Evaluation of Healthcare Facilities and Services Provided for Tuberculosis and Zoonotic Tuberculosis in Kajiado County, Kenya

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A thesis submitted in partial fulfillment of the requirements for the Master of Science degree in Pathology and Laboratory Medicine

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Abstract

There are key challenges people face due to the burden of tuberculosis (TB), caused by *Mycobacterium tuberculosis*, and zoonotic tuberculosis (ZTB), caused by *Mycobacterium bovis*, in Kenyan Maasai rural communities. Thus, the objective of this study was to describe current capacities/infrastructure for diagnosis/treatment of TB/ZTB, and knowledge, attitudes, and practices among healthcare workers in rural Kajiado, Kenya. A questionnaire was delivered by Talaku - a Community Based Organization – with data being received from 25 healthcare facilities (HCFs) and 69/75 healthcare workers during January-February, 2022. The data was descriptively analyzed. It was found that only 12% of HCFs listed bacterial culture as an available diagnostic technique, and among healthcare workers, only 19% correctly identified pyrazinamide as a drug that is inherently resisted by *M. bovis*. We are confident that the data collected will increase awareness for TB, ZTB and provide information to local stakeholders for further work.

Keywords

Tuberculosis, zoonotic tuberculosis, one health, infectious disease, bovine tuberculosis, Kenya, Maasai
Summary for Lay Audience

Annually, the World Health Organization reports over 10 million cases of TB disease among people, globally. TB is caused by the bacterium *Mycobacterium tuberculosis*, and approximately 1.4 million deaths are caused by TB every year. An unknown number of TB cases are caused by bacteria (*Mycobacterium bovis*) transmitted from animals, mostly bovines, to humans, which causes zoonotic TB (ZTB) in humans. Kajiado County is home to a significant population of Maasai who are at higher risk for ZTB due to sociocultural practices that increase direct and indirect contact with infected animals and with food products containing *M. bovis*. These include consumption of raw milk and milk products. The burden of TB/ZTB are ultimately facilitated by a lack of knowledge and resources surrounding TB/ZTB. Thus, there is substantial need to evaluate the ability of Kajiado County healthcare facilities (HCFs) to diagnose, treat, and understand TB/ZTB. With help from Talaku – A Community Based Organization, a survey was delivered to 69 individuals at 25 local HCFs where we found that many HCFs are lacking in the diagnostic techniques necessary for diagnosis of TB in humans, and many healthcare workers are unaware of key knowledge surrounding *M. bovis* drug resistance and contact at the human-animal interface. The results of the project will contribute to providing key, new information to local stakeholders, and to create awareness for a neglected disease in rural Kenya.
Co-Authorship Statement

MSc candidate Joel Zhang, was primarily tasked with the design of the questionnaire to be used for data collection by Talaku in Kenya during fieldwork, completing quantitative/qualitative data analysis, facilitating the reporting, write-up of the project results, and developing the scientific manuscript for publication. The main supervisor (Dr. Francisco Olea Popelka) was responsible with overseeing this project, initiating collaboration, guiding the in-field work conducted by Talaku, and supervising Joel during the data analysis phase of this study. Talaku- a Community Based Organization in Kenya, headed by Ms. Timpiyian Leseni, delivered the questionnaire to be completed, entered data electronically, and provided key insight and guidance in regards to health in rural Kajiado County. Ms. Leseni will co-author all manuscripts and presentations.
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First and foremost, I would like to acknowledge and greatly thank, my supervisor, Dr. Francisco Olea Popelka, for his unparalleled support and assistance in the progression of my Master’s project, graduate, and undergraduate thesis, especially given the unique circumstances of COVID-19. Without his countless hours reviewing manuscripts, proposals, presentations, and statistics, I am not sure where I would be right now. He really has never “missed a beat.” As a professional, he has pushed me, a young man wanting to make a sliver of difference in the world. More importantly, as a friend, he has been everything I could have asked for.

Dr. Francisco Olea Popelka introduced me to One Health. This concept has given me a sense of purpose in the scientific world, at a major crossroad in my life, and at a time of uncertainty, he helped me feel certain. One health has become the forefront of my life and has changed the way I have looked at science, forever.

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My family. My mother, my father, and my sister. It must have been hard having your son away from home these six years and your brother living out his life during your formative years. I have a feeling. Maybe because, contrary to your belief, it was hard for me to be away from home for six years too. Phone calls became increasingly spread apart. As I got older, I wanted to shoulder those hardships myself. I hope you can forgive me. Blindly supporting your kid in an endeavor, one so foreign, must have been difficult. No matter where I end up, I want to make you proud, and when I feel like I failed myself, I feel like I failed you too.

Finally, I would like to recognize the people who suffer from tuberculosis, zoonotic tuberculosis, and other infectious diseases in Kenya, Africa, and across the globe. To those people, I want to say that we still have so much work to do in science, especially in the often overlooked discipline of collaboration. We are so busy in our own fields that we forget we should be working towards common goals to solve crucial global health issues together. I hope that our generation can become the stalwarts in recognizing the necessity of collaboration in health disciplines; in the words of my mentor Dr. Olea Popelka: “To improve health, scientific/medical knowledge is 100% necessary, but not sufficient.”
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Chapter 1: Literature Review

1.1 Tuberculosis

1.1.1 Tuberculosis in Humans caused by Mycobacterium tuberculosis

Infectious diseases are the leading causes of morbidity and mortality, globally\(^1\). Of these diseases, Tuberculosis (TB) is the number one infectious “killer” in the world and has, in the past decades, caused more deaths annually than any other single infectious agent. According to the 2020 Global Tuberculosis Report published by the World Health Organization (WHO), 10 million individuals were newly diagnosed with TB, and 1.4 million individuals died due to the disease in 2019, globally\(^2\). The WHO estimates that about 25% of the entire global population has been infected with the causal agent of TB - *Mycobacterium tuberculosis* (M. *tb*) - at some point in their lives\(^2\).

Factors that can exacerbate *M. tb* transmission between people include lack of proper ventilation and close-quarters, extensive contact with an infected person\(^3\). While the disease mainly affects the lungs, it can also present as an extra-pulmonary disease in other organs\(^2\) including the lymph nodes, bones, joints, the genitourinary tract\(^4\), and the brain\(^3\) in the potentially fatal form of meningitis TB\(^4\).

In the lungs, disease-causing *Mycobacterium* interact with host macrophages, where the *Mycobacterium* are phagocytosed and are met with mechanisms aiming to kill the bacteria, including acid pH, reactive oxygen intermediates, lysosomal enzymes, and toxic peptides. Some of the *Mycobacterium* may survive and form granulomas where the bacterial agent can remain dormant for decades. The aim of the granulomas is to contain the bacterial agent, even after the macrophage is killed, causing the caseous center that is seen clinically\(^4\). This controlled state of the bacteria is known as latent TB infection (LTBI), typically does not cause clinical symptoms\(^5\), and the *Mycobacterium* cannot be transmitted to other individuals. However, immune-compromised individuals, such as those co-infected with HIV/AIDS are at a great risk for re-infection\(^5\) in which the bacterial agent can continue to multiply and spread, bypassing macrophage control, causing active pulmonary TB and extra-pulmonary TB\(^4\). Approximately 5%-15% of people infected with *M. tb* progress to
active TB disease. Without treatment, TB is a disease with a high mortality rate and a vast majority of patients who are HIV-positively, co-infected with TB will die from the disease.

1.1.2 Tuberculosis in Humans caused by *Mycobacterium tuberculosis* in Kenya

Africa represents 24% of global TB cases and 32% of deaths worldwide due to TB. In Kenya, where 46% of the population lives below the poverty line, TB is the fourth overall leading cause of death. According to the WHO, Kenya is one of the top twenty burdened countries in terms of TB, TB/HIV co-infection, and Multi-Drug Resistant TB (MDR-TB). One study stated that Kenya was one of the top ten most burdened countries globally due to TB, and, in 2019, there were an estimated 140,000 incident cases of TB in Kenya. Due to large gaps in screening, diagnosis, and reporting, there is a high possibility that the current burden of TB in Kenya is much greater than reported, as numbers vary highly depending on the literature reviewed. A study through the National TB Programme in Kenya noted that almost 50% of persons with TB have not notified public health authorities for the disease. The 2016 Kenya TB prevalence survey also estimated that 54% of infected Kenyans are not notified for TB, 67% of individuals with symptoms did not seek care, and only 46% of those diagnosed for TB were put on treatment regiments for the disease. Yet another study investigating patients starting TB treatment in 2015 estimates that over 75% of MDR-TB cases and 65% of paediatric TB cases are missed. Overall prevalence of the disease is also not well documented, especially in areas of rural Kenya. A study of high-burden counties in Kenya between 2012 and 2015, including Kisumu and Siaya, noted that the notification incidence per 100,000 individuals was 370.4 and 248.8 respectively, with some areas having incidence rates of up to 563.9 per 100,000 individuals.

1.2 Bovine Tuberculosis

1.2.1 Bovine Tuberculosis in Cattle caused by *Mycobacterium bovis*

Bovine tuberculosis (BTB) caused by *Mycobacterium bovis* (*M. bovis*) in cattle, is a common, widespread disease that continues to impact cattle, and other mammals, including
people, worldwide\textsuperscript{13}. According to a 2019 report by the World Organization for Animal Health (WOAH), formerly known as the International Organisation for Animal Health (OIE), fifty-one countries reported presence of \textit{M. bovis} in livestock\textsuperscript{14}. In a previous WOAH report that included 179 countries, it was estimated that more than half of these had presence of BTB in livestock, wildlife or both\textsuperscript{15}. Based on skin test results, the global prevalence of the disease in livestock is estimated to be 9\%, although the true prevalence is largely unknown due to large cattle populations in developing countries\textsuperscript{16}. Even in high-income countries, it is estimated that BTB cases are increasing, despite regular test-and-slaughter programs\textsuperscript{17} in cattle. However, there is severe underreporting for this disease globally. Underreporting at the national level is likely to occur in places, such as Kenya, as there are reports of BTB as part of specific studies in this country\textsuperscript{18-24}, however, routine, nationwide estimates are lacking.

In brief, cattle are commonly screened for BTB using the Caudal Fold Test (CFT) or the Single Cervical Intradermal test (CIT), where only a purified protein derivative (PPD) of \textit{M. bovis} is injected. The Single Intradermal Comparative Cervical Test (SICCT), where cattle are injected with PPD of both \textit{Mycobacterium avium} (\textit{M. avium}) and \textit{M. bovis} is used in certain countries with the difference in duration between the two being measured after 72 hours\textsuperscript{25}. If the reaction of the \textit{M. bovis} site is >4mm compared to the \textit{M. avium} site, cattle are considered positive for BTB\textsuperscript{26}. The injection of \textit{M. avium} increases the specificity of the test, preventing false positive due to \textit{Mycobacterium avium sub. Paratuberculosis} or non-tuberculous mycobacteria\textsuperscript{27}. Overall, the SICCT has a sensitivity that varies between 68\% and 95\% and a specificity that varies between 99.8\% and 99.9\%, according to studies completed in Ireland. This is dependent, among other factors, on how advanced BTB is in the animal\textsuperscript{26, 28}.

\subsection*{1.2.2 Bovine Tuberculosis in Cattle caused by \textit{Mycobacterium bovis} in Kenya}

In Africa, only seven countries list BTB as a notifiable disease, in which there are regular control strategies implemented. Forty-eight countries in Africa inadequately or do not control BTB at all. As a result of this, almost 85\% of cattle and 82\% of the human
population in Africa reside in areas where BTB is not controlled or controlled inadequately\(^29\).

According to Kenya’s Animal Diseases Act, BTB is a notifiable disease\(^30\). Yet, when considering Kenya as a whole, there are only a few studies discussing prevalence, incidence or burden of BTB nationwide. Although WOAH reports, published in 2019, state that there is an absence of BTB in Kenya\(^14\), other literature indicates that BTB is prevalent in Kenya. One study examining 929 cattle carcasses from a period of July to November 2009, in a post-mortem abattoir inspection using molecular analysis, found that *M. bovis* was present in 2.05% of these samples\(^18,19\). A study published in 2016, found that in Migori, West Pokot, and Laikipia Counties, there was a prevalence of 4.03%, 6.41%, and 4.96% within 124, 78, and 262 cattle screened with the SICCT\(^20\). Another study published in 2020, found *M. bovis* in 3% of 1000 isolated lesions in slaughtered cattle in Kenya\(^21\). A final study conducted between 2011 and 2013 noted that of 276 bovine blood samples there was an individual prevalence of *M. bovis* in cattle of 3.9% and a herd prevalence of 58.3%\(^22\). Most studies noting BTB detection in Kenya utilized a post-mortem diagnosis or SICCT test, as opposed to culture or molecular techniques, thus, several cases could be missed\(^22\). Another study, published in 2018, reported a prevalence of 3.57% in lactating, domestic camels in Isiolo County, Kenya\(^23\). BTB is also reported in wildlife, as in one study, it was found that 1.69% of 1,600 slaughtered camels in Samburu County were positive for *M. bovis*\(^24\).

There are high economic impacts in Kenya due to *M. bovis* infection in cattle. A Food and Agriculture Organization of the United Nations (FAO) report, published in 2018, evaluating the burden of zoonotic diseases on the Kenyan economy, reported that Kenya spends nearly 175.6 million US dollars to combat losses due to BTB, accounting for 6% of cattle value added to annual gross domestic product (GDP), with each death due to BTB in cattle resulting in a loss of 963 US dollars\(^31\).

### 1.3 Zoonotic Tuberculosis

#### 1.3.1 Zoonotic Tuberculosis in Humans caused by *Mycobacterium bovis*

While TB is most commonly caused by *M. tb* in humans, there is increasing evidence to suggest a substantial amount of human TB cases are caused by other members of the
*Mycobacterium tuberculosis* complex (MTBC), namely *Mycobacterium bovis* (*M. bovis*) and *Mycobacterium africanum* (*M. africanum*), although other members of the MTBC may also cause disease in humans.

Zoonotic TB (ZTB) occurs when *M. bovis* is transmitted from cattle to humans through three main transmission routes. The first, and most important, is through the consumption of unpasteurized milk or milk products containing *M. bovis*. Prior to pasteurization of milk, TB caused by *M. bovis* in the United States accounted for approximately 10% of all TB cases and a quarter of TB cases in children. Other transmission routes include the consumption of raw or undercooked meat containing *M. bovis*, or aerosolized particles containing the bacterial agent. There is also increasing evidence indicating that *M. bovis* can enter the body through exposure to cuts in the skin, where regular protective equipment is absent or improperly used.

It is unknown how much *M. bovis* contributes to the burden of TB in humans worldwide. Various publications have estimated that anywhere from 1.4%-10% of all cases of TB are due to *M. bovis* in humans. In the developing world, an estimated 10% of TB cases are due to *M. bovis*, with that number being as high as 37.7% of TB cases in Tanzania based on a systematic literature review completed in 2001. Four separate regional studies conducted in Tanzania that used molecular techniques and bacterial culture stated that an average of 30% of human TB cases could be caused by *M. bovis*. A reason for this uncertainty is that clinically, pathologically, radiologically, and anatomically, ZTB is indistinguishable from TB caused by *M. tuberculosis* (*M. tb*), although it is known that ZTB is more likely to cause extra-pulmonary disease in humans. Like TB caused by *M. tb*, all organs can be infected by *M. bovis*; most commonly, the lungs, the lymph nodes, genitourinary systems, bones, joints, intestines, peritoneum, and central nervous system.

There is a considerable risk for zoonotic transmission of *M. bovis* to humans from cattle, especially for farmers, veterinarians, abattoir workers and those working with infected animals. The true burden of the disease is largely unknown due to the lack of routine surveillance for this disease in many low-income countries globally, and the inability of the diagnostic techniques that detect TB in humans to properly differentiate between *M. tuberculosis* and *M. bovis*. 
1.3.2 Zoonotic Tuberculosis caused by *Mycobacterium bovis* in Kenya

The 2019 WHO Global Tuberculosis Report estimates 69,800 cases of ZTB in Africa, along with 9100 deaths\(^4\). As well, the 2018 FAO report that examined BTB in Kenya, also included information regarding the burden of zoonotic transmission of *M. bovis* to humans. This report found that *M. bovis* caused 1,168 deaths in humans in 2016; 70% of these being from livestock keepers, accounting for 41,590 years of life lost. This cost the country 336.5 million US dollars, which is nearly 12% of the yearly budget for the Kenyan Ministry of Health (MOH)\(^3\).

Due to lack of surveillance, there is no current, accurate estimate of ZTB in Kenya\(^4\). One study, published in 2018, examined eleven slaughterhouse workers, and found that nine of them were infected with *Mycobacterium* subspecies, with one (1.9%) of these nine being confirmed to be *M. bovis*\(^4\). This indicates that zoonotic transmission of *M. bovis*, which causes ZTB, is present in Kenya.

The unprocessed milk trade is rampant in Kenya, with its consumption being a major risk factor for ZTB transmission. It is estimated that over 85% of milk on the market in Kenya is sold in its raw form. There is a high percentage of raw milk consumption, even in urban areas, but is an especially important cultural practice in rural Kenya. Kenyans prefer the consumption of raw milk due to a variety of factors: 1) it is 20-50% cheaper than processed milk, 2) preferred taste, 3) raw milk can be sold in varying quantities, depending on individual financial situation, 4) it is accessible and readily available, and 5) there is a cultural and historical norm of consuming raw milk\(^4\). With raw milk being the main transmission risk factor for zoonotic transmission\(^5\), that puts a large proportion of the Kenyan population at risk for ZTB.

1.4 Multi-Drug Resistant Tuberculosis caused by *Mycobacterium bovis* in Humans

Drug resistance has become increasingly important within the context of TB in humans. While there are key literature discussing the implications of increasing drug resistance in *M. tb*, globally, the knowledge base for drug-resistant *M. bovis*, specifically in rural Kenya
is non-existent. Without data on drug resistant *M. bovis* specifically in rural Kenya, it is important to note the available data and studies regarding *M. bovis* drug-resistance in other areas around the world.

*M. bovis* is naturally resistant to pyrazinamide\(^{15}\), one of the main, first-line drugs used to treat TB in humans. In terms of *M. bovis* resistance to other anti-microbials, studies focus on tests conducted with *M. bovis* samples obtained from cattle. One study in Sardinia, Italy that was published in 2001, isolated 22 *M. bovis* strains from cattle, and 14 of these samples were found to be resistant to rifampicin and isoniazid, both first-line drugs in the treatment of TB in humans\(^{50}\). Another study in Brazil, conducted between August 2014 and May 2015, where 67 *M. bovis* isolates were obtained, found that 11 (16\%), 8 (12\%), and 2 (3\%) of the samples were found to be resistant to multiple first-line drugs (isoniazid and rifampicin together), isoniazid by itself, and rifampicin by itself, respectively\(^{51}\). Another study completed in Tunisia, published in 2018, had tested and found one sample (of 36) that was resistant to streptomycin, another first-line drug in TB treatment\(^{52}\). While these studies evaluated *M. bovis* samples obtained from cattle, all three of these studies stated that there could be important and unknown implications for human TB cases in these regions, with studies in these regions not evaluating zoonotic transmission of *M. bovis* to humans\(^{50-52}\).

Studies that have utilized *M. bovis* samples obtained from humans in Mexico indicate that there is evidence that *M. bovis* strains in humans have developed multi-drug resistance (MDR) (rifampicin and isoniazid)\(^{53}\). One case study in Mexico City surveyed 306 ZTB cases in a 15-year period, between 2000 and 2014, and found that 10.9\% of these samples were resistant to streptomycin, and 7.6\% were resistant to rifampicin and isoniazid\(^{54}\). A study in Madrid, Spain, examined nineteen cases of MDR-TB caused by *M. bovis* in HIV positive patients. These samples showed resistance to eleven antimicrobials; first-line drugs such as: isoniazid, rifampicin, ethambutol, pyrazinamide, streptomycin, as well as second-line drugs like amino salicylic acid, clarithromycin, ethionamide, ofloxacin, capreomycin, and amikacin\(^{55}\). Another study evaluating *M. tb* and *M. bovis* samples in Pakistan conducted between June 2012 and June 2014, examined 91 MDR *Mycobacteria* from TB cases, in which half the samples (n=46) were tested for drug sensitivity using a
drug proportion (DP) method, and another half (n=45) were tested using the MGIT (mycobacterial growth indicator tube) method. Using the DP method, 32.6% were found resistant to rifampicin, 28% resistant to isoniazid and rifampicin, and 37% found resistant to the combination of rifampicin, isoniazid, and ethambutol. Utilizing the MGIT method, 82.2% of the samples were found resistant to the isoniazid, rifampicin, ethambutol combination.

1.5 Socio-Cultural-Economic Factors Related to TB, BTB, and ZTB in the Maasai Community

Kajiado County in Kenya is home to a significant population of people from the Maasai ethnic group, who are at a high risk for TB due to a variety of socio-cultural-economic factors. A large percentage of the Maasai community are protected as part of a United Nations Educational, Scientific, and Cultural Organization (UNESCO) heritage site, thus making them unique and culturally important. The Maasai are a semi-nomadic group that practices pastoralism, thus their livelihoods are strongly tied to their close relationship with animals. A large majority of land in Kenya is utilized by pastoralists (70%) and of the 1.5 million pastoralists in Kenya, 300,000 are ethnic Maasai. In terms of diet, milk and milk products account for over 60% of their dietary energy, coupled with meat and blood, all substantial risk factors for disease transmission.

Much of Kajiado County is composed of pastoral communities. The sharing of micro-environments between livestock and people (sometimes sleeping in the same quarters) exacerbates the potential for zoonotic transmission in pastoralist communities. Pastoralist communities are largely neglected when exploring health surveillance, which is problematic within the context of emerging zoonotic diseases. The semi-nomadic nature of the Maasai community make them difficult to measure within the context of healthcare in an entire region or country, as larger settlements may split up depending on the time of year. New measurement techniques, including geospatial techniques are promising, but preliminary.

Also, of special interest to Maasai communities, is their housing situation. Traditionally, the Maasai community live in small, single-room houses known as Manyatas, which lack
proper ventilation, and are almost completely closed off to the outside. This, coupled with large amounts of people living in these Manyatas, allows diseases, such as TB, to spread easily and quickly\(^{64}\).

The Maasai community faces other unique barriers to healthcare access. One study\(^{65}\) noted three categories of qualitative factors that were significant barriers to healthcare access:

1. Individual/family/community factors including women not having a “say” in healthcare decisions, misconceptions about services provided at healthcare facilities (HCFs), fear of HCFs, and a preference for traditional healing methods.

2. Geographic factors including distance to the facilities, lack of transportation, poor terrain, and lack of night-time services and

3. Health facility factors including lack of appropriate care (drugs and supplies)\(^{65}\).

Many of the barriers regarding TB in the Maasai community are based upon certain knowledge, attitudes, assumptions, and practices regarding the disease, its transmission, prevention, detection, and treatment. Many Maasai community members are aware of the dangers of TB, symptoms, and the treatable nature of the disease\(^{66, 67}\). This is noted in qualitative and quantitative studies alike. Yet, the causes of TB are not well understood in these communities, with some individuals attributing the disease to punishment from god, sun exposure, promiscuity, excessive exercise\(^{67}\), drinking, cold air, witchcraft, physical trauma, genetic factors\(^{66}\), smoking, and dust exposure\(^{66, 67}\). Many Maasai correctly indicate that TB can be spread through coughing, but also incorrectly indicate that the sharing of utensils can cause TB as well\(^{66}\). The connection between TB and HIV is well understood, except many community members indicate that they are the same disease, rather than two separate diseases\(^{67}\).

In terms of treatment, traditional healers are often the preferred practitioner for treatment due to tradition and availability. Of note, one treatment technique included drinking raw blood from the meat of animals such as cattle, potentially causing zoonotic diseases\(^{67}\). Thus, there is still a substantial barrier to a full understanding of these diseases in rural Maasai communities.
1.6 Diagnosis and Treatment of Tuberculosis in Humans

1.6.1 Diagnosis of Tuberculosis in Humans

TB is commonly screened using the tuberculin skin test (TST). The current test used is known as the Mantoux method, in which a protein purified derivative (PPD) of tuberculin is injected intradermally. A positive result for this test is based on the size of the induration within 48-72 hours of the injection\(^68\). However, the Mantoux method has poor specificity, as exposure to non-tuberculous *Mycobacterium* (NTM), those which do not cause TB, and previous bacille Calmette-Gueren (BCG) vaccination (which utilizes a derivative of *M. bovis*) will accrue a false positive under the Mantoux test. This can be a major issue, as many countries with high TB burden apply the BCG vaccination regularly, and exposure to some NTM can be high\(^27\). One other test utilized in TB screening is the interferon gamma release assay (IGRA), which looks to detect antibodies specific to the MTBC in the blood\(^69\). While this test is more specific than the TST, blood samples must be processed within 30 hours, is expensive, and depends on correct handling of samples and interpretation of the assay\(^70\). While the Kenyan MOH reports availability of the IGRA test, it is not considered readily available and its availability is dependent on the affordability of the test in different regions of the country\(^71\).

Screening aside, there are some key diagnostic techniques for TB in humans. The first of these, most commonly used as a simple, and inexpensive method to detect TB in humans, is a sputum smear using an acid-fast bacillus stain, the Ziehl-Neelsen (ZN) stain, with conventional light microscopy\(^72\). This technique is the main technique approved by the WHO for middle-low-income countries\(^73\). Yet, there are some limitations associated with acid-fast bacillus stains, namely that it can display poor sensitivity\(^74\), depending on the skill of the technician involved\(^73\). One systematic review of microscopy methods indicated sensitivities as low as 32\% for conventional microscopy and 52\% for other stains utilizing fluorescent microscopy, respectively\(^72\). Another study stated that only 30\%-60\% of culture- positive TB cases can be detected by ZN stain, with fluorescent microscopy raising this to 78\% at its highest capacity\(^75\). As well, microscopy cannot differentiate between *M. tb*, and other disease-causing mycobacteria, such as *M. bovis*\(^74\).
The “gold-standard” method for diagnosing TB in humans is the use of bacterial culture to grow the bacterial agent. The sensitivity for detecting pulmonary TB utilizing three sputum cultures is greater than 90%. Yet, utilizing culture to detect TB is not without several limitations. Bacterial culture can take 6 to 12 weeks to finalize a positive or negative result. As well, glycerol, used in Lowenstein-Jensen culture medium, inhibits the growth of certain members of the MTBC, most notably, M. bovis. The use of culture mediums containing pyruvate (i.e Stonebrink or Middlebrook), are more accurate to detect ZTB, as they do not inhibit M. bovis growth, yet are much less available in human TB diagnostic centers. Additionally, to work with the MTBC complex, a biosafety level 3 (BCL-3) hazard, stringent guidelines are required as it is an infectious agent that can be transmitted through the air that can cause a potentially deadly infection. Thus, all laboratories should be adhering to key international and national guidelines in terms of laboratory safety as this can spread the disease to laboratory workers.

With the rise of MDR-TB, there is a higher demand for drug-susceptibility testing (DST) of TB. DST can be rapidly carried out in a liquid medium and produces accurate results. One study by the WHO and the International Union Against Tuberculosis and Lung Disease (The Union), showed that when testing for drug resistance to isoniazid, rifampin, streptomycin, and ethambutol, some of the main drugs in TB treatment, sensitivities were 98.7%, 97.2%, 90.8% and 89.3%, respectively. Thus, DST is a highly recommended technique by the WHO for middle-low-income countries. However, DST is not readily available in these middle-low-income countries.

Molecular techniques such as the polymerase chain reaction (PCR) can also be used to diagnose TB in humans. Recent use of PCR techniques suggests a rapid test with relatively high sensitivity and specificity when compared to traditional techniques utilized to detect TB causing bacterial agents. A relatively new diagnostic tool, a PCR-based approach known as the Gene Xpert, can detect members of the MTBC, as well as resistance to one drug in less than two hours, with minimal requirement for technical training, the latter of which is a key limitation in traditional PCR techniques. However, Gene Xpert cannot differentiate between members of the MTBC and this test is not readily available in many global contexts because it is expensive. While the use of Gene Xpert is readily increasing
in low-income countries, such as Kenya (70 sites as of 2014), most of these are not available in rural areas and of these units may be severely underutilized\textsuperscript{76}.

Despite awareness for the global burden of TB, there is still a considerable amount of underreporting and underdiagnosing of individuals suffering from the disease. Of the 10 million estimated incident cases of TB globally, only 7.1 million of these were notified cases. Thus, over 3 million incident cases are estimated to be missed annually. Of these 7.1 million notified cases, only 57\% of these were confirmed bacteriologically, with most of the underreporting and underdiagnosing occurring in low-income countries\textsuperscript{2}.

Underreporting of TB can occur for a variety of reasons. According to one WHO report, this can be due to an inability to seek care at healthcare centres that regularly report TB cases, a lack of law or mandate for reporting, inappropriate management practices for TB, as well as geographical, financial, healthcare, or medical knowledge barriers\textsuperscript{84}.

\textbf{1.6.2 Treatment of Tuberculosis in Humans}

TB is a treatable disease when detected early and treated properly. The current, standard treatment for TB is a drug cocktail containing isoniazid, rifampicin, ethambutol, and pyrazinamide that is usually taken for six months\textsuperscript{2}. This would typically be followed by maintenance with isoniazid and rifampicin\textsuperscript{85}. The total cost of treatment is approximately $40 USD per individual. MDR-TB drug regiments are longer and can cost over $1000 USD\textsuperscript{2}. Some TB causing agents, such as \textit{M. bovis}, are naturally resistant to pyrazinamide, which can further complicate and prolong treatment. The current treatment is prolonged from six months to nine months when the patient is infected with \textit{M. bovis}\textsuperscript{85}. The recommendation of a 9-month regiment for \textit{M. bovis}, as opposed to a 6-month regiment for \textit{M. tb} decreases patient adherence and increases cost\textsuperscript{86}. An epidemiologic study investigating pyrazinamide-resistance in the United States during a period of 1999-2009 had stated that pyrazinamide resistance had significantly increased from 2.0\% to 3.3\% in a total of 79 321 MTBC cases. Of these 3.3\% of cases, only 50.3\% of cases were \textit{M. bovis}, indicating that pyrazinamide resistance is also present in other members of the MTBC\textsuperscript{87}. While some studies suggest that this just compromises treatment, other studies state that this may have fatal outcomes and higher mortality rates in patients who receive incorrect or insufficient treatment\textsuperscript{88}, although a study published by the Center for Disease Control
and Prevention (CDC) state that the culture-negative conversion rate after three months of the same treatment completed for \textit{M. tb} and \textit{M. bovis} were not significantly different\textsuperscript{89}.

### 1.7 Availability of Diagnosis and Treatment in Kenyan Healthcare Facilities

As of 2014, there were a total of five TB culture labs, 150 LED microscopes, 1,860 Acid Fast Bacillus microscopy sites, and 70 Gene Xpert sites in Kenya. There were goals to implement over 340 LED microscopes by 2017 and 250 Gene Xpert machines by 2018\textsuperscript{76}. There are no reports to follow up on this goal. There is still limited access to Gene Xpert, due to high cost for patients, poor specimen referral, doubts surrounding mortality and morbidity, low trust in clinicians, false negatives, and a lack of specimens\textsuperscript{11}. Many of these factors may contribute to Gene Xpert machines only being utilized at 20% capacity\textsuperscript{76}.

The national electronic recording system for reporting of TB cases is known as TIBU,\textsuperscript{76} but at what capacity this system is utilized is unknown. Even with these programs in place, as it stands, the National Tuberculosis, Leprosy, and Lung Disease Program (NTLD) faces a 200 million US dollar financial gap in terms of helping to reduce the burden of these diseases in the country\textsuperscript{76}.

A 2017 study concluded that Gene Xpert was available in, at the least, 14\% of Level 2 facilities (dispensaries and clinics), and at the most, 41\%\textsuperscript{90}. Yet, many people suffering from TB (42\%) sought care at level 1 facilities such as community health/traditional healer facilities, which have no capacity for TB diagnosis. In some counties, only 17\% of individuals have access to any diagnostic services\textsuperscript{90}. More diagnostic tools are available at private health clinics, yet only 22\% of patients from rural areas sought care at private HCFs\textsuperscript{90}. The National Strategic Plan for Tuberculosis, Leprosy, and Lung Health by the Kenya MOH states that while 91\% of hospitals and 78\% of health centres can diagnose TB, only 20\% of dispensaries and 15\% of health clinics have diagnostic capacity for TB. Even when diagnosed, there is a poor notification rate of the disease in Kenya from many counties. As notification rate is related inversely to poverty, many cases among the poorer population are likely missed\textsuperscript{76} and an estimated 33\% of cases are un-notified. This is likely\textsuperscript{7} due to:
1. A low number of people being sent for diagnostic detection (healthcare providers do not believe patients have TB)

2. Private sector cases not being notified

3. People living with the disease not seeking care

4. The prevalence of TB being lower than average

5. Inflated estimates of access to diagnostic tests

6. Facilities having a lack of supply or functional equipment

In Kenya, the National TB and Leprosy Programme (NTP) provides free TB treatment, and this treatment is available at over 3 000 facilities in Kenya. Yet, over 35% of patients sought treatment at dispensaries (level 2 facilities) and fewer than 40% of dispensaries can provide treatment for TB. In 27 counties across Kenya, the success rate of TB treatment is less than 88%, suggesting incorrect or insufficient treatment methods being utilized in these Counties.

1.8 Barriers to Healthcare Access in Kenyan Rural Communities

An important issue with TB treatment success is non-adherence to treatment regimens. One study investigating treatment adherence between 2005 and 2011 explored use of Community Health Workers (CHWs) and their importance in many areas of rural Kenya. In rural areas, the use of CHWs increases treatment adherence from 63% to 83%. The conclusion of this study, however, stated that CHW influence did not significantly improve treatment adherence. This may be because very few treatment centres still utilize CHWs in their treatment plans. As well, CHWs can only make twenty visits a month in rural areas, compared to 50 visits a month in urban areas, due to a variety of socio-cultural factors. Thus, their use is less robust in rural areas.

People affected with TB face many other restraints for healthcare access, one of the main barriers being the cost of diagnosis and treatment. TB patients carry the costs for various health services, transport, accommodation, as well as income and time lost due to treatment.
By visiting HCFs for diagnosis and treatment, these individuals are often unable to work, and these direct and indirect costs can accrue anywhere up to $294 US dollars in losses over the entire time of the illness. As well, 15% of these individuals may not be able to work after they are infected with the disease. Testing can also be expensive, as more than one trip is often required to be taken for diagnosis and treatment of the disease. Additionally, wait times can be up to 12 hours for diagnosis. In this study, 57% of patients had to borrow money or sell personal assets to offset direct and indirect costs of TB diagnosis and treatment.

Distances to HCFs are also a major barrier to healthcare access. A previous study in Kenya, published in 2006, modelled transport distances to government HCFs to assess the World Bank’s standards of having individuals within one hour of health services. This study estimated that 68% of people are within an hour travel of healthcare services, as opposed to 82% sampled by a separate study in 2001-2002. The 2001-2002 study failed to accommodate for the lack of straight-line travel available. These patients were assumed to be travelling in a straight line directly to each HCF, when, in reality, this is unfeasible and impossible. Thus, the coverage of healthcare is likely vastly overestimated.
1.9 Critical Need

Due to the current scenario regarding TB and ZTB in Kenyan Maasai rural communities there is a critical need to investigate current capacities, protocols and the logistics in place at rural HCFs, related to these diseases.
1.10 Objectives

1.10.1 General Objectives

The main objective of this pilot study is to collect preliminary data to describe the current capacities, logistics, and infrastructure in place for diagnosis and treatment of TB and ZTB in rural Kajiado County, Kenya. In addition, this pilot study collects data and describes the knowledge, attitudes, and practices (KAPs) of healthcare workers at these HCFs regarding TB and ZTB.

1.10.2 Specific Objectives

1) Describe techniques and capacities in place for diagnosis and screening of TB/ZTB at HCFs

2) Describe anti-microbial treatment provided and treatment capacities for TB/ZTB at HCFs

3) Quantify the number of people that seek TB/ZTB services/care at rural HCFs in Kajiado, monthly, based upon existing knowledge at HCFs

4) Describe KAPs regarding TB/ZTB among healthcare workers at these HCFs
Chapter 2: Evaluation of healthcare facilities and knowledge, attitudes, and practices for tuberculosis and zoonotic tuberculosis in Kajiado County, Kenya

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2.1 Introduction and Context

Tuberculosis (TB) has long been the leading cause of morbidity and mortality by a single infectious agent, globally. In 2019, there were 10 million incident cases of TB, and 1.4 million deaths due to the disease according to the 2020 Global Tuberculosis Report published by the World Health Organization (WHO)². The WHO estimates that a quarter of the world’s population has been infected with *M. tb* - at some point in their lives². In Kenya, TB is the fourth overall leading cause of death⁷ and Kenya itself is one of the top ten most burdened countries due to TB⁸. In 2019, there were an estimated 140 000 incident cases² of TB in Kenya but due to current gaps in screening, diagnosis, and reporting, it is likely that the current burden of TB in Kenya is much greater than reported⁶.

Bovine tuberculosis (BTB) caused by *Mycobacterium bovis* (*M. bovis*) in cattle, is a widespread disease that continues to impact cattle, and other mammals, including people, worldwide¹³ as it is estimated that cases of BTB in cattle are increasing globally¹⁷. Like human TB, there is severe underreporting for this disease, especially in countries like Kenya, where routine nationwide estimates are lacking, despite BTB being an official notifiable disease, as stated on the Kenyan Animal Diseases Act³⁰. Although the World
Organisation for Animal Health (WOAH) reports published in 2019 state that there is an absence of BTB in Kenya, literature indicates that BTB is present in Kenya. Furthermore, a report published by FAO, in 2018, evaluating the burden of zoonotic diseases on the Kenyan economy, reported that Kenya spends nearly 175.6 million US dollars to combat losses due to BTB.

ZTB caused by *M. bovis* in humans occurs mostly as the result of direct or indirect contact with infected cattle or their food products. The 2019 WHO Global Tuberculosis Report estimates 69,800 cases of ZTB in Africa, along with 9100 deaths. Despite literature regarding ZTB in Kenya being largely non-existent, due to lack of surveillance, the 2018 FAO report that examined BTB in Kenya, also included information regarding the burden of zoonotic transmission of *M. bovis* to humans. This report found that *M. bovis* caused 1,168 deaths in humans due to the transmission of the bacterial agent in 2016. This transmission is facilitated mostly through the consumption of unpasteurized milk or milk products containing *M. bovis*. The unprocessed milk trade is common in Kenya, with over 85% of milk on the market in Kenya being sold in its raw form.

Kajiado County in Kenya is home to a significant population of people from the Maasai ethnic group, who are at a high risk for TB due to a variety of socio-cultural-economic factors. The Maasai are a semi-nomadic group that practices pastoralism, thus, their livelihoods are strongly tied to their close relationship with cattle. As such, over 60% of their dietary energy is dependent on milk and milk products. The semi-nomadic nature of the Maasai community make them difficult to measure within the context of healthcare in the country. New measurement techniques, including geospatial techniques are promising, but preliminary. To compound the difficulty of measuring health in these communities, the Maasai community also faces other unique barriers to healthcare access. Some of these include misconceptions about services provided at HCFs, fear of HCFs, preference for local, traditional healing, lack of transportation, poor terrain, high indirect costs and HCFs lacking the appropriate drugs and supplies for treatment.

TB is commonly diagnosed through an acid-fast bacillus stain combined with microscopy, bacterial culture, drug-susceptibility testing, and molecular techniques, such as GeneXpert. In Kenya, these diagnostic techniques are largely unavailable or are not
sufficient for screening and diagnosis of *M. bovis*. As of 2014, there were a total of five TB culture laboratories, 150 LED microscopes, 1,860 Acid Fast Bacillus microscopy sites, and 70 Gene Xpert sites in Kenya. There were goals to implement over 340 LED microscopes by 2017 and 250 Gene Xpert machines by 2018 \(^7\) but there are no reports to follow up on this goal. Even so, most of these Gene Xpert machines are not available in rural areas and are likely only being utilized at 20% capacity \(^7\). The Lowenstein-Jensen culture mediums used to growth Mycobacterium Tuberculosis Complex (MTBC) species, contains glycerol, which inhibits the growth of certain members of the MTBC, most notably, *M. bovis*. The use of mediums containing pyruvate (i.e. Stonebrink or Middlebrook), are recommended for *M. bovis*, but are not being used routinely in National TB Programmes \(^7\). In some counties only 17% of individuals have access to any diagnostic service \(^9\). To compound this, by themselves, these techniques are insufficient to differentiate between *M. tb* and *M. bovis* \(^7\), \(^4\). This differentiation is crucial as *M. bovis* is inherently resistant to pyrazinamide, a first-line drug for the treatment of TB. Inability to differentiate these causal agents may result in fatal outcomes and higher mortality rates in patients who receive incorrect or insufficient treatment \(^8\). When seeking treatment, the Kenyan National TB and Leprosy Programme provides free treatment for TB. Even with free TB treatment available, a limited number of individuals are seeking care at HCFs with treatment \(^7\).

It is due to these unique factors regarding TB and ZTB in Kenyan Maasai rural communities that there is a critical need to investigate current capacities, protocols and the logistics in place at rural HCFs related to these diseases. Thus, in this pilot study, we aimed to describe current capacities, logistics, and infrastructure, knowledge, attitudes, and practices in place for diagnosis and treatment of TB and ZTB in rural Kajiado County, Kenya.
2.2 Materials & Methods

2.2.1 Sample Size

Non-probability sampling methods were used for this pilot study. HCFs and healthcare workers were selected purposively. Twenty-five HCFs were selected as the study units, and 3 health care workers were selected to complete the questionnaire per HCF. The 25 HCFs were previously known to Talaku- A Community Based Organization and were accessible by the workers at Talaku to conduct this pilot project. The health care workers to complete the questionnaire were selected among personnel working with TB and ZTB.

2.2.2 Inclusion Criteria for the Healthcare Facilities

The twenty-five HCFs were selected from the five major Sub-Counties in Kajiado: Kajiado North, Kajiado West, Kajiado East, Kajiado Central, and Kajiado South, in which an uneven distribution were selected for study in each of the sub-counties (based on accessibility) during a period of [January-February 2022]. Level 2-4 facilities were selected for sampling, the “levels” of which were defined by the Kenyan MOH, and the number of samples per “level” chosen due to selection criteria set by Talaku. Level 1 facilities were not selected as these healthcare facilities do not have capacity for diagnosis or treatment of TB.

2.2.3 Inclusion Criteria for the Healthcare Workers

Participants in this study were individuals working at HCFs that handled TB and ZTB cases based off previous knowledge acquired from Talaku. The three individuals selected per HCF were as follows: the facility head, known as the “in-charge” person, one TB nurse, and one laboratory technician, where applicable.

2.2.4 Questionnaire Design

All participants that were given the questionnaire were:

1. informed about the study in advance
2. provided an opportunity to provide consent (or decide not to) participate in this study
3. advised that they could withdraw from the study at any point

The questionnaire (see appendix A) was developed in collaboration with Timpiyian Leseni at Talaku, and included 26 questions divided into two parts and four sections were as follows:

**Part 1: Characteristics of the 25 HCF**

Section A. Healthcare Centre Characteristics: containing questions 1 through 5

Section B. Healthcare Access: containing questions 6 through 14

Section C. Characteristics of people receiving diagnosis and treatment for TB at Kajiado County HCFs: containing questions 15 through 20, and,

**Part 2: Knowledge, Attitudes, and Practices Among Healthcare Workers**

Section D. Healthcare worker knowledge, attitudes, and practices regarding BTB, ZTB, and TB: containing questions 21 through 26

### 2.2.5 Research Ethical Reviews and Approvals

This study was reviewed and approved by the Health Science Research Ethics Board (HSREB) through the Western Research Ethics Manager (WREM) at Western University. The project identification number is 118897 (Appendix B). Additionally, research and ethical approval was also obtained from the Kenyan Medical Research Institute (KEMRI) (PROTOCOL NO. NON KEMRI 4299) (Appendix C) and a Research License was obtained from the Kenya National Commission for Science, Technology and Innovation (NACOSTI) (Reference Number 895605) (Appendix D). The approval documentation was shared with the Local Government in Kajiado County by Talaku, which also approved the implementation of this study in Kajiado.

### 2.2.6 Statistical Analysis

Data were collected on a paper copy. Then, the data were transferred into an Excel Sheet (Microsoft® Excel®) and subsequently imported into STATA/IC 16.1 (StataCorp LLC, Texas) for statistical analysis. Descriptive analysis was conducted on continuous variables
using interquartile ranges, means, standard deviations, minimums, medians, and maximum values. For categorical variables, frequencies, counts and proportions were used. The data were displayed using standard descriptive tables. 95% confidence intervals were obtained for all the data.
2.3 Results

All selected HCFs participated in the study (100% response rate), and 69 questionnaires (92% response rate) were completed by healthcare workers in these HCFs.

2.3.1 Characteristics of the Twenty-Five Healthcare Facilities - Tuberculosis Diagnostic Capabilities

Overall, an average of 6.2 (SD 15.5) people are diagnosed for TB at these HCFs, per month (Min= 1, Median= 3, Max= 80). Only 12% [95% CI: 4-33%] of the HCFs (n=3) had access to bacterial culture as a diagnostic technique (table 1). Molecular techniques, such as GeneXpert, and microscopy using an Acid-Fast Bacillus (MAFB) stain were the most commonly utilized methods to diagnose TB among the twenty-five facilities at 76% [55-89%] and 72% [51-87%], respectively (table 1). None of the facilities had the intradermal skin test available as a screening technique (table 1).

Table 1. Availability of tuberculosis diagnostic techniques at 25 Kajiado County healthcare facilities (HCFs)

<table>
<thead>
<tr>
<th>Diagnostic Technique</th>
<th>Frequency</th>
<th>Percentage [95% Conf. Interval]</th>
</tr>
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<tbody>
<tr>
<td>Intradermal Skin Test</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Microscopy/Acid Fast Bacillus (MAFB) Stain</td>
<td>18</td>
<td>72% [51-87%]</td>
</tr>
<tr>
<td>Bacterial Culture</td>
<td>3</td>
<td>12% [4-33%]</td>
</tr>
<tr>
<td>Molecular Techniques</td>
<td>19</td>
<td>76% [55-89%]</td>
</tr>
<tr>
<td>Clinical Symptoms</td>
<td>16</td>
<td>64% [43-81%]</td>
</tr>
</tbody>
</table>
2.3.2 Characteristics of the Twenty-Five Healthcare Facilities - Tuberculosis Treatment Capabilities

Overall, an average of 5.1 (SD 3.3) patients are treated for TB at these HCFs, per month (Min= 1, Median= 5, Max= 11).

First-line TB drugs were available at 88% [67-96%] of the twenty-five HCFs, with three of the HCFs not having drugs available for treatment at the time the questionnaire was implemented. Rifampicin and isoniazid were available at 88% [67-96%] of the HCFs (table 2), while pyrazinamide and ethambutol were available at 80% [59-92%] of the HCFs (table 2). Streptomycin, a less common first-line TB drug, was only available at 12% [3.7-33%] of the HCFs (table 2).

Table 2. Availability of first-line tuberculosis drugs at 25 Kajiado County healthcare facilities (HCFs)

<table>
<thead>
<tr>
<th>Drug</th>
<th>Frequency</th>
<th>Percentage</th>
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<td></td>
<td></td>
<td>[95% Conf. Interval]</td>
</tr>
<tr>
<td>Rifampicin</td>
<td>22</td>
<td>88% [67-96%]</td>
</tr>
<tr>
<td>Isoniazid</td>
<td>22</td>
<td>88% [67-96%]</td>
</tr>
<tr>
<td>Pyrazinamide</td>
<td>20</td>
<td>80% [59-92%]</td>
</tr>
<tr>
<td>Ethambutol</td>
<td>20</td>
<td>80% [59-92%]</td>
</tr>
<tr>
<td>Streptomycin</td>
<td>3</td>
<td>12% [3.7-33%]</td>
</tr>
</tbody>
</table>

Age of people receiving treatment varied among these twenty-five HCFs. Among the 25 healthcare facilities, the average age for the younger patient receiving TB treatment at these HCFs was 9.4 years old [(SD 7.56) (Min= 1, Median= 10, Max= 30)] while the average age for the older patient receiving TB treatment among these 25 HCFs was 64 years old [(SD 14.6) (Min= 24, Median= 68, Max= 90)].
2.3.3 Barriers to TB and ZTB Diagnosis and Treatment

On average, those suffering from TB travelled 37.7 (SD 31) kilometres for diagnosis or treatment of TB (Min= 10 km, Median= 30 km, Max= 150 km), and 88% [67-96%] of the HCFs stated that distance is a barrier for TB diagnosis or treatment.

There were few costs associated with diagnosis or treatment of TB at these HCFs. These costs were 564 Kenyan Schillings, which corresponds to approximately $4.89 US dollars (Min= 0, Median= 0, Max= 10 000 Kenyan Schillings) for diagnosis and 20 Kenyan Schillings (Min= 0, Median= 0, Max= 500) which corresponds to approximately $0.17 US dollars, for treatment. Only 16% [5.8-37] (n=4) and 12% of HCFs [3.7-33%] (n=3) stated that cost is a barrier for diagnosis and treatment, respectively.

On average, the wait time for diagnosis of TB at these twenty-five HCFs was 38.5 (SD 39) hours (Min= 0.5, Median= 24, Max= 180), while the wait time for treatment also averaged out to 38.5 hours (SD 149) (Min= 0, Median= 0.5, Max= 720) (n=23). Some individuals had to wait anywhere up to 180 hours (7.5 days) for diagnosis and 720 hours (29 days) for TB treatment. Despite this, wait times were only considered a major barrier for diagnosis or treatment of TB in 24% [11-45%] (n=6) of the HCFs surveyed.

2.3.4 Knowledge, Attitudes, and Practices Among Sixty-Nine Healthcare Workers - Bovine tuberculosis

Among the sixty-nine healthcare workers that completed the questionnaire, 39% [28-51%] (n=27) stated that bovine TB does not exist within the communities receiving TB care at these HCFs (table 3), while another 28% [18-39%] (n=19) were unaware or are not sure of the presence of bovine TB within the community (table 3). A third (n=23) of the healthcare workers 33% [23-45%] were aware of bovine TB within the community (table 3).
Forty one percent [30-53%] of healthcare workers (n=28) stated that TB patients have not been in contact with any livestock (table 4).

Table 4. Healthcare worker knowledge of animal contact among TB patients (n=69)

<table>
<thead>
<tr>
<th>Patient Contact with Animals</th>
<th>Frequency</th>
<th>Percentage [95% Conf. Interval]</th>
</tr>
</thead>
<tbody>
<tr>
<td>No</td>
<td>28</td>
<td>41% [30-53%]</td>
</tr>
<tr>
<td>Yes</td>
<td>30</td>
<td>43% [32-56%]</td>
</tr>
<tr>
<td>I don’t know</td>
<td>11</td>
<td>16% [9.0-27%]</td>
</tr>
</tbody>
</table>

Forty nine of the healthcare workers, or 71% [59-81%], have not diagnosed or treated any people for ZTB (table 5), with another 19% [11-30%] (n=13) of healthcare workers unsure or unaware if ZTB had been diagnosed or treated at the HCF (table 5). There was knowledge of people being treated for ZTB from seven healthcare workers in these HCFs.
but overall, 90% (n=62) of responders did not think or were unsure of ZTB diagnosis or treatment within their HCF (table 5).

Table 5. Healthcare workers that have diagnosed or treated zoonotic tuberculosis (n=69)

<table>
<thead>
<tr>
<th>Have patients been treated with zoonotic tuberculosis?</th>
<th>Frequency</th>
<th>Percentage [95% Conf. Interval]</th>
</tr>
</thead>
<tbody>
<tr>
<td>No</td>
<td>49</td>
<td>71% [59-81%]</td>
</tr>
<tr>
<td>Yes</td>
<td>7</td>
<td>10% [4.8-20%]</td>
</tr>
<tr>
<td>I don’t know</td>
<td>13</td>
<td>19% [11-30%]</td>
</tr>
</tbody>
</table>

The majority of healthcare workers knew about the main transmission risk factors of *M. bovis*. In total, 85% [73-92%] (n=55) (table 6) and 77% [65-87] (n=50) of healthcare workers knew that consumption of raw milk/milk products, and consumption of raw meat are the main transmission risk factors for ZTB in humans, respectively (table 6). Another 62% [49-73%] (n=40) (table 6) of healthcare workers identified aerosols as a transmission risk factor for ZTB in humans.

Table 6. Healthcare worker knowledge regarding zoonotic tuberculosis transmission risk factors (n=65)

<table>
<thead>
<tr>
<th>Risk Factor</th>
<th>Frequency</th>
<th>Percentage [95% Conf. Interval]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Consumption of Raw Milk/Milk Products</td>
<td>55</td>
<td>85% [73-92%]</td>
</tr>
<tr>
<td>Consumption of Raw Meat</td>
<td>50</td>
<td>77% [65-87%]</td>
</tr>
<tr>
<td>Airborne (Aerosol)</td>
<td>40</td>
<td>62% [49-73%]</td>
</tr>
<tr>
<td>Contact with Open Wound</td>
<td>26</td>
<td>40% [29-53%]</td>
</tr>
</tbody>
</table>
Forty-seven of the sixty-nine healthcare workers responded to the survey question regarding *M. bovis* drug resistance (table 7). Only 19% [10-33%] (n=9) healthcare workers correctly identified pyrazinamide as a drug that is resisted by *M. bovis* (table 7).

### Table 7. Healthcare worker knowledge regarding zoonotic tuberculosis drug resistance (n=47)

<table>
<thead>
<tr>
<th>Drug used for Treatment</th>
<th>Frequency</th>
<th>Percentage [95% Conf. Interval]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rifampicin</td>
<td>17</td>
<td>36% [23-51%]</td>
</tr>
<tr>
<td>Isoniazid</td>
<td>7</td>
<td>15% [7.1-29%]</td>
</tr>
<tr>
<td>Pyrazinamide</td>
<td>9</td>
<td>19% [10-33%]</td>
</tr>
<tr>
<td>Ethambutol</td>
<td>8</td>
<td>17% [8.6-31%]</td>
</tr>
</tbody>
</table>
2.4 Discussion

Due to the current scenario of TB and ZTB in Kenyan Maasai rural communities, there is a critical need to look at current capacities, protocols and the logistics in place at rural HCFs in order to plan and implement actions to reduce the burden and improve the diagnosis and treatment of TB and ZTB. In Kajiado County, it is unclear how many cases of TB are zoonotic in nature; thus, the true burden of ZTB is unknown, largely due to lack of proper surveillance aiming to identify *M. bovis* as the causal agent of ZTB in humans\textsuperscript{15}. This is of concern as socio-cultural practices specific to the Maasai Community\textsuperscript{60-62}, especially their close relationship with animals as pastoralists, put them at higher risk for ZTB.

Globally, there is increasing evidence to suggest that an important amount of human TB cases are caused by *M. bovis*\textsuperscript{29, 36, 37}. One study, conducting a systematic literature search in 2001 found that in Tanzania\textsuperscript{38}, TB caused by *M. bovis* may account for anywhere up to 37.7\% of cases. Four separate regional studies conducted in Tanzania that used molecular techniques and bacterial culture stated that an average 30\% of human TB cases could be caused by *M. bovis*. Kenya neighbors Tanzania, which also has a large ethnic Maasai population with similar socio-cultural practices\textsuperscript{68}. Mobility of people between these two countries does occur. Despite this information, further studies investigating *M. bovis* have not been conducted in this area. With the Maasai people’s close relationship with livestock, lifestyle choices and dietary requirements\textsuperscript{60-62}, it is crucial that research is further conducted in this region in terms of diagnosing TB/ZTB, as proper diagnostic techniques are essential for these communities.

2.4.1 Characteristics of the 25 Healthcare Facilities

Responses among different health care workers within a HCF were consistent in the vast majority of the 25 HCFs. For each question asked, and in the few instances where a discrepancy was observed, the most extreme value reported was utilized for the analysis. For example, in one facility, the three responders answered that 80, 76, and 75 individuals were diagnosed per month; in this case, we used 80 for the analysis. The rationale for the
approach used in this scenario was to capture the ‘maximum’ number of people seeking TB services for communications with field stakeholders that will plan and design strategies and policies to address needs in these facilities. The minor discrepancies observed were deemed expected as part of asking questions on KAPs and did not deviate nor affect the results of this study. These discrepancies were communicated and shared internally with our local collaborators in Kajiado county.

We assessed TB and ZTB diagnostic capacities as a part of this study. Molecular techniques, such as Gene Xpert, as well as AFB microscopy were available at a majority of these HCFs (76% and 72%, respectively). While Gene Xpert equipment was present in 76% of these 25 HCFs, this technology is likely underutilized due to low coverage and inaccessibility. Additionally, GeneXpert is not able to differentiate between *M. bovis* and *M. tb*. Microscopy also cannot differentiate between causal agents of TB in humans, thus, while microscopy is readily available, the availability of microscopy alone is insufficient to diagnose ZTB.

Interestingly, only three of the surveyed HCFs had access to bacterial culture for diagnosis of TB, despite this being the “gold standard” for TB diagnosis and members of the MTBC. A majority of reports detecting TB in Kenya are not utilizing the correct bacterial culture medium to detect *M. bovis*, when it is used. The standard Lowenstein-Jensen bacterial culture medium used for TB diagnosis in humans, containing glycerol as a growth medium, inhibits growth of *M. bovis*. Bacterial culture containing pyruvate (i.e Stonebrink or Middlebrook), more commonly used to diagnose TB in cattle, would need to be implemented to understand the true burden of *M. bovis* induced ZTB in Kajiado County. The last nation-wide prevalence survey utilized a Lowenstein-Jensen Medium, containing glycerol, thus cases of ZTB are likely missed. Due to the lack of availability of the proper medium to detect *M. bovis* nationally, it would be beneficial to ensure that the three facilities with access to bacterial culture in Kajiado County are using Stonebrink or Middlebrook mediums.

A majority of the HCFs (n=22) had first-line TB drugs available for treatment, and only three of the HCFs did not have any drugs available for TB treatment at the time the
questionnaire was implemented in the field. These three HCFs obtain drugs for treatment from other facilities once a person is diagnosed with TB (T. Leseni, personal communications, April, 2022). Thus, all 25 HCFs have the capacity to treat TB when required (T. Leseni, personal communications, April, 2022).

On average, on a monthly basis, 6.2 cases are diagnosed, and 5.1 are treated for TB at these HCFs. Thus, there may be a substantial gap between people diagnosed for TB and treated for TB that exists in rural Kajiado County. The maximum number of diagnosed individuals with TB at one HCF was 80 per month, while the corresponding number of individuals treated at that facility was 11. On average, more people were diagnosed with TB at these HCFs than treated for TB. While a majority (88%) of the HCFs have capacity to treat TB cases, antimicrobial treatments are not being readily distributed based on the results of this study, despite low costs associated with treatment due to the National TB and Leprosy Programme in Kenya. Additional research should explore the discrepancy between individuals diagnosed and those treated for TB. According to a 2017 report, there were 844 bacteriologically confirmed cases of TB in Kajiado County alone. This number is likely an underestimate due to the large amount of unreported TB cases that occur in Kenya due to distance or other key barriers to healthcare access that may explain low treatment numbers relative to diagnosis.

2.4.2 Barriers to Healthcare Access for TB and ZTB Services in Kajiado

There are limited studies discussing distance as a barrier to healthcare access in Kenya, and even fewer that explore TB and ZTB specifically. We found that the average distance travelled for diagnosis or treatment of TB was nearly 38 kms, which would likely present a challenge for those patients travelling by foot or small motorized vehicles like motorcycles, especially considering the poor road conditions that hindered access to our enumerators in the months of data collection (T. Leseni, personal communications, April, 2022). Thus, it is not surprising that 88% of HCFs identified distance as a major barrier to healthcare access. Studies that exist examining distance travelled for patients to HCFs to receive care are underestimating the true challenges patients face due to distance travelled. The Euclidean distance model is commonly used to look at the proportion of a population
within an hour of health services. This model only considers straight-line travel, and the use of the closest health services, a reality that likely vastly overestimates availability of health services based upon distance. A previous study in Kenya evaluating distance as a barrier for HIV/AIDS, TB, and malaria estimated average distances travelled by patients to government health services based on a model different from the traditional Euclidean model. Over six million Kenyans were incorrectly placed within one hour of effective healthcare. Despite this, the study also did not explore travel to private or smaller-level HCFs. The results of these studies align with the results of our study that distance to a HCF presents major challenges in receiving treatment for diseases such as TB and ZTB.

Most (73%) HCFs in our study did not state wait times as a major barrier to people’s healthcare. The average wait time for diagnosis or treatment was approximately 38.5 hours. The maximum wait times were approximately 7.5 days for diagnosis and 29 days for treatment. It is clear there is a gap between diagnosis and treatment, corroborated by the results in this study. The average wait times of our study were much longer than the twelve hours reported in a study conducted in 2008. It is important to note that this question was asked to healthcare workers within the facilities and may not be representative of the views and experiences of people seeking health services. In the future, healthcare access can be assessed from the perspective of the people seeking services, rather than the healthcare workers.

### 2.4.3 Healthcare Worker Knowledge, Attitudes, and Practices Surrounding TB, BTB, and ZTB

Healthcare worker KAPs were also assessed as part of this pilot study, focusing on key topics related to the prevention, diagnosis, and treatment of TB and ZTB, and overall knowledge on BTB. Healthcare workers stated that BTB either does not exist (39%) or they were unaware of the existence of BTB (28%) within the community. A FAO report, published in 2018, reported that the Kenyan economy reported losses of 175.6 million US dollars due to BTB, thus, this is an area that also needs more focus, as there is a gap in knowledge between healthcare workers and the communities which they work in. At the time of the project implementation, there were no scientific manuscripts found that
investigated BTB in Kajiado County, but there are several studies that indicate that BTB does exist within cattle herds in Kenya\textsuperscript{18,22}. Thus, future work should focus on prevalence studies in Kajiado County specifically.

It is worth noting that in our study, 43\% of healthcare workers had no knowledge of livestock contact within the TB cases. Given the close relationship of the Maasai people and their livestock as pastoralists, there is a disconnect between healthcare workers and the Maasai Community. Although there is still work to be done in understanding the true link between pastoralism in the Maasai community and ZTB, it is surprising to see that less than half of the healthcare workers are aware of livestock contact. Based on knowledge shared by collaborators at Talaku, this could be influenced by the fact that healthcare workers are not of Maasai origin and have limited knowledge of the Maasai lifestyle (T. Leseni, personal communications, April, 2022). Understanding the sharing of macro and micro-environments between the Maasai people and their livestock represents a key factor in further understanding of ZTB transmission within the community. Furthermore, healthcare should account for the traditional cultural practices of the Maasai people. As a result, there needs to be further investigation into zoonotic diseases, such as ZTB, which are exacerbated by contact with livestock.

Preventing BTB in cattle is the most important step in reducing risk for human transmission\textsuperscript{13,39,86}. A majority of healthcare workers correctly identified the main transmission risk factors causing ZTB in humans, this being consumption of raw milk/milk products (85\%), and consumption of raw or undercooked meat (77\%). Thus, a majority of responders had understood the animal-human link in terms of transmission risk. Despite this, healthcare workers have likely not diagnosed or treated people for ZTB based on previous diagnostic capability results conducted in our study. This is compounded with the fact that there is currently no surveillance for this disease in Kajiado County. Thus, these results are surprising, especially considering the lack of knowledge in regards to other knowledge, attitudes and practices among healthcare workers based on results of our study.

For example, only 19\% (n=9) of healthcare workers identified pyrazinamide as a drug that \textit{M. bovis} is inherently resistant to. Pyrazinamide resistance can complicate and prolong treatment\textsuperscript{88}, although a study published in 2017 evaluating TB and ZTB cases in the United
States, suggests that there is no significant difference between treatment containing pyrazinamide or without pyrazinamide in TB cases caused by *M. bovis* and may result in similar patient outcomes\(^9\). Despite this, the recommendation of a 9-month regimen, opposed to a 6-month regimen for *M. tb* decreases patient adherence and increases cost\(^6\) and may be associated with higher mortality rates\(^8\). It is clear that more work needs to be done in regard to the lasting impacts due to inappropriate usage of first-line drugs and health outcomes from ZTB patients who incorrectly take pyrazinamide in treatment. In our study, when asked about drug resistance, only forty-seven responders had responded to this question on the survey. This represents the question that received the lowest number of responses, thus, additionally, more knowledge surrounding pyrazinamide resistance and *M. bovis* needs to be available at rural HCFs.

Several key findings of this study describe the importance of understanding key socio-cultural practices of the unique Maasai Community, and how these factors influence the health of people suffering from TB and ZTB within areas such as rural Kajiado County. There is still much work that needs to be completed so that healthcare workers are aware and have sufficient knowledge on how these socio-cultural practices influences the link between disease and members of the Maasai Community.
Chapter 3: Overall Conclusions

3.1 Significance, the One Health Approach, and Future Directions

This project was made possible due to the extensive and active involvement of Talaku- a Community Based Organization, their leader, Timpiyian Leseni, and her workers. Talaku is a local organization that aims to provide resources and knowledge to the Maasai regarding TB and ZTB. Timpiyian Leseni herself is a Maasai woman and a ZTB survivor and has been active in advocating and creating awareness for TB and ZTB. This project is a prime example that outlines the importance of collaboration with local stakeholders such as Talaku, local healthcare workers and local and national TB coordinators who are primarily impacted by health issues such as TB and ZTB. It is this collaboration that allowed this study to be the first of its kind that investigated TB and ZTB in rural Kajiado County, focusing on the diagnostic and treatment capacities, as well as KAPs among these HCFs. This project highlights the need for more work on TB and ZTB to be implemented not only in Kajiado County, but in other rural areas of Kenya and other developing countries. Research for ZTB, a largely neglected disease in rural areas, is critically important to understand the true burden of the disease on a global scale. This type of research cannot be completed without the help and expertise of local organizations and individuals.

Additionally, this project touches upon the ever-important human-animal-socio-cultural-economic connection that all contributes heavily to the current burden of TB and ZTB in rural Kenya. The need for a collaborative and multisectoral approach across multiple disciplines to properly tackle human, animal and environmental health issues is quickly and widely being recognized. In order to ensure the correct types of diagnostic techniques and treatment for TB and ZTB are available at rural HCFs, there needs to be collaboration with major health and governmental organizations within Kenya. Surveillance and control of BTB in cattle herds are also an essential next step. Due to the pastoral nature of the Maasai community, there is a unique relationship between cattle and humans, exacerbating the issue of zoonotic transmission of pathogens between cattle and humans. Providing
better diagnostic techniques for BTB in cattle in these rural areas will be the most important step to reducing the burden of ZTB in humans.

Along with proper diagnosis and treatment, a proper knowledge base for diagnosis, treatment, and key socio-cultural factors in rural areas, especially in the case of ZTB, are needed. There are key gaps in knowledge that exist between healthcare workers and those suffering from TB and ZTB especially when it comes to the awareness of animal contact and the importance of BTB. There needs to be key communication and knowledge transfer for healthcare workers to fully understand key connections between BTB and largely Maasai TB cases. To compound this, work needs to be done within these communities, through local stakeholders, such as Talaku, to further understand key transmission risk factors due to social practices within these communities.

The data used in this study were collected purposively from accessible HCFs working with TB. Further work must be completed to understand the full scope of capacities, logistics, and KAPs among all (other) rural HCFs in Kenya. Future work should also focus on understanding KAPs among people seeking care at these HCFs to further understand barriers and experiences from those seeking care as it pertains to healthcare access. Furthermore, detailed focus group sessions could target those healthcare workers who have knowledge of ZTB diagnosis to further understand the current gaps in knowledge that exist among healthcare workers.

Despite being mentioned in the third edition of the United Nations Sustainable Development Goals, there is still inadequate attention on ZTB as part of these goals\textsuperscript{95}, thus, ZTB literature is still limited\textsuperscript{47} despite the WHO classifying ZTB as a neglected zoonotic disease\textsuperscript{96}. Beginning in March of 2014, international meetings between key global organizations commenced as the issues surrounding ZTB were discussed to be addressed\textsuperscript{19}. As a result of these meetings, in 2017, a Roadmap for ZTB was published in collaboration with the WHO, the OIE, the Union, and FAO. The completed project in Kajiado County is in direct response to the guidelines and priorities set out by this important document. This roadmap listed ten main priorities required to reduce the burden of ZTB globally, using the
One Health approach to reach a list of milestones, cumulating with the stop of the TB epidemic by 2030. These ten priorities were listed under three main pillars:

A) Improve the Scientific Evidence Base:

1. Collect and report more complete and accurate data
2. Improve diagnosis in people
3. Address research gaps

B) Reduce Transmission at the Animal-Human Interface

4. Ensure safer food
5. Improve animal health
6. Reduce the risk to people

C) Strengthen Intersectoral and Collaborative Approaches:

7. Increasing awareness, engagement, and collaboration
8. Developing policies and guidelines
9. Implementing joint interventions
10. Advocating for investment.

As a result of these global and local efforts, in Uganda, ZTB was included for the first time in the National Strategic Plan for Tuberculosis for the period of 2020/2021-2024/2025. We are confident that this pilot project in Kajiado, Kenya will contribute in a similar manner to include ZTB in official, and important government documents and TB programmes. This project has also contributed significantly to the available scientific evidence and has strengthened intersectoral and collaborative approaches.

Overall, to better prevent, diagnose, and treat TB and ZTB among the Maasai community in Kenya, a multi-disciplinary and multi-sectoral approach is required, in which collaboration among stakeholders in rural Kajiado County is utilized. Only in this way can the current knowledge base between TB and ZTB be utilized to its full capacity. A One
Health approach is being increasingly recognized internationally to tackle health issues, including frameworks published by the Global Fund (2021)\(^7\), and at the One Health workshop that occurred on June 2\(^{nd}\), 2022, in preparation for the G20 meeting (F Olea Popelka, personal communications, June 2022). The results of this project have been shared with various local, regional, and national government stakeholders that will cement the importance of a multi-disciplinary, One Health approach to solving major health issues such as those presented by TB and ZTB, going forward. We are confident that the data collected from these rural HCFs, will positively contribute to 1) increasing awareness for TB and ZTB, diseases that continue to be neglected, especially in marginalized rural communities, and 2) providing new information to local stakeholders in Kenya, thereby providing knowledge to guide further work towards addressing the challenges posed by TB and ZTB, and improve HCF services and capabilities provided to people in these rural and marginalized communities in Kenya.

### 3.2 Conclusions

This pilot study collected initial data regarding diagnostic capabilities and logistics among HCFs and KAPs among healthcare workers in rural Kajiado County. Through this study, valuable and novel information was collected that shed light on the various gaps in knowledge that exist and current TB and ZTB diagnostic/treatment capabilities. The results of this study can and will be used to encourage future work and education in collaboration with local community organizations such as Talaku, while also collaborating with Local County and National TB programmes, individuals within the Maasai Community, healthcare workers at these HCFs, as well as major animal health sectors to reduce the burden of BTB in cattle herds that will begin to clear the burden that we face due to ZTB. There needs to be a greater priority given to the key diagnostic and treatment challenges that marginalized communities, such as the Maasai, face due to ZTB; a disease that continues to impact health and the economy in these communities. Future work implemented, aiming to improve the prevention, detection, and control of TB and ZTB must continue to focus on local knowledge, attitudes, and practices to understand local realities at the human-animal-environmental interface and account for socio-cultural-economic factors relevant to the local context. Work in these areas must include local
stakeholders and consider strategies that are effective under local circumstances. Education targeting prevention of TB and ZTB needs to be tailored within the context of the Maasai community. Future projects of this nature need to also consider the knowledge, attitudes, and practices from members of the community seeking TB and ZTB care, especially those people actively and directly working with livestock. It is evident through this project, that a collaborative, One Health approach is necessary to prioritize and complete the goals set out by the 2017 Roadmap and the goals of the WHO to eliminate the burden of TB globally by the year 2035.
References or Bibliography


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Appendix A: Approved Questionnaire Implemented at 25 Healthcare Facilities

Evaluation of healthcare facilities and services provided for tuberculosis and zoonotic tuberculosis within the rural Maasai Community in Kajiado County, Kenya

The purpose of this pilot study questionnaire is to evaluate current capacities, logistics, and infrastructure in place for diagnosis and treatment of TB and ZTB in rural Kajiado County, Kenya. In addition, the KAP of healthcare workers at these healthcare facilities will be evaluated. The information you provide will help us to understand where where gaps of knowledge in terms of diagnosis, treatment, and other practices are in your local healthcare facilities in regards to TB and ZTB. Your opinion matters.

Section A. Healthcare Centre Characteristics

1) Participant Number (to be assigned by researcher):
____________________________________________________________________

2) How many cases of tuberculosis (TB) are received by the healthcare centre per month? _____

3) Currently, how is TB detected at this healthcare facility?

   i) Tuberculosis Skin Test                   Yes ☐    No ☐
   ii) Microscopy/Acid Fast Bacillus (AFB) stain Yes ☐    No ☐
   iii) Bacterial culture                    Yes ☐    No ☐
   iv) GeneXpert/molecular techniques        Yes ☐    No ☐
   v) symptomatic                            Yes ☐    No ☐
4) Are there local laboratories used to submit sputum samples? Yes ☐ No ☐
   If the answer was yes to question 4, please answer the next questions (4.a, 4.b, 4.c)

4.a) To which laboratory are sputum samples submitted?
   ______________________________________

4.b) How long (time) does it take from the collection of sputum for the sputum sample to arrive at the laboratory?
   ______________________________________

4.c) How are sputum samples handled for storage and transportation?
   ______________________________________

5) Are antimicrobial drugs available for TB treatment at this healthcare facility?
   Yes ☐ No ☐
   If the answer is yes to question 5, which drugs are available?
   i) Rifampicin Yes ☐ No ☐
   ii) Isoniazid Yes ☐ No ☐
   iii) Pyrazinamide Yes ☐ No ☐
   iv) Ethambutol Yes ☐ No ☐
   v) Streptomycin Yes ☐ No ☐
   vi) Other (specify) ☐
   ______________________________________

Section B. Healthcare Access

6) Is distance to this healthcare centre a relevant patient barrier for TB diagnosis or treatment?
   Yes ☐ No ☐

7) What is the average distance (km) people travel to attend this healthcare centre?
   _________
8) What is the typical cost for diagnosis of tuberculosis?
____________________________

9) What is the typical cost of treatment of tuberculosis using anti-microbial drugs?
_______

10a) Are people less likely to receive diagnosis due to cost? Yes □ No □
10b) Are people less likely to receive treatment due to cost? Yes □ No □

11) How many people affected by TB are typically assigned to one healthcare worker at a time? ____________

12) On average, how long are wait times for diagnosis of TB? _____________________

13) On average, how long are wait times for acquiring treatment for TB?
____________

14) Are people less likely to receive diagnosis/treatment due to wait times? Yes □ No □

Section C. Characteristics of people receiving diagnosis and treatment for tuberculosis at Kajiado County healthcare facilities

15) How many people are diagnosed on a monthly basis with TB?
____________

16) What proportion of people diagnosed with TB are i) men: _____ ii) women: ______

17) What proportion of people diagnosed with TB are children (<15 years old)
__________

18) How many people are treated on a monthly basis for TB? ___________________
19i) What is the youngest age of a person receiving diagnosis or treatment? 
_________________

ii) What is the oldest age of a person receiving diagnosis or treatment? 
_________________

20) Does TB present as an extra-pulmonary disease (outside of the lungs)? Yes ☐ No ☐

Section D. Healthcare worker knowledge, attitudes, practices regarding bovine tuberculosis, zoonotic tuberculosis, and tuberculosis.

D.1 Bovine Tuberculosis (in animals)

21) Are you aware of the presence of bovine TB within local communities? 
Yes ☐ No ☐ I don’t know ☐

22) Are you aware of any direct contact with livestock (cattle, sheep, goats) by people diagnosed with TB? Yes ☐ No ☐ I don’t know ☐

D.2 Zoonotic Tuberculosis

23) What is zoonotic TB?
________________________________________________________________________
____
________________________________________________________________________
____
________________________________________________________________________
____

24) Have you diagnosed or treated people with zoonotic TB? Yes ☐ No ☐ I don’t know ☐

25) Which one of the following are risk factors for the transmission of zoonotic TB from animals to humans?

   i) Consumption of Raw Milk/Milk products? Yes ☐ No ☐

   ii) Consumption of Raw Meat? Yes ☐ No ☐

   iii) Aerosol transmission? Yes ☐ No ☐
iv) Coming into contact through open wounds? Yes □ No □

v) Other

26) *Mycobacterium bovis* (cause of zoonotic TB) is resistant to which of the following drugs?

i) Rifampicin □ Yes □ No □

ii) Isoniazid □ Yes □ No □

iii) Pyrazinamide □ Yes □ No □

iv) Ethambutol □ Yes □ No □

v) Streptomycin □ Yes □ No □

vi) other □
Appendix B: Ethical approval from Western University HSREB

Date: 6 July 2021
To: Dr. Francisco Oleta Pepelka

Project ID: 118897

Study Title: Evaluation of healthcare facilities and services provided for tuberculosis and zoonotic tuberculosis within the rural Mauesa Community in Kajiado County, Kenya

Application Type: HSREB Initial Application

Review Type: Delegated

Meeting Date / Full Board Reporting Date: 20 July/2021

Date Approval Issued: 06 Jul/2021

REB Approval Expiry Date: 06 Jul/2022

Dear Dr. Francisco Oleta Pepelka

The Western University Health Science Research Ethics Board (HSREB) has reviewed and approved the above mentioned study as described in the WREIM application form, as of the HSREB Initial Approval Date noted above. This research study is to be conducted by the investigator noted above. All other required institutional approvals and mandated training must also be obtained prior to the conduct of the study.

Documents Approved:

<table>
<thead>
<tr>
<th>Document Name</th>
<th>Document Type</th>
<th>Document Date</th>
<th>Document Version</th>
</tr>
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<tr>
<td>1.10 Research Protocol HCF_Resubmission 1 Clean</td>
<td>Protocol</td>
<td>22 Jun/2021</td>
<td>2</td>
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<tr>
<td>12.9 Telephone Script for healthcare worker recruitment HCF_Resubmission 1, clean</td>
<td>Telephone Script</td>
<td>22 Jun/2021</td>
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<tr>
<td>Healthcare Facility Questionnaire, HCF_Resubmission 2 Clean</td>
<td>Paper Survey</td>
<td>30 Jun/2021</td>
<td>3</td>
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</table>

Documents Acknowledged:

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<th>Document Name</th>
<th>Document Type</th>
<th>Document Date</th>
<th>Document Version</th>
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<tbody>
<tr>
<td>Budget for TALAKU Healthcare Facility Survey Project</td>
<td>Study Budget</td>
<td>05 May/2021</td>
<td>Final</td>
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<tr>
<td>Reference List HCF</td>
<td>References</td>
<td>05 Jun/2021</td>
<td>2</td>
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</tbody>
</table>

No deviations from, or changes to, the protocol or WREIM application should be implemented without prior written approval of an appropriate amendment from Western HSREB, except when necessary to eliminate immediate hazard(s) to study participants or when the change(s) involves only administrative or logistical aspects of the trial.

REB members involved in the research project do not participate in the review, discussion or decision.

The Western University HSREB operates in compliance with and is constituted in accordance with, the requirements of the TriCouncil Policy Statement: Ethical Conduct for Research Involving Humans (TOPS 2), the International Conference on Harmonisation Good Clinical Practice Consolidated Guideline (ICH-GCP); Part C; Division 5 of the Food and Drug Regulations; Part 4 of the Natural Health Products Regulations; Part 3 of the Medical Devices Regulations and the provisions of the Ontario Personal Health Information Protection Act (PHIPA 2004) and its applicable regulations. The HSREB is registered with the U.S. Department of Health & Human Services under the IRB registration number IRB 00000540.

Please do not hesitate to contact us if you have any questions.

Sincerely,
Appendix C: Ethical approval from Kenyan Medical Research Institute (KEMRI)

KENYA MEDICAL RESEARCH INSTITUTE

P.O. Box 54840-00200, Naïrobí, Kenya
Tel: (254) 7722541, 7713349, 0722-206901, 07733-400003, Fax: (254) (020) 3720030
Email: director@kemri.org, info@kemri.org, Website: www.kemri.org

KEMRI/RES/7/3/1

TO: DR. FRANCISCO OLEA POPELKA,
PRINCIPAL INVESTIGATOR,
UNIVERSITY OF WESTERN ONTARIO.

Dear Sir,

RE: PROTOCOL NO. NON KEMRI 4299 (RESUBMISSION OF INITIAL SUBMISSION): EVALUATION OF HEALTHCARE FACILITIES AND SERVICES PROVIDED FOR TUBERCULOSIS AND ZOONOTIC TUBERCULOSIS IN KAJIADO COUNTY, KENYA (VERSION 2.0 DATED 08 OCTOBER 2021)

Reference is made to your letter dated October 08, 2021. The KEMRI Scientific and Ethics Review Unit (SERU) acknowledges receipt of the revised study documents on October 14, 2021.

This is to inform you that the Committee notes that the issues raised during 315th Committee B meeting of the KEMRI Scientific Ethics Review Unit (SERU) held on September 22, 2021 have been adequately addressed.

Consequently, the study is granted approval for implementation effective this day, October 29, 2021 for a period of one (1) year. Please note that authorization to conduct this study will automatically expire on October 28, 2022. If you plan to continue with data collection or analysis beyond this date, please submit an application for continuation approval to SERU by September 16, 2022.

Please note that only approved documents including (informed consents, study instruments, Material Transfer Agreement) will be used. You are required to submit any proposed changes to this study to SERU for review and the changes should not be initiated until written approval from SERU is received. Any unanticipated problems resulting from the implementation of this study should be brought to the attention of SERU and you should advise SERU when the study is completed or discontinued.

Prior to commencing your study, you will be expected to obtain a research license from National Commission for Science, Technology and Innovation (NACOSTI) https://pris.nacosti.or.ke and also obtain other clearances needed.

Yours faithfully,

PROF. CHARLES OMONYO,
THE ACTING HEAD,
KEMRI SCIENTIFIC AND ETHICS REVIEW UNIT.

In Search of Better Health
Appendix D: Ethical approval from National Commission for Science, Technology & Innovation (NACOSTI)

This is to certify that Dr. Francisco Javier Olasa Popokha of Western University, has been licensed to conduct research in Kajiado County, Kenya for the period ending 20/November/2022. The research is on the topic: Evaluation of healthcare facilities and services provided for tuberculosis and atypical tuberculosis in Kajiado County, Kenya for the period ending 20/November/2022.

License No: NACOSTI/P/21/14414
Applicant Identification Number: 895605

NOTE: This is a computer generated license. To verify the authenticity of this document, scan the QR code using a QR scanner application.

Ref No: 895605
Date of Issue: 20/November/2021
Director General
Curriculum Vitae

Name:  Joel Zhang

Post-secondary Education and Degrees:
The University of Western Ontario
London, Ontario, Canada
2016-2020, BMSc

Honours and Awards:
Dr. Frederick Winnet Luney Graduate Research Award
2021-2022

One Health Graduate Research Award
2021-2022, 2020-2021

Related Work Experience:
Teaching Assistant
The University of Western Ontario
2021-2022, 2020-2021