umesema na kutenda na kamwe hatutatoa habari

yako kwingineko.

Iwapo unaelewa kile unachoulizwa ufanye hapa na umeamua kusaidia katika utafiti huu, unaulizwa uandike jina lako na utie sahihi hapo chini.

Jina na sahihi ya kijana..... Tarehe.....

Sahihi ya Mtafiti..... Tarehe.....

Mshiriki alipata nakala hii ☐

Kiamabatisho Cha X: Fomu ya Maswali kwa Mshiriki

Maswali Kwa washiriki (Ijazwe na mtafiti kwa wale tayari wamepeana idhini).

Unaulizwa utusaidie katika mradi wa utafiti ambao umekwisa ulezewa hapo awali. Utaulizwa maswali kuhusu afya yako, hautalazimizwa kujibu maswali, uko huru kuuliza maswali yoyote.

Mada ya Utafiti: Viwango vya juu vya kifua kikuu kisichosikia dawa miongoni mwa wagonjwa walioambukizwa Virusi Vya Ukimwi wanaopokea huduma katika kituo cha afya cha Maryland, Mathare 4A. Nairobi nchini Kenya.

Watafiti: Lucy Nyanga'u: Chuo Kikuu cha Kilimo Na Teknolojia Jomo Kenyatta, Dkt. Amukoye: KEMRI-CRDR, Prof. Zipporah: JKUAT

1. Je umri wako ni? ☐

2. Je unakohoa? Ndio ☐ La ☐

Kama ndio, umekohoa kwa muda gani?

Zaidi ya wiki mbili ☐ Chini ya wiki mbili ☐

3. Ni wakati gani unakohoa sana?

Mchana ☐ Usiku ☐ Kila Wakati ☐

4. Je unatoa kikohozi?

Ndio ☐ La ☐

Kama ndio hicho kikohozi kinaonekana aje?

Kimakamasi ☐ Mate Mate☐ Damu ☐

5. Je unatokwa jasho usiku?

Ndio ☐ La ☐

6. Je usawahi kaa na mtu ambaye ameugua kifua kikuu

Ndio ☐ La ☐

7. Je usawahi tibiwa kwa ajili ya kifua kikuu?

Ndio ☐ La ☐

8. Je unaumwa na kifua?

Ndio ☐ La ☐

9. Jinsia ya mshiriki? Mume ☐ Mke ☐

Msimamo wa mshiriki

Baada ya kushiriki kikamilivu katika kujaza hii fomu ya maswali, mimi nimekubali/ mlezi kutia sahihi kwa ajili ya mtoto au mtu yeyote asiyeweza kusoma.

Jina la Mshiriki/Mtoto..... Tarehe.....

Jina na sahihi ya Mshiriki/Mlezi..... Tarehe.....

Jina na sahihi ya Mtafiti Mkuu..... Tarehe.....

Tazama: Kwa kutia sahihi yako hapa, unakubali kuwa; umehuzizwa kikamilivu katika kujaza hii fomu ya maswali na umepewa nafasi ya kuuliza maswali.

Shahidi:

Sahihi……. …..

Tarehe…….

Mshiriki alipata nakala hii ☐