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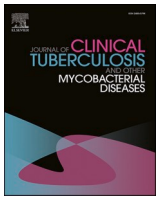

Factors associated with tuberculosis drug resistance among presumptive multidrug resistance tuberculosis patients identified in a DRTB surveillance study in western Kenya

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ABSTRACT

Multidrug-resistant tuberculosis (MDR-TB) is caused by *M. tuberculosis* (*Mtb*) with resistance to the first-line anti-TB medicines isoniazid (INH) and rifampicin (RIF). In Western Kenya, there is reported low prevalence of drug resistant strains among HIV tuberculosis patients, creating a need to determine factors associated with drug resistance patterns among presumptive MDR-TB patients. To determine factors associated with drug resistance patterns among presumptive MDR-TB patients in western Kenya. Three hundred and ninety (390) sputum sample isolates from among presumptive multidrug TB patients, were analyzed for TB drug resistance as per Ministry of Health (MoH) TB program diagnostic algorithm. Frequency and percentages were used to summarize categorical data while median and interquartile range (IQR) were used for continuous data. Multivariable logistic regression was carried out to identify factors associated with TB drug resistance. Out of 390 participants enrolled, 302/390 (77.4 %) were males, with a median age of 34 years. The HIV-infected were 118/390 (30.3 %). Samples included 322 (82.6 %) from presumptive patients, while 68/390 (17.4 %) were either lost to follow-up patients, failures to first-line treatment or newly diagnosed cases. A total of 64/390 (16.4 %) of the isolates had at least some form of drug resistance. Out of 390, 14/390 (3.6 %) had MDR, 12 (3.1 %) were RIF mono-resistance, 34 (8.7 %) had INH, while 4 (1 %) had ethambutol resistance. The category of previously treated patients (those who received or are currently on TB treatment) had a 70 % reduced likelihood of resistance (aOR: 0.30; 95 % CI: 0.13–0.70). In contrast, older age was associated with an increased likelihood of resistance to INH and RIF, with an adjusted odds ratio of 1.04 per year (95 % CI: 1.00–1.08). Prompt MDR-TB diagnosis is essential for appropriate patient care, management, and disease prevention and control. We recommend active surveillance on drug resistant TB in these regions to detect drug resistance patterns for rapid disease management.

1. Introduction

Tuberculosis (TB) an infectious disease, is both curable and preventable, but continues to wreak havoc as a public health threat, thus second leading global mortality cause, after COVID-19, but twice as much deaths as HIV/AIDS [61]). In 2022, the WHO said that 10.6 million people became ill of TB, 7.5 million were newly diagnosed of TB while 450,000 developed MDR-TB, however, unfortunately, 1.30 million died from TB. Tuberculosis is primarily a disease of the lungs, caused by very closely related mycobacteria belonging to the Mycobacterium tuberculosis complex (MTBc) [7]. The presence of the drug-resistant gene in M. tuberculosis is the major challenge of controlling TB [24]. Bacteria resistant to isoniazid (INH) and RIF, the 2 most effective first-line anti-TB drugs[73], has been an increasing challenge to global TB control in recent years[43,20]. Recent reports indicate that TB is still the most common cause of death from a single infectious pathogen[10].

Despite the global TB control interventions, it continues to be a public health concern and is currently ranked among the top three killer

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infectious diseases worldwide [47]. Among the TB and human immunodeficiency virus (HIV) co-infected patients, a high mortality of TB disease was predominantly observed [72]potentially since both diseases poses a lethal combination, towards each other's progress[49]. Unfortunately, HIV co-infection with MDR-TB did not get key attention until recently in East Africa, where the TB prevalence and risk factors are highest [63]. While TB is curable [70], patients with MDR-TB are practically challenging to treat by standard first-line TB drugs, i.e. Isoniazid (INH) and Rifampicin (RIF), [12], thus it remains a source of infection for a longer time [68]. Due to the spread of MDR-TB, TB may once again become an "incurable disease" [65]. This may be due to TB bacteria acquiring mutations because of specific genes that are associated with drug resistances, [62]. Tuberculosis bacteria's fitness may be impaired by administered resistance antibiotics,[9,35]. The drug resistance TB strains could eventually acquire some mutations known as compensatory mutations, which end up aiding their survival and ultimately drug resistance [65]. Gagneux et al., further suggests that drug-resistant TB may be transmitted as pan-sensitive strains [17].

While TB prevalence surveys has been carried out across the globe, it has been reported that in Africa, only half of the countries have carried out at least a formal TB prevalence surveys by 2018 [23]. Of the countries that have conducted surveys, both Nigeria and South Africa had a combination of 42 % cases of drug resistance from an estimated combined 92,829 cases [23]. MacNeil reports that Africa records a mortality rate of either 21 % for MDR-TB, and 43 % for extensively-drug-resistant (XDR)-TB respectively [34]. Both MDR- and XDR-TB-related morbidity and mortality occurs predominantly in middle- and low-income countries, such as in East Africa [34]. This phenomenon could be possibly attributed to poor adoption of policies on new diagnostic tools and lack of implementation strategies, possibly leading to increased drug-resistant tuberculosis in the continent [23].

In East Africa, Kenya is among the 30 high MDR-TB burden countries [3])[32]and is ranked 13th among 30 high TB burden countries globally [60]. The prevalence of resistance to both first- and second-line TB drugs has been reported to be 1.5 % in Kenya [46]while her neighbours, Uganda and Tanzania had a MDR-TB prevalence of 2.3 % and 1.1 %, respectively [53]. The Kenya's national TB/HIV co-infection rate stands at 23 %, while in western region, where the study was carried out, it ranges from 10 % to 56 % [45]. In other studies carried out, some factors have been shown to be associated with MDR-TB risks [5]. These factors include low socio-economic status, anti-TB treatment history, HIV co-infection, sex, other comorbidities, alcohol use and malnutrition [5]. Additional studies have further identified them as critical risk factors associated with the development of MDR-TB in the region [4],(Gyamfi–Gyimah, 2019).

In 2019, 56 % of global TB cases were among men [10]. Other studies show that the burden of TB by sex ratio is 1:1.5 in women to men in low- and middle-income countries [22]. Sex has also been shown to be a biological risk factor for TB infection [60], since males were predictors of poor treatment outcomes [25]while women were known to represent treatment success [67].

Characterizing sex differences in drug resistance tuberculosis/HIV immunodeficiency virus (DR-TB/HIV) co-infection is important in sub-Saharan Africa where women have a disproportionally higher prevalence of HIV and a higher risk of TB/HIV-related mortality than men [6]. Identifying sex differences can guide implementation of sex-specific interventions to improve DR-TB treatment outcomes [6].

There are several tests available for diagnosis of TB depending on the type of TB suspected, whether pulmonary or extrapulmonary. These tests range from sputum collection and testing using various diagnostic platforms such as phenotypic (microscopy and cultures) or genotypic techniques [GeneXpert Mtb RIF and Line Probe Assay (LPA)] [33]. Other non-laboratory-related tests include chest X-rays, Magnetic Resonance Imaging (MRI), Computed Tomography (CT) scans and ultrasounds [6,39].

Early diagnosis of TB is a critical step towards its control[58]and bacteriological confirmation of MTB and subsequent drug resistance testing[14]aids in MDR-TB detection. While MDR-TB diagnosis has made significant strides[29], it is largely compounded by lack of or poor laboratory infrastructures and other resources in many instances resulting into delays in diagnosis, therapy initiation or completion[50]. Systematic surveillance and tracking of MDR-TB helps in understanding the overall burden of the disease [40]and can inform research and practice in diagnosis, treatment, and infection control. More efficient strategies and increased investment and commitment to MDR-TB control will be needed[51]. In order to meet the World Health Organization (WHO) goal/target to end TB by 2035, countries must address the emergence of drug-resistant strains effectively[38].

The serious epidemic of MDR-TB is considered an important public health issue in many nations and constitutes a huge factor hampering effective TB control worldwide, which then highlights the usefulness of universal drug susceptibility testing[59]. This study sought to determine factors associated with MDR-TB among presumptive multidrug resistance tuberculosis patients identified in a drug resistance TB surveillance study in western Kenya with an overall aim of providing evidence for planning of targeted programmatic interventions to reduce burden of multidrug-resistant tuberculosis in Kenya.

2. Materials and methods

2.1. Study Area

The study was conducted in western Kenya, a region that has a high TB/HIV burden [41]. The samples were collected from the Ministry of Health [3] peripheral health facilities as part of routine workup and transported to KEMRI-CGHR TB Laboratory in Kisumu, Kenya, for culture and drug susceptibility testing (DST) for either first line or second-line DST, and archival for MTBc isolates identified.

2.2. Study design

This was a health facility-based cross-sectional study conducted between November 1st 2021 to March 31st 2022.

2.3. Target population

The study included all presumptive TB patients visiting TB clinics at the health facility and provided sputum specimens for TB diagnosis.

2.4. Sampling (Patient selection) method

Patients presenting with suggestive TB clinical signs and symptoms with or without prior TB treatment history, positive smear relapses, or a possible treatment failure, and those who were lost to follow-up.

2.5. Sample size

Each presumptive TB patient provided one sputum sample during their TB clinic visit at the peripheral health facilities during the study period. The total sample size (N=390) used in this study was calculated using Fisher's sample size formula for one proportion,[3]assuming 95 % level of significance, 0.558 % prevalence of TB in the general population [13], and 5 % margin of error.

2.6. Data collection

The facility's clinicians at the TB clinics collected and entered participant information using the Kenya National TB program's MoH laboratory requisition forms available at the health facilities. The information included socio-clinical and demographic information patient's age, sex, HIV status, clinical history that included coughs for more than two weeks, night sweats, fever, loss of breath, loss of weight, patient type (i.e., either new, presumptive, lost to follow-up, treatment

failure), and TB type (either pulmonary or extra pulmonary TB). This information collected were subsequently entered onto the laboratory's information management system (LIMS).

2.7. Laboratory procedures

Sputum samples (3-5mls collected in sterile 50 ml falcon tubes) were processed as per the standard culture processing algorithm, as provided for in the MGIT™ manual [55]. The samples were decontaminated using 2 % NALC-NaOH/Na citrate concentration of equal volumes (resulting into a final concentration of 1 %), washed using phosphate buffered saline (PBS), vortexed, and centrifuged (at 3000 rpm), pellets (deposits) resuspended in 0.5mls PBS. A resultant 0.1 was thereafter obtained, which was adequate for both inoculation onto BACTEC *Mycobacterium* growth indicator tube (MGIT) for culture and fluorescent microscopy (FM). Subsequently, all positive growths from MGIT underwent MTBc identification using Brain Heart Infusion (BHI) culture, Ziehl Nielsen (ZN) microscopy and BD's MTBc identification kits of which MTBc positives received drug susceptibility tests (DST) against the first line TB treatment drugs.

Drug sensitivity testing, (a laboratory diagnostic technique for determining ability of a drug to inhibit the growth of *M. tuberculosis*) to first line TB drugs was performed by dilution of 1:100 for each of the first line drugs (Isoniazid, Rifampicin, and ethambutol). Onto the diluted drugs in the individual tubes, 0.8 ml of DST supplement and 0.5 ml of inoculum from the growth obtained from liquid culture MGIT culture tubes were added, incubated into BACTEC 960 MGIT instrument and observed daily for growth for 12–14 days.

2.8. Study Variables

In the regression analyses, the dependent variable was drug resistant TB measured on a binary scale as resistant or non-resistant to each of the drug being tested. The independent variables were age of patients in years, sex, HIV status categorized as positive, negative, and unknown, county of residence, and patient type (new, smear positive relapse, treatment failure, and lost to follow up).

2.9. Statistical Analysis

All the data were entered into Microsoft Excel spreadsheet and transferred into STATA version 15.0 (Stata Corporation, College Station, Texas, USA) for statistical analyses. The continuous variables were summarized using median [interquartile range (IQR)] and frequencies and percentages for categorical variables. Multivariable logistic regression was conducted controlling for all the independent variables to identify factors associated with the TB drug resistant for each drug. All the variables included in this study are traditional/known risk factors of drug resistance to TB and were included in the regression model irrespective of their value. All $P \leq 0.05$ were considered statistically significant.

2.10. Ethical Considerations

The study was approved by School of Graduate Studies of Maseno University and Kenya Medical Research Institute's Scientific and Ethical Review Unit (SERU), protocol no. SERU 4247, prior to commencing the study.

Informed consent was not sought from the study participants as we used secondary data. Confidentiality of the information collected was maintained by omitting their names and other personal identifiers from extraction sheet.

3. Results

3.1. Socio-demographic Characteristics of the study participants

A total of 390 isolates from sputum samples were included in this analysis. The median age of patients was 34 (IQR: 25–43) years and 302 (77.4 %) of the participants were males. Most patients were from Kisii County (40.5 %; n = 158); 322 (82.6 %) had been previously treated (presumptive MDR-TB), 28 (7.2 %) had smear positive relapses, 26 (6.7 %) were lost to follow–up (LTFU), 7 (1.8 %) were first line treatment failures and 7 (1.8 %) were new. The prevalence of MDR-TB was 3.6 % (95 % CI: 2.0 %-5.9 %) and HIV 30.3 % (95 % CI: 25.7 %-35.1 %) (Table 1).

A total of 64/390 (16.4 %) isolates had a form of resistance. The RIF resistance was identified in 12 (3.1 %), INH resistance in 34 (8.7 %), 4 (1.0 %) had ethambutol resistance and 14 (3.6 %) had multidrug resistance.

3.2. Drug resistance by patient characteristics

Of the various drug resistances patterns witnessed, HIV uninfected had 34/64 (53.1 %) cases comprising of 17/64 (26.5 %) INH, 7/64 (10.9 %) MDR, 6/64 (9.4 %) RIF and 4/64 (6.3 %) ETH, respectively. The HIV infected had 26/64 (40.6 %) cases comprising of 15/64 (23.4 %) INH, 6/64 (9.4 %) MDR and 5/64 (7.8 %) RIF, respectively. For those individuals in which the HIV status was unknown, had 4/64 (6.3 %) cases of drug resistances that comprised 2/64 (3.1 %) INH, 1 (1.6 %) MDR, and 1 (1.6 %) RIF (Table 2).

Table 1
Summarizes the socio-demographics characteristics of patients for the TB surveillance study performed at the KEMRI TB laboratory, Kisumu (N=390).

Variable	Frequency/median (IQR)	Percentage (%)
Age in years	34 (25 – 43)	
Sex		
Female	88	22.6
Male	302	77.4
County of Residence		
Busia	3	0.8
Elgeyo Marakwet	19	4.9
Homa Bay	60	15.4
Kakamega	6	1.5
Kericho	2	0.5
Kisii	158	40.5
Kisumu	8	2.1
Migori	41	10.5
Nandi	21	5.4
Siaya	21	5.4
Trans Nzoia	1	0.3
Uasin Gishu	28	7.2
Vihiga	22	5.6
Patient Type		
Failure of First Line	7	1.8
New Cases	7	1.8
Previously Treated	322	82.6
Relapse Smear Positive	28	7.2
Lost to Follow Up	26	6.7
HIV Status		
Negative	246	63.1
Not Done	26	6.7
Positive	118	30.3
Resistance		
Non-resistance	326	83.6
INH	34	8.7
MDR	14	3.6
RIF	12	3.1
ETHA	4	1.0

Data presented in median (IQR), frequency and percentages. INH=Isoniazid, MDR=multi-drug resistance, RIF=rifampicin, ETHA=ethambutol.

The prevalence of resistance across all the drugs were higher among males 55/64 (85.9 %), comprising resistances to INH 31/34 (91.2 %), MDR 11/14 (78.6 %), RIF 10/12 (83.3 %) and ETH 3/4 (75 %) cases as compared to 9/64 (14.1 %) among females with INH 3/34 (8.8 %), MDR 3/14 (21.4 %), RIF 2/12 (16.7 %) and ETH 1/4 (25 %), respectively. While considering the patient types, the category of previously treated for TB disease had increased resistance patterns, INH 21/34 (61.8 %), MDR 8/14 (57.1 %), RIF 8/12 (66.7 %) and ETH 4/4 (100 %), followed by patients who had smear positive relapses with INH 6/34 (17.6 %), MDR 3/14 (21.4 %), and RIF 1/12 (8.3 %), respectively. The patients reported as lost to follow up, had 5/34 (14.7 %) INH resistance, and 1/12 (8.3 %) RIF resistance, those reported as failure to first line treatment had 1/34 (2.9 %) INH, 1/14 (7.1 %) MDR and 1/12 (8.3 %) RIF resistances Table 3.

Among the counties of residences, Siaya had 7/34 (20.5 %) INH resistance, Homabay, Kisii, and Migori reported 6/34 (17.6 %) cases each, Vihiga had 3/34 (8.8 %), Elgeyo Marakwet and Nandi 2/34 (5.9 %) each, and lastly Kisumu and Uasin Gishu had 1/34(2.9 %) cases each. For MDR cases, both Homabay and Migori had 4/14 (28.6 %), Kisii had 3/14(21.4 %), Uasin Gishu had 2/14(14.3 %) and Nandi reported 1/14 (7.1 %). For RIF resistance, Kisii had 5/12 (41.7 %), 2/12 (16.7 %) in Homabay and Kisii, and 1/12(8.3 %) for Elgeyo Marakwet, Migori and Siaya respectively. Lastly, for Ethambutol resistance, Nandi had 2/4(50 %), with both Migori and Uasin Gishu reporting ¼ (25 %) cases each. Overall, Siaya County had a higher proportion of patients resistant to INH (n = 7; 20.6 %), however, Homabay and Migori counties had 4 (28.6 %) MDR patients each, whilst majority in Kisii County were resistance to RIF (n = 5; 41.7 %) and half of the patients' resistance to ethambutol were from Nandi County (n = 2; 50.0 %). Table 2.

3.3. Factors associated with drug resistance

To identify factors associated with MDR-TB, multivariate logistic regression analyses was used. Results show that the category of previously treated (patients who received or currently on TB treatment) had a 70 % decrease likelihood of resistance (aOR: 0.30; 95 % CI: 01.3–0.70) while other categories (lost to follow-up, relapses, first-line treatment failures and new cases) had a 3.33 times increased likelihood of resistance [aOR: 3.33 (1.42–7.44)] to INH. Further analyses demonstrated that older age in years had an increased likelihood to resistance to RIF (aOR: 1.04; 95 % CI: 1.00–1.08) while younger age experienced less resistance. All the rest of the analyses were non-significant.

4. Discussion

The current study has shown more TB distribution among males than females, congruent to previous studies carried out earlier, [47]with males showing higher TB infections over females [13]. In Kenya, studies have shown the prevalence to both MDR TB (firstline) and second line TB drugs resistance is 1.5 % [46]as well as having an estimated national HIV prevalence rates of 5.6 % and 6.3 %, [13].

The HIV prevalence in the study area (western region) was diverse, ranging from 2.5 % to 27 %, compared to the national rate of 4.9 % [42], however the current study had a HIV infection of 30.3 %, which fairly contrasts a similar study in, western Kenya, [27].

The current study found a higher prevalence rate for MDR-TB (firstline drug resistance), at 3.6 %, probably due to intensified or enhanced active case findings over the recent past. The current study also observed an overall prevalence of 3.1 % RIF monoresistance, contrary to a study conducted in South Africa [37], which had RIF (11 %) but comparable to its INH monoresistance of 8.7 % to 8.5 %, respectively. Similarly, it also showed a difference to a study from Uganda [8] that reported both RIF and INH monoresistance at 4.8 % and 3.2 %, respectively.

The factors considered for this current study (i.e., sex and HIV status) were not associated with MDR-TB, except the age, where participants of age group 25 – 43 years had TB, comparing favorably to other studies [16,52,63]and patient type where among those previously treated [28], a significant association with INH resistance was observed. In addition, aged individuals were more likely to show resistance to RIF relative to younger groups, probably because they may have not been exposed to Rifampicin, since it was recently introduced into use [28].

Tuberculosis prevalence among this age group (25–43 years) could be attributed to their probable socio-economic activities [44],.e.g., mobility for search of, or working, strict and unfavorable job schedules, conflicting time of taking medicines, or poor health seeking behaviour altogether [57].

Despite the global efforts to combat and eliminate TB which is 100 % curable, it continues to cause deaths among adults and infects close to a third of global populations [63]. A major challenge to these efforts has

Table 2
Summaries of the demographic and clinical characteristics of patients with different drug resistances, N=64.

Characteristic	INH n = 34	%	MDR n = 14	%	RIF n = 12	%	ETHA n = 4	%
Age in years, Median (IQR)	37 (28 – 44)		39 (33 – 43)		38 (35 – 46)		NA	
Sex								
Female	3	8.8	3	21.4	2	16.7	1	25.0
Male	31	91.2	11	78.6	10	83.3	3	75.0
County of Residence								
Elgeyo Marakwet	2	5.9	0	0.0	1	8.3	0	0.0
Homa Bay	6	17.6	4	28.6	2	16.7	0	0.0
Kisii	6	17.6	3	21.4	5	41.7	0	0.0
Kisumu	1	2.9	0	0.0	0	0.0	0	0.0
Migori	6	17.6	4	28.6	1	8.3	1	25.0
Nandi	2	5.9	1	7.1	2	16.7	2	50.0
Siaya	7	20.6	0	0.0	1	8.3	0	0.0
Uasin Gishu	1	2.9	2	14.3	0	0.0	1	25.0
Vihiga	3	8.8	0	0.0	0	0.0	0	0.0
Patient Type								
Failure of First Line	1	2.9	2	14.3	1	8.3	0	0.0
New	1	2.9	1	7.1	1	8.3	0	0.0
Previously Treated	21	61.8	8	57.1	8	66.7	4	100.0
Relapse Smear Positive	6	17.6	3	21.4	1	8.3	0	0.0
Treatment after Loss to Follow Up	5	14.7	0	0.0	1	8.3	0	0.0
HIV Status								
Negative	17	50.0	7	50.0	6	50.0	4	100.0
Not Done	2	5.9	1	7.1	1	8.3	0	0.0
Positive	15	44.1	6	42.9	5	41.7	0	0.0

Table 3
Summaries of the multivariable logistic regression of factors associated with drug resistance patterns.

Variables	MDR aOR	95 %CI	INH aOR	95 %CI	RIF aOR	(95 %CI)
Age in year	1.02	0.97–1.06	1.01	0.98–1.03	1.04	1.00–1.08
Sex						
Females	0.76	0.11–3.12	0.32	0.07–0.96	0.79	0.12–3.27
Males	1.32	0.32–8.90	3.10	1.04–13.35	1.26	0.31–8.52
Type of patients						
Previously treated	0.31	0.09–1.26	**0.30**	**0.13–0.70**	0.47	0.13–2.26
*Others	3.18	0.79–11.19	3.33	1.42–7.44	2.12	0.44–7.86
HIV status						
Negative	0.44	0.12–1.54	0.57	0.26–1.22	0.55	0.15–2.03
Positive	2.27	0.65–8.24	1.77	0.82–3.79	1.82	0.49–6.54

*Includes: Failure of first line, new cases, relapse smear positive, Treatment after lost to follow-up;CI: confidence interval.

been the emergence or presence of resistance, of a severe form also referred to as multidrug resistance tuberculosis. The MDR-TB poses a huge challenge to public health systems in terms of treatment costs and disease control [56].

This current work determined the factors associated with TB drug resistance among MDR-TB presumptive patients, such as age, sex, HIV status, patient type, geographical locations, and TB types. The majority of participants (82.6 %), were from presumptive MDR-TB patients, with an age group between 25 and 43 years similar to a study conducted in India [54]. In the current study, we obtained only a limited number of samples from children, none of which had MDR-TB consistent with previous findings [71]. The low numbers are likely due to challenges with collecting samples from children. More effort and emphasis should be made towards children with regards to sample collection to have comprehensive analyses that can guide interventions in this group.

The current study observed an association of TB with being male, similar to a study from China [66]), as well as other previously conducted studies in both Ethiopia [21,64]but contrasting to a Nigeria study, [48]and this may be due to a lower number of males seeking health care [25], as well as biological differences in response to *Mycobacterium*.

The current study also shows an association of MDR-TB among the older population, similar to a study from Nigeria [48], but contrasting to STOP TB, where majority are within the middle age groups of 25–44 years, as well as among the HIV infected, [16,21]. For example, in the multivariable logistic regression analysis, it showed that HIV infection was not associated with drug resistant TB, similar to previous studies [13,30].

The study did not find any overt significant associations between sex, HIV status, patient and/or TB types as well as geographical locations, and TB drug resistance except age, which had a significant risk factor for MDR-TB, and in concurrent with studies carried out in Bangladesh and China [16,66]). We also observed a significant association between age and RIF resistance among the patients. The high frequencies of MDR-TB among older age groups may indicate the possibility of propagation of MDR-TB in the community, due to delays in seeking health care services, lack of resources among other reasons [1,2].

In establishing clinical characteristics of patients with different drug resistances, our findings show that all the drug resistances were higher among the previously treated patients as compared to other patient categories, of whom RIF and INH monoresistance was 66.7 % and 61.8 %, respectively, while MDR was at 57.1 %, consistent with other studies [31,26,69,15].

The proportion of females presenting with MDR-TB was 21.4 %, INH and RIF monoresistance was 8.8 % and 16.7 %, respectively, to males at MDR-TB at 78.6 %. Monoresistance to both INH and RIF were at 91.2 % and 83.3 %, respectively, in line with a study conducted in India[54]. The current study found a significant difference in the MDR-TB prevalence between previously treated and other patient categories, i.e., lost to follow-up, smear relapses treatment failures and new cases, contrary to a previous study conducted in India[54].

Among the sex characteristics, the current study observed more resistances to INH, RIF and MDR with men, in line with previous findings [19]. In the current study, isolates from previously treated patients had INH (8.7 %) monoresistance, while isolates from males had a higher risk of INH monoresistance as compared to females which was similar to other findings[6].

Isoniazid mono-resistance had a significant association with previous treatment, similar to the rate found in studies conducted in Uganda [37] and Ethiopia [36], and higher than the outcomes as observed in the recent 2016, Kenya Tuberculosis Prevalence Survey (KTBPS, 2016) of 2.1 % [13].

The current study observed that at least 16.4 % of all isolates had a form of resistance. Mono-RIF resistance and mono-INH resistance were observed to be the most prevalent among this population. In addition, of all the RIF resistance, 57 % were equally multidrug resistance in close concordance with the global value of between 60 to 70 % [54].

Among the HIV-infected individuals, failure of first-line treatment was associated with MDR-TB, consistent to previous studies[11], however, this was contrary to a similar study from Uganda[37], which found out that the prevalence of TB has been increasing among patients who are HIV-negative since 2005. While there were no strong associations with individual factors such as HIV infection, history of previous treatment towards MDR-TB infections, there was however, an association between age on RIF mono-resistance and previous treatment towards INH mono-resistance.

In the multivariate regression analysis performed, we observed no significant association between MDR and INH resistance with regards to age, with an exception to RIF resistance, which had a P=0.048. Equally important observation realized was a significant association between INH resistance (P=0.004) in patients who had been previously treated with anti-TB therapy, of which include the first line treatment of tuberculosis, comprising of RIF and INH among other drugs. Our study did not observe any significant associations between sex and HIV status and MDR-TB resistance, in line with other studies [37].

The current study has provided pertinent information about factors associated with MDR-TB which can form part of heightened implementation towards a decreased burden of TB in western Kenya for the national TB program.

There are many differences among studies assessing risk factors for MDR-TB because of region and sample size differences, scattered factors, and inconsistent results among others [65]. The current study observed a high MDR-TB prevalence in Homabay and Migori counties, both at 28.6 %, and the regions are considered to have high HIV prevalence as per the recent Kenya Aids Indicator Survey (2018) report [18], therefore MDR-TB prevalence may be associated with regions with a higher prevalence of both HIV and TB.

5. Strengths and weaknesses

The major strength of this study was inclusion of all MDR-TB patients' samples from the regions of Western Kenya.

5.1. Study Limitation

Our study included very few isolates from children with TB, so we were unable to draw any conclusion about this age group. The study was not able to conduct second-line drug susceptibility nor sequencing tests for patients showing resistance to first line regimens and for possible mutations associated with RIF and INH resistance and possibly understanding the genotypic diversity.

The current study only involved presumptive MDR-TB participants. The study did not include all TB patients, such as those on susceptible TB treatment regimens. Our multivariable analysis did not include other behavioural factors that may be associated with MDR-TB e.g. smoking, alcohol intake, poor adherence etc. However, the study included renown demographic and clinical characteristics such age, sex, HIV status, TB treatment history, and lost to follow-up which also determines the behavioural characteristics.

Despite these limitations, we provide preliminary analyses on the risk factors associated with MDR-TB which are important information for health policy and planning of targeted programmatic interventions for future improvements of MDR-TB prevention and control in western region or in Kenya as a whole.

6. Conclusion

The study showed MDR-TB prevalence of 3.6 % among presumptive MTB patients in western Kenya, and HIV infection prevalence of 30.3 %, this requires a concerted effort to combat both TB and HIV infections among these population. While the median age of current study's participants was 34 years, the study showed the age group of 25–43 years as predominantly infected with TB, drug resistances to first line, and among these, males are the majority, there is therefore the need to develop strategies targeting these age group and males in general.

Previously treated TB patients constituted most MDR-TB in the current study; hence a lot needs to be done towards these patients, e.g., better education on importance of adherence, use of right drugs appropriately and emphasis on changes to wards health seeking behaviour. Geographically, current study demonstrated high MDR-TB prevalence, (28.6 %) for both Migori and Homabay, with (21.4 %) in Kisii. Migori and Homabay counties with shared boundaries have both higher MDR and RIF resistance rates, while Siaya poses challenge with INH resistance (20.5 %).

7. Recommendations

There is hence a heightened need to develop MDR-TB mop up strategies that should be employed in both counties simultaneously, to aid in reducing TB and MDR in these areas. Therefore, planning for continuous drug resistance screening for all TB patients, as well as the strengthening of drug resistance control measures in these regions as well as the rest of the country, is recommended.

Other studies should consider behavioural characteristics of their prospective participants too.

CRediT authorship contribution statement

Albert Okumu: . **James Orwa:** Data curation, Methodology. **Ruth Sitati:** Investigation, Methodology. **Isaiah Omondi:** Data curation, Formal analysis. **Ben Odhiambo:** Data curation, Formal analysis. **Jeremiah Ogoro:** Supervision. **George Oballa:** Supervision. **Benjamin Ochieng:** Investigation, Supervision, Writing – review & editing. **Steve Wandiga:** Funding acquisition, Supervision, Writing – review & editing. **Collins Ouma:** Conceptualization, Investigation, Methodology, Supervision, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Author Contributions

They contributed to developing and writing of the study design, data analysis, writing and reviewing of the manuscript.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jctube.2024.100466.

References

[1] Abdella K, Abdissa K, Kebede W, Abebe G. Drug resistance patterns of Mycobacterium tuberculosis complex and associated factors among retreatment cases around Jimma. Southwest Ethiopia *BMC public health* 2015;15(1):1–7.
[2] Adane K, Ameni G, Bekele S, Abebe M, Aseffa A. Prevalence and drug resistance profile of Mycobacterium tuberculosis isolated from pulmonary tuberculosis patients attending two public hospitals in East Gojjam zone, northwest Ethiopia. BMC Public Health 2015;15(1):1–8.
[3] Alelign A, Zewude A, Mohammed T, Tolosa S, Ameni G, Petros B. Molecular detection of Mycobacterium tuberculosis sensitivity to rifampicin and isoniazid in South Gondar Zone, northwest Ethiopia. BMC Infect Dis 2019;19(1):1–8.
[4] Ambaye GY, Tsegaye GW. Factors associated with multi-drug resistant tuberculosis among TB patients in selected treatment centers of Amhara Region: a case-control study. Ethiop J Health Sci 2021;31(1).
[5] Asgedom, S. W., Teweldemedhin, M., & Gebreyesus, H. (2018). Prevalence of multidrug-resistant tuberculosis and associated factors in Ethiopia: a systematic review. *Journal of pathogens*, *2018*.
[6] Baluku, J. B., Mukasa, D., Bongomin, F., Stadelmann, A., Nuwagira, E., Haller, S.,… Turyahabwe, S. (2021). Gender differences among patients with drug resistant tuberculosis and HIV co-infection in Uganda: a countrywide retrospective cohort study. *BMC Infectious Diseases*, *21*(1), 1-11.
[7] Barbosa, C. d. B., Lazzarini, L., Elias, A., Leung, J., Ribeiro, S., Da Silva, M.,… Kritski, A. (2012). Tuberculosis caused by RDRio Mycobacterium tuberculosis is not associated with differential clinical features. *The International journal of tuberculosis and lung disease*, *16*(10), 1377-1382.
[8] Bazira J, Asiimwe BB, Joloba ML, Bwanga F, Matee MI. Mycobacterium tuberculosis spoligotypes and drug susceptibility pattern of isolates from tuberculosis patients in South-Western Uganda. BMC Infect Dis 2011;11(1):1–9.
[9] Billington O, McHugh TD, Gillespie SH. Physiological cost of rifampin resistance induced in vitro in Mycobacterium tuberculosis. Antimicrob Agents Chemother 1999;43(8):1866–9.
[10] Chakaya, J., Khan, M., Ntoumi, F., Aklillu, E., Fatima, R., Mwaba, P.,…Katoto, P. D. (2021). Global Tuberculosis Report 2020–Reflections on the Global TB burden, treatment and prevention efforts. *International Journal of Infectious Diseases*, *113*, S7-S12.
[11] Dean AS, Cox H, Zignol M. Epidemiology of drug-resistant tuberculosis. Strain variation in the Mycobacterium tuberculosis complex: its role in biology, epidemiology and control 2017:209–20.
[12] Dean AS, Zignol M, Cabibbe AM, Falzon D, Glaziou P, Cirillo D, et al. Prevalence and genetic profiles of isoniazid resistance in tuberculosis patients: a multicountry analysis of cross-sectional data. PLoS Med 2020;17(1):e1003008.
[13] Enos, M., Sitienei, J., Ong'ang'o, J., Mungai, B., Kamene, M., Wambugu, J.,… Nyaboke, D. (2018). Kenya tuberculosis prevalence survey 2016: challenges and opportunities of ending TB in Kenya. *PLoS One*, *13*(12), e0209098.
[14] Falzon D, Schünemann HJ, Harausz E, González-Angulo L, Lienhardt C, Jaramillo E, et al. World Health Organization treatment guidelines for drug-resistant tuberculosis, 2016 update. Eur Respir J 2017;49(3).

[15] Faustini A, Hall AJ, Perucci CA. Risk factors for multidrug resistant tuberculosis in Europe: a systematic review. Thorax 2006;61(2):158–63.
[16] Flora M, Amin M, Karim M, Afroz S, Islam S, Alam A, et al. Risk factors of multi-drug-resistant tuberculosis in Bangladeshi population: a case control study. Bangladesh Med Res Counc Bull 2013;39(1):34–41.
[17] Gagneux, S., DeRiemer, K., Van, T., Kato-Maeda, M., de Jong, B. C., Narayanan, S., …Small, P. M. (2006). Variable host-pathogen compatibility in Mycobacterium tuberculosis. *Proc Natl Acad Sci U S A*, *103*(8), 2869-2873.
[18] Gichangi, A., Wambua, J., Mutwiwa, S., Njogu, R., Bazant, E., Wamicwe, J.,… Mudany, M. (2018). Impact of HIV self-test distribution to male partners of ANC clients: results of a randomized controlled trial in Kenya. *Journal of acquired immune deficiency syndromes (1999)*, *79*(4), 467.
[19] Guenaoui K, Harir N, Ouardi A, Zeggai S, Sellam F, Bekri F, et al. Use of GeneXpert Mycobacterium tuberculosis/rifampicin for rapid detection of rifampicin resistant Mycobacterium tuberculosis strains of clinically suspected multi-drug resistance tuberculosis cases. Annals of translational medicine 2016;4(9).
[20] Harausz EP, Garcia-Prats AJ, Law S, Schaaf HS, Kredo T, Seddon J, et al. Treatment and outcomes in children with multidrug-resistant tuberculosis: a systematic review and individual patient data meta-analysis. PLoS Med 2018;15(7):e1002591.
[21] Hirpa S, Medhin G, Girma B, Melese M, Mekonen A, Suarez P, et al. Determinants of multidrug-resistant tuberculosis in patients who underwent first-line treatment in Addis Ababa: a case control study. BMC Public Health 2013;13:782.
[22] Horton KC, MacPherson P, Houben RM, White RG, Corbett EL. Sex differences in tuberculosis burden and notifications in low-and middle-income countries: a systematic review and meta-analysis. PLoS Med 2016;13(9):e1002119.
[23] Ismail N, Omar SV, Ismail F, Blows L, Koornhof H, Onyebujoh PC, et al. Drug resistant tuberculosis in Africa: Current status, gaps and opportunities. African journal of laboratory medicine 2018;7(2):1–11.
[24] Jain A, Mondal R. Extensively drug-resistant tuberculosis: current challenges and threats. FEMS Immunol Med Microbiol 2008;53(2):145–50.
[25] Jeon DS, Shin DO, Park SK, Seo JE, Seo HS, Cho Y, et al. Treatment outcome and mortality among patients with multidrug-resistant tuberculosis in tuberculosis hospitals of the public sector. J Korean Med Sci 2011;26(1):33–41.
[26] Kigozi E, Kasule GW, Musisi K, Lukoye D, Kyobe S, Katabazi F, et al. Prevalence and patterns of rifampicin and isoniazid resistance conferring mutations in Mycobacterium tuberculosis isolates from Uganda. PLoS One 2018;13(5): e0198091.
[27] Kwena, Z. A., Njuguna, S. W., Ssetala, A., Seeley, J., Nielsen, L., De Bont, J.,… Team, L. V. C. f. H. R. HIV prevalence, spatial distribution and risk factors for HIV infection in the Kenyan fishing communities of Lake Victoria. PLoS One 2019;14 (3):e0214360.
[28] Law W, Yew W, Chiu Leung C, Kam K, Tam C, Chan C, et al. Risk factors for multidrug-resistant tuberculosis in Hong Kong. Int J Tuberc Lung Dis 2008;12(9): 1065–70.
[29] Li, B.-Y., Shi, W.-P., Zhou, C.-M., Zhao, Q., Diwan, V. K., Zheng, X.-B.,…Xu, B. (2020). Rising challenge of multidrug-resistant tuberculosis in China: a predictive study using Markov modeling. *Infectious Diseases of Poverty*, *9*(03), 57-64.
[30] Lomtadze N, Aspindzelashvili R, Janjgava M, Mirtskhulava V, Wright A, Blumberg H, et al. Prevalence and risk factors for multidrug-resistant tuberculosis in the Republic of Georgia: a population-based study. Int J Tuberc Lung Dis 2009; 13(1):68–73.
[31] Lukoye, D., Adatu, F., Musisi, K., Kasule, G. W., Were, W., Odeke, R.,…Joloba, M. L. (2013). Anti-tuberculosis drug resistance among new and previously treated sputum smear-positive tuberculosis patients in Uganda: results of the first national survey. *PLoS One*, *8*(8), e70763.
[32] Lukoye D, Ssengooba W, Musisi K, Kasule GW, Cobelens FG, Joloba M, et al. Variation and risk factors of drug resistant tuberculosis in sub-Saharan Africa: a systematic review and meta-analysis. BMC Public Health 2015;15(1):1–13.
[33] Machado D, Couto I, Viveiros M. Advances in the molecular diagnosis of tuberculosis: from probes to genomes. Infect Genet Evol 2019;72:93–112.
[34] MacNeil A, Glaziou P, Sismanidis C, Maloney S, Floyd K. Global epidemiology of tuberculosis and progress toward achieving global targets—2017. Morb Mortal Wkly Rep 2019;68(11):263.
[35] Mariam DH, Mengistu Y, Hoffner SE, Andersson DI. Effect of rpoB mutations conferring rifampin resistance on fitness of Mycobacterium tuberculosis. Antimicrob Agents Chemother 2004;48(4):1289–94.
[36] Mekonnen F, Tessema B, Moges F, Gelaw A, Eshetie S, Kumera G. Multidrug resistant tuberculosis: prevalence and risk factors in districts of metema and west armachiho. Northwest Ethiopia *BMC Infectious Diseases* 2015;15(1):1–6.
[37] Micheni LN, Kassaza K, Kinyi H, Ntulume I, Bazira J. Rifampicin and isoniazid drug resistance among patients diagnosed with pulmonary tuberculosis in southwestern Uganda. PLoS One 2021;16(10):e0259221.
[38] Migliori, G. B., Dheda, K., Centis, R., Mwaba, P., Bates, M., O'Grady, J.,…Zumla, A. (2010). Review of multidrug-resistant and extensively drug-resistant TB: global perspectives with a focus on sub-Saharan Africa. *Tropical Medicine & International Health*, *15*(9), 1052-1066.
[39] Munawar F, Azmat S, Iqbal T, Grönlund C, Ali H. Segmentation of lungs in chest X-ray image using generative adversarial networks. IEEE Access 2020;8:153535–45.
[40] Musa BM, Adamu AL, Galadanci NA, Zubayr B, Odoh CN, Aliyu MH. Trends in prevalence of multi drug resistant tuberculosis in sub-Saharan Africa: A systematic review and meta-analysis. PLoS One 2017;12(9):e0185105.
[41] Musyoki, H., Bhattacharjee, P., Blanchard, A. K., Kioko, J., Kaosa, S., Anthony, J., …Olwande, C. (2018). Changes in HIV prevention programme outcomes among key populations in Kenya: data from periodic surveys. *PLoS One*, *13*(9), e0203784.
[42] NACC. (2018). *Kenya HIV estimate report 2018*.
[43] Nathanson, E., Nunn, P., Uplekar, M., Floyd, K., Jaramillo, E., Lönnroth, K.,… Raviglione, M. (2010). MDR tuberculosis—critical steps for prevention and control. *New England Journal of Medicine*, *363*(11), 1050-1058.
[44] Nduba, V., Van't Hoog, A. H., Mitchell, E., Onyango, P., Laserson, K., & Borgdorff, M. (2015). Prevalence of tuberculosis in adolescents, western Kenya: implications for control programs. *International Journal of Infectious Diseases*, *35*, 11-17.
[45] NTLDP. (2022). *Annual Report 2022*. NTLDP. nltp.co.ke/wp-content/uploads/2024/01/Annual-Report_2022_22_12_2023.pdf.
[46] Ogari CO, Nyamache AK, Nonoh J, Amukoye E. Prevalence and detection of drug resistant mutations in Mycobacterium tuberculosis among drug naïve patients in Nairobi. Kenya *BMC infectious diseases* 2019;19(1):1–7.
[47] Ogwang MO, Imbuga M, Ngugi C, Mutharia L, Magoma G, Diero L. Distribution patterns of drug resistance Mycobacterium tuberculosis among HIV negative and positive tuberculosis patients in Western Kenya. BMC Infect Dis 2021;21(1):1–15.
[48] Olanrewaju OP, Omosalewa OE, Okoro L, Olawale O-R. Co-epidemics: implications of COVID-19 outbreak associated with human immune-deficiency virus, tuberculosis and Lassa fever in a low resource economy-a call for proactive measures. International Journal of Community Medicine and Public Health 2023; 10(2):824.
[49] Pan, Z., Zhang, J., Bu, Q., He, H., Bai, L., Yang, J.,…Lyu, J. (2020). The Gap Between Global Tuberculosis Incidence and the First Milestone of the WHO End Tuberculosis Strategy: An Analysis Based on the Global Burden of Disease 2017 Database. *Infection and drug resistance*, *13*, 1281.
[50] Parsons LM, Somoskövi Á, Gutierrez C, Lee E, Paramasivan C, Abimiku A, et al. Laboratory diagnosis of tuberculosis in resource-poor countries: challenges and opportunities. Clin Microbiol Rev 2011;24(2):314–50.
[51] Reid, M. J., Arinaminpathy, N., Bloom, A., Bloom, B. R., Boehme, C., Chaisson, R., …Ditiu, L. (2019). Building a tuberculosis-free world: The Lancet Commission on tuberculosis. *The Lancet*, *393*(10178), 1331-1384.
[52] Rifat, M., Milton, A. H., Hall, J., Oldmeadow, C., Islam, M. A., Husain, A.,… Siddiquea, B. N. (2014). Development of multidrug resistant tuberculosis in Bangladesh: a case-control study on risk factors. *PloS one*, *9*(8), e105214.
[53] Senkoro, M. F. (2016). Burden of pulmonary tuberculosis and its major determinants: A national prevalence survey in Tanzania.
[54] Shivekar SS, Kaliaperumal V, Brammacharry U, Sakkaravarthy A, Raj C, Alagappan C, et al. Prevalence and factors associated with multidrug-resistant tuberculosis in South India. Sci Rep 2020;10(1):1–11.
[55] Siddiqi, S. (2006). MGIT Procedure Manual; For BACTEC MGIT 960 TB System, Specially Prepared for FIND MGIT demonstration Project. In.
[56] Tanimura T, Jaramillo E, Weil D, Raviglione M, Lönnroth K. Financial burden for tuberculosis patients in low-and middle-income countries: a systematic review. Eur Respir J 2014;43(6):1763–75.
[57] Thompson AE, Anisimowicz Y, Miedema B, Hogg W, Wodchis WP, Aubrey-Bassler K. The influence of gender and other patient characteristics on health care-seeking behaviour: a QUALICOPC study. BMC Fam Pract 2016;17:1–7.
[58] Van Cutsem G, Isaakidis P, Farley J, Nardell E, Volchenkov G, Cox H. Infection control for drug-resistant tuberculosis: early diagnosis and treatment is the key. Clin Infect Dis 2016;62(suppl_3):S238–43.
[59] Wang, D.-M., Li, Q.-F., Zhu, M., Wu, G.-H., Li, X., Xu, Y.-H.,…Ying, B.-W. (2020). Epidemiological, clinical characteristics and drug resistance situation of culture-confirmed children TBM in southwest of China: a 6-year retrospective study. *BMC Infectious Diseases*, *20*(1), 1-7.
[60] WHO. (2021). *Global tuberculosis report 2021*. World Health Organization.
[61] WHO. (2023). Global Tuberculosis report 2023. In.
[62] WHO, Programme, W. H. O. G. T., & World Health Organization. Communicable Diseases, C. (2004). Global tuberculosis control : surveillance, planning, financing : WHO report 2004. In. Geneva: World Health Organization.
[63] Workicho A, Kassahun W, Alemseged F. Risk factors for multidrug-resistant tuberculosis among tuberculosis patients: a case-control study. Infection and drug resistance 2017;10:91.
[64] Worku S, Derbie A, Mekonnen D, Biadglegne F. Treatment outcomes of tuberculosis patients under directly observed treatment short-course at Debre Tabor General Hospital, northwest Ethiopia: nine-years retrospective study. Infect Dis Poverty 2018;7:1–7.
[65] Xi Y, Zhang W, Qiao R-J, Tang J. Risk factors for multidrug-resistant tuberculosis: A worldwide systematic review and meta-analysis. PLoS One 2022;17(6): e0270003.
[66] Yang Y, Zhou C, Shi L, Meng H, Yan H. Prevalence and characterization of drug-resistant tuberculosis in a local hospital of Northeast China. Int J Infect Dis 2014; 22:83–6.
[67] Yunusbaeva, M., Borodina, L., Alekseev, P., Davydov, R., Yunusbaev, U., Sharipov, R.,…Yunusbayev, B. (2019). Treatment efficacy of drug-resistant tuberculosis in Bashkortostan, Russia: a retrospective cohort study. *International Journal of Infectious Diseases*, *81*, 203-209.
[68] Zhang Z, Pang Y, Wang Y, Liu C, Zhao Y. Beijing genotype of Mycobacterium tuberculosis is significantly associated with linezolid resistance in multidrug-resistant and extensively drug-resistant tuberculosis in China. Int J Antimicrob Agents 2014;43(3):231–5.
[69] Zhao, Y., Xu, S., Wang, L., Chin, D. P., Wang, S., Jiang, G.,…Ou, X. (2012). National survey of drug-resistant tuberculosis in China. *New England Journal of Medicine*, *366*(23), 2161-2170.
[70] Zhou C, Chu J, Geng H, Wang X, Xu L. Pulmonary tuberculosis among migrants in Shandong, China: factors associated with treatment delay. BMJ Open 2014;4(12): e005805.

[71] Zignol M, Sismanidis C, Falzon D, Glaziou P, Dara M, Floyd K. Multidrug-resistant tuberculosis in children: evidence from global surveillance. Eur Respir J 2013;42 (3):701–7.
[72] Zumla A, George A, Sharma V, Herbert RHN, Oxley A, Oliver M. The WHO 2014 global tuberculosis report—further to go. Lancet Glob Health 2015;3(1):e10–2.
[73] Zuur MA, Pasipanodya JG, van Soolingen D, van der Werf TS, Gumbo T, Alffenaar J-W-C. Intermediate susceptibility dose-dependent breakpoints for high-dose rifampin, isoniazid, and pyrazinamide treatment in multidrug-resistant tuberculosis programs. Clin Infect Dis 2018;67(11):1743–9.