A COMPARISON OF TREATMENT OUTCOMES IN TUBERCULOSIS PATIENTS WITH AND WITHOUT CONCURRENT DIABETES MELLITUS AT THE UNIVERSITY TEACHING HOSPITAL, LUSAKA

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A dissertation submitted to the University of Zambia, School of Medicine, in partial fulfilment of the requirement for the award of Master of Medicine, Internal Medicine and Infectious Diseases Degree.

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CERTIFICATE OF APPROVAL

This dissertation titled “A comparison of treatment outcomes in tuberculosis patients with and without concurrent diabetes mellitus at the University Teaching Hospital, Lusaka” by Sombo Fwoloshi is approved as fulfilling the requirements of the Degree of Master of Medicine in Internal Medicine and Infectious Diseases of the University of Zambia.

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ABSTRACT

Diabetes Mellitus (DM) is known to be associated with active tuberculosis (TB). Zambia remains a high TB burden country with reports of increasing DM amongst developing countries. However, to date, few relevant studies have originated from Zambia. The strength of the association remains unexplored in Zambia and may be eclipsed by high HIV prevalence. This study in Lusaka, Zambia aims to determine the prevalence of DM among individuals with active TB and thus determine treatment outcomes in this population. In the research setting, Acid Fast Bacillus (AFB) smear status and culture at 8 weeks is a clinically validated surrogate end point for TB treatment outcome.

A prospective cohort study with limited follow-up was done among adult TB cases at University Teaching Hospital in Lusaka, Zambia over a 17 month long period from October, 2014 to February, 2016. A smear status positive for AFB defines a TB case, while a fasting blood sugar (FBS) ≥7 measured at the time of TB diagnosis, or a known DM individual on medication defines a DM case and was the exposure of interest. Participants were followed up at 8 weeks to determine outcomes which included smear status for culture AFB and death after 8 weeks on Anti Tuberculosis Treatment (ATT). Descriptive statistics were used to analyse the baseline characteristics, bivariate logistic regression to assess crude associations by determining crude odds ratios, multivariate logistic regression to assess adjusted associations by determining adjusted odds ratios.

A total of 127 individuals were enrolled in the study. Mean age was 36.9 years for non-DM versus 33years for DM participants, 6 were considered diabetic. Of these, 3 (50%) were known diabetics on medication while 3 (50%) were a new diagnosis as per study definition. The prevalence of DM among smear positive TB cases was 4.72 %. The mean FBS among the diabetics was 8.05mmol/l while for the individuals without DM the mean FBS was 5.15mmol/l. The risk factors associated with DM among TB patients was low education level (p = 0.001 and 95% CI, 0.001 – 0.148). At 8 weeks, cure and failure were similar in both groups (p-value=0.283). By 8 weeks, 11(8.66%) patients had died, all deaths were among patients without DM.

Overall, the prevalence of DM among individuals with smear positive TB was similar to that of the general population in Zambia; this is less than expected. In this study, there was no evidence to suggest that TB treatment outcomes differed between TB patients with and without DM, though the number of participants with DM was small. To explore this further, studies need to be done in settings with a higher prevalence of DM.

Keywords: Diabetes Mellitus, Tuberculosis, Treatment outcome, University Teaching Hospital, Zambia.

DEDICATION

To the two Annette's –my mother and my daughter.

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ABBREVIATIONS

AFB	Acid Fast Bacilli
AIDS	Acquired Immune Deficiency Syndrome
ATT	Anti-Tuberculosis Treatment
CAMP	Collection of Antimicrobial Peptide
CCL	Chemokine Ligands
DEF A and B	Defensins Alpha and Beta
DM	Diabetes Mellitus
DNA	Deoxyribonucleic Acid
FBS	Fasting Blood Sugar
HbA1c	Glycosylated Haemoglobin
HIV	Human Immune Deficiency Virus
IFNγ	Interferon Gamma
IRB	Institutional Review Board
MTB	Mycobacterium Tuberculosis
RHZE	Rifampicin, Isoniazid, Pyrazinamide, Ethambutol
RNA	Ribonucleic Acid
SSA	Sub-Saharan Africa
TB	Tuberculosis
UTH	University Teaching Hospital
W.H.O.	World Health Organization
ZAMBART	Zambia AIDS Related Tuberculosis

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CHAPTER 1

INTRODUCTION

1.1 Background

One third of the world is infected with Tuberculosis (TB), but only 10% will develop active TB in their lifetime (1).

TB is caused by *Mycobacterium tuberculosis (MTB), Mycobacterium bovis, Mycobacterium africanum* and *Mycobacterium microti, collectively* called Mycobacterium tuberculosis complex. TB is an airborne disease spread via the respiratory route; inhalation of a small number of bacilli is enough to develop infection. On the other hand, progression to active disease is dependent on several factors which include but is not limited to the host immune response. Both genetic and acquired risk factors lead to loss of immune response or failure to mount an effective immune response. Immunodeficiency states such as Human Immunodeficiency Virus (HIV) infection, chronic kidney disease, malnutrition and Diabetes Mellitus (DM) allow the bacilli to thrive and establish disease.

DM is a state of chronic hyperglycaemia as a consequence of relative insulin deficiency or insulin resistance and in some cases both (2). Individuals with overt symptoms are easily diagnosed with DM; on the other hand it can be a challenge to make a diagnosis in an asymptomatic individual especially in resource-constrained settings. According to estimates by the World Health Organisation (WHO), in 2014, DM affected more than 422 million people worldwide (3). This was projected to increase to 366 million people by the year 2030 (4, 5). In the early phase individuals have been known to lead a relatively normal life, however in the long term the complications of DM include premature death and reduced quality of life. Epidemiological studies have shown that for every person known to be diabetic there is another undiagnosed in the general population (2). The WHO diagnostic criteria for diabetes is based on figures that have been shown to be associated with high risk of micro vascular complications particularly retinopathy and the distribution of glucose across the population.

The number of adults with diabetes in sub-Saharan Africa is predicted to rise from 14.7 million in 2011 to 28.0 million in 2030 (6-9). A population-based cross-sectional study by Bailey *et al.* (10) done in Zambia and the Western Cape of South Africa showed the prevalence of DM to be 3.2% and 9.4% respectively. High risk groups for diabetes were found to be those with obesity, females and those of older age.

1.2 TB-DM Association

The mutually beneficial association between TB and DM is well established (11). Several studies have shown that people with diabetes are three times more likely to develop TB than the general population (12-15). In addition, these people suffer worse TB treatment outcomes, have a higher risk of treatment failure and have a higher mortality (16-19).

In Zambia, DM and its effect on the incidence, prevalence and severity of tuberculosis has become an important public health issue. Epidemiological studies estimate that in the next 20 years 75% of diabetics will live in countries with high prevalence of TB (20). With 70% of TB patients in Zambia having co-morbid HIV, a growing burden of diabetes presents a unique clinical and public health challenge, one which is easily overshadowed by HIV. Furthermore, this is a challenge which may be overlooked, as physicians are not often aware of the possible co-morbidity between infectious and non- infectious diseases. A study at UTH in Lusaka found sputum-producing diabetes patients to have a 6-fold increase in the odds of having culture positive TB compared to patients without diabetes (21).

With the economy of Zambia improving it is expected that lifestyle related diseases like DM will start rising in addition to known and undiagnosed diabetics. Unfortunately, this is the very terrain in which numerous studies have demonstrated high prevalence of infectious diseases. Data on the prevalence and pattern of non-communicable disease in Zambia is lacking, there is even less data on impact of communicable diseases on non-communicable disease or vice versa. HIV infected individuals who are exposed to TB infected persons are regarded as a high risk population and are screened for TB, with intention to administer chemoprophylaxis as prevention for TB disease. A study in Russia reported a 2-3 times reduction in TB among diabetics when Ftivazid (a Russian analogue of isoniazid), while a study in

Germany showed that provision of chemoprophylaxis after initial TB treatment in a group of DM patients was not associated with TB relapse (15).

1.3 Immune Response to TB in DM

Cellular and humoral immunity are both impaired in patients with DM (22, 23). Immunologic studies have shown reduced expression of antimicrobial peptides CAMP, DEFα1, DEFβ4 and DEFβ103A which have been noted to increase progression of active TB. This suggests that there is impaired production in DM patients, promoting the risk of reactivation of latent TB. Mice studies have shown a delay in the priming of the adaptive immune response in response to virulent MTB when compared to controls (24). Interferon gamma (IFN-γ) producing T-cells arise earlier in controls than in the mice with DM. There is also a late delivery of antigen bearing antigen-presenting cells from the lungs to the lymph nodes, due to reduction in the leukocyte chemo attractants such as CCL2 and CCL5 at the sites of early infection (24). Generally, in patients with TB the population of hypodense alveolar macrophages is increased as compared to non-TB infected controls, while the activation status of the alveolar macrophages and T-cells is similar among subjects with DM and normal subjects. This population of cells is key in the elimination of mycobacterial infections and control of disease severity.

It has been observed that in TB patients with co-morbid DM, the alveolar macrophages are activated to a lesser extent than in patients with TB alone and that the ratio of alveolar macrophages is inversely related to bacterial load and disease extent as determined by chest radiograph (25). A retrospective analysis of patients with TB and DM in Texas demonstrated a 5 day delay in the clearance of mycobacterium from sputum in the intensive phase (26), hence DM seems to interfere with the pulmonary sterilizing power of the anti-tuberculosis drugs. This translates into additional days of potential infectivity.

1.4 Statement of the Problem

The slow decline of TB and the upsurge of DM in low and middle-income countries is a potential epidemic of Active TB. TB-DM patients have higher rates of treatment failure and death. In Zambia this double burden of disease is unexplored. No study in Zambia has explored the unique association between DM and TB. Understanding the

pattern of disease and response to standard treatment in this subpopulation can facilitate the provision of better quality care.

Currently TB patients are routinely screened for HIV, which is a well-recognised driver of the TB epidemic; however DM has been shown to negatively and significantly affect TB in multiple ways in other parts of the world. It is only prudent to attempt to investigate this issue so as to explore the pattern of disease in our country as great variation in disease prevalence and associations can exist in different regions of the world. Examination of prior studies reveals that sufficient data does not currently exist to answer the questions identified in section 3.1. The study will highlight the need for an intensified focus on diabetes among TB populations in Zambia regardless of their HIV status. This study will potentially serve for incremental studies in the future on the TB-DM associations; for instance, of the few studies that have shown that there is reduced rifampicin levels in TB patients co-afflicted with DM, few researchers have set out to specifically compare rifampicin levels to TB treatment outcomes (49, 53) let alone the pharmacogenomics which may influence this phenomenon.

1.5 Study Question

In adults on treatment for active pulmonary TB at UTH in Lusaka, do treatment outcomes differ between those with and without DM?

1.6 Rationale

This study is expected to not only compliment previous and current studies into TB-DM association's elsewhere in the world, but is anticipated to justify and propel further research by quantifying the magnitude and strength of associations and the impact on TB treatment outcome. The study will highlight the need for an intensified focus on diabetes among TB populations in Zambia regardless of their HIV status. The use of a surrogate endpoint for TB treatment outcome is a feasible way of assessing the factor under investigation in a timely and cost-effective way.

1.7 Hypothesis

1.7.1 Primary Hypothesis

1.7.1.1 Null Hypothesis

The proportions of death and treatment failure in adults on treatment for active pulmonary TB are the same in those with and without concurrent DM.

1.7.1.2 Alternate Hypothesis

The proportions of both death and treatment failure are higher in adults on treatment for active pulmonary TB who have concurrent DM compared to those without diabetes.

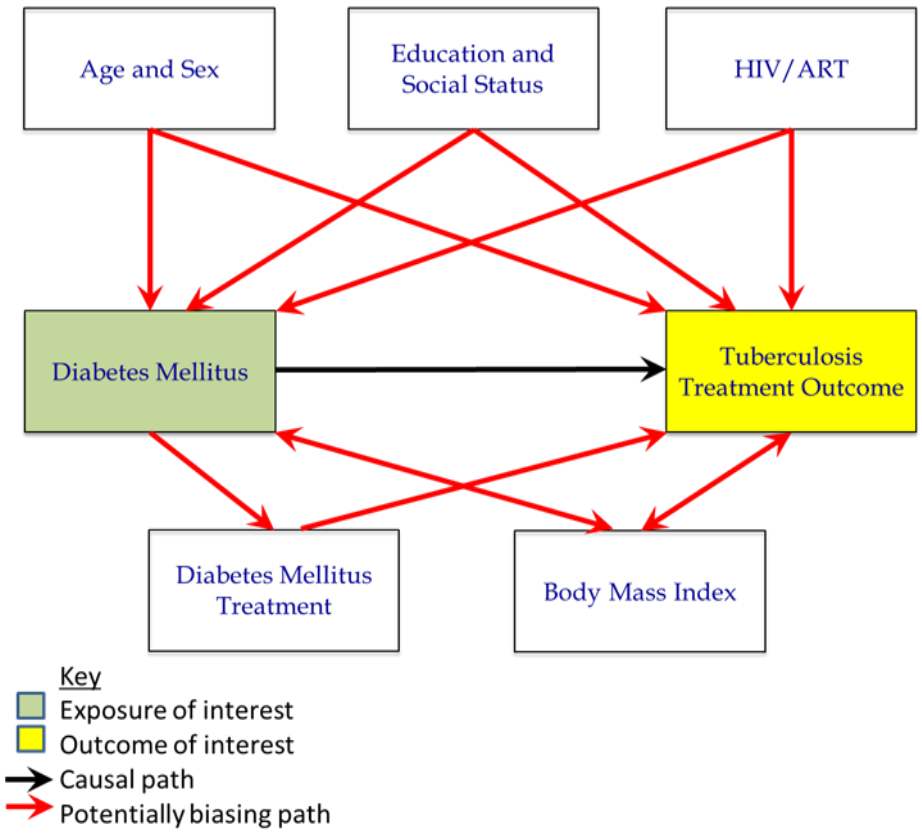


Figure 1.1: Conceptual framework

The exposure of interest is DM while the outcomes of interest are TB smear status and death when compared to controls with TB only. Potential measureable confounding factors in this setting are age, sex, household, socio-economic position, education and HIV status.

Body disposition has a unique relationship with TB and DM. Low body disposition has a causal association with TB incidence and is also an effect of TB disease. Low body disposition is a risk factor for poor TB outcome, but is also a consequence of severe TB disease which is independently a risk factor for poor TB outcome. High body disposition (particularly central obesity) has a causal association with insulin

resistance and diabetes, and thus secondarily with hyperglycaemia, but low body disposition can be an effect of hyperglycaemia from uncontrolled DM.

DM is more common among older individuals (54) and advanced age is a risk factor for Active TB(55), thus may influence the response to treatment. Active TB has been associated with male's preponderance (56). However the sex ratio may be modified by the presence of DM.

HIV is an immunosuppressive driver and by far the biggest effect modifier expected in the study. It is an independent risk factor for TB. On the other hand, being on effective Anti-retroviral (ARV) therapy may reduce the frequency of TB. Contrariwise, some ARVs especially from the class of protease inhibitors (PI) may lead to DM and may alter TB treatment outcome negatively (57).

1.8 Objectives

To determine if active pulmonary TB treatment outcomes at UTH in Lusaka between individuals with or without DM.

1.8.1 Specific Objectives:

i. To estimate the prevalence of DM among adults on treatment for smear positive TB at the UTH, in Lusaka.
ii. To study the association between DM and treatment failure in those on therapy for active pulmonary TB at the UTH, Lusaka.
iii. To determine if active pulmonary TB treatment outcomes at UTH in Lusaka differ between individuals with or without DM.

CHAPTER 2

LITERATURE REVIEW

2.1 DM Trends

According to epidemiological studies, it is estimated that between 2010 and 2030 there will be a 69% and 20% increase in DM in developing and developed countries respectively (27). They also estimated that by 2030, DM would affect up to 439 million people worldwide, however these figures may actually be an underestimation as reported by the International Diabetes Federation which estimates that by 2030 the prevalence of DM will be 530 million people worldwide. A population based prevalence study carried out in Lusaka Urban (28) found a combined prevalence of impaired glucose tolerance and diabetes of 4.0%, while Bailey *et al.* (in press, personal communication) found a diabetes prevalence of 3.2%.

On 20th December 2006 the United Nations General Assembly declared DM an international public health issue. The Assembly also declared World Diabetes Day as a United Nations day on 14th November 2011. This was in view of the serious negative impact of DM on individuals and economies across the globe (29).

2.2 TB Trends

According to the WHO report 2013, there were 8.6 million new cases of TB in 2012, with resultant 1.3 million deaths. Interestingly most deaths (1 million) were in the non-HIV infected individuals. The report also stated that Africa has a quarter of the world's TB cases; it also has the highest rates of cases and deaths relative to the population, 255 per 100,000, this is double the global average of 122 per 100, 000 people. It is widely acknowledged that the TB epidemic is mostly fuelled by HIV. On the other hand in order to achieve the set targets of reducing incidence and mortality from TB by 2015, other drivers of the TB crisis like DM need to be addressed.

For centuries the marriage between TB and Diabetes has provoked much thought. Root H in 1934 published a treatise on the patients affected with both TB and DM, among other things he found that TB was curiously common among adults with DM while at post-mortem, the findings were similar for TB patients with and without DM (30).

This was more than 10 years after insulin was discovered, and before anti-tubercular drugs were discovered. A systematic analysis of 13 observational studies (14) showed that DM was associated with an increased risk of TB, (RR3.11, 95% CI, 2.27 to 4.26 OR range of 1.16 to 7.83), some studies have demonstrated that patients with diabetes and tuberculosis have a higher likelihood of TB treatment failure compared to patients with only TB (16-18). A study in Egypt showed a 3.9 times increased risk of treatment failure in patients suffering from diabetes. Currently the overall burden of communicable diseases is concentrated within low income countries particularly those in sub-Saharan Africa. The burden of non-communicable diseases which has been low so far is also rising (31). Prevalence of diabetes is expected to rise from 12.1 million people infected in 2010 to 23.9 million in 2030 (11). Increasing industrialisation and urbanisation are leading to higher rates of obesity and diabetes as more people in developing countries are adopting a sedentary lifestyle. Diabetes poses a large financial burden on countries with very limited resources. Concomitantly the highest burden of tuberculosis is present in sub-Saharan Africa with Zambia having an incidence of 506 per 100, 000 population.

The association between diabetes and tuberculosis has been shown in many studies (12, 13, 21). In another study by Mboussa and colleagues at Brazzaville University Hospital, treatment failure was found in 41% of the patients with tuberculosis and diabetes but in only 13% of those with tuberculosis alone (17). The negative influence of DM on TB is attributed to monocyte chemo attraction, alveolar macrophage activity and type 1 cytokine phenotype. Both innate and adaptive immune responses are compromised in DM patients hence making them susceptible to TB. Unlike patients with TB alone hypodense alveolar macrophages are less activated in TB patients with diabetes, hypodense macrophages may contribute to susceptibility of TB in DM patients (32). Diabetes has been significantly associated with multidrug resistance TB (33). Diabetes increases the risk of treatment failure, relapse and death (34). Diabetes is one of the poor prognostic indicators in patients with TB (19). Time to culture conversion is longer in diabetic patients than in patients without diabetes (18, 35).

2.3 Use of Surrogate Endpoints

Validated biomarkers can be used as surrogate markers of endpoints. In short, they are especially useful in the development of drugs. Time to culture positivity and

conversion are two biomarkers of TB that have been validated for use as surrogate endpoints (19). Studies looking at culture conversion show a mixed picture depending on the outcome variable being measured with patients suffering from both diseases seeming to have longer sputum-culture conversion times than patients suffering from just tuberculosis but with culture positivity rates at 3 months being similar (19, 26, 36). Previous studies from outside Zambia suggest that there is an increased risk of tuberculosis treatment failure and an increased risk of mortality among diabetes patients infected with tuberculosis when compared to other tuberculosis patients (8, 9, 11). Other studies assessing time to sputum culture conversion show longer culture conversion times for tuberculosis patients with diabetes compared to those without diabetes which has been suggested may lead to higher rates of relapse. This presents questions that could greatly affect the clinical management of these diseases and these questions have not been adequately studied in sub-Saharan Africa.

Several studies have addressed TB/HIV co-infection in the world and Sub-Saharan Africa; however there is scanty data on the convergence of TB and diabetes in our region and in particular Zambia. TB is the leading cause of mortality among patients with HIV/AIDS, the risk in these patients is associated with CD4+ T cell deficiency. Patients presenting to the health services with TB are routinely screened for HIV as an entry point to HIV care services, there are well outlined protocols and policies based on large studies on the management of TB/HIV co-infection.

In Zambia, data is lacking on the association between TB and diabetes. A look at records in TB clinics has shown that most do not indicate if a patient is known to have DM or not, a clear indication that the magnitude of the problem is not appreciated in Zambia.

2.4 TB and Diabetes: Past, Present and Future

With the finding of increased risk of TB among patients with diabetes, it appears that the greater risk is among populations with DM type 1 (T1DM) (24, 37) and patients with high glycosylated haemoglobin (HbA1c) (19). Using these as markers of disease severity, uncontrolled DM is associated with increased incidence of TB. It is relatively unknown if improving glycaemic control may actually alter treatment outcomes in patients with DM. Studies have shown that mortality and treatment failure is higher among DM patients when compared to controls (16, 38). However only 1 study has

documented the cause of death among these patients (17), thus there is a knowledge gap as to whether co-afflicted individuals demise due to severity of the TB infection or complications of DM. This information would be useful to determine if these deaths are actually due to preventable causes. There is mixed evidence as to whether patients with co-morbid DM have a more severe form of TB than controls based on chest x-ray. Some studies concluded that TB patients with DM had more diffuse lung involvement and more cavitatory lesions on chest imaging (39-42). However most were retrospective and mostly used chest x-ray while only one study used CT imaging (39). However contrary to these findings, other studies found that radiographic appearance did not differ between the two patient groups (43, 44). It has been observed that TB patients with and without DM will have similar rates of sputum culture conversion to negativity by 2 to 3 months (18, 45-47) despite individuals co-infected with DM having a higher initial TB bacillary burden (17, 25, 48). However it has been observed that co-afflicted patients have a significantly longer time to negative culture conversion and a shorter time to culture positivity (26, 36, 45). The cause of this has not been fully explored. Similarly, it remains unknown if this phenomenon places DM patients at higher risk of relapse.

Reduced drug levels are implicated in the development of drug resistance and treatment failure. The pharmacokinetics of anti-tuberculosis drugs is a likely contributory factor to the slower response to anti-tubercular therapy (ATT) in DM patients. Rifampicin is a bactericidal agent and a very potent sterilizer (a member of the rifamycins). It exerts its antimicrobial activity by inhibiting bacterial DNA–dependent RNA polymerase, suppressing RNA synthesis. It only requires one step to develop resistance via RNA genome mutation. Rifampicin exposure has been found to be lower in subjects with co-existent TB and DM when compared to patients with TB alone; a study in Indonesia found serum rifampicin concentrations in patients with both TB and DM to be half of that TB controls (49). On the other hand, hyperglycaemia is related to reduced rifampicin exposure, as this increases the gastric pH by reducing the production of gastric hydrochloric acid (50, 51). In this way, the absorption of rifampicin is significantly reduced as it is a pH dependant process (52). Another factor that may potentially affect ATT drug levels and dosing is the increased incidence of obesity among DM afflicted individuals.

What has not been established is if these reduced rifampicin levels are directly related to treatment outcomes. If this is found to be the case in follow up studies then prolonged treatment and higher doses of anti-tuberculosis treatment (49) may be considered to attain better treatment outcomes in TB patients with DM (37).

In a study in Indonesia that screened for DM among TB patients, up to 60% of those diagnosed with DM were not previously known to have DM (38). In such instances, it may prove to be a worthwhile undertaking to use TB services as a point of entry into DM care. Conversely, data is still lacking to establish if screening for latent TB in DM patients would be beneficial or feasible. Despite evidence from other parts of the world showing we are drawing closer to a possible double epidemic of TB and DM, literature searched revealed no documented study done in Zambia on the TB-DM association, let alone any research to attempt to fill the knowledge gap that exists in the understanding of the potentially mortal interaction of these two diseases.

CHAPTER 3

RESEARCH METHODOLOGY

3.1 Research Methods

3.1.1 Operational Definitions

- Diabetes Mellitus (DM) patient: Individual with a fasting blood sugar of 7 mmol/l as confirmed by a point of care test or self-report of diagnosis of DM or on treatment for DM.
- Fasting blood sugar: blood sugar measurement after at least 8 hours of not eating or drinking anything excluding water.
- TB Patient: individual with Smear positive TB.
- Sputum positive TB: laboratory confirmed presence of TB on smear/culture.
- TB-DM patient: Individual co-afflicted with both TB and DM.
- Treatment Outcome: sputum status (positive or negative) after 2 months of start of ATT or death in a patient who was initially sputum positive.
- Died: A patient who was initially sputum positive dies for any reason during the initial 2 months course of TB treatment.
- TB treatment: Anti-tuberculosis drugs as prescribed by attending physician.

3.1.2 Study Design

The study was an observational prospective cohort study which was conducted from 17th October 2014 to February 2016. Participants were recruited consecutively in a parallel manner (as illustrated in Figure 3.1). The study set out to recruit 114 individuals with a diagnosis of smear positive TB about to be commenced on ATT or already on ATT for not more than 7 days.

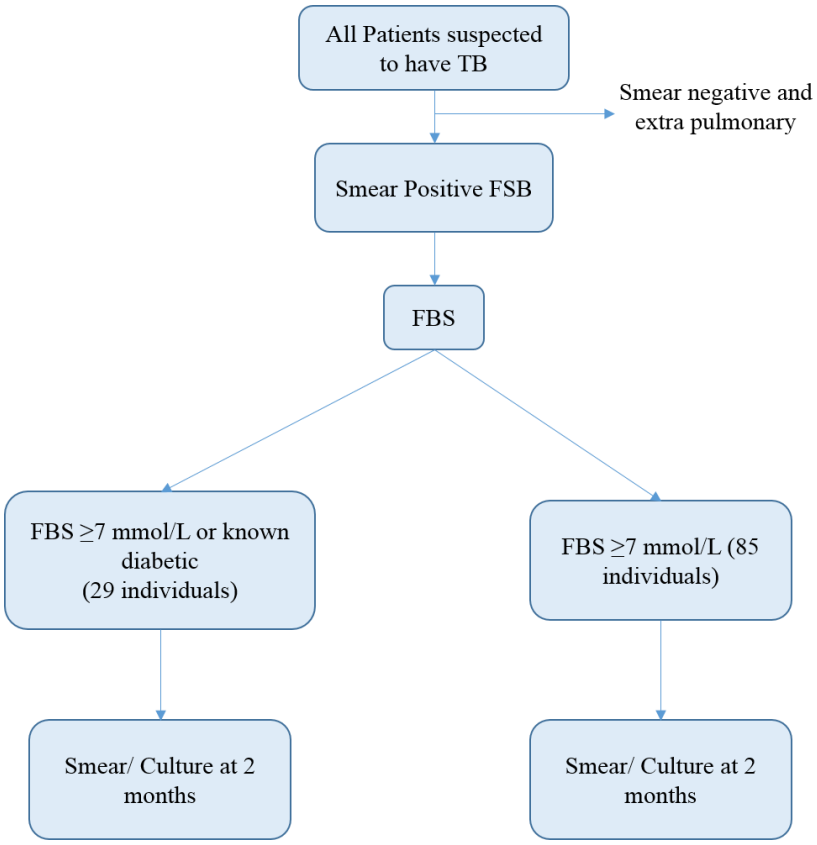


Figure 3.1: Study diagram

3.1.3 Study Site

The patients were recruited from the chest clinic and adult inpatient wards at the University Teaching Hospital (UTH).

3.1.4 Study Population

Individuals who met the following eligibility criteria were recruited to the study. A total of 127 participants were recruited as below.

Table 3.1: Eligibility criteria

Inclusion criteria	Exclusion Criteria
Smear positive or culture pulmonary TB case within 7 days of starting ATT FBS >7.0mmol/L for diabetic arm Known diabetics	Less than 18 years of age Smear-negative pulmonary TB Extra pulmonary tuberculosis

3.1.5 Sample Size

OpenEpi was used to calculate the sample size, based on formulas for cross sectional and cohort studies from Kelsey *et al.* Methods in Observational Epidemiology 2nd Edition. Previous studies estimated a prevalence of smear positivity of about 30% in diabetic patients with TB and about 8% of TB controls. At 80% power and at a 3:1 ratio between TB controls patients and TB/DM individuals, using 1-α of 0.95. A calculated sample size of 102 participants, assuming a dropout rate of 10%, a total of 114 individuals needed to be enrolled (therefore 85 TB individuals, 29 TB-DM individuals). The chest clinic attends to approximately 5,000 patients in a year, of which approximately 350 are smear positive. This sample size calculation was preferred for convenience and feasibility. However during the course of the study it was evident that the number of participants who were eligible for the diabetic arm were considerably less than suggested from previous studies therefore the study extended screening up to 131 patients to enhance the pickup of individuals in the diabetic arm in order to achieve the calculated 29. Despite this extension only 6 individuals were eligible for the diabetic arm

3.1.6 Study Variables

The following study variables will be collected during the study.

Table 3.2: Study variables

Independent variables	Dependent variables
Age Sex Weight and height Socio-economic status FBS HIV status AFB Smear	AFB smear and culture status at 2 months Death Loss to follow-up

3.1.7 Study Procedure

All the participants were on standard Anti-tuberculosis treatment (ATT) as was routinely prescribed by the attending clinician according to the National Treatment Guidelines for Tuberculosis. At the time of the study, the recommended ATT treatment regimen by the Ministry of Health included Pyrazinamide, Ethambutol, Isoniazid, Rifampicin and Streptomycin (added only during re-treatment regimen), which are given as a fixed dose combinations according to weight bands (Appendix XIII).

Fasting blood sugar (FBS) was checked on all the patients using a point of care glucometer. Participants who were known to be diabetic or with a FBS of more than or equal to 7mmol/l were enrolled into the arm with TB patients with co-morbid DM (cases) while the control arm was TB patients without DM i.e. patients who had a fasting blood sugar less than 7mmol/l. The initial data set was collected to meet Objective a. (section 6.1) as it was expected that more than 114 of the smear positive TB patients needed to be screened for DM to attain the target for the arm with co-afflicted DM. At recruitment, all participant data was collected using a validated interviewer administered questionnaire. All data collected from participants was entered via PDA into a secure database specifically designed for the study with appropriate privacy safeguards. To minimise on the loss of follow-up participants, participants were contacted 2 months into their ATT to remind them to come for their subsequent routine review at the hospital. To meet Objective b. (section 6.1), sputum samples were collected to check for TB status at 2 months. Participants and caregivers were contacted or contacted the study team in case of death. Hospital documents were also reviewed to check for possible deaths.

This study was an observational prospective cohort study with limited follow-up. The researcher was not responsible for administering the ATT and the study used a clinically validated surrogate endpoint (2 months).

Table 3.3: Study activities

At enrolment	At 8 weeks (2 months)
Sputum AFB	Sputum AFB
capillary blood for FBS	sputum culture

3.2 Data Collection Methods: Definitions and Measurement

The exposure, DM, was measured by a single capillary fasting blood sugar concentration using an Accu-Chek® point-of-care glucometer. This test is simple, quick, inexpensive, minimises participant inconvenience (the test requires one finger-prick drop of blood). The potential effect modifier, HIV status, was taken from TB clinic records. As part of the Zambian National Tuberculosis Control Programme, HIV infection is routinely tested for in all new TB cases. In order to adjust for potential confounding factors, a structured questionnaire (clinically validated) was administered to all cases by a member of the research team to obtain information on age, sex, household, socio-economic position and education level. In addition to checking the records for HIV status, the questionnaire also dedicated a section to collecting data in relation to the patients HIV status, other possible immune deficient states like malignancies or drugs (such as steroids if known to the patient) will be noted. However, the spectrum of immunodeficiency states is vast. Other known immune deficiency states such as cyclic neutropenia, hypogammaglobulinemia, leukocyte adhesion defects are rare especially in adults and were not expected to significantly bias the study. This information was entered directly onto a pre-programmed Personal Digital Assistant (PDA) by the researcher.

To allow for sub-group analyses, the TB register and laboratory records of all cases was reviewed at enrolment to determine if microbiological confirmation was obtained for the diagnosis, based on routinely collected specimens processed by the TB clinic. The TB register was reviewed again at 2 months into The TB treatment.

Research staff were trained by the principal investigator (PI) on all aspects of data collection and were required to follow standard operating procedures for glucose, and for measurement of anthropometrics. Research staff were monitored daily by the PI for quality control through repetition of non-invasive data collection on 2 randomly selected participants, and observation of invasive sample collection in another 2 randomly selected participants. A pilot study was performed on the first 10 participants recruited to test all data collection tools and procedures.

Accounting for weekends and public holidays, a research assistant was required to assist in the recruitment of patients for a year.

3.3 Sample Archiving

Biological (Blood and sputum) specimen samples were stored for 5 years to facilitate for future research on TB and DM. Written consent was obtained for storage of such samples (see information sheet and consent form). Left over samples will be destroyed or sent to the National Repository as provided for in the National Health Research Act.

3.4 Data Analysis Plan

The collected data was analysed using Stata version 12. The analysis focused on establishing the relationship between:

- The treatment outcomes in patients with TB alone.
- The treatment outcomes in patients with TB-DM.

Descriptive statistics will be used to analyse the baseline characteristics, Univariate logistic regression to assess crude associations by determining crude Odds Ratios, Multivariate logistic regression to assess adjusted associations by determining adjusted Odds Ratios.

3.5 Ethical Issues

The study followed all procedures in accordance with the Helsinki declaration. All the study recruits received care in line with current routines and guidelines in the hospital and benefited from having an assigned nurse to monitor the progress and outcomes of their treatment. Currently, TB culture is not routinely done at 2 months and 6 months, which is the time when sputum AFB is done to see if the course of treatment is effective. Patients in the study received all required tests and procedures in line with the DOTS strategy recommended by the W.H.O and currently in use in Zambia. Patients experienced minimal bruising and discomfort during blood draws, which was be done by trained staff. Standard operating procedures, which have been included in the annexes, were used to ensure safe and correct collection of all biological specimens from participants. No other hazards were related to this study.

The participants' interests were prioritized over those of science or society. Formal consent was required of every participant. Each participant received an information sheet detailing the aims and methods of the study and were asked if they wanted to be included in the study. Participants who responded favourably were formally consented

and were required to sign two copies of a consent form, one of which was retained, in order to participate. There was no coercion of those not inclined to participate. Medical care was not withheld from participants who decided to withdraw from the study. Strict confidentiality was maintained throughout the study. Ethics approval was secured prior to study commencement from ERES Converge IRB (Institutional Review Board).

CHAPTER 4

RESULTS

4.1 Research Findings

4.1.1 Study Process

131 participants were screened, 1 was excluded as he had no evidence of a smear result while three did not meet study criteria as they were not in a fasted state. A total of 127 were finally enrolled into the study. Figure 4.1 illustrates the recruitment and enrolment process.

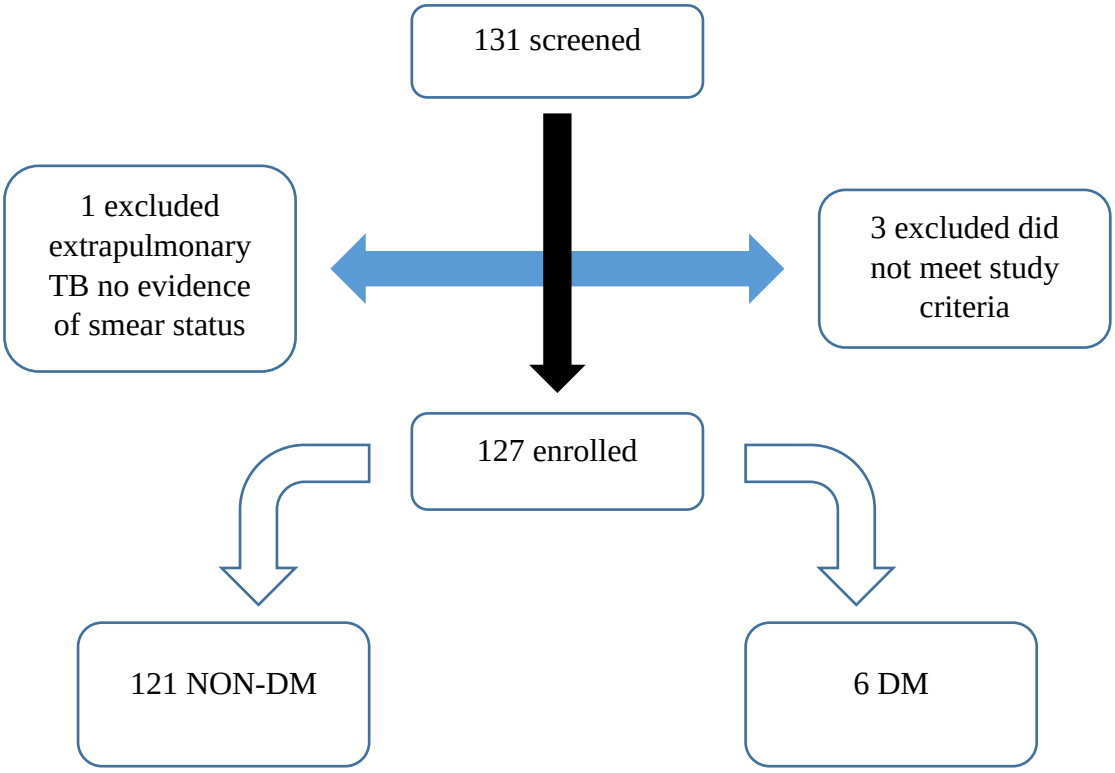


Figure 4.1: Study flow diagram

4.1.2 Participants

Of the 127 recruits, only 6 were considered diabetic as per study protocol. Of these, 3 (50%) were known diabetics on treatment while the other 3 (50%) were newly identified. As shown in Figure 4.2, the exploratory prevalence of Diabetes during the period of the study (October 2014 through February 2016) in this group was 4.72%. More than two thirds were out patients from both groups (p=0.993).

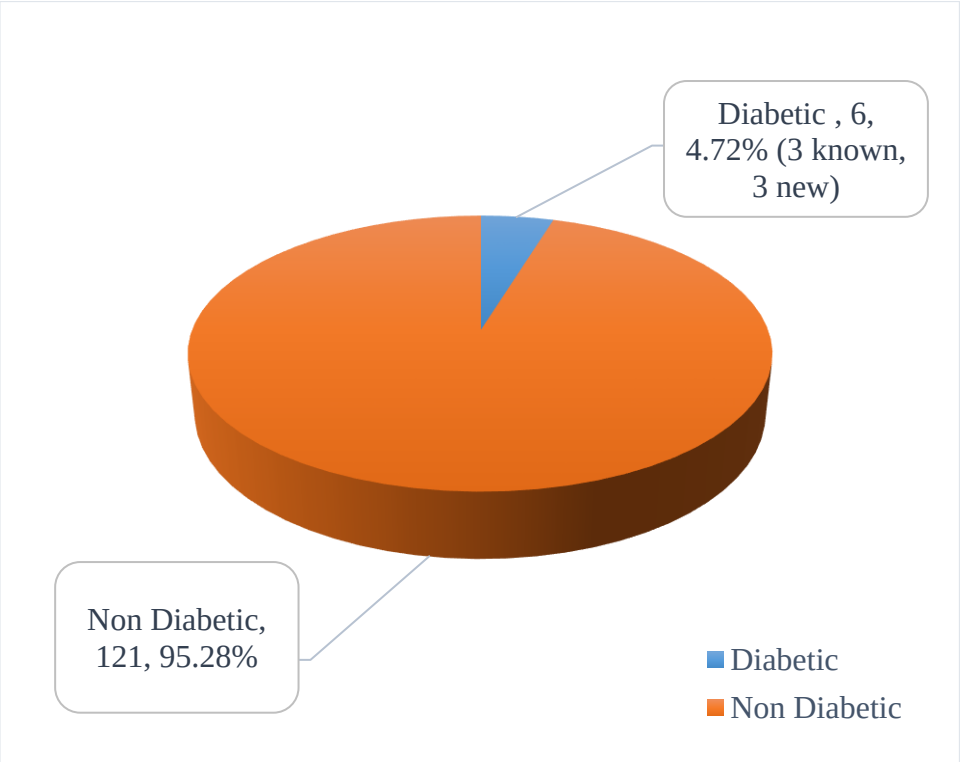


Figure 4.2: Prevalence of DM

4.1.3 Baseline Characteristics of participants

Table 4.1 summarises the baseline demographic and clinical characteristics. No significant differences in age were observed between the two groups. The mean age for the diabetics was 33 years versus 36.9 years for the non-diabetics. However, among the participants with DM, there were no patients over 50 contrasted with years of age while 17 (14.05%) were between 51 and 85 years among the non-diabetics.

The median FBS for the Diabetic group was 5.2 (IQR 5.9-4.6) versus 8.05 (IQR 10.8-7). All blood sugars that were measured were less than 10mmol/l aside for one individual who had a FBS of 19.5mmol/l.

Table 4.1: Background characteristics of 127 pulmonary tuberculosis patients

Characteristic	**Patients without diabetes N (%)** 121 (95.28)	**Patients with diabetes N (%)** 6 (4.72)	**p-value**
Age			0.491
18 – 35	69 (57.02)	3 (50.00)	
36 – 50	35 (28.93)	3 (50.00)	
51 – 85	17 (14.05)		
Sex			**0.017**
Male	38 (31.4)	1(16.67)	
Female	83 (68.6)	5 (83.33)	
HIV infection*			0.658
No	45 (37.19)	5 (83.33)	
Yes	25 (20.66)	1(16.67)	
Missing	51 (42.15)		
ART*			0.841
No	5 (4.13)	0 (0.00)	
Yes	20 (16.53)	1 (16.67))	
Fasting Blood Sugar (median IQR)	5.2 (4.6 – 5.9)	8.05 (7 – 10.8)	
Education			**<0.001**
Uneducated	2 (1.71)	4 (66.67)	
Primary	39 (33.33)	1 (16.67)	
Secondary	46 (39.32)	1 (16.67)	
Tertiary	30 (25.64)	0 (0.00)	
Smoked			0.840
No	108 (92.31)	2 (100)	
Yes	9 (7.69)	0 (0.00)	
Body Mass Index (BMI)			0.817
Under weight	31 (27.68)	1 (16.67)	
Normal Weight	70 (62.50)	5 (83.33)	
Over weight	9 (8.04)		
Obese	2 (1.79)		
Abdominal circumference mean (SD)	64.27 (10)	64.5 (13.90)	
Treatment outcome*			0.283
Cured	19 (45.24)	5 (83.33)	
Failed	12 (28.57)	1 (16.67)	
Death	11 (26.19)		
Location of recruitment			0.993
Inpatient	20 (16.53)	1 (16.67)	
Outpatient	101 (83.47)	5 (83.33)	

*missing values

Data are n (%) unless otherwise specified

Females were more likely to have diabetes than their male counterparts (p=0.017) at univariate analysis this was still statistically significant (p=0.032, CI=1.233-96.72) however adjustment for baseline BMI changed the group differences in sex. Table 4.2 illustrates the univariate logistic regression analysis.

Table 4.2: Unadjusted factors associated with Diabetes in Tuberculosis patients

Characteristic	Unadjusted Odds Ratio (OR)	P Value	95% CI
Age			
18 – 35	1		
36 – 50	1.971	0.420	0.378 – 10.28
Sex			
Male	1		
Female	10.92	**0.032**	**1.233 – 96.72**
HIV infection*			
No	1		
Yes	0.36	0.363	0.040 – 3.255
Education			
Uneducated	1		
Primary	**0.013**	**0.001**	**0.001 – 0.175**
Secondary	**0.011**	**0.001**	**0.001 – 0.148**
Body Mass Index (BMI)			
Under weight	1		
Normal Weight	2.114	0.476	0.248 – 19.75
Treatment outcome*			
Failed	1		
Cured	0.345	0.359	0.036 – 3.350
Location of recruitment			
Inpatient	1		
Outpatient	0.990	0.993	0.1097 – 8.935

All participants with DM knew their HIV status while over half of the non-diabetics (79.34%) did not know or were unwilling to disclose their status.

Over 60% DM participants had no formal education while more than 50% of the non-DM participants were educated (p<0.001). Further analysis at univariate logistic regression found that the difference in education levels was statistically significant (p< 0.001) for both primary and secondary education. Being overweight and obesity are associated with incident DM but may be protective of TB, while being underweight is associated with TB. On the other hand, poorly controlled DM also induces weight loss. Therefore to determine if the effect of education was influenced by BMI, correcting

for BMI using multivariate logistic regression did not appreciably alter the overall results (p=0.003 and 95% CI, 0.001 – 0.2217) for primary education and (p=0.002 and 95% CI, 0.001 – 0.2068) for secondary education.

Table 4.3: Multivariate logistic regression assessing factors associated with diabetes in Tuberculosis patients

Characteristic	Adjusted Odds Ratio (AOR)	*P* Value	95% CI
Body Mass Index (BMI)			
Under weight	-		
Normal Weight	-	-	-
Sex			
Male	1		
Female	9.986	0.097	0.659 – 151.3
Education			
Uneducated	**1**		
Primary	**0.012**	**0.003**	**0.001 – 0.2217**
Secondary	**0.011**	**0.002**	**0.001 – 0.2068**

4.1.4 Treatment outcomes

As shown in Table 4.1, by 8 weeks, 11 (26.19%) participants had died; all mortalities were among participants without diabetes. The Two groups had similar proportions of failure and cure as determined by negative AFB and MGIT after 8 weeks on ATT. Table 4.4 presents the lab results of the 8 weeks sputum samples. Some participants were lost to follow up or did not have sputum results despite submitting a sample at 8 weeks as show in Table 4.5.

Table 4.4: Follow-up sputum results

	Non-DM		DM	
AFB	Positive 12	Negative 19	Positive 1	Negative 5
MGIT	Positive 12	Negative 19	Positive 1	Negative 5
Capillia	Positive 12	-	Positive 1	
	12	19	1	5

Table 4.5: Non availability of sputum results at 8 weeks

	Non-DM	DM
Samples submitted but rejected at the lab	12	0
Sample not collected participant was LTFU	67	0
Sample not collected as participant died before study end point	11	0

CHAPTER 5

DISCUSSION

5.1 Discussion

In this study, very few individuals were considered diabetic among participants with smear positive TB and even fewer were actually a new diagnosis of DM. This is less than reported from studies in other parts of the world (12, 14). However, this finding is similar to Zambia's national prevalence of diabetes (4.2%) (58). Several factors may explain this result. In general, diabetes tends to affect people who are older; in this study most participants were in the age range of 18 to 35 years. In contrast to a study by Bates *et al.* (21) done at the University Teaching Hospital in Lusaka which found a significantly high prevalence of TB among patients with diabetes (P = 0.006, OR 6.571, 95%CI: 1.706–25.3), this study was done among patients with a diagnosis of TB made by the attending healthcare worker. This may be a reflection of the lack of active investigation for TB among patients who are known to be diabetic. This is also supported by the fact that in the Bates *et al.* (21) study, among the patients who were found to have TB, a good number (13.4%%) were actually not being evaluated for TB by the admitting healthcare providers.

The results of this study suggest that DM is no more common among TB patients than the rest of the population. Therefore, it seems TB is not significantly driven by DM in our setting. However, the results must be viewed cautiously due to the small sample size. In the absence of significant numbers of participants with the exposure of interest in the study group, a number of factors must be considered in collectively including the statistical significance of the baseline characteristics and the results of the primary outcome analysis. The adherence to TB treatment and possible drug resistance should also be taken into account. To date, only 1 study in Zambia has assessed for DM among TB patients and the results are similar to this study. A recent study by Bailey *et al.* enrolled 3131 participants. This was a much larger cohort in comparison to this study. The study used a cut off of ≥11.1 to define hyperglycaemia. In that study, a hyperglycaemia prevalence of 1.3% was found and an even smaller proportion of DM after 12 weeks of treatment of TB. No evidence of association between DM and active TB was found in that study. Aside the larger study population, other strengths

of that study were that it was conducted at 3 large primary health centres as opposed to this study which was at 1 centre; at the time of TB diagnosis they also measured glycosylated haemoglobin (HbA1c) and repeated a FBS at 12 weeks of ATT which allowed for distinction of participants who may have had stress induced hyperglycaemia as opposed to DM. In the same study, FBS was found to be as accurate as the HbA1c for DM diagnosis at the time of TB diagnosis.

In other studies, income and education levels are predictors of mortality among patients with DM (59). In this study, lower education level was significantly associated with being diabetic. This is similar to findings in 2012 by Sacerdote *et al*. (60) in which 340 234 participants were followed up for 11.6 years and 12 403 cases of incident diabetes were found. On analysis, incident of diabetes was inversely associated with lower education levels. DM may be associated with health in a number of ways; higher level educated people may have better access to healthcare and be more knowledgeable on the need to seek medical assistance in certain situations thereby less likely to have undiagnosed disease and get communicable diseases such as TB. Several studies that have explored the relationship between education and DM were done in high income countries. A significant number of these have found that DM is more common among people with lower education levels (60, 61). Findings in this study should be taken in the context of the wide confidence intervals and further studies with larger sample sizes are needed to confirm this result. There is reason to believe the opposite may be true in our setting; the upsurge of non-communicable diseases is expected with change in dietary habits and lifestyle.

As expected, we had more males enrolled in the study than females as TB is more common among males (56). In contrast, wide variations have been reported in the gender distribution of DM. This study had more females than males in the DM group. This can be attributed to the fact that the population was younger, so there may be a genetic basis to DM. With older populations, men are affected more as type 2 DM becomes more common. However, the statistical significance of this was attenuated at multivariate logistic regression analysis. The few studies that have been done in Africa have not specifically shown any gender biasness (62).

Not all the data was available on the HIV sero status of the participants but over 50% was a known status, of these, more than half were HIV negative. This is consistent

with the fact that HIV negative individuals are more likely to have a positive smear for TB than their HIV positive counterparts (63). Only 1 of the DM patients was HIV positive. Therefore the remainder had other risk factors for getting TB other than HIV. However there was insufficient evidence to suggest DM was the risk factor for TB.

Rates of cure and failure was similar in both groups while all deaths that occurred were among non-diabetic participants. This is in contrast to studies that have shown more treatment failure and death among TB patients with diabetes. This is also in contrast to findings of a systematic review done by Baker *et al.* (34) which showed increased risk of death, failure and relapse among TB patients with DM. The difference can be attributed to the fact that only one of the thirty-three studies that were reviewed was actually done in Africa. Additionally, HIV is known to negatively impact TB treatment outcomes. Participants in the non-diabetic arm were more likely to have HIV or not know there HIV sero-status which might explain at least mortality rates and treatment failures in that group. Our findings suggest that TB and subsequent treatment outcomes were driven by other factors other than DM. Determining the differences or similarities in treatment outcomes in these two groups was challenging as the initial assumption that diabetic patients among individuals with TB would be fairly well represented was not appropriate for this particular population. In order to answer this question in our setting, a very large number of participants may need to be recruited. In the bid to curb the TB and DM, the WHO collaborative framework for TB has recommended the screening of all TB patients for DM despite little evidence to support this recommendation in our population. Since another study with a bigger population also drew few diabetics, testing all TB patients for DM is unlikely to be a worthwhile undertaking.

While all the participants with DM came back for review at 8 weeks, some participants were loss to follow up. This may be attributed to various factors including the fact that there were more participants in the non-diabetic arm. Some chose to go to a clinic nearer to their home for subsequent reviews, a few chose not to come back due loss of interest in participating in the study, while a few had relocated. Disappointingly, some samples were unfit for lab processing due to spillage during the transportation process. A more robust retention plan would have improved the retention rate. For the questions that the study set out to answer, this loss to follow up did not bias the study as there were very few individuals with the exposure of interest in the first place.

The strengths of this study included use of clinically validated tests for measuring blood sugar and was carried out at a tertiary hospital.

5.2 Study Limitations

The study was observational with limited follow-up, therefore difficult to establish a true causal relationship between the predictors and outcomes. In reality, numbers of DM patients with TB were considerably fewer than expected based on studies in other parts of the world. To meet the study objectives, it would take much longer than anticipated. Some patients may have had stress-induced hyperglycaemia rather than DM, so repeat testing after TB treatment would be necessary to assess for this phenomena. In addition, the study did not access for levels of control of DM. The results of this study may not be applicable to the rest of the population in Zambia as the participants were all seen at only one centre. Two months follow up is a surrogate endpoint by completion of TB treatment thus some patients would have converted to sputum negativity if they were followed up to standard 6 months. This study did not have a measure of adherence to ATT which greatly impacts treatment outcome.

CHAPTER 6

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusion

Overall, the prevalence of DM among individuals with smear positive TB was similar to that of the general population in Zambia. This is by and large less than expected. In order to determine treatment outcomes for individuals co-afflicted with TB and DM, studies need to be done in settings with a higher prevalence of DM and therefore more TB-DM comorbidity. Findings in this study suggest that at present it would not be worthwhile to screen all TB patients for DM in our setting but must be done on a case by case basis. However, there is still need to actively assess for possible TB among patients with DM. As the incidence of DM is expected to keep rising in low and middle income countries, there will still be need to duplicate similar studies in future.

6.2 Recommendations

6.2.1 University Teaching Hospital

Create deliberate policy to screen patients with DM (particularly those with poor control) for TB and provide the necessary laboratory support for detection in those that meet criteria for sputum examination.

6.2.2 Primary Health Care

Ministry of Health to make deliberate efforts to raise awareness in the community through talks, public health messages and TV programmes on the increased risk of having TB if one has DM.

6.2.3 Recommendations for Future Research

Further research is needed to characterise the pattern of disease in TB-DM comorbid affected individuals in Zambia and the rest of Africa. As Africa develops, deliberate efforts must be made by the Ministry of Health and other stakeholders must facilitate active surveillance of non-communicable diseases like DM among individuals who present with communicable diseases like TB and vice versa as overlap of these is bound to increase in the future. Thus through such implementations, a database can be

created which can be used as basis for further research. However before such an undertaking, a cost-benefit analysis will need to be done.

REFERENCES

1. Kondratieva T, Azhikina T, Nikonenko B, Kaprelyants A, Apt A. Latent tuberculosis infection: what we know about its genetic control? Tuberculosis (Edinb). 2014;94(5):462-8.
2. Kumar P CM. Kumar And Clarks Clinical Medicine. In: Kumar P CM, editor. Diabetes Mellitus and other endocrine disorders. 8 ed. United Kingdom: Saunders (Elsevier); 2012. p. 1142-45.
3. Mathers CD, Loncar D. Projections of Global Mortality and Burden of Disease from 2002 to 2030. PLOS Medicine. 2006;3(11):e442.
4. Wild S, Roglic G, Green A, Sicree R, King H. Global prevalence of diabetes: estimates for the year 2000 and projections for 2030. Diabetes care. 2004;27(5):1047-53.
5. Meetoo D, McGovern P, Safadi R. An epidemiological overview of diabetes across the world. Br J Nurs. 2007;16(16):1002-7.
6. Miranda JJ, Kinra S, Casas JP, Davey Smith G, Ebrahim S. Non-communicable diseases in low- and middle-income countries: context, determinants and health policy. Trop Med Int Health. 2008;13(10):1225-34.
7. Mbanya JC, Motala AA, Sobngwi E, Assah FK, Enoru ST. Diabetes in sub-Saharan Africa. Lancet. 2010;375(9733):2254-66.
8. Hall V, Thomsen RW, Henriksen O, Lohse N. Diabetes in Sub Saharan Africa 1999-2011: epidemiology and public health implications. A systematic review. BMC Public Health. 2011;11:564.
9. Whiting DR, Guariguata L, Weil C, Shaw J. IDF diabetes atlas: global estimates of the prevalence of diabetes for 2011 and 2030. Diabetes Res Clin Pract. 2011;94(3):311-21.
10. Bailey SL, Ayles H, Beyers N, Godfrey-Faussett P, Muyoyeta M, du Toit E, et al. Diabetes mellitus in Zambia and the Western Cape province of South Africa: Prevalence, risk factors, diagnosis and management. Diabetes Research and Clinical Practice. 2016;118:1-11.
11. Kelly. E. Dooley REC. Tuberculosis and diabetes mellitus: convergence of two epidemics. Lancet Infect Dis. 2009;9(12):737-74.

12. Dooley KE, Chaisson RE. Tuberculosis and diabetes mellitus: convergence of two epidemics. Lancet Infect Dis. 2009;9(12):737-46.
13. Stevenson CR, Forouhi NG, Roglic G, Williams BG, Lauer JA, Dye C, et al. Diabetes and tuberculosis: the impact of the diabetes epidemic on tuberculosis incidence. BMC Public Health. 2007;7:234.
14. Jeon CY, Murray MB. Diabetes mellitus increases the risk of active tuberculosis: a systematic review of 13 observational studies. PLoS Med. 2008;5(7):e152.
15. Jeon CY, Harries AD, Baker MA, Hart JE, Kapur A, Lonnroth K, et al. Bi-directional screening for tuberculosis and diabetes: a systematic review. Trop Med Int Health. 2010;15(11):1300-14.
16. Morsy AM ZH, Hassan MH, Shouman A. Predictors of treatment failure among tuberculosis patients under DOTS strategy in Egypt. East Mediterr Health J. 2003;9:689-701.
17. Mboussa J MH, Kombo M, Yokolo D, Yoka-Mbio A, Yala F. Course of pulmonary tuberculosis in diabetics. Rev Pneumol Clin. 2003;59:39-44.
18. Alisjahbana B, Sahiratmadja E, Nelwan EJ, Purwa AM, Ahmad Y, Ottenhoff TH, et al. The effect of type 2 diabetes mellitus on the presentation and treatment response of pulmonary tuberculosis. Clin Infect Dis. 2007;45(4):428-35.
19. Leung CC, Lam TH, Chan WM, Yew WW, Ho KS, Leung GM, et al. Diabetic control and risk of tuberculosis: a cohort study. American journal of epidemiology. 2008;167(12):1486-94.
20. King H, Aubert RE, Herman WH. Global burden of diabetes, 1995-2025: prevalence, numerical estimates, and projections. Diabetes care. 1998;21(9):1414-31.
21. Bates M, O'Grady J, Mwaba P, Chilukutu L, Mzyece J, Cheelo B, et al. Evaluation of the burden of unsuspected pulmonary tuberculosis and co-morbidity with non-communicable diseases in sputum producing adult inpatients. PloS one. 2012;7(7):e40774.
22. Edwards D, Kirkpatrick CH. The immunology of mycobacterial diseases. The American review of respiratory disease. 1986;134(5):1062-71.

23. Dunlap NE, Briles DE. Immunology of tuberculosis. The Medical clinics of North America. 1993;77(6):1235-51.
24. Olmos P, Donoso J, Rojas N, Landeros P, Schurmann R, Retamal G, et al. [Tuberculosis and diabetes mellitus: a longitudinal-retrospective study in a teaching hospital]. Revista medica de Chile. 1989;117(9):979-83.
25. Wang CS, Yang CJ, Chen HC, Chuang SH, Chong IW, Hwang JJ, et al. Impact of type 2 diabetes on manifestations and treatment outcome of pulmonary tuberculosis. Epidemiol Infect. 2009;137(2):203-10.
26. Restrepo BI F-HS, Smith B, et al. Mycobacterial clearance from sputum is delayed during the first phase of treatment in patients with diabetes. Am J Trop Med Hyg. 2008;79:541-44.
27. Shaw JE, Sicree RA, Zimmet PZ. Global estimates of the prevalence of diabetes for 2010 and 2030. Diabetes Res Clin Pract. 2010;87(1):4-14.
28. Nsakashalo-Senkwe M, Siziya S, Goma FM, Songolo P, Mukonka V, Babaniyi O. Combined prevalence of impaired glucose level or diabetes and its correlates in Lusaka urban district, Zambia: a population based survey. Int Arch Med. 2011;4(1):2.
29. Deshpande AD, Harris-Hayes M, Schootman M. Epidemiology of Diabetes and Diabetes-Related Complications. Physical Therapy. 2008;88(11):1254-64.
30. Root H. The association of diabetes and tuberculosis. New Engl J Med. 1934;210:1-13.
31. Dalal S BJ, Volmink J, Adebamowo C, Bajunirwe F, et al. Non-communicable diseases in sub-Saharan Africa: what we know now. Int J Epidemiol. 2011;40:885-901.
32. Prasad HB, Borse RT, Avate AN, Palasdeokar A. Prognostic Indicators of Response to Plasmapheresis in Patients of Guillain Barre Syndrome. The Journal of the Association of Physicians of India. 2017;65(4):32-6.
33. Harries AD, Murray MB, Jeon CY, Ottmani SE, Lonnroth K, Barreto ML, et al. Defining the research agenda to reduce the joint burden of disease from diabetes mellitus and tuberculosis. Trop Med Int Health. 2010;15(6):659-63.
34. Baker MA, Harries AD, Jeon CY, Hart JE, Kapur A, Lonnroth K, et al. The impact of diabetes on tuberculosis treatment outcomes: a systematic review. BMC medicine. 2011;9:81.

35. Harries A. Some clinical aspects of HIV infection in Africa. Afr Health. 1992;14(5):10-1.
36. Guler M, Unsal E, Dursun B, Aydln O, Capan N. Factors influencing sputum smear and culture conversion time among patients with new case pulmonary tuberculosis. Int J Clin Pract. 2007;61(2):231-5.
37. Mugusi F, Swai AB, Alberti KG, McLarty DG. Increased prevalence of diabetes mellitus in patients with pulmonary tuberculosis in Tanzania. Tubercle. 1990;71(4):271-6.
38. Alisjahbana B, van Crevel R, Sahiratmadja E, den Heijer M, Maya A, Istriana E, et al. Diabetes mellitus is strongly associated with tuberculosis in Indonesia. Int J Tuberc Lung Dis. 2006;10(6):696-700.
39. Ikezoe J, Takeuchi N, Johkoh T, Kohno N, Tomiyama N, Kozuka T, et al. CT appearance of pulmonary tuberculosis in diabetic and immunocompromised patients: comparison with patients who had no underlying disease. AJR American journal of roentgenology. 1992;159(6):1175-9.
40. Umut S, Tosun GA, Yildirim N. Radiographic location of pulmonary tuberculosis in diabetic patients. Chest. 1994;106(1):326.
41. Perez-Guzman C, Torres-Cruz A, Villarreal-Velarde H, Vargas MH. Progressive age-related changes in pulmonary tuberculosis images and the effect of diabetes. American journal of respiratory and critical care medicine. 2000;162(5):1738-40.
42. Perez-Guzman C, Torres-Cruz A, Villarreal-Velarde H, Salazar-Lezama MA, Vargas MH. Atypical radiological images of pulmonary tuberculosis in 192 diabetic patients: a comparative study. Int J Tuberc Lung Dis. 2001;5(5):455-61.
43. Morris JT, Seaworth BJ, McAllister CK. Pulmonary tuberculosis in diabetics. Chest. 1992;102(2):539-41.
44. Bacakoglu F, Basoglu OK, Cok G, Sayiner A, Ates M. Pulmonary tuberculosis in patients with diabetes mellitus. Respiration; international review of thoracic diseases. 2001;68(6):595-600.
45. Dooley KE, Tang T, Golub JE, Dorman SE, Cronin W. Impact of diabetes mellitus on treatment outcomes of patients with active tuberculosis. Am J Trop Med Hyg. 2009;80(4):634-9.

46. Singla R, Khan N, Al-Sharif N, Ai-Sayegh MO, Shaikh MA, Osman MM. Influence of diabetes on manifestations and treatment outcome of pulmonary TB patients. Int J Tuberc Lung Dis. 2006;10(1):74-9.
47. Banu Rekha VV, Balasubramanian R, Swaminathan S, Ramachandran R, Rahman F, Sundaram V, et al. Sputum conversion at the end of intensive phase of Category-1 regimen in the treatment of pulmonary tuberculosis patients with diabetes mellitus or HIV infection: An analysis of risk factors. Indian J Med Res. 2007;126(5):452-8.
48. Hendy M, Stableforth D. The effect of established diabetes mellitus on the presentation of infiltrative pulmonary tuberculosis in the immigrant Asian community of an inner city area of the United Kingdom. British journal of diseases of the chest. 1983;77(1):87-90.
49. Nijland HM, Ruslami R, Stalenhoef JE, Nelwan EJ, Alisjahbana B, Nelwan RH, et al. Exposure to rifampicin is strongly reduced in patients with tuberculosis and type 2 diabetes. Clin Infect Dis. 2006;43(7):848-54.
50. Groop LC, Luzi L, DeFronzo RA, Melander A. Hyperglycaemia and absorption of sulphonylurea drugs. Lancet. 1989;2(8655):129-30.
51. Angervall L, Dotevall G, Lehmann KE. The gastric mucosa in diabetes mellitus. A functional and histopathological study. Acta medica Scandinavica. 1961;169:339-49.
52. Kenny MT, Strates B. Metabolism and pharmacokinetics of the antibiotic rifampin. Drug metabolism reviews. 1981;12(1):159-218.
53. Ruslami R, Nijland HMJ, Adhiarta IGN, Kariadi SHKS, Alisjahbana B, Aarnoutse RE, et al. Pharmacokinetics of Antituberculosis Drugs in Pulmonary Tuberculosis Patients with Type 2 Diabetes. Antimicrobial Agents and Chemotherapy. 2010;54(3):1068-74.
54. Kirkman MS, Briscoe VJ, Clark N, Florez H, Haas LB, Halter JB, et al. Diabetes in Older Adults. Diabetes Care. 2012;35(12):2650-64.
55. Byng-Maddick R, Noursadeghi M. Does tuberculosis threaten our ageing populations? BMC Infectious Diseases. 2016;16(1):119.
56. Neyrolles O, Quintana-Murci L. Sexual Inequality in Tuberculosis. PLOS Medicine. 2009;6(12):e1000199.

57. Kalra S, Kalra B, Agrawal N, Unnikrishnan A. Understanding diabetes in patients with HIV/AIDS. Diabetology & Metabolic Syndrome. 2011;3(1):2.
58. (WHO) WHO. Diabetes country profiles, 2016, Zambia. WHO: WHO; 2016 2016.
59. Saydah S, Lochner K. Socioeconomic Status and Risk of Diabetes-Related Mortality in the U.S. Public Health Reports. 2010;125(3):377-88.
60. Sacerdote C, Ricceri F, Rolandsson O, Baldi I, Chirlaque M-D, Feskens E, et al. Lower educational level is a predictor of incident type 2 diabetes in European countries: The EPIC-InterAct study. International Journal of Epidemiology. 2012;41(4):1162-73.
61. Agardh EE, Sidorchuk A, Hallqvist J, Ljung R, Peterson S, Moradi T, et al. Burden of type 2 diabetes attributed to lower educational levels in Sweden. Population Health Metrics. 2011;9:60-.
62. Hilawe EH, Yatsuya H, Kawaguchi L, Aoyama A. Differences by sex in the prevalence of diabetes mellitus, impaired fasting glycaemia and impaired glucose tolerance in sub-Saharan Africa: a systematic review and meta-analysis. Bulletin of the World Health Organization. 2013;91(9):671-82D.
63. Campos LC, Rocha MVV, Willers DMC, Silva DR. Characteristics of Patients with Smear-Negative Pulmonary Tuberculosis (TB) in a Region with High TB and HIV Prevalence. PloS one. 2016;11(1):e0147933.

APPENDICES

Appendix I: World Health Organization definition of a tuberculosis case

Taken from: Treatment of tuberculosis: guidelines – 4th edition.
WHO/HTM/TB/2009.420. P23-25.
http://whqlibdoc.who.int/publications/2010/9789241547833_eng.pdf
The TB case definitions below are based on the level of certainty of the diagnosis and on whether or not laboratory confirmation is available.

Definition of definite case of tuberculosis: A patient with *Mycobacterium tuberculosis* complex identified from a clinical specimen, either by culture or by a newer method such as molecular line probe assay. In countries that lack the laboratory capacity to routinely identify *M. tuberculosis*, a pulmonary case with one or more initial sputum smear examinations positive for acid-fast bacilli (AFB) is also considered to be a "definite" case, provided that there is a functional external quality assurance (EQA) system with blind rechecking.

Pulmonary tuberculosis (PTB) refers to a case of TB (defined above) involving the lung parenchyma. Miliary TB is classified as pulmonary TB because there are lesions in the lungs. Tuberculosis intrathoracic lymphadenopathy (mediastinal and/or hilar) or tuberculosis pleural effusion, without radiographic abnormalities in the lungs, constitutes a case of extra-pulmonary TB. A patient with both pulmonary and extra-pulmonary TB should be classified as a case of *pulmonary* TB.

Appendix II: World Health Organization criteria for DM diagnosis

Taken from: Definition and diagnosis of DM and intermediate hyperglycaemia. Report of a WHO/IDF consultation. WHO Document Production Services, Geneva, Switzerland. 2006. P3

Recommendation for diagnosis of DM: Currently HbA_{1c} is not considered a suitable diagnostic test for diabetes or intermediate hyperglycaemia. The following Table summarises the 2006 WHO recommendations for the diagnostic criteria for diabetes and intermediate hyperglycaemia.

Diabetes	
Fasting plasma glucose	≥7.0mmol/l (126mg/dl)
OR	
2–h plasma glucose*	≥11.1mmol/l (200mg/dl)

Impaired Glucose Tolerance (IGT)	
Fasting plasma glucose	<7.0mmol/l (126mg/dl)
AND	
2–h plasma glucose*	≥7.8 and <11.1mmol/l (140mg/dl and 200mg/dl)

Impaired Fasting Glucose (IFG)	
Fasting plasma glucose	6.1 to 6.9mmol/l (110mg/dl to 125mg/dl)
AND (if measured)	
2–h plasma glucose*	<7.8mmol/l (140mg/dl)

** Venous plasma glucose 2–h after ingestion of 75g oral glucose load*

** If 2–h plasma glucose is not measured, status is uncertain as diabetes or IGT cannot be excluded*

Appendix III: Participant Information Sheet

INFORMATION FOR POTENTIAL PARTICIPANTS OF A STUDY TO COMPARE TREATMENT OUTCOMES IN TUBERCULOSIS PATIENTS WITH AND WITHOUT CONCURRENT DM AT THE UNIVERSITY TEACHING HOSPITAL, LUSAKA

You are invited to take part in a study to improve our understanding of how diabetes (sugar disease) affects tuberculosis and its outcome in Zambia. Individuals who are diagnosed with smear-positive tuberculosis at UTH will take part in this study. However, out of these patients, only 114 (85 of which are non-diabetic, 29 diabetic) will be considered for follow-up. Information on the study is supplied in this leaflet. A trained research assistant will be on hand to explain and answer all your questions. Please check that you understand everything in this document. If you decide to take part, you will be asked to give written consent before you take part.

Who is doing the study?

This study is being done by Dr Fwoloshi, a Master of Medicine student from the School of Medicine, Department of Internal Medicine at the University Teaching Hospital here in Lusaka. The results to this research will contribute to the thesis for my Master of Medicine degree and also add to knowledge about the care of TB patients with DM. Dr Fwoloshi and research assistants will be responsible for the day to day running of the study. Contact details are listed below:

Principal Investigator	**Supervisors**	**Research Assistants**
Dr Fwoloshi 0977319994	Dr Hachaambwa 976435603 Dr Chiyenu 0955998207 Dr Bailey 0969276263	Name:…….. …… ……

If you have any questions about the study, feel free to contact:

The Chairperson
ERES CONVERGE IRB
33 Joseph Mwilwa Road, Rhodes Park
Lusaka
Phone: 0955155633, 0955155634, 0966765503
Email:eresconverge@yahoo.co.uk

What is the purpose of the study?

The main aim of this study is to improve our understanding of how diabetes affects tuberculosis (TB) and its control in Zambia. We know from other studies that have been done in other countries around the world, that people who have diabetes are more likely to develop tuberculosis than people who don't have diabetes. We also know that once they have tuberculosis, the treatment for tuberculosis doesn't work as well as it does in people who don't have diabetes. But Zambia and other similar countries in Africa may be different to elsewhere in the world, and we want to know how diabetes affects tuberculosis here in Zambia. This will in turn help to improve the way diabetes and tuberculosis is dealt with in our country.

Confidentiality

Data for this study will be kept confidential under lock and key. Only the researcher and the supervisors will have access to the information. In the event of publication of the research, no personally identifying information will be disclosed.

Participants Rights

By consenting to participate in this study, you do not waiver any of your legal rights. Giving consent means you have read or heard the information about the study and you agree to participate. You will not suffer any penalty or lose any benefits to which you are entitled by participating in the study

Right to refuse or withdraw

Participation in this study is voluntary; you can choose to either be part of the study or not to be part of the study. If you feel uncomfortable with any of the questions in the study you have the right to refuse to take part in the study .If you decide to be in the study and then change your mind; you can withdraw from the study at any point. Your

decision to withdraw will not affect the standard of care that you will receive from the hospital.

Procedure (What taking part in the study will involve)

If you agree to take part in the general study to just check for blood sugar:

1. We will do a finger prick test to test for blood sugar
    - If you are 1 of the 114 to be enrolled into the main study, in addition to a finger prick test to check for blood sugar.
2. We will get information about you and the duration of your illness
3. We will measure the height and weight
4. A finger prick test to measure your blood sugar will be done
5. if your sugar is more than 7 mmol/l(above expected in someone who has not eaten for more than 8 hours) ,About 2 mls (half a teaspoon) of venous blood will be drawn to get an idea how your sugar has been in the past 3 months by measuring a sugar on your red blood cells called glycosylated haemoglobin $HbA_{1c,}$
6. You will also be required to come back 8 weeks 2months into your TB treatment,
    A. when About 4-6mls(2 tablespoons) of blood will be drawn to :
        I. check for CD4 count (how active your immune system is) if you are HIV positive and have no recent CD4 result
        II. store to test for a TB drug (rifampicin) levels in future ,
    B. You will also be required to submit sputum to check for tuberculosis

Potential risks and discomfort

- The procedures mentioned above carry minima (very little) risks:
- Blood collection and needle pricks may cause pain at the collection site
- Blood collection and needle pricks will be done by the doctor or trained research assistant
- Infection at the site of collection rarely occurs (<1%), to prevent such problems the doctor or trained research assistant will collect blood or perform needle pricks.
- Your blood samples and sputum collected will be kept in a sample bank with only a bar code on (not your name). These samples can be used in future for further tests on diabetes or TB.

Appendix IV: Participant Consent Form

Participant No.:

INFORMED CONSENT FORM FOR STUDY TO COMPARE TREATMENT OUTCOMES IN TUBERCULOSIS PATIENTS WITH AND WITHOUT CONCURRENT DM AT THE UNIVERSITY TEACHING HOSPITAL, LUSAKA (PERSONS AGED 18 YEARS AND OLDER)

1. I confirm that I have read the information sheet, and that the information and procedures involved in my taking part in this study have been explained to me.
2. I confirm that I have had the opportunity to ask questions about the study and that I am satisfied with the answers provided.
3. I have been given time and opportunity to read the information carefully, to discuss it with others and to decide whether or not to take part in this study.
4. I understand that the results of the blood sugar test will be given to me and that I will be referred to the clinic for appropriate care if my blood sugar is too high.
5. I understand that if the blood sugar test is positive, the TB clinic will be informed.
6. I understand that my blood specimens will be kept in a sample bank with only a bar code on (not my name) and that these samples can be used in future for further diabetes tests.
7. I understand that my sputum specimens will be kept in a sample bank with only a bar code on (not my name) and that these samples can be used in future for further TB tests.
8. I understand that the researchers will keep all my personal information confidential.
9. I understand that I will not get any financial reward for taking part in this study.
10. I understand that the results of this study will be published in scientific journals but that my name will not be used.
11. I understand that I may in future be requested to participate in follow-up studies.
12. I agree to take part in the study.

Participant's signature/thumbprint: _____ Date _____

Participant's name: _____ (please print)

The person who obtains the informed consent discussion must also sign and date this form.

Signature: _____Date _____

Name: _____
(please print)

(If participant is unable to consent, next-of- kin can consent)

Name of next of kin:
_____

Signature: _____

Or Thumb print:_____

Date: _____

Signature of witness (if applicable)

Signature of witness: _____Date_____

Witnessed by (print name): _____

Appendix V: Diabetes Fact Sheet

What is diabetes?

Diabetes is a disease commonly known as sugar disease. A person suffering from diabetes has too much blood sugar.

What causes Diabetes?

There are two major types of diabetes:

Type 1: In type 1 (also called juvenile-onset or insulin-dependent) diabetes, the body completely stops producing any insulin, a substance in the blood which the body needs to use sugar. People with type 1 diabetes must take daily insulin injections to survive. This form of diabetes usually develops in children or young adults, but can occur at any age.

Type 2: Type 2 (also called adult-onset or non-insulin-dependent) diabetes results when the body doesn't produce enough insulin and/or is unable to use insulin properly (insulin resistance). This form of diabetes usually occurs in people who are over 40, overweight, and have a family history of diabetes, although it is increasingly occurring in younger people, particularly adolescents.

How do people know if they have diabetes?

People with diabetes frequently experience certain symptoms. These include:

- being very thirsty and the mouth feels dry
- frequent urination which is more than normal
- weight loss
- increased hunger
- inability to see clearly
- itching around the genitals
- tingling or numbness in the hands or feet
- frequent skin, bladder or gum infections
- wounds that don't heal
- extreme unexplained fatigue

In some cases, there are no symptoms — this happens at times with type 2 diabetes. In this case, people can live for months, even years without knowing they have the disease. This form of diabetes comes on so gradually that symptoms may not even be recognized.

Appendix VI: Initial Data Sheet for Interviewers

Participant No.:

Initial Data Form Investigators (2)

After consent, document the following

1. SEX

2. AGE

M	F
1	2

3. Blood Glucose. Write actual Results below

		.	

4. When did you last have anything to eat or drink (except water)? Write Number of hours ago

5. Patient location:

inpatient	outpatient
1.	2.

6. If available; HIV status.....

Negative	0
Positive	1

7. Last CD4 count and date

CD4	
Date	

If to be enrolled in main study: **RECORD CONTACT DETAILS** and proceed to questionnaire. If not, **STOP HERE**

Appendix VII: Participant Questionnaire

SECTION 1

ALL QUESTIONS IN THIS SECTION MUST BE ANSWERED

Q01_INC **Interviewer's code**

Q02_DAT **Date today**

D	D	M	M	Y	Y	Y	Y

Q03_SEN **Participant Number**

Q04_SEX **Sex**

M	F
1	2

Q05_AGE **Age**

Q06_DIS **Disability?**

No Disability	1
Sight(blind/ severe visual impairment)	2
Hearing (deaf/ profoundly hard of hearing)	3
Communication(speech impairment)	4
Physical(needs wheelchair/ crutches)	5
Mental disability	6

Q07_CON **Consent**

No	Yes	Absent	Excluded
0	1	2	3

ONLY CONTINUE IF CONSENT IS GIVEN

SECTION 2 – FILL THIS AND SUBSEQUENT SECTIONS IN ONLY IF PERSON HAS GIVEN CONSENT

Q8_DOB **Date of Birth** (01/01/1800 if unknown) **If not known, what was your age in Years at your last birthday?** (999 if unknown)

D	D	M	M	Y	Y	Y	Y

Q9_CMS What is your current marital Status?

Never married	1
Currently married or living as married	2
Divorced or Separated	3
Widowed	4

Q10_MOY What has been your main occupation during the past year?

Unemployed/working on own land	1
Occasional/seasonal employment	2
Employed (Formal employment or self-employed making money)	3
Unable to work	4
Student	5
Housewife/ home-maker	6

I would like to ask you about your current drinking and smoking habits

Q11_CSH How would you classify your smoking habits?

Have never smoked	1
Daily smoker	2
Occasional smoker	3
Ex-smoker	4

Q12_CDH How would you classify your drinking habits?

Have never drunk	1
Daily drinker	2
Occasional drinker	3
Ex-drinker	4

Now I will ask questions about your education

Q13_HEA What is the highest level of education you have attained?

No Formal Education	0
Grade 1-12(Indicate actual grade)Note Grade 8-12 is also Form 1 –form 5	
College	20
University	30

I would like to ask you about your health. (Current TB questions)

Q14_DTS **Date TB treatment started**

D	D	M	M	Y	Y	Y	Y

Q15_TTN **TB treatment Number**

Q16_CAT **Category of TB?**

Category 1	1
Category 2	2
Extra-pulmonary	3
Unknown/not recorded	4

Questions about previous TB treatment

Previous TB treatment

Q17_TTB Have you ever been on TB treatment before?
If yes continue, if no go to Q37

No	Yes	Unk
0	1	9

Q18_HMT How many times?

Once	1
Twice	2
Three times	3
More than three times	4
Unknown	9

Now I will ask questions about Diabetes and HIV

Q19_THD Have you ever been told you have diabetes
If Yes continue, if No go to Q55

No	Yes
0	1

Q20_CAT If yes, are you currently on any treatment for diabetes?
If yes continue If no go to Q55

No	Yes
0	1

Q21_TON What treatment are you on?

Dietary only	1
Tablets	2
Insulin injections	3

Q22_KHS Do you know your HIV status?

No	Yes
0	1

Q23_DHS Are you willing to disclose your HIV status?
If yes continue, if not willing to discuss and male go to Q60.

No	Yes
0	1

Q24_HIV What was the result?

Negative	0
Positive	1

If HIV status is Positive, continue, if negative go to Q60

Q25_ART Are you on Antiretroviral treatment(ART)
If yes continue, if no go to Q60

No	Yes
0	1

Q26_LAR How long have you been on ART? Write down actual number of months

SECTION 4

RECORD BODY HABITUS MEASUREMENTS

Q27_WEI Weight? Record weight in Kilograms (one decimal point)
If not done, write 999.9 | | | | . | | Kg

Q28_HEI Height? Record weight in centimetres
If not done, write 999.9 | | | | cm

Q29_ABC Abdominal Circumference? Record in centimetres
If not done, write 999.9 | | | | cm

SECTION 5

RECORD BLOOD SUGAR AND HIV RESULTS HERE

Q30_BLG Blood Glucose. Write actual Results below

		.	

Q31_LAG When did you last have anything to eat or drink (except water)? Write Number of hours ago

Q32_HIV_DET HIV Test result (Determine).

Negative	0
Positive	1
Not Done	9

Q33_HIV_UNI Confirmatory HIV Test result (Unigold)

Negative	0
Positive	1
Not Done	9

Q34_KHS Does study participant want to know his/her HIV Result? No Yes

0	1

Q35_GHB HIV test results given to study participant?

No	Yes
0	1

THANK YOU FOR YOUR HELP

Appendix VIII: Daily Task List for Investigators

Initial procedures for study participants

1. Identify newly diagnosed tuberculosis patients at UTH chest clinic and in patients from TB lab through consultation with lab staff and records
2. Obtain individual written informed consent from those meeting inclusion criteria and willing to participate
3. Administer structured questionnaire
4. Measure fasting blood glucose concentration
5. Measure height, weight and waist circumference
6. Record HIV status. If no current HIV status available, perform an HIV test with pre- and post-test counselling
7. For HIV +ve participants, record CD4 count. If no current CD4 count available, measure CD4 count at 8 weeks

Initial procedures for individuals with ≥7.0mmol/L

8. Take a venous blood sample for measurement of HbA_{1c} concentration
9. Inform participants of procedures for fasting blood glucose measurement

For study participants returning 8 weeks post-enrolment

10. Inform participants not to take that days dose of ATT, the dose will be taken on the same day after collection of blood specimens
11. Collect sputum specimens

For study participants at 8 weeks post-enrolment

12. Collect sputum specimens
13. Take a second venous blood sample for measurement of CD4 If no current CD4 count available, measure CD4 count for HIV +ve participants

For all specimens collected during day

14. Transport to ZAMBART lab for processing or storage

Appendix IX: Blood Glucose Testing Standard Operating Procedures

Instructions for blood glucose testing using a glucometer

In this study blood glucose measurement for fasting blood glucose will be carried out using a glucometer through the collection of a capillary blood sample. The glucometer is a rapid method of determining the blood sugar level. This SOP outlines procedures for collection of a capillary blood sample, handling and discarding of the test strips, use of the glucometer and giving results back to the participants.

Materials for the test

- **Disposable gloves**
- **Sharps and biohazards disposable container**
- **Alcohol swabs**
- **Gauze/cotton pads**
- **Auto-lancets**
- **Glucose test strips**
- **Glucometer**
- **Batteries**

Performing the test

- Prepare a place where the test can be done. This should be a flat surface and there should be no clutter around.
- Wash your hands thoroughly. If water is not available, rub your hands with gel or with an alcohol swab.
- Use standard precautions – wear gloves.
- Confirm that the test strip is within the expiry date.
- Turn on the glucometer and place the test strip in the glucometer.
- Touch drop but not finger to test strip. Press the skin puncture site with dry cotton/gauze.
- Wait for the glucometer to read the test strip.
- Dispose of waste in biohazard waste container.
- Read the result given on the glucometer. Note whether the result falls outside the normal or reportable range.
- Record the result on the questionnaire (on the PDA) or separate sheet provided at the relevant position.

Quality control and maintenance policy

This is performed automatically by the glucometer. No additional measures are required.

Giving results to the study participant

- Explain the results as given on the glucometer.
- If the fasting blood glucose level is outside the normal range explain that the participant needs to return to see a clinic doctor. Fill in a referral letter with them and arrange an appointment with the doctor at the earliest opportunity.
- Give all study participants diabetes information sheets.

Interpreting blood glucose results

Abnormal glucose levels if:

- Fasting blood glucose is ≥7.0mmol/L

Any study participant with either an abnormal fasting blood glucose level should be referred to the doctor for further screening.

Appendix X: Diabetes Follow-up Letter

FOLLOW-UP – BLOOD SUGAR IS ABNORMAL

Name:.....

Date:.....

Your blood has been tested for the presence of sugar and the test results show that you have a sugar level of …….in your blood (this test was a fasting blood sugar test) .These tests suggest that you may have diabetes.

We have therefore booked an appointment for you to see a doctor .The doctor will explain more about diabetes to you and you will be able to ask the doctor any questions that you may have about diabetes. The doctor will assess your health and arrange for any treatment or further management that may be needed. In the meantime please read our diabetes fact sheet to find out more about diabetes and if you have any concerns please speak to a member of the study team.

Your appointment is on ..... (date) at ..... (time).

Appendix XI: Height, Weight and Abdominal Circumference Standard Operating Procedures

INSTRUCTIONS FOR WEIGHT, HEIGHT AND ABDOMINAL CIRCUMFERENCE RECORDING

Step 1: Eligibility

Height, Weight and Abdominal circumference should only be measured in individuals who are eligible. The groups of people that are not eligible to have their measurements done are:

- Chair bound or bedbound participants
- Individuals who are unsteady on their feet
- Individuals who find it painful to stand straight
- Pregnant women
- Individuals who cannot stand without support
- Individuals with very high hair styles

Step 2: Site for taking measurements

It is preferable that weight and height are measured on a floor which is level and not carpeted (hard flat surface).

Step 3: Height Measurement

Equipment: Stadiometer- This is a portable collapsible device with a sliding head plate, a base plate and three contacting rods marked with a measuring scale.

The Protocol

1. Ask the participant to remove his/her shoes in order to obtain a measurement that is as accurate as possible.
2. Assemble the stadiometer and raise the head plate to allow sufficient room for the participant to stand underneath it. Double check that you have assembled the stadiometer correctly.
3. The participant should stand with his/her feet flat on the centre of the base plate, feet together and heels against the rod but not leaning on it. He/she

should have his/her arms hanging loosely by their sides. He/she should be facing forwards.

4. Move the participant's head so that the Frankfort Plane is in a horizontal position (i.e. parallel to the floor). The Frankfort Plane is an imaginary line passing through the external ear canal and across the top of the lower bone of the eye socket, immediately under the eye. This position is important if an accurate reading is to be obtained. An additional check is to ensure that the measuring arm rests on the crown of the head, i.e. the top back half. To make sure that the Frankfort Plane is horizontal, you can use the Frankfort Plane Card to line up the bottom of the eye socket with the flap of skin on the ear. The Frankfort Plane is horizontal when the card is parallel to the stadiometer arm.
5. Instruct the participant to keep his/her eyes focused on a point straight ahead, to breathe in deeply and to stretch to their fullest height. If after stretching up the participant's head is no longer horizontal, repeat the procedure. It can be difficult to determine whether the stadiometer head plate is resting on the participant's head. If so, ask the participant to tell you when she/he feels it touching their head.
6. Ask the participant to step forward. If the measurement has been done correctly the participant will be able to step off the stadiometer without ducking their head. Make sure that the head plate does not move when the participant does this.
7. Look at the bottom edge of the head plate cuff. There is a red arrowhead pointing to the measuring scale. Take the reading from this point and record the participant's height in centimetres and millimetres, at *Height* space in the Questionnaire (on the PDA).
8. Height must be recorded in centimetres and millimetres, e.g. 176.5 cms. If a measurement falls between two millimetres, it should be recorded to the nearest even millimetre. E.g. if a participant's height is between 176.4 and 176.5 cm, you should round it down to 176.4. Likewise, if the participant's height is between 176.5 and 176.6 cm, you should round it up to 176.6 cms.
9. Push the head plate high enough to avoid any member of the household hitting his/her head against it when getting ready to be measured.

Step 4: Weight Measurements

Equipment: Electronic bathroom scales

The protocol

1. Turn the display on by pressing firmly with your hand or foot on the top of the scales (the scales will turn themselves off after a short while). The readout should display 888.8 momentarily as a check for the operation. If this is not displayed check the batteries, if this is not the cause you may need to report the problem to the Principal Investigator. **While the scales read 888.8 do not attempt to weigh anyone.**
2. Ask the participant to remove shoes, heavy outer garments such as jackets and cardigans, heavy jewellery, loose change and keys.
3. Turn the scales on with your foot again. Wait for a display of 0.0 before the participant stands on the scales.
4. Ask the participant to stand with their feet together in the centre and their heels against the back edge of the scales. Arms should be hanging loosely at their sides and head facing forward. Ensure that they keep looking ahead - it may be tempting for the participant to look down at their weight reading. Ask them not to do this and assure them that you will tell them their weight afterwards if they want to know.

   The posture of the participant is important. If they stand to one side, look down, or do not otherwise have their weight evenly spread, it can affect the reading.
5. The scales will take a short while to stabilize and will read 'C' until they have done so. If the participant moves excessively while the scales are stabilizing you may get a false reading. If you think this is the case re-weigh the participant.
6. The scales have been calibrated in kilograms and 100 gram units (0.1 kg). Record the reading in the space provided for weight on the questionnaire.

WARNING: The maximum weight registering accurately on the scales is 130 kg (20 stone). If you think the participant exceeds this limit do not attempt to weigh them. Tick the box 'Weight exceeds scale maximum' on the Questionnaire.

Step 5: Abdominal circumference

Equipment: Insertion (SECA) measuring tape calibrated in mm, with a plastic buckle at the end.

The Protocol

1. Prepare the participant by asking him/her to;
    - Remove all outer clothing such as jackets, cardigans, waist coats, and heavy or baggy jumpers.
    - Remove shoes with high heels.
    - Remove tight garments that are intended to alter ones shape such as corsets or body suits.
    - Belts should be removed or loosened.
    - Pockets should be emptied.

If the study participant is not willing to remove bulky outer garments or tight garments and you are of the opinion that this will significantly affect the measurement, do not proceed with the measurement

2. Ask the participant to stand erect in a relaxed manner. The arms should be hanging loosely at their sides.
3. If possible, sit or kneel on a chair to the side of the participant.
4. Pass the tape around the body of the participant and insert the clip into the holder. Ensure that the tape is horizontal by peering around the participant.
5. Measure the abdominal circumference at the level of the navel (belly button)
6. Hold the buckle flat against the body and flatten the end of the tape to read the measurement from the outer edge of the buckle. Do not pull the tape towards you as this will lift away from the participant's body, affecting the measurement.
7. Record the measurement to the nearest centimetre on the questionnaire.

Appendix XII: Mycobacteria Culture Standard Operating Procedures

1. Introduction

This document deals with the processing of sputum samples for culture and smears

1.1. Endpoints

For each sputum specimen received by the laboratory the following should be prepared:

I. Sputum archive
II. Inoculation on two MGIT tubes
III. Capilia TB test

Note: All results (Mtb, NTM, contaminated) should be reported back to the PI

Safety Precautions

All sputum specimens should be treated with utmost care as they may be infectious. Universal safety precautions towards infectious materials should be followed. All processing must be done in a Bio-Safety cabinet.

This SOP is targeted at staff that are qualified to work in a TB culture laboratory

1.2. Abbreviations

AFB	Acid Fast Bacilli
AMGIT	Automated Mycobacteria Growth Indicator Tube
BA	Blood Agar
BSC	Bio-Safety Cabinet
MMGIT	Manual Mycobacteria Growth Indicator Tube
MOTT	Mycobacteria other than tuberculosis
Mtb	Mycobacteria tuberculosis
NTM	Non Tuberculosis Mycobacteria
RT	Room temperature
SOP	Standard Operating Procedure
ZAMBART	Zambia AIDS Related Tuberculosis

2. Specimen Handling

2.1. Collection

Specimens must be collected into 50ml falcon tubes with a tight fitted lid. They should be placed in a leak proof plastic bag for transportation to the laboratory. One spot samples should be collected from each patient.

2.2. Transportation

Specimens should be transported to the laboratory as soon as possible after collection. Delays in transportation may increase the risk of contamination of cultures. Specimens

should be transported in a cooler box with ice packs, in which temperature is maintained as low as possible.

Specimens must be packaged to withstand leakage of contents, shocks, pressure changes and other conditions incident to ordinary handling practices. Primary specimen container (containing actual specimen) must be placed first in simple plastic bag that is tied, and then placed in self-zipped bags which is then placed in secondary container.

2.3. Storage

Samples should be processed as soon possible after receipt. If samples are not processed on the same day of receipt, they should be refrigerated at 2-8^{o}C. **DO NOT FREEZE SPECIMENS**

All specimens must be processed within 4 days of submission.

3. Specimen Receipt and log in

Samples will be received at ZAMBART laboratory. All samples should arrive with a request form packed in the side pocket of the specimen transfer bag with a barcode stuck on the request form. The samples from one patient will be packed in one sample transfer bag.

3.1. Sample receipt

- Check for sample examination request form
- Remove from bag and check that barcode on request form matches that on samples
- Samples where barcodes are not matching: Field staff should be contacted for verification. Samples should be discarded where the mismatch cannot be verified or resolved
- Leaked out samples should be discarded if they cannot be salvaged
- Indicate number of samples received by ticking appropriately on the request form
- Sign request form to indicate that samples have been checked and received
- Place signed off request form in designated Box folder (pending registration)
- Place samples in appropriate drawer in the Fridge if they are not to be processed immediately
- Samples that have not been received and checked should be placed in the electric cool box until someone is available to receive them

Note: Samples that have not been received as described above should not be placed in the fridge. If samples cannot be appropriately received at the time of arrival, they should be placed in the electric cool box marked <u>UN RECEIVED</u> Samples.

3.2. Sample registration

- Remove sample examination request forms from folder

- Scan in barcode from request forms
- Enter date sample collected for each sample that is indicated on the request form
- Enter all samples to be processed on that day
- Once all samples are registered, print sample registration form
- Samples can now be moved from the fridge into BSC1
- After registration, request forms are placed in appropriately labelled box folder "Request forms, registered samples"

Note: Take care that only samples indicated as received are registered. Once all samples for processing on the day are registered, print sample registration form.

3.3. Unpacking samples from transfer bag

This should be done in BSC1

- Wipe BSC1 surface with appropriate disinfectant
- Place samples in BSC1 and carefully check samples for breakages or leakages before removing from bag
- Broken or leaked out samples should be recorded on the registration form and then discarded

Note: Samples that are not completely leaked out can be salvaged by transferring into another falcon tube

- Remove samples from the sample transfer bag and place on a rack
- Wipe down with disinfectant and place samples in centrifuge bucket
- Spin down samples for 1 minute
- Remove from centrifuge, wipe down with disinfectant
- Check and record volume and sample quality on the registration form
- Enter sample volume and quality into data base

3.4. Specimen rejection

- Sputum is leaked out completely and cannot be salvaged
- Sputum container is broken
- Sputum container is empty

3.5. Batch Creation

- Group samples into batches of maximum 26 (fewer if insufficient number of samples) with 2 control tubes included
- For each specimen place one blue top screw cap vial (sputum archive vial) and slide and two manual MGIT tubes on a rack
- Label each MGIT tube with an individual barcode and another unique barcode
- Label the sputum archive and slide with unique barcodes
- Place two extra MGIT tubes on the rack for negative and positive controls. These should have unique barcodes

- Paste a red/pink sticker on top of the positive control tube and a green sticker on the negative control tube.
- Write "P" on the positive control tube and an "N" on the negative control tube

- Take an empty MGIT box rack and paste a sticker and label with date of sample processing. All batches (samples) run on the same day use the same rack for incubation
- Create batch, enter batch number automatically
- Scan in individual barcode for sample followed by the MGIT tube barcodes, slide and sputum archive
- Scan in MGIT positive and negative control (every batch should have an accompanying positive and negative control tube)

Warnings!

- **From this point forward all techniques must be carried out in the biosafety lab(40, 43)**
- **No material may leave the lab unless it has been decontaminated or autoclaved**
- **Procedures that can cause the generation of an aerosol must be minimised and carried out in a bio-safety cabinet**
- **In order to minimize cross-contamination of cultures the following measures must be adhered to:**

♦ A single hood is dedicated to the isolation of mycobacteria from sputum. Fully-grown cultures (MGIT) are to be dealt with in a separate hood or later in the day after sample processing is complete. If a second hood is not available an interval of at least 30 minutes should be allowed between processing sputum samples and fully-grown cultures and handling of cultures should always be done after sputum samples have been processed and before to reduce cross contamination

♦ After shaking or spinning, aerosols are to be allowed to settle for 5 minutes.

4. Adding PANTA and OADC to each MGIT tube

PANTA and OADC are mixed together before it is added to the MGIT tube. A mixture of one vial of PANTA (3ml) with one bottle of OADC (15ml) is enough for 30 MGIT tubes.

Note: All reagents should be at room temperature before use. PANTA/OADC mixture should be added to MGIT tubes just before adding samples

- Add the entire 15ml of OADC into the vial containing PANTA
- Mix by carefully shaking
- Put a sterile 10 ml syringe onto the multi-pipette
- Add 0.5ml of the PANTA/OADC mixture into each MGIT tube
- Do not store MGIT tubes after addition of PANTA/OADC mix

Note: MGIT tubes to which PANTA/OADC has been added should be used within 30 minutes of adding PANTA/OADC

Left over PANTA/OADC should be used within 5 days of opening but should be kept at 4°C. Label the PANTA/OADC mixture with the date mixed

6. Preparing positive and negative control

1. Label one empty falcon tube with and "N" for negative control and "P" for positive control
2. Add 3 ml of saline[1] to the negative control container, and add 2 ml of saline to the positive control container
3. IN BSC2 add 1000ul of the vial with the positive control culture H37Rv to the positive control container
4. Nothing is added to the negative control container

NOTE: positive control samples are always placed at the end of the sample row and will only be opened if handling of the other clinical samples and negative control is done for the particular step of the protocol

7. Specimen Decontamination

- Processing should be done in batches. The Batch size is a maximum of 26 specimens plus 2 control tubes.
- For actual decontamination, the batch is split into 2 if the maximum number is reached so that only a maximum of 13 samples is processed for maximum batch size.

7.1. Materials and Methods

- Disposable 50ml plastic tubes(falcon tubes)
- Sterile NaOH-NALC-sodium citrate solution-Mycoprep
- Phosphate buffer pH 6.8 (aliquoted into McCartney bottles)
- Graduated pasture pipettes
- Centrifuge
- Vortex mixer
- Timer
- Gloves
- Waste bags
- Media(2 MGIT tubes)
- Normal saline

7.2. Prepare fresh digestant

1. Place one flask of the MycoPrep into BSC1
2. Open the lid of the flask to squeeze the air out
3. Close the lid
4. Break the glass vial inside the flask
5. Open the lid again to bring the air back into the flask
6. Ensure that all powder is completely dissolved

[1] See Appendix…. For preparation of Normal saline

7. Add 4ml of 10M NaOH to the flask to create a final concentration of 3% NaOH

Note: Label the mycoprep bottle with date and time activated. Keep refrigerated and use within 24 hours of activation.

7.3. Decontamination procedure

1. Place a batch of sputum samples into BSC 1
2. Place Negative and positive control tubes at the end of the rack
3. Add the MycoPrep solution in equal volume to that of the specimen using a sterile pipette. Tighten the cap
4. Start timer when you have added Mycoprep to the first sample
5. Put samples in orbital shaker and vortex the tube for 15-30 seconds and invert the tube to ensure that the mycoprep solution contacts all surfaces of the tube
6. Wait 20 minutes after adding the NaOH-NALC solution. Vortex lightly every 5-10 minutes or put tubes on a shaker and shake lightly during whole decontamination time
8. Make sure specimen is completely liquefied. If specimen is still mucoid, add a small quantity NALC-powder (30-35 grams) directly to the specimen tube, mix well
9. At the end of 20 minutes, add phosphate buffer (pH 6.8) up to the top ring 50ml mark on the centrifuge tube. Tighten tube. Mix well by light vortexing or inverting several times
10. Place tubes in centrifuge and close lid tightly. Centrifuge at 3800 X g for 20 minutes at 4-16oc Let the tube stand for 5 minutes after centrifuging to allow aerosol to settle but do not leave to stand for too long after centrifuge. If refrigerated centrifuge is not available use, use cold (2-6^{o}c buffer)
11. Remove tubes from centrifuge
12. Decant supernatant from tubes into bin containing mycobactericidal disinfectant, leaving only sample pellet in the tube
13. ***NB at this stage it is especially critical that only one specimen be open at a time and generation of aerosols be minimised to avoid cross-contamination of samples***
14. Re-suspend the pellet sediment with 2mls of phosphate buffer pH 6.8 using a sterile Pasteur pipette to achieve a final volume of 1- 3mls. Vortex the suspended sample
15. Wipe down the container containing the pellet, move the rack to BSC2
16. Spray the centrifuge buckets with 1% Virkon. Move to sink in readiness for washing
17. Clean the BSC with 1% Virkon

Note: Decontamination should be done on maximum sample volume of 5ml. Samples that are greater than 5ml should be aliquoted into another falcon tube to achieve volume of less or equal to 5 ml.

Note: Inoculation will be done in BSC2 only

8. Inoculation

For each specimen two MGIT cultures will be inoculated. Manual Reading will be done

Materials and reagents

a. **MGIT Media**
b. **MGIT growth Supplement (enrichment)**
c. **MGIT PANTA**
d. **Blood Agar**

Note; for reconstitution of PANTA/OADC see section 0 above

8.1. Inoculation of MGIT media

1. Place specimens back in order with the corresponding MGIT tubes, sputum archive vial and slide
2. Using a sterile transfer pipette;
    a. Add 0.5 ml of well mixed processed/concentrated specimen to the appropriately labelled 2 MMGIT tubes
    b. Add 1ml to the appropriately labelled sputum archive vial

Note: Wipe tubes and caps with a mycobactericidal (1% virkon) disinfectant and leave inoculated tubes at room temperature for 30 minutes before incubating

8.2. Incubation

1. Place the MGIT tubes into the MGIT rack
2. Fill out a reading form. This form contains the date of inoculation
3. Place the form on the rack
4. Place the MGIT rack in the incubator according to the day and reading week
5. Incubate to a maximum of 6 weeks
6. The MMGIT tubes should be read once a day using the manual reader until they are positive, up to a maximum of 42 days
7. MGIT cultures showing no growth at 42 days are captured as negative
8. Cultures flagged positive by the manual reader are entered into the data base using the mass positive button.
9. Cultures that are 6 weeks must be inspected for any growth. Any showing any growth-cloudy, crumby particles etc. must be captured as positive and entered as positive and worked up for positive samples
10. Cultures that are 6 weeks old are entered as mass negatives if not showing any growth
11. Mass negative cultures must be autoclaved before discarding. They should be discarded in the bag destined for incineration

9. Capilia TB test (MPB64 antibody test)

1. Positive cultures are further identified to differentiate between ***Mtb*** and MOTT or contamination using the Capilia TB Test
2. Positive control culture should have a Capilia test done

9.1. Performing Capilia TB test MGIT isolates and archiving of isolates

- Apply 1 ml of culture broth to DNA archive tube
- Apply 0.5 ml of culture broth to MGIT culture archive tube(tube should have 0.5ml 20% glycerol)
- Apply 100µl of the positive media to the sample well on the slide directly without any manipulation
- Leave for 15minutes (*all results should be read within 1 hour after dispensing the sample into the sample well)*
- Check for the presence or absence of red to pink bands on both the test and control bands
- Enter results onto results onto the Capilia work sheet
- Enter Capilia TB test results
- Print results and send results to the PI

9.2. Interpretation of Capilia results

1. Positive for *Mycobacterium tuberculosis* complex

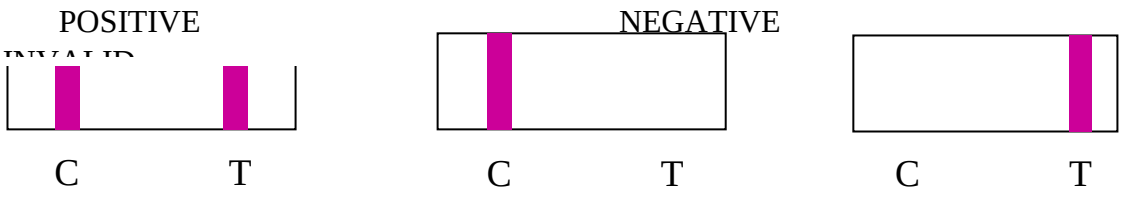
- Red-purple colour appears in both the T and C result windows. The result is read as positive if window T shows red-purple colour that is lighter than, the same as, or darker than the colour window C

2. Negative for *mycobacterium tuberculosis* complex

- Red-purple colour appears in results Window C but NOT in results window T

3. Invalid Test

- A test is invalid, if red-purple colour does not appear in result Window C
- If a test is invalid, report as invalid on results form. Repeat test using a new test Plate, preferably from a newly opened foil pouch



9.3. Quality Control for Capilia Test

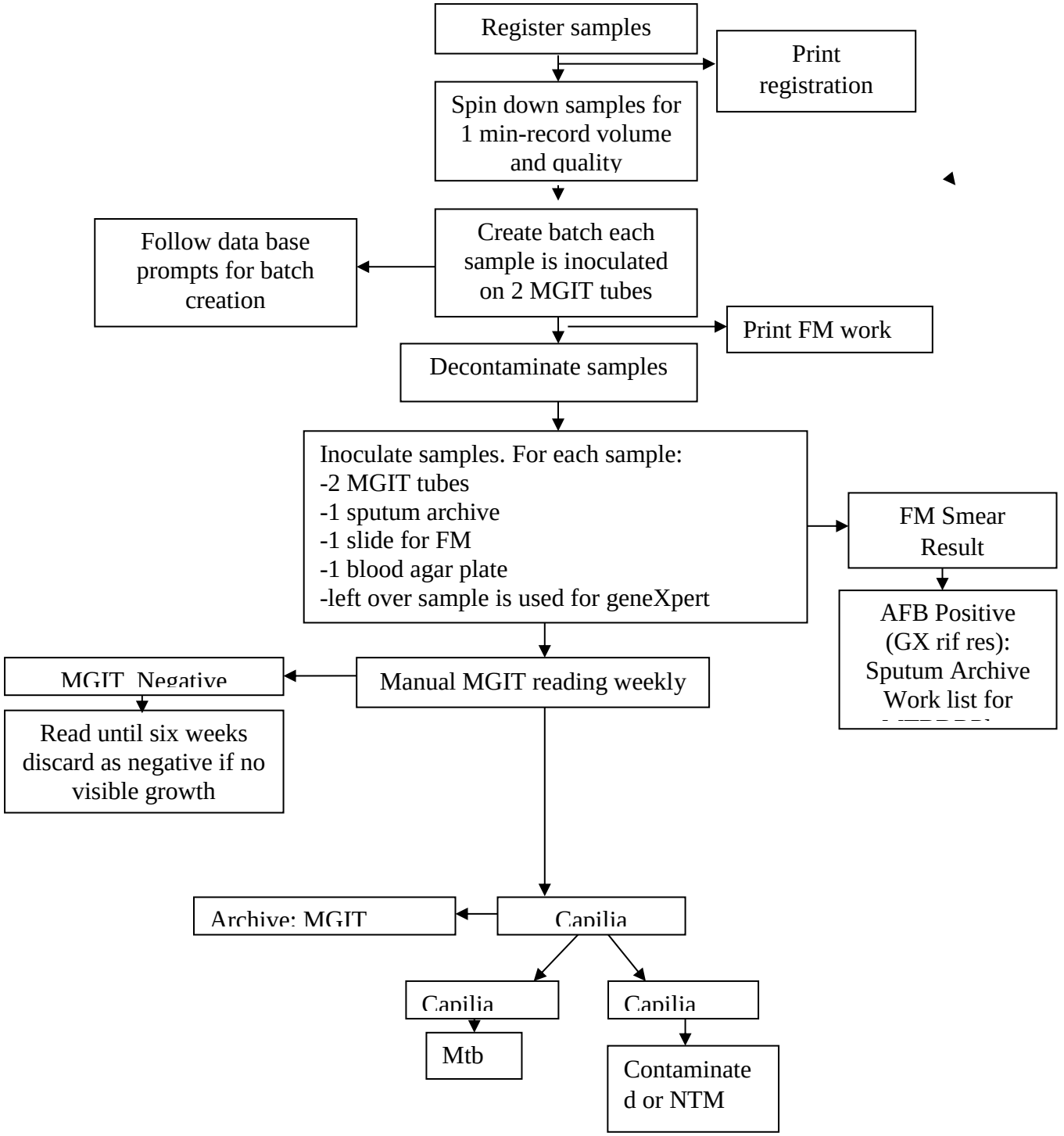
1. Reagents should not be used beyond the expiration date as indicated by the manufacturer
2. The following strains should be included as controls, with results recorded, for each batch of TAUNS test that is used:
   - Positive Control: *M.tuberculosis* reference strain
   - Negative control: M. avium or *intracellulare* reference strain
3. For an invalid test, the lot number, expiration date, date of testing and operator/technologist should be recorded

10. Storage of archived sputum

- Place the sputum archive vials in a blue storage box (use one box that is labelled by week number and date)
- Store at -20^{o}C

Note: One sample is collected from each patient at each time point. Each sample is inoculated on two MGIT tubes.

Flow diagram Culture process



Appendix XIII: Treatment Regimens in Adults

Category I Patients:

All new patients (Smear positive, negative and extra pulmonary).

Intensive Phase: 2 (RHZE)
Continuation Phase: 6 (HE) or 4 (RH)

Table 1: Dose-body weight relation for patients treated with category I

Weight in Kg	**Intensive Phase 2 months**	**Continuation Phase 4 months**
	RHZE 150/75/400/275	**RH** 150/75
30-37	**2**	**2**
38-54	**3**	**3**
55-70	**4**	**4**
>71	**5**	**5**

Category II Patients:

All previous treated patients including smear positive retreatment, smear negative retreatment and treatment failures, treatment after default and relapse cases.

Intensive Phase: 2S (RHZE)/ 1 (RHZE)
Continuation Phase: 5 (RHE)

Table 2: Dose-body weight relation for patients treated with category II

Weight in Kg	**Intensive Phase 3 months**			**Continuation Phase 5 months**
	2 months		**1 month**	
	RHZE 150/75/400/275	**S** 1 g	**RHZE** 150/75/400/275	**RHE** 150/75/275
30-37	**2**	**0.5 g**	**2**	**2**
38-54	**3**	**0.75g**	**3**	**3**
55-70	**4**	**1g**	**4**	**4**
>71	**5**	**1g**	**5**	**5**

Appendix XIV: Translated Documentation

Zenizeni zoona pa matenda a shuga.[Diabetes]

Kodi Diabetes n'ciani?
"Diabetes" ndi matenda ozibika , kambiri, ndi dzina lakuti "matenda a shuga"". Muntu odwala matende a shuga apedzeka ndi shuga yambiri mu magazi.

Ndi ciani cizetsa matenda a shuga?

Kuli mitundu iwiri ya matenda a shuga.
Mutundu waciyambi: Uyu mutundu umachedwa kuti 'Oyambila ku ufana' kapena 'Odalila pa mankhwala ya "insulin"'; thupi lilekelatu kupanga "insulin", zipangizo za m'thupi, zomwe zifunikira mumagazi pakuti thupi likwanilitse kusebenzesa shuga. Bantu bali na uyu mutundu wa matenda a shuga, afunikila nsingano za "insulin" matsiku onse kuti akhale moyo. Uyu mutundu wa matenda kambiri uma pezeka mu bana ban'gono/aci cepele m'sinkhu, ngakhale kuti unga pedzeke pa m'sinkhu uli onse.
Mutundu waciwiri: Umacedwa kuti 'Oyambira mu ukulu' kapena 'Osadalira pa mankhwala a insulin'; uyu mutundu umadza ngati thupi silikwanilisa kupanga 'insulin' yambiri kapena thupi lilephera kusebenzesa bwino 'insulin'. Uyu mutundu wa matenda a shuga upezeka kambiri mubakulu bazaka makhumi anai ndi pa mwamba, onenepa kwambiri ndi iwo ali ndi mbiri pa banja yokhala na matenda a shuga; ngakhale kuti matsiku ano , matenda achuka mu anyamata ndi asikana.
Kodi bantu badziwa bwanji ngati bali ndi matenda a shuga?
Bantu bali ndi matenda a shuga kambiri baona ndikumbvera zizindikiro mwa izi:

1. Kumva ludzu/njota kwambiri ndi kuuma m'kamwa.
2. Kutunda-tunda kuphyola mu m'pimo.
3. Kuonda.
4. Kumva njala kwambiri.
5. Kusaona/kusapenyena bwino.
6. Kunyeleza kumalo obitsika.
7. Kulasa-lasa kapena kumva kuzizira m'manja ndi kuma phadzi.
8. Kabili-kabili matenda yakhungu, mucitsungilo ca mitundo ndi muvi gama va mano.
9. Zilonda zosapola.
10. Kulema kwambiri kosaziwika bwino.

Nthawi zina, kuma khala kulibe zizindikilo – izi zima theka mu m'tundu waciwiri wa matenda a shuga. Zika khala tero, bantu banga khale minyezi , kapena zaka kosaziwa kuti bali ndi matenda a shuga. Uyu mutundu wa matenda umabwera pan'gono-pan'gono, mwina kosaziwika.
Ngati muzimbvera kuti muli nazo zisonyezo/zizindikilo za matenda ya shuga, pitani ku cipatala cili pafupi ndi komwe mukhala kuti apime magazi yanu kuona ngati muli ndi shuga yopitilira malile.
Ndani ali m'ciophyedzo cokhala ndi matenda a shuga?
Aliyense, m'nsinkhu uli onse ndi okhala m'malo ali onse, angathe kukhala ndi matenda a shuga. Koma, matenda niyo wanda muli:

- Bantu omwe akhala cabe osa gwira ncito zina ziri zonse.
- Bantu onenepa kwambiri.
- Nkalamba.
- Baja bali ndi mbiri pa banja ya matenda a shuga.

Kodi matenda aciritsidwa?
Yayi. Koma kuli mankhwala ya mphamvu yo thandidzira, ndipo na kudziwa mozi samalira we mwine, kuma zetsa umoyo wa thanzi.

Zocita ngati mwapezeka ndimatenda ya shuga.

1. Pewani kudya vakudya muli shuga yambiri.
2. Pewani kudya va kudya va mafuta yambiri.
3. Idyani va kudya mupezeka vikamba-kamba monga nyemba, unga wa m'gaiwa ndi 'brown' bread.
4. Kudya kulinganiza zakudya zofunikira m'thupi kuphatikizapo zipaso ndi ndiyo za masamba.
5. Pewani vinyo kapena moba.
6. Pewani kuina koposa.
7. Khalani opikitika, kugwira-gwira tu ncito ndithu, osa khala cabe ndwii.
8. Kumbukilani nthawi zonse kumwa mankhwala monga aku uzilani ancito za umoyo.
9. Pitani kawiri-kawiri kukapimisa, kusata momwe anya ncito za umoyo aku uzirani.

N'zobvuta zotani zotuluka mu matenda ya shuga?

Zapafupi: Kuli zobvuta zapafupi ziwiri zotuluka mu matenda ya shuga ndipo zicedwa ngati zofunika thandizo la cipatala mwa m'sanga.

A. **Kukwera kwa shuga mu magazi.**

Kukwera kwa shuga mumagazi kunga citike ngati matenda a shuga salandira thandizo la mankhwala mokwana kapena mu muntu omwe saziba kuti ali ndi matenda a shuga ndipo sali pa mankhwala.

Zinzindikilo za kuti shuga yakwera mu magazi zili monga izi:

1. Kuyamba ndi kutunda-tunda kawiri-kawiri, m'tsogolo mwake osa tunda konse cifukwa caku cepela kwa madzi m'thupi.
2. Kucepekera kwa madzi m'thupi ndi ku uma m'kamwa.
3. Ku komoka.

Satani bwino malamulo omwe akupatsani ancito za umoyo momwela mankhwala. Pitani kucipatala ngati muli ndi matenda a shuga ndipo muganizira kuti shuga mu magazi yakwela.

B. **Kucepekela kwa shuga mu magazi.**

Kucepekera kwa shuga mu magazi kungacitike mu muntu ozibika kunti ali ndi matenda a shuga ngati:

- Azilansa nsingano ya 'insulin' kapena akumwa mankhwala ya shuga, asanadye vakudya vo kwanilira.
- Agwira ncito ya mphamvu pambuyo pozilasa nsingano ya 'insulin' kapena kumwa mankhwala ya shuga.

Zizindikilo za kuti shuga yacepekela mu magazi zili mwa izi:

1. Kututuma ndi kunjenjemera.
2. Kumva mantha.
3. Kumva cizwezwe.
4. Kumva njala.
5. Kumva m'tima kugunda ndi mphamvu ndipo mwa m'sanga.
6. Kumva cibe.

Ngati mwaona/mwamva zina mwa izi, conde mwa m'sanga idyani (monga switi) kapena mwani conzuna, cakumwa conzuna monga zigoro.

Kodi matenda a shuga anga cinjilizidwe?

INDE!! Nthawi zina. N'zambiri zomwe muntu angathe kucita kecedwetsa kapena kucinjiliza kuyambika kwa matenda a shuga.

- Sungani thupi la udongo; osanenepesa.
- Khalani onyang'anya- nyang'anya kawiri-kawiri/*osakonda kukhala cabe kosa gwira ncito za manja.*
- Ngati muli ndi m'bale ali nao matenda a shuga, muzipita kawiri-kawiri kukapimisa.
- Kumadya zakudya zokwanilitsa zofunikira za m'thupi, kuphatikizapo zipaso ndi ndiyo za masamba.

PEPALA YOBVOMEKEDZA.

PEPALA YOBVOMEKEDZA PA KUFUFUDZA KWA MATENDA A SHUGA NDI TB (BANTU BALI NADZAKA KHUMI LIMODZI, ZISANU NDI ZITATU KUPITA PA MWAMBA)

1. Ndi simikidza kuti ndawerenga mau o unikira ndi kuti zonse zofunikira m'kutengamo mbali mukufufudzaku, zafotokozedwa kwa ine.
2. Ndapatsidwa nthawi ndi m'pata kuwerenga mau o unikira mo dekha mutima, kukambitsanapo ndi bena ndiku sankha kutengamo kapena kusa tengamo mbali muli uku kufufudza.
3. Ndamvetsa kuti magazi ndi vikolala vanga, viza sungidwa/sungiwa kumalo osungila zimenedzo, ndi cidziwitso ca nambala cabe, osati dzina langa, ndikuti zosungidwazo zingathe kusebenzesedwa m'tsogolomu, pakufufudza kwina kwa matenda a shuga.
4. Ndamva kuti zopezeka muli uku kufufudza kuza falitsidwa mumapepala yolemba/ yowerenga ya akaswili, koma kuti dzina langa siliza sebenzesedwa/kuulusidwa.
5. Ndabvomera kutengamo mbali muli uku kufufudza.

Kusaina kwa otengamo mbali/kufwatika cikumo:_____ Tsiku: _____

Dzina la otengamo mbali: _____(conde lembani mwa udongo/malemba akulu).

Othandizira kukambirana ndi kutenga cibvomelezo ayenera kusaina ndi kulemba tsiku pomwe zonsezo zacitika papepala ili.

Kusaina kwa othandizira: _____

Tsiku:_____

Dzina: _____ (conde lembani mwaudongo/malemba akulu)

Kusaina kwa ocitila umboni (ngati kufunika)

Kusaina kwa mboni: _____ Tsiku: _____

Dzina: _____ (conde lembani mwaudongo/malemba akulu).

MAU KWA OTENGAKO MBALI PA FUFUZO LOYANG'ANA PAZA KACHIRISIDWE KA ANTHU OMWE ALI NDI TB PAMODZI NDI MATENDA A SHUGA NDI OPANDA MATENDA A SHUGA PA CHIPATALA CHA UNIVERSITY TEACHING HOSPITAL, LUSAKA

Muitanidwa kutengako mbali muphunziro imene izatithandiza kumvetsetsa momwe matenda aShuga akhuzana ndi TB ndimomwe zimaonekela mu Zambia. Azatengako mbali ndi anthu omwe azapezedwa ndi TB pa University Teaching Hospital. Komabe, mwa anthu amenewa, 114 (85 opanda matenda a Shuga ndi 29 ali ndi matenda aShuga) ndiwo azasankhidwa kuti tiwalondole. Mau apa punziro iyi apasidwa mupepala iyi. Othandizira pa punziro iyi azakhalapo kufotokoza ndi kuyankha mafunso anu onse omwe mungakhale nawo. Chonde onani kuti mwamvetsetsa zones zili mupepala iyi. Ngati mwasankhula kutengako mbali, muzapemphedwa kupatsa chibvomekezo chanu musanatengeko mbali.

Ndani akuchita punziro iyi?

Phunziro iyi ili kuchitidwa ndi a Dotolo Fwoloshi, omwe akuchita maphnziro apatali kuchokela ku School of Medicine, muchigawo cha Internal Medicine pa UTH muno mu Lusaka. Mayankho a phunziro iyi azathandizira kuonesa maphunziro anga apamwamba ndiponso kuonjezera nzeru pakasamalidwe ka anthu amene ali ndi TB ndi Shuga. A Dotolo Fwoloshi ndi owathandizira ndiwo azayanganira momwe phunziro iyi ikuyendela tsiku ndi tsiku. Oona ndi:

Principal Investigator	**Supervisors**	**Research Assistants**
Dr Fwoloshi 0977319994	Dr Hachaambwa 976435603 Dr Chiyenu 0955998207 Dr Bailey 0969276263	Name:…….. …… ……

Ngati muli ndi masunso ena pa zapunziro iyi mungathe kuziwisa:

The Chairperson

ERES CONVERGE IRB

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Lusaka

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Lingo la phunziro iyi ndi lotani?

Lingo leni leni la phinziro iyi ndilofuna kuonjezera kumvetsetsa momwe matenda aShuga akhuzana ndi TB ndi katetezedwe kawo muno mu Zambia. Tidziwa kuchokera mumaphunziro ena omwe anachitidwa mumaiko m'dziko lapansi kuti anthu amene ali ndi shuga ali ndi mpata waukulu kukhala ndi TB kupambana anthu opanda shuga. Tidziwanso kuti ngati akhala ndi TB mankhwala aTB sama sewenza bwino monga momwe amasewenzera mu anthu opanda shuga. Koma mu Zambia di maiko ena amu Afrika zingakhale zosiyana ndi kumalo ena ace m'dziko lapansi, ndipo ife tifunakudziwa kuti shuga ikhuzana bwanji ndi TB muno mu Zambia. Izi zizalenga kuti tionjezere njira zosamalira shuga ndi TB m'dziko lathu

Chisinsi

Mau amukufufuza uku azasungidwa mwachisinsi pobisala. Ogwira nchito iyi ndi owayanganira ndiwo chabe azakhala ndi mpata kuona mauwa. Ngati kuzakhala zolembedwa za phinziro iyi,palibe chosonyeza inu chomwe chizapekekapo pazolemba izi.

Ufulu wa otengako mbali

Kutengako mbali muphunziro iyi sikutanthauza kuti mwataya ufulu wanu. Kupasa chibvomerezo kutanthauza kuti mwawerenga kapena mwamva mau aphunziro iyi ndipo mwabvomera kutengako mbali. Simuzapeza bvuto ili yonse kapena kusapisidwa zomwe muyenela kupasidwa potengako mbali muphunziro iyi.

Ufulu okana kapena kuleka

Kutengako mabli muphunziro iyi ndi kufuna kwanu. Ngati mfunso ena alengetsa kusamva bwino muhunziro iyi muli ndi ufulu okana kutengako mbali. Ngati mwasankha kutengako mbali ndikuzasintha pasogolo, muli omasuka kusatengako mbali panthwi ili yonse. Sankho lanu kusatengako mbali sikuzaletsa kasamalidwe kanu ka nthawi zones pa chipatala.

Zizachitika

Ngati mwabvomera kutengako mbali muphunziro lalikulu loona za shuga mumagazi chabe:

- Tizalasa kanyeleti pachala ndikupima shuga mumagazi

1. Ngati muli wina wa anthu114 ozatengako mbali muphunziro lalikulu, kuonjezera kulasiwa pachala kupima shuga mumagazi

   Tizatenga mau paza inu ndi nthawi yomwe mwakhala muli kudwala

   Tizapima kutalimpha ndi sikelo

   Tizalasa chala kupima shuga mumagazi anu

7. Ngati yapitilila pa 7 mmol/l, tizatenga magazi okwana 2 mls (monga hafu teaspoon) kuti tione momwe shuga yanu inakhalira pamyezi itatu yapita popima zochedwa glycosylated haemoglobin mumagazi anu.($HbA_{1c)}$

   Muzafunidwa kubweranso mutatha milungu 8 pa mankhwala a TB,

   pomwe teaspoon 4-6mls (teaspoon limodzi ndi hafu) la magazi lizatengedwa kuti:

   tiyangane CD4 ngati muli ndi kadoyo kaHIV ndipo mulibe CD4 latsopano apa

   tizasunga kuzapima kuona kuchuluka kwa mankhwala aTB (Rifampicin) kusogolo

   Muzafunikanso kupasa zikolala kuti tipime TB

Ziyopezo ndi kusamvera bwino

Zochitika zaululidwa pamwamba zili ndi ziyopezo zazing'ono

Kuchosa magazi ndi kulasidwa pachala kungabweretse kuphweteka pamalo omwe titengela magazi

Kutenga magazi ndi kulasa pachala kuzachitidwa ndi adotolo kapena othandizira awo.
Kukhala ndi matenda pamalo omwe titengelapo magazi n'chapatali, kuletsa mabvuto amenewa adotolo ndi owathandizira ndiwo chabe azatenga magazi kapena kulasa chala