1. **An overview of tuberculosis outbreaks reported in the years 2011-2020.**

BMC Infect Dis. 2023 Apr 20;23(1):253. doi: 10.1186/s12879-023-08197-w.

Żukowska L(1)(2), Zygała-Pytlos D(1)(2), Struś K(1)(3), Zabost A(4), Kozińska M(4), Augustynowicz-Kopeć E(4), Dziadek J(1), Minias A(5).

BACKGROUND: In many countries tuberculosis (TB) remains a highly prevalent disease and a major contributor to infectious disease mortality. The fight against TB requires surveillance of the population of strains circulating worldwide and the analysis of the prevalence of certain strains in populations. Nowadays, whole genome sequencing (WGS) allows for accurate tracking of TB transmission. Currently, there is a lack of a comprehensive summary of the characteristics of TB outbreaks.
METHODS: We systematically analyzed studies reporting TB outbreaks worldwide, monitored through WGS of Mycobacterium tuberculosis. We 1) mapped the reported outbreaks from 2011- 2020, 2) estimated the average size of the outbreaks, 3) indicated genetic lineages causing the outbreaks, and 4) determined drug-resistance patterns of M. tuberculosis strains involved in the outbreaks.
RESULTS: Most data originated from Europe, Asia, and North America. We found that TB outbreaks were reported throughout the globe, on all continents, and in countries with both high and low incidences. The detected outbreaks contained a median of five M. tuberculosis isolates. Most strains causing the outbreaks belonged to lineage four, more rarely to lineage two. Reported outbreak isolates were often drug resistant.
CONCLUSIONS: We conclude that more WGS surveillance of M. tuberculosis outbreaks is needed. Globally standardized procedures might improve the control of M. tuberculosis infections.

DOI: 10.1186/s12879-023-08197-w
PMCID: PMC10116450
PMID: 37081448 [Indexed for MEDLINE]

Conflict of interest statement: The authors declare no conflict of interest.

3. **Evolution of tuberculosis diagnostics: From molecular strategies to nanodiagnostics.**

Tuberculosis (Edinb). 2023 Apr 5;140:102340. doi: 10.1016/j.tube.2023.102340. Online ahead of print.

Mukherjee S(1), Perveen S(2), Negi A(2), Sharma R(3).

Tuberculosis has remained a global concern for public health affecting the lives of people for ages. Approximately 10 million people are affected by the disease and 1.5 million succumb to the disease worldwide annually. The COVID-19 pandemic has highlighted the role of early diagnosis to win the battle against such infectious diseases. Thus, advancement in the diagnostic approaches to provide early detection forms the foundation to eradicate and manage contagious diseases like tuberculosis. The conventional diagnostic strategies include microscopic examination, chest X-ray and tuberculin skin test. The limitations associated with sensitivity and specificity of these tests demands for exploring new techniques like probe-based assays, CRISPR-Cas and microRNA detection. The aim of the current review is to envisage the correlation between both the conventional and the newer approaches to enhance the specificity and sensitivity. A significant emphasis has been placed upon nanodiagnostic approaches manipulating quantum dots, magnetic nanoparticles, and biosensors for accurate diagnosis of latent, active and drug-resistant TB. Additionally, we would like to ponder upon a reliable method that is cost-effective, reproducible, require minimal infrastructure and provide point-of-care to the patients.

DOI: 10.1016/j.tube.2023.102340
PMCID: PMC10072981
PMID: 37031646

Conflict of interest statement: Declaration of competing interest Authors declares no conflict of interest.

4. **Pyrazinamide-resistant Tuberculosis Obscured From Common Targeted Molecular Diagnostics.**

Drug Resist Updat. 2023 May;68:100959. doi: 10.1016/j.drup.2023.100959. Epub 2023 Apr 6.

Modlin SJ(1), Mansjö M(2), Werngren J(2), Ejike CM(1), Hoffner SE(3), Valafar F(4).

Here, we describe a clinical case of pyrazinamide-resistant (PZA-R) tuberculosis (TB) reported as PZA-susceptible (PZA-S) by common molecular diagnostics. Phenotypic susceptibility testing (pDST) indicated PZA-R TB. Targeted Sanger sequencing reported wild-type PncA, indicating PZA-S TB. Whole Genome Sequencing

(WGS) by PacBio and IonTorrent both detected deletion of a large portion of pncA, indicating PZA-R. Importantly, both WGS methods showed deletion of part of the primer region targeted by Sanger sequencing. Repeating Sanger sequencing from a culture in presence of PZA returned no result, revealing that 1) two minority susceptible subpopulations had vanished, 2) the PZA-R majority subpopulation harboring the pncA deletion could not be amplified by Sanger primers, and was thus obscured by amplification process. This case demonstrates how a small susceptible subpopulation can entirely obscure majority resistant populations from targeted molecular diagnostics and falsely imply homogenous susceptibility, leading to incorrect diagnosis. To our knowledge, this is the first report of a minority susceptible subpopulation masking a majority resistant population, causing targeted molecular diagnostics to call false susceptibility. The consequence of such genomic events is not limited to PZA. This phenomenon can impact molecular diagnostics' sensitivity whenever the resistance-conferring mutation is not fully within primer-targeted regions. This can be caused by structural changes of genomic context with phenotypic consequence as we report here, or by uncommon mechanisms of resistance. Such false susceptibility calls promote suboptimal treatment and spread of strains that challenge targeted molecular diagnostics. This motivates development of molecular diagnostics unreliant on primer conservation, and impels frequent WGS surveillance for variants that evade prevailing molecular diagnostics.

DOI: 10.1016/j.drup.2023.100959
PMID: 37043916 [Indexed for MEDLINE]

Conflict of interest statement: 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.

5. **Epidemiology of extensively drug-resistant tuberculosis among multidrug resistant tuberculosis patients: A systematic review and meta-analysis.**

Int J Infect Dis. 2023 Apr 16:S1201-9712(23)00524-6. doi: 10.1016/j.ijid.2023.04.392. Online ahead of print.

Diriba G(1), Alemu A(2), Yenew B(2), Tola HH(3), Gamtesa DF(2), Mollalign H(2), Eshetu K(4), Moga S(2), Abdella S(2), Tollera G(2), Kebede A(5), Dangisso MH(2).

OBJECTIVE: To estimate the pooled proportion of extensively drug-resistant tuberculosis (XDR-TB) and pre-extensively drug-resistant tuberculosis

(pre-XDR-TB) in multidrug-resistant TB (MDR-TB) patients.
METHODS: We systematically searched articles from electronic databases: MEDLINE (PubMed), ScienceDirect, and Google Scholar. We also searched grey from different literature sources main outcome of the review was either XDR-TB or pre-XDR-TB in MDR-TB patients. We used the random-effects model considering substantial heterogeneity among studies. Heterogeneity was assessed by subgroup analyses. STATA version 14 was used for analysis.
RESULTS: A total of 64 studies reported on 12,711 MDR-TB patients from 22 countries were retrieved. The pooled proportion of pre-XDR-TB was 26% (95%CI: 22%-31%), while XDR-TB in MDR-TB cases was 9% (95%CI: 7%-11%) in patients treated for MDR-TB. The pooled proportion of resistance to fluoroquinolones was 27% (95%CI: 22%-33%) and second-line injectable-drugs was 11% (95%CI: 9%-13%). Whereas the pooled resistance proportion to bedaquiline, clofazimine, delamanid, and linezolid was 5% (95%CI: 1%-8%), 4% (95%CI: 0%-10%), 5% (95%CI; 2%- 8%), and 4% (95%CI: 2%-10%), respectively.
CONCLUSIONS: The burden of pre-XDR-TB and XDR-TB in MDR-TB were considerable. The high burdens of pre-XDR-TB and XDR-TB in patients treated for MDR-TB is suggesting the need of strengthen TB program and drug resistance surveillance.

DOI: 10.1016/j.ijid.2023.04.392
PMID: 37072053

6. **Post-tuberculosis lung impairment: systematic review and meta-analysis of spirometry data from 14 621 people.**

Eur Respir Rev. 2023 Apr 19;32(168):220221. doi: 10.1183/16000617.0221-2022. Print 2023 Jun 30.

Ivanova O(1)(2)(3), Hoffmann VS(4)(3), Lange C(5)(6)(7)(8), Hoelscher M(9)(2), Rachow A(9)(2).

BACKGROUND: A substantial proportion of tuberculosis patients remain with pulmonary symptoms and reduced physical capacity despite successful treatment. We performed a systematic review to analyse the burden of post-tuberculosis lung impairment measured by lung function testing.
METHODS: We searched the PubMed database for articles published between database inception and November 2020 and performed meta-analyses to estimate the prevalence, type and severity of lung impairment among drug-susceptible and multidrug-resistant tuberculosis survivors. Methodological quality of included studies was assessed using the Newcastle-Ottawa scale.
RESULTS: 54 articles were included in this review. For subjects with former

drug-susceptible tuberculosis, the combined estimated mean was 76.6% (95% CI 71.6-81.6) of predicted for forced expiratory volume in 1 s (FEV1) and 81.8% (95% CI 77.4-86.2) for forced vital capacity (FVC). In former patients with multidrug-resistant tuberculosis, it was 65.9% (95% CI 57.1-74.7) for FEV1 and 76.0% (95% CI 66.3-85.8) for FVC, respectively. The analysis of impairment types in former patients with drug-susceptible and multidrug-resistant tuberculosis showed that 22.0% versus 19.0% had obstructive, 23.0% versus 22.0% restrictive and 15.0% versus 43.0% had mixed impairment type, respectively. In the majority of studies, at least 10-15% of tuberculosis survivors had severe lung impairment.
CONCLUSIONS: This systematic review showed long-term abnormal spirometry results in a significant proportion of tuberculosis survivors.

DOI: 10.1183/16000617.0221-2022
PMCID: PMC10113954
PMID: 37076175 [Indexed for MEDLINE]

Conflict of interest statement: Conflict of interest: The authors declare no conflict of interest.

7. **Linezolid Pharmacokinetics and Its Association with Adverse Drug Reactions in Patients with Drug-Resistant Pulmonary Tuberculosis.**

Antibiotics (Basel). 2023 Apr 6;12(4):714. doi: 10.3390/antibiotics12040714.

Padmapriyadarsini C(1), Solanki R(2), Jeyakumar SM(1), Bhatnagar A(3), Muthuvijaylaksmi M(1), Jeyadeepa B(1), Reddy D(1), Shah P(2), Sridhar R(4), Vohra V(5), Bhui NK(6).

We evaluated the relationship between the pharmacokinetic parameters of linezolid (LZD) and development of adverse drug reactions (ADRs) in patients with pulmonary drug-resistant tuberculosis. A prospective cohort of adults with pulmonary multidrug-resistant tuberculosis with additional resistance to fluoroquinolone (MDR-TBFQ+) received treatment with bedaquiline, delamanid, clofazimine, and LZD. Blood samples were collected during weeks 8 and 16 at eight time points over 24 h. The pharmacokinetic parameters of LZD were measured using high-performance liquid chromatography and associated with ADRs. Of the 165 MDR-TBFQ+ patients on treatment, 78 patients developed LZD-associated anemia and 69 developed peripheral neuropathy. Twenty-three patients underwent intense pharmacokinetic testing. Plasma median trough concentration was 2.08 µg/mL and 3.41 µg/mL, (normal <2 µg/mL) and AUC0-24 was 184.5 µg/h/mL and 240.5 µg/h/mL at

weeks 8 and 16, respectively, showing a linear relationship between duration of intake and plasma levels. Nineteen patients showed LZD-associated ADRs-nine at week 8, twelve at week 16, and two at both weeks 8 and 16. Thirteen of the nineteen had high plasma trough and peak concentrations of LZD. A strong association between LZD-associated ADRs and plasma LZD levels was noted. Trough concentration alone or combinations of trough with peak levels are potential targets for therapeutic drug monitoring.

DOI: 10.3390/antibiotics12040714
PMCID: PMC10135341
PMID: 37107075

Conflict of interest statement: The authors declare no conflict of interest. The sponsors had no role in the design, execution, interpretation, or writing of the study.

8. **The relative transmission fitness of multidrug-resistant Mycobacterium tuberculosis in a drug resistance hotspot.**

Nat Commun. 2023 Apr 8;14(1):1988. doi: 10.1038/s41467-023-37719-y.

Loiseau C(#)(1)(2), Windels EM(#)(3)(4), Gygli SM(1)(2), Jugheli L(1)(2)(5), Maghradze N(1)(2)(5), Brites D(1)(2), Ross A(1)(2), Goig G(1)(2), Reinhard M(1)(2), Borrell S(1)(2), Trauner A(1)(2), Dötsch A(1)(2), Aspindzelashvili R(5), Denes R(6), Reither K(1)(2), Beisel C(6), Tukvadze N(1)(2)(5), Avaliani Z(5), Stadler T(6)(7), Gagneux S(8)(9).

Multidrug-resistant tuberculosis (MDR-TB) is among the most frequent causes of death due to antimicrobial resistance. Although only 3% of global TB cases are MDR, geographical hotspots with up to 40% of MDR-TB have been observed in countries of the former Soviet Union. While the quality of TB control and patient-related factors are known contributors to such hotspots, the role of the pathogen remains unclear. Here we show that in the country of Georgia, a known hotspot of MDR-TB, MDR Mycobacterium tuberculosis strains of lineage 4 (L4) transmit less than their drug-susceptible counterparts, whereas most MDR strains of L2 suffer no such defect. Our findings further indicate that the high transmission fitness of these L2 strains results from epistatic interactions between the rifampicin resistance-conferring mutation RpoB S450L, compensatory mutations in the RNA polymerase, and other pre-existing genetic features of L2/Beijing clones that circulate in Georgia. We conclude that the transmission fitness of MDR M. tuberculosis strains is heterogeneous, but can be as high as drug-susceptible forms, and that such highly drug-resistant and transmissible strains contribute to the emergence and maintenance of hotspots of MDR-TB. As

these strains successfully overcome the metabolic burden of drug resistance, and given the ongoing rollout of new treatment regimens against MDR-TB, proper surveillance should be implemented to prevent these strains from acquiring resistance to the additional drugs.

DOI: 10.1038/s41467-023-37719-y
PMCID: PMC10082831
PMID: 37031225 [Indexed for MEDLINE]

Conflict of interest statement: The authors declare no competing interests.

9. **Optimizing mycobacteria molecular diagnostics: No decontamination! Human DNA depletion? Greener storage at 4 °C!**

Front Microbiol. 2023 Apr 11;14:1104752. doi: 10.3389/fmicb.2023.1104752. eCollection 2023.

Prajwal P(1)(2)(3), Neary T(1), Rohrbach K(1), Bittel P(1), Göller PC(1)(4), Buch T(2), Dümcke S(3), Keller PM(1).

INTRODUCTION: Tuberculosis (TB) is an infectious disease caused by the group of bacterial pathogens Mycobacterium tuberculosis complex (MTBC) and is one of the leading causes of death worldwide. Timely diagnosis and treatment of drug-resistant TB is a key pillar of WHO's strategy to combat global TB. The time required to carry out drug susceptibility testing (DST) for MTBC via the classic culture method is in the range of weeks and such delays have a detrimental effect on treatment outcomes. Given that molecular testing is in the range of hours to 1 or 2 days its value in treating drug resistant TB cannot be overstated. When developing such tests, one wants to optimize each step so that tests are successful even when confronted with samples that have a low MTBC load or contain large amounts of host DNA. This could improve the performance of the popular rapid molecular tests, especially for samples with mycobacterial loads close to the limits of detection. Where optimizations could have a more significant impact is for tests based on targeted next generation sequencing (tNGS) which typically require higher quantities of DNA. This would be significant as tNGS can provide more comprehensive drug resistance profiles than the relatively limited resistance information provided by rapid tests. In this work we endeavor to optimize pre-treatment and extraction steps for molecular testing.
METHODS: We begin by choosing the best DNA extraction device by comparing the amount of DNA extracted by five commonly used devices from identical samples.

Following this, the effect that decontamination and human DNA depletion have on extraction efficiency is explored.
RESULTS: The best results were achieved (i.e., the lowest Ct values) when neither decontamination nor human DNA depletion were used. As expected, in all tested scenarios the addition of decontamination to our workflow substantially reduced the yield of DNA extracted. This illustrates that the standard TB laboratory practice of applying decontamination, although being vital for culture-based testing, can negatively impact the performance of molecular testing. As a complement to the above experiments, we also considered the best Mycobacterium tuberculosis DNA storage method to optimize molecular testing carried out in the near- to medium-term. Comparing Ct values following three-month storage at 4 °C and at -20 °C and showed little difference between the two.
DISCUSSION: In summary, for molecular diagnostics aimed at mycobacteria this work highlights the importance of choosing the right DNA extraction device, indicates that decontamination causes significant loss of mycobacterial DNA, and shows that samples preserved for further molecular testing can be stored at 4 °C, just as well at -20 °C. Under our experimental settings, human DNA depletion gave no significant improvement in Ct values for the detection of MTBC.

DOI: 10.3389/fmicb.2023.1104752
PMCID: PMC10126496
PMID: 37113238

Conflict of interest statement: This work was funded by an Innosuisse project whose industrial partner is Clemedi AG, a Medtech company that develops in vitro molecular diagnostic solutions. SD, PK, TB, and PP hold shares in the company, and TB is on the board of directors. SD is Clemedi CEO, PP is Clemedi CSO, TN and PK are part of Clemedi's R&D team, PG worked for Clemedi in the past. The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

10. **Molecular typing and drug sensitivity profiles of M. Tuberculosis isolated from refugees residing in Ethiopia.**

J Clin Tuberc Other Mycobact Dis. 2023 Apr 15;31:100371. doi: 10.1016/j.jctube.2023.100371. eCollection 2023 May.

Meaza A(1)(2), Diriba G(2), Girma M(1), Wondimu A(2), Worku G(1)(3), Medhin

G(1), Ameni G(1)(4), Gumi B(1).

BACKGROUND: Refugees in developing countries have poor access to Tuberculosis (TB) care and control services. The understanding of genetic diversity and drug sensitivity patterns of M. tuberculosis (MTB) is important for the TB control program. However, there is no evidence that shows the drug sensitivity profiles and genetic diversity of MTB circulating among refugees residing in Ethiopia. This study aimed to investigate the genetic diversity of MTB strains and lineages, and to identify the drug sensitivity profiles of MTB isolated from refugees residing in Ethiopia.
METHODS: A cross-sectional study was conducted among 68 MTB positive cases isolated from presumptive TB refugees from February to August 2021. Data and samples were collected in the refugee camp clinics and both rapid TB Ag detection and region of difference (RD)-9 deletion typing were used to confirm the MTBs. Drug susceptibility test (DST) and molecular typing were done using Mycobacterium Growth Indicator Tube (MGIT) method and spoligotyping respectively.
RESULTS: DST and spoligotyping results were available for all 68 isolates. The isolates were grouped into 25 spoligotype patterns, which consisted of 1-31 isolates with 36.8% strain diversity. The international shared type (SIT)25 was predominant spoligotype pattern consisting of 31 (45.6%) isolates, followed by SIT24 comprising 5 (7.4%) isolates. Further investigation showed that 64.7% (44/68) of the isolates were belonged to CAS1-Delhi family and 75% (51/68) of the isolates were belonged to lineage(L)-3. Multi-drug resistance (MDR)-TB was observed only in one isolate (1.5%) for first-line anti-TB drugs and the highest level of mono-resistance, 5.9% (4/68), was observed for PZA(Pyrazinamide). Mono-resistance was observed in 2.9 % (2/68) and while 97.0% (66/68) of the MTB positive cases were susceptible to the second-line anti-TB drugs.
CONCLUSION: The findings are useful evidence for the TB screening, treatment and control in refugee populations and surrounding communities in Ethiopia.

DOI: 10.1016/j.jctube.2023.100371
PMCID: PMC10127110
PMID: 37113677

Conflict of interest statement: 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.

11. **Comparative effectiveness of adding delamanid to a multidrug-resistant tuberculosis regimen comprised of three drugs likely to be effective.**

PLOS Glob Public Health. 2023 Apr 28;3(4):e0000818. doi: 10.1371/journal.pgph.0000818. eCollection 2023.

Rodriguez CA(1)(2), Lodi S(3), Horsburgh CR(1)(3)(4)(5), Mitnick CD(2)(6)(7), Bastard M(8), Huerga H(8), Khan U(9), Rich M(6)(7), Seung KJ(6)(7), Atwood S(2), Manzur-Ul-Alam M(10), Melikyan N(8), Mpinda S(11), Myint Z(12), Naidoo Y(13), Petrosyan O(14), Salahuddin N(15), Sarfaraz S(15), Vilbrun SC(16), Yae K(17), Achar J(18), Ahmed S(19), Algozhina E(20), Beauchamp J(21), de Guadelupe Perea Moreno S(22), Gulanbaeva M(23), Gergedava M(24), Indah Sari CY(25), Hewison C(26), Khan P(9), Franke MF(2).

Clarity about the role of delamanid in longer regimens for multidrug-resistant TB is needed after discordant Phase IIb and Phase III randomized controlled trial results. The Phase IIb trial found that the addition of delamanid to a background regimen hastened culture conversion; the results of the Phase III trial were equivocal. We evaluated the effect of adding delamanid for 24 weeks to three-drug MDR/RR-TB regimens on two- and six-month culture conversion in the endTB observational study. We used pooled logistic regression to estimate the observational analogue of the intention-to-treat effect (aITT) adjusting for baseline confounders and to estimate the observational analogue of the per-protocol effect (aPP) using inverse probability of censoring weighting to control for time-varying confounding. At treatment initiation, 362 patients received three likely effective drugs (delamanid-free) or three likely effective drugs plus delamanid (delamanid-containing). Over 80% of patients received two to three Group A drugs (bedaquiline, linezolid, moxifloxacin/levofloxacin) in their regimen. We found no evidence the addition of delamanid to a three-drug regimen increased two-month (aITT relative risk: 0.90 (95% CI: 0.73-1.11), aPP relative risk: 0.89 (95% CI: 0.66-1.21)) or six-month culture conversion (aITT relative risk: 0.94 (95% CI: 0.84, 1.02), aPP relative risk: 0.93 (95% CI: 0.83, 1.04)). In regimens containing combinations of three likely effective, highly active anti-TB drugs the addition of delamanid had no discernible effect on culture conversion at two or six months. As the standard of care for MDR/RR-TB treatment becomes more potent, it may become increasingly difficult to detect the benefit of adding a single agent to standard of care MDR/RR-TB regimens. Novel approaches like those implemented may help account for background regimens and establish effectiveness of new chemical entities.

DOI: 10.1371/journal.pgph.0000818

PMCID: PMC10146539
PMID: 37115740

Conflict of interest statement: The endTB Consortium coordinated donations of delamanid (Otsuka Pharmaceutical) and bedaquiline (Janssen) to be used for treatment by some of the patients included in the endTB Observational Study. All authors report no additional potential conflicts of interest. There are no patents, products in development or marketed products associated with this research to declare. This does not alter our adherence to PLOS ONE policies on sharing data and materials.

12. **Socioeconomic disparities and multidrug-resistant tuberculosis in South Korea: Focus on immigrants and income levels.**

J Microbiol Immunol Infect. 2023 Apr;56(2):424-428. doi: 10.1016/j.jmii.2022.08.014. Epub 2022 Sep 7.

Jeong HE(1), Bea S(2), Kim JH(1), Jang SH(3), Son H(4), Shin JY(5).

Risk factors of MDR-TB remain unclear in South Korea, despite being an important public health issue. Findings from this study, which included ≥50,000 patients with TB from South Korea, suggests that immigrants and patients with lower income levels were strong predictors of MDR-TB in a high-income, high TB incidence country.

DOI: 10.1016/j.jmii.2022.08.014
PMID: 36115791 [Indexed for MEDLINE]

Conflict of interest statement: Declaration of competing interest All authors declare no competing interests.

13. **FLASH-TB: an Application of Next-Generation CRISPR to Detect Drug Resistant Tuberculosis from Direct Sputum.**

J Clin Microbiol. 2023 Apr 20;61(4):e0163422. doi: 10.1128/jcm.01634-22. Epub 2023 Apr 3.

Tram TTB(#)(1), Ha VTN(#)(1), Trieu LPT(1), Ashton PM(1)(2)(3), Crawford ED(4), Thu DDA(1), Quang NL(1), Thwaites GE(1)(2), Walker TM(1)(2), Anscombe C(1)(2), Thuong NTT(1)(2).

Offering patients with tuberculosis (TB) an optimal and timely treatment regimen depends on the rapid detection of Mycobacterium tuberculosis (Mtb) drug resistance from clinical samples. Finding Low Abundance Sequences by Hybridization (FLASH) is a technique that harnesses the efficiency, specificity, and flexibility of the Cas9 enzyme to enrich targeted sequences. Here, we used FLASH to amplify 52 candidate genes probably associated with resistance to first- and second-line drugs in the Mtb reference strain (H37Rv), then detect drug resistance mutations in cultured Mtb isolates, and in sputum samples. 92% of H37Rv reads mapped to Mtb targets, with 97.8% of target regions covered at a depth ≥ 10X. Among cultured isolates, FLASH-TB detected the same 17 drug resistance mutations as whole genome sequencing (WGS) did, but with much greater depth. Among the 16 sputum samples, FLASH-TB increased recovery of Mtb DNA compared with WGS (from 1.4% [IQR 0.5-7.5] to 33% [IQR 4.6-66.3]) and average depth reads of targets (from 6.3 [IQR 3.8-10.5] to 1991 [IQR 254.4-3623.7]). FLASH-TB identified Mtb complex in all 16 samples based on IS1081 and IS6110 copies. Drug resistance predictions for 15/16 (93.7%) clinical samples were highly concordant with phenotypic DST for isoniazid, rifampicin, amikacin, and kanamycin [15/15 (100%)], ethambutol [12/15 (80%)] and moxifloxacin [14/15 (93.3%)]. These results highlighted the potential of FLASH-TB for detecting Mtb drug resistance from sputum samples.

DOI: 10.1128/jcm.01634-22
PMCID: PMC10117099
PMID: 37010411 [Indexed for MEDLINE]

Conflict of interest statement: The authors declare no conflict of interest.

14. **Genomic Sequencing Profiles of Mycobacterium tuberculosis in Mandalay Region, Myanmar.**

Trop Med Infect Dis. 2023 Apr 21;8(4):239. doi: 10.3390/tropicalmed8040239.

Phyu AN(1)(2), Aung ST(3), Palittapongarnpim P(4), Htet KKK(2), Mahasirimongkol S(5), Ruangchai W(4), Jaemsai B(4), Aung HL(6), Maung HMW(2), Chaiprasert A(7), Pungrassami P(8), Chongsuvivatwong V(2).

This study aimed to characterize whole-genome sequencing (WGS) information of Mycobacterium tuberculosis (Mtb) in the Mandalay region of Myanmar. It was a cross-sectional study conducted with 151 Mtb isolates obtained from the fourth nationwide anti-tuberculosis (TB) drug-resistance survey. Frequency of lineages 1, 2, 3, and 4 were 55, 65, 9, and 22, respectively. The most common sublineage was L1.1.3.1 (n = 31). Respective multi-drug resistant tuberculosis (MDR-TB)

frequencies were 1, 1, 0, and 0. Four clusters of 3 (L2), 2 (L4), 2 (L1), and 2 (L2) isolates defined by a 20-single-nucleotide variant (SNV) cutoff were detected. Simpson's index for sublineages was 0.0709. Such high diversity suggests that the area probably had imported Mtb from many geographical sources. Relatively few genetic clusters and MDR-TB suggest there is a chance the future control will succeed if it is carried out properly.

DOI: 10.3390/tropicalmed8040239
PMCID: PMC10141229
PMID: 37104364

Conflict of interest statement: The authors declare no conflict of interest.

15. **Clinical characteristics and drug resistance profile of patients with endobronchial tuberculosis in South Korea: single-center experience.**

Ann Palliat Med. 2023 Apr 13:apm-22-1218. doi: 10.21037/apm-22-1218. Online ahead of print.

Moon SM(1), Lee WY(2), Shin B(1).

BACKGROUND: Tuberculosis (TB) is a major infectious disease worldwide; there has been a significant increase in the number of elderly patients with TB, largely contributing to TB-related mortality. Although endobronchial tuberculosis (EBTB) is a unique form of pulmonary TB, available data on the clinical characteristics and drug susceptibility (DST) patterns of patients with EBTB are scarce.
METHODS: We evaluated the clinical characteristics of patients with EBTB in South Korea and the culturebased DST patterns of EBTB. Further, the DST patterns were compared between elderly (≥65 years) and young (<65 years) patients. We retrospectively reviewed data of patients with EBTB who had the results of DST and were diagnosed between January 2013 and December 2019 at a tertiary referral hospital in South Korea. Phenotypic DST of 15 first-line and second-line anti-TB drugs was performed using Mycobacterium tuberculosis isolates prior to treatment.
RESULTS: Of the 230 patients with EBTB, 69% were in elderly patients (≥65 years). Any-resistance occurred in 24 patients (10.4%), while multi-drug resistance (MDR) and extensive drug resistance (XDR) were observed in six patients (2.6%). Compared to that of the elderly treatment-naïve patients, previously treated elderly patients had a significantly higher proportion of resistance to rifampin (14.3% vs. 2.2%; P=0.031), ethambutol (9.5% vs. 0.7%; P=0.046), and pyrazinamide (9.5% vs. 0.7%; P=0.046). Further, MDR/XDR was observed more frequently in the previously treated elderly patients than that in the treatment-naïve elderly patients (14.3% vs. 1.4%; P=0.017). A relatively

small number of drug-resistant cases (5.6%) were observed in young patients.
CONCLUSIONS: Elderly EBTB patients with previous Anti-tuberculous medications had a significantly higher proportion of drug-resistant TB. These patients should be carefully assessed using DST analysis before treatment.

DOI: 10.21037/apm-22-1218
PMID: 37081701

16. **Factors affecting successful antituberculosis treatment: a single-center experience.**

Rev Assoc Med Bras (1992). 2023 Apr 14;69(4):e20221054. doi: 10.1590/1806-9282.20221054. eCollection 2023.

Gonçalves MC(1), Aguiar AAS(2), Biadola AP(3), Mazaro PJM(4), Rodrigues MVP(3), Prado RLD(3), Peresi-Lordelo E(3).

OBJECTIVE: The identification of factors that influence a favorable antituberculosis treatment outcome could be of great use for the promotion of specific health actions to increase the success rate. Thus, the objective of this study was to investigate the factors affecting successful antituberculosis treatment in patients seen at a reference service in the Western region of São Paulo State/Brazil.
METHODS: A retrospective study was carried out from 2010 to 2016 based on the data obtained from the Notification Disease Information System of TB patients treated at a reference service in Brazil. The study included patients with treatment outcomes and excluded those from the penitentiary system or with resistant or multidrug-resistant TB. Patients were categorized as having a successful (cured) or unsuccessful (treatment default and death) treatment outcome. The association between TB treatment outcomes and social and clinical factors was analyzed.
RESULTS: A total of 356 cases of TB were treated between 2010 and 2016. Among the cases, the majority were cured and the overall treatment success rate was 85.96%, with a range between 80.33% (2010) and 97.65% (2016). After the exclusion of resistant/multidrug-resistant TB, 348 patients were analyzed. In the final logistic regression model analysis, education less than 8 years (OR 1.66; $p<0.0001$) and people living with human immunodeficiency virus/acquired immunodeficiency syndrome (OR 0.23; $p<0.0046$) were found to be significantly related to an unfavorable treatment outcome.
CONCLUSION: Low education and being a person living with human immunodeficiency virus/acquired immunodeficiency syndrome are vulnerability factors that can affect the successful outcome of antituberculosis treatment.

DOI: 10.1590/1806-9282.20221054
PMID: 37075442 [Indexed for MEDLINE]

17. **QcrB inhibition as a potential approach for the treatment of tuberculosis: A review of recent developments, patents, and future directions.**

J Infect Public Health. 2023 Apr 13;16(6):928-937. doi: 10.1016/j.jiph.2023.04.011. Online ahead of print.

Imran M(1), Abida(2), Alotaibi NM(3), Thabet HK(4), Alruwaili JA(5), Asdaq SMB(6), Eltaib L(7), Alshehri A(8), Alsaiari AA(9), Almehmadi M(9), Alshammari ABH(10), Alshammari AM(10).

The unmet medical need for drug-resistant tuberculosis (DRTB) is a significant concern. Accordingly, identifying new drug targets for tuberculosis (TB) treatment and developing new therapies based on these drug targets is one of the strategies to tackle DRTB. QcrB is an innovative drug target to create treatments for DRTB. This article highlights QcrB inhibitors and their therapeutic compositions for treating TB. The literature for this article was gathered from PubMed and free patent databases utilizing different keywords related to QcrB inhibitor-based inventions. The data was collected from the conceptualization of telacebec (2010) QcrB to December 2022. A little interesting and encouraging research has been performed on QcrB inhibitors. Telacebec and TB47 are established QcrB inhibitors in the clinical trial. The inventive QcrB inhibitor-based drug combinations can potentially handle DRTB and reduce the TB therapy duration. The authors anticipate great opportunities in fostering QcrB inhibitor-based patentable pharmaceutical inventions against TB. Drug repurposing can be a promising strategy to get safe and effective QcrB inhibitors. However, developing drug resistance, drug tolerance, and selectivity of QcrB inhibitors for Mtb will be the main challenges in developing effective QcrB inhibitors. In conclusion, QcrB is a promising drug target for developing effective treatments for active, latent, and drug-resistant TB. Many inventive and patentable combinations and compositions of QcrB inhibitors with other anti-TB drugs are anticipated as future treatments for TB.

DOI: 10.1016/j.jiph.2023.04.011
PMID: 37086552

Conflict of interest statement: 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.

18. **Practice regarding tuberculosis care among physicians at private facilities: A cross-sectional study from Vietnam.**

PLoS One. 2023 Apr 27;18(4):e0284603. doi: 10.1371/journal.pone.0284603. eCollection 2023.

Ngo DM(1), Doan NB(1), Tran SN(2), Hoang LB(3), Nguyen HB(4), Nguyen VD(1).

OBJECTIVES: To evaluate the practice of TB care among physicians at private facilities.
METHODS: A cross-sectional study was conducted using questionnaires on knowledge, attitude, and practice related to TB care. The responses to these scales were used to explore latent constructs and calculate the standardized continuous scores for these domains. We described the percentages of participant's responses and explored their associated factors using multiple linear regression.
RESULTS: A total of 232 physicians were recruited. The most important gaps in practice included requesting chest imaging to confirm TB diagnosis (~80%), not testing HIV for confirmed active TB cases (~50%), only requesting sputum testing for MDR-TB cases (65%), only requesting follow-up examination at the end of the treatment course (64%), and not requesting sputum testing at follow-up (54%). Surgical mask was preferred to N95 respirator when examining TB patients. Prior TB training was associated with better knowledge and less stigmatizing attitude, which were associated with better practice in both TB management and precautions.
CONCLUSION: There were important gaps in knowledge, attitude, and practice of TB care among private providers. Better knowledge was associated with positive attitude towards TB and better practice. Tailored training may help to address these gaps and improve the quality of TB care in the private sector.

DOI: 10.1371/journal.pone.0284603
PMCID: PMC10138252
PMID: 37104504 [Indexed for MEDLINE]

Conflict of interest statement: The authors have declared that no competing interests exist.

19. **Public health concerns about Tuberculosis caused by Russia/Ukraine conflict.**

Health Sci Rep. 2023 Apr 17;6(4):e1218. doi: 10.1002/hsr2.1218. eCollection 2023 Apr.

Paul IK(1), Nchasi G(1), Bulimbe DB(2), Mollel MK(2), Msafiri GG(2), Mbogo A(3), Mswanzari MB(2), Joseph M(2), Mahmoud A(4), Volkova A(5).

According to WHO, Ukraine has the fourth-highest Tuberculosis (TB) incidence in the WHO European region while globally has the fifth-highest number of confirmed cases of extensively drug-resistant TB. Before the Russian invasion in Ukraine several interventions have been employed to mitigate the TB epidemic in the country. However, the ongoing war has demolished meticulous efforts and subsequently worsen the situation. WHO in collaboration with the Ukraine government and other organizations such as EU and UK are required to take up arms against the situation. In this work, implications brought up from the war, efforts, and recommendations to battling TB epidemic due to the war are highlighted.

DOI: 10.1002/hsr2.1218
PMCID: PMC10108851
PMID: 37077183

Conflict of interest statement: The authors declare no conflict of interest

20. **Tuberculosis infection prevention and control in rural Papua New Guinea: an evaluation using the infection prevention and control assessment framework.**

Antimicrob Resist Infect Control. 2023 Apr 12;12(1):31. doi: 10.1186/s13756-023-01237-9.

Marme G(1), Kuzma J(2), Zimmerman PA(3), Harris N(4), Rutherford S(4).

BACKGROUND: Papua New Guinea (PNG) is one of the 14 countries categorised as having a triple burden of tuberculosis (TB), multidrug-resistant TB (MDR TB), and TB-human immunodeficiency virus (HIV) co-infections. TB infection prevention and control (TB-IPC) guidelines were introduced in 2011 by the National Health Department of PNG. This study assesses the implementation of this policy in a sample of district hospitals in two regions of PNG.

METHODS: The implementation of TB-IPC policy was assessed using a survey method based on the World Health Organization (WHO) IPC assessment framework (IPCAF) to implement the WHO's IPC core components. The study included facility assessment at ten district hospitals and validation observations of TB-IPC practices.
RESULTS: Overall, implementation of IPC and TB-IPC guidelines was inadequate in participating facilities. Though 80% of facilities had an IPC program, many needed more clearly defined IPC objectives, budget allocation, and yearly work plans. In addition, they did not include senior facility managers in the IPC committee. 80% (n = 8 of 10) of hospitals had no IPC training and education; 90% had no IPC committee to support the IPC team; 70% had no surveillance protocols to monitor infections, and only 20% used multimodal strategies for IPC activities. Similarly, 70% of facilities had a TB-IPC program without a proper budget and did not include facility managers in the TB-IPC team; 80% indicated that patient flow poses a risk of TB transmission; 70% had poor ventilation systems; 90% had inadequate isolation rooms; and though 80% have personal protective equipment available, frequent shortages were reported.
CONCLUSIONS: The WHO-recommended TB-IPC policy is not effectively implemented in most of the participating district hospitals. Improvements in implementing and disseminating TB-IPC guidelines, monitoring TB-IPC practices, and systematic healthcare worker training are essential to improve TB-IPC guidelines' operationalisation in health settings to reduce TB prevalence in PNG.

DOI: 10.1186/s13756-023-01237-9
PMCID: PMC10092912
PMID: 37046339 [Indexed for MEDLINE]

Conflict of interest statement: The authors declare that they have no competing interests.

21. **Evaluation of Xpert (®) MTB/XDR test for susceptibility testing of Mycobacterium tuberculosis to first and second-line drugs in Uganda.**

medRxiv. 2023 Apr 5:2023.04.03.23288099. doi: 10.1101/2023.04.03.23288099. Preprint.

Katamba A, Ssengooba W, Sserubiri J, Semugenze D, William KG, Abdunoor N, Byaruhanga R, Turyahabwe S, Joloba ML.

BACKGROUND: Drug-Resistant Tuberculosis (DR-TB) is one of the key challenges toward TB control. There is an urgent need for rapid and accurate drug susceptibility tests (DST) for the most commonly used 1 st and 2 nd line TB

drugs.
DESIGN AND METHODS: In a blinded, laboratory-based cross-sectional study, we set out to validate the performance of the Xpert ® MTB/XDR test for DST of M. tuberculosis . Sputum samples or culture isolates collected between January 2020 and December 2021 from patients with rifampicin resistance -TB and/or with higher suspicion index for isoniazid (INH) resistance and/or 2 nd line fluoroquinolones (FQ) and injectable agents (IAs) were tested using the Xpert ® MTB/XDR test from 11/September 2021 to 26/May /2022. Diagnostic accuracy and factors for laboratory uptake of Xpert ® MTB/XDR test were compared to MGIT960 and the Hain Genotype® MTBDR plus and MDRsl assays (LPA) as reference DST methods.
RESULTS: A total of 100 stored sputum samples were included in this study. Of the samples tested using MGIT960, 65/99 (65.6%) were resistant to INH, 5/100 (5.0%) resistant to FQ and none were resistant to IAs. The sensitivity and specificity, n (%; 95%Confidence Interval, CI) of Xpert ® MTB/XDR test for; INH were 58 (89.2; 79.1-95.5) and 30 (88.2; 72.5-96.6), FQ; 4 (80.0; 28.3-99.4) and 95 (100; 96.2-100), respectively. The specificity for AIs was 100 (100; 96.3-100). Using LPA as a reference standard, a total of 52/98 (53.1%) were resistant to INH, 3/100 (3.0%) to FQ, and none to IA. The sensitivity and specificity, n (%; 95%CI) of Xpert ® MTB/XDR test compared to LPA for; INH was 50 (96.1; 86.7-99.5) and 34 (74.0; 58.8-85.7) and FQ 3 (100; 29.2-100) and 96 (99.0; 94.3-99.9) respectively. The specificity of IAs was 96 (100; 96.2-100). The factors for laboratory uptake and roll-out included; no training needed for technicians with previous Xpert-ultra experience and one day for those without, recording and reporting needs were not different from those of Xpert ultra, the error rate was 4/100 (4%), no uninterpretable results reported, test turn-around-time was 1hr/45 minutes and workflow similar to that of the Xpert-ultra test.
CONCLUSION: There is high sensitivity and specificity of Xpert ® MTB/XDR test for isoniazid, fluoroquinolones, and Injectable agents. There are acceptable Xpert ® MTB/XDR test attributes for test uptake and roll-out.

DOI: 10.1101/2023.04.03.23288099
PMCID: PMC10104194
PMID: 37066316

Conflict of interest statement: Conflict of interest: The authors have declared that no competing interests exist

22. **Incidence and predictors of acquired resistance to second-line antituberculosis drugs during the course of multi-drug resistant tuberculosis treatment: protocol for a systematic review and meta-analysis.**

BMJ Open. 2023 Apr 5;13(4):e070143. doi: 10.1136/bmjopen-2022-070143.

Alemu A(1)(2), Bitew ZW(3), Diriba G(4), Gashu E(5), Seid G(4)(2), Eshetu K(6), Kebede A(7), Gumi B(2).

INTRODUCTION: To date, acquired resistance to second-line antituberculosis drugs (SLDs) during multi-drug resistant tuberculosis (MDR-TB) treatment is becoming a public health concern. Different studies have assessed the incidence of acquired resistance to SLDs. However, the findings are inconsistent and there is limited global evidence. Thus, we are going to assess the incidence and predictors of acquired resistance to SLDs during MDR-TB treatment.
METHODS AND ANALYSIS: We designed this protocol following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses checklist. Electronic databases and grey literature sources will be searched systematically for articles published up to 25 March 2023. Studies reporting the incidence and predictors of acquired resistance to SLDs in MDR-TB patients will be explored. The studies will be managed using Endnote X8 citation manager and a stepwise approach will be followed to select studies. Data will be summarised using Microsoft Excel 2016 spreadsheet. A Newcastle-Ottawa Scale quality assessment and cochrane risk-of-bias tools will be used to assess the study's quality. The authors will independently search databases, select studies, assess the study's quality and extract data. Data will be analysed using STATA V.17 software. We will estimate the pooled incidence of acquired resistance with 95% CI. In addition, the pooled effect measures (OR, HR, risk ratio) with their 95% CI will be estimated. Heterogeneity will be assessed using the I2 statistics.
Publication bias will be assessed using funnel plot and Egger's test. A subgroup analysis will be conducted for the primary outcome (acquired resistance) per each study characteristics such as WHO regional category, country's TB/MDR-TB burden, data collection period and per the specific second-line anti-TB drug.
ETHICS AND DISSEMINATION: Since this study will be based on data extraction from published studies, ethical approval is not mandatory. The study will be published in peer-reviewed scientific journals and the findings will be presented at different scientific conferences.
PROSPERO REGISTRATION NUMBER: CRD42022371014.

DOI: 10.1136/bmjopen-2022-070143
PMCID: PMC10083796
PMID: 37019479 [Indexed for MEDLINE]

Conflict of interest statement: Competing interests: None declared.

23. **Protocol for a feasibility randomized controlled trial to evaluate the efficacy, safety and tolerability of N-acetylcysteine in reducing adverse drug reactions among adults treated for multidrug-resistant tuberculosis in Tanzania.**

Pilot Feasibility Stud. 2023 Apr 1;9(1):55. doi: 10.1186/s40814-023-01281-7.

Mpagama SG(1)(2), Mvungi HC(3)(4), Mbelele PM(3)(5), Semvua HH(4), Liyoyo AA(3), de Guex KP(6), Sloan D(7), Kibiki GS(8), Boeree M(9), Phillips PPJ(10), Heysell SK(6).

BACKGROUND: Adverse drug reactions (ADRs) frequently occur in patients using second-line anti-tuberculosis medicine for treatment of multidrug resistant tuberculosis (MDR-TB). ADRs contribute to treatment interruptions which can compromise treatment response and risk acquired drug resistance to critical newer drugs such as bedaquiline, while severe ADRs carry considerable morbidity and mortality. N-acetylcysteine (NAC) has shown promise in reducing ADRs for medications related to TB in case series or randomized controlled trials in other medical conditions, yet evidence is lacking in MDR-TB patients. TB endemic settings have limited capacity to conduct clinical trials. We designed a proof-of-concept clinical trial primarily to explore the preliminary evidence on the protective effect of NAC among people treated for MDR-TB with second-line anti-TB medications.
METHODS: This is a proof-of-concept randomized open label clinical trial with 3 treatment arms including a control arm, an interventional arm of NAC 900 mg daily, and an interventional arm of NAC 900 mg twice-daily administered during the intensive phase of MDR-TB treatment. Patients initiating MDR-TB treatment will be enrolled at Kibong'oto National Center of Excellence for MDR-TB in the Kilimanjaro region of Tanzania. The minimum anticipated sample size is 66; with 22 participants in each arm. ADR monitoring will be performed at baseline and daily follow-up over 24 weeks including blood and urine specimen collection for hepatic and renal function and electrolyte abnormalities, and electrocardiogram. Sputum will be collected at baseline and monthly thereafter and cultured for mycobacteria as well as assayed for other molecular targets of Mycobacterium tuberculosis. Adverse drug events will be analysed over time using mixed effect models. Mean differences between arms in change of the ADRs from baseline (with 95% confidence intervals) will be derived from the fitted model.
DISCUSSION: Given that NAC promotes synthesis of glutathione, an intracellular antioxidant that combats the impact of oxidative stress, it may protect against medication induced oxidative damage in organs such as liver, pancreas, kidney, and cells of the immune system. This randomized controlled trial will determine if NAC leads to fewer ADRs, and if this protection is dose dependent. Fewer ADRs among patients treated with MDR-TB may significantly improve treatment outcomes for multidrug regimens that necessitate prolonged treatment durations. Conduct

of this trial will set the needed infrastructure for clinical trials.
TRIAL REGISTRATION: PACTR202007736854169 Registered 03 July 2020.

DOI: 10.1186/s40814-023-01281-7
PMCID: PMC10066962
PMID: 37005695

Conflict of interest statement: The authors declare that they have no competing interests.

24. **Assessing the knowledge, attitude and practice (KAP) measures against tuberculosis in patients in the ambulatory department facilities in Pakistan: a cross-sectional analysis.**

Monaldi Arch Chest Dis. 2023 Apr 12. doi: 10.4081/monaldi.2023.2500. Online ahead of print.

Ahmad S(1), Khawaja UA(2), Haider SM(3), Mowlabaccus WB(4), Mohan A(5), Ansari A(6), Ahmad M(7), Garg T(8), Ahmed H(9), Ahmad S(10), Essar MY(11), Perez-Fernandez J(12), Yatzkan GD(13).

Tuberculosis (TB), at present, is the leading infectious aetiology of death globally. In Pakistan, there are approximately 510,000 new cases annually, with more than 15,000 of them developing into drug resistant TB, making the nation the fifth leading country in TB prevalence in the world. Due to the ongoing COVID-19 pandemic, focus has drifted away from TB screening, diagnostic, health awareness campaigns and therapeutic measures endangering KAP (knowledge, attitude and practices) towards TB in our population. We conducted a cross-sectional descriptive study in Pakistan to assess the knowledge, attitude and practices of Pakistani residents attending the adult outpatient departments of public hospitals for any health-related concern. Our sample size was of 856 participants, with a median age of 22 years. Occupation-wise, those who were employed had better knowledge of TB than those who were unemployed [odds ratio (OR): 1.011; 95% CI :1.005-1.8005]. No differences were observed in TB knowledge between those adherents to common preventive practices versus not adherent (OR 0.875, 95% CI: 0.757-1.403). More than 90 % of participants agreed that TB is dangerous for the community and a majority opted against stigmatising TB patients (79.1%). People who could read and write were 3.5 times more likely to have a good attitude towards TB compared to those who could not (OR: 3.596;95% CI: 1.821-70.230; p=0.037). Similarly, employed subjects had better attitude compared to unemployed ones (p=0.024), (OR: 1.125; 95% CI: 0.498, 1.852) and

those having better knowledge of TB had a better attitude grade (OR:1.749; 95% CI: 0.832-12.350), p=0.020). Age, occupation, and educational status were statistically significant among the two groups (p=0.038, p=0.023, p=0.000) respectively. Literate subjects had thrice good practice towards TB than illiterate (OR: 3.081; 95% CI: 1.869-4.164; p=0.000). Future education and awareness programs should target specific groups such as the unemployed and illiterate with practice-focused approaches. Our study outcomes can enable the concerned officials and authorities taking appropriate evidence-based steps to direct the efforts in an efficient manner to curtail the burden of TB in Pakistan and to limit its progression that could potentially lead our nation to become an MDR-TB endemic territory.

DOI: 10.4081/monaldi.2023.2500
PMID: 37052048

25. **Global prevalence of hepatitis B or hepatitis C infection among patients with tuberculosis disease: systematic review and meta-analysis.**

EClinicalMedicine. 2023 Apr 6;58:101938. doi: 10.1016/j.eclinm.2023.101938. eCollection 2023 Apr.

Olaru ID(1)(2)(3), Beliz Meier M(1), Mirzayev F(4), Prodanovic N(1), Kitchen PJ(1), Schumacher SG(4), Denkinger CM(1)(5).

BACKGROUND: There is a substantial overlap in the epidemiology of chronic hepatitis B (HBV), hepatitis C (HCV) and tuberculosis (TB) due to overlapping risk factors. Testing for viral hepatitis is not widely recommended for patients with TB. The aim of this systematic review was to evaluate the global prevalence of chronic viral hepatitis infection among patients with TB.
METHODS: MEDLINE, EMBASE, Web of Science, Cochrane Library, African Journals Online, LILACS, and country TB reports were searched for studies published between January 1st, 2011 and June 17th 2021. Random-effects meta-analyses for proportions were conducted to obtain pooled prevalences. The prevalence of chronic HBV/HCV infection among patients with TB was also compared to that in the general population. The protocol was registered on PROSPERO (CRD42021276468).
FINDINGS: This analysis included 127 studies (83 for both HBV and HCV, 28 for HBV only, and 25 for HCV only) and data from 94,936 patients. The global pooled seroprevalence was 5.8% (95% CI 5.0-6.8) for HBs-antigen and 10.3% (95% CI 8.4-12.3) for HCV-antibodies. Pooled prevalence was highest in the WHO African Region for HBV at 7.8% (95% CI 5.2-10.9) and in the WHO European Region at 17.5% (95% CI 12.2-23.5) for HCV. In studies among TB patients who inject drugs, HCV prevalence was 92.5% (95% CI 80.8-99.0). Pooled HCV-antibody seroprevalence

among patients with TB was higher than in the general population in all six WHO regions while HBs-antigen seroprevalence was higher in 3/6 regions.
INTERPRETATION: This review highlights the syndemicity of chronic viral hepatitis and TB and suggests that routine testing for hepatitis upon TB diagnosis may be justified. The prevalence of chronic HBV and HCV infections was higher among patients with TB than in the general population.
FUNDING: This study was study was funded by the Global Tuberculosis Programme, World Health Organization.

DOI: 10.1016/j.eclinm.2023.101938
PMCID: PMC10113747
PMID: 37090436

Conflict of interest statement: CMD serves as advisor to the World Health Organization and PLOS Med academic editor. She received grants from a number of donors for work unrelated to the subject of this paper. IDO was funded by the Wellcome Trust for work unrelated to this manuscript. The other authors have no competing interests.

26. **Radiomics analysis of lung CT for multidrug resistance prediction in active tuberculosis: a multicentre study.**

Eur Radiol. 2023 Apr 1:1-10. doi: 10.1007/s00330-023-09589-x. Online ahead of print.

Li Y(1), Xu Z(1), Lv X(1), Li C(1), He W(1), Lv Y(1), Hou D(2).

OBJECTIVES: Multidrug-resistant TB (MDR-TB) is a severe burden and public health threat worldwide. This study aimed to develop a radiomics model based on the tree-in-bud (TIB) sign and nodules and validate its predictive performance for MDR-TB.
METHODS: We retrospectively recruited 454 patients with proven active TB from two hospitals and classified them into three training and testing cohorts: TIB (n = 295, 102), nodules (n = 302, 97), and their combination (n = 261, 81). Radiomics features relating to TIB and nodules were separately extracted. The maximal information coefficient and recursive feature elimination were used to select informative features per the two signs. Two radiomics models were constructed to predict MDR-TB using a random forest classifier. Then, a combined model was built incorporating radiomics features based on these two signs. The capability of the models in the combined training and testing cohorts was validated with ROC curves.

RESULTS: Sixteen features were extracted from TIB and 15 from nodules. The AUCs of the combined model were slightly higher than those of the TIB model in the combined training cohort (0.911 versus 0.877, $p > 0.05$) and testing cohort (0.820 versus 0.786, $p < 0.05$) and similar to the performance of the nodules model in the combined training cohort (0.911 versus 0.933, $p > 0.05$) and testing cohort (0.820 versus 0.855, $p > 0.05$).
CONCLUSIONS: The CT-based radiomics models hold promise for use as a non-invasive tool in the prediction of MDR-TB.
CLINICAL RELEVANCE STATEMENT: Our study revealed that complementary information regarding MDR-TB can be provided by radiomics based on the TIB sign and nodules. The proposed radiomics models may be new markers to predict MDR in active TB patients.
KEY POINTS: • This is the first study to build, validate, and apply radiomics based on tree-in-bud sign and nodules for the prediction of MDR-TB. • The radiomics model showed a favorable performance for the identification of MDR-TB. • The combined model holds potential to be used as a diagnostic tool in routine clinical practice.

DOI: 10.1007/s00330-023-09589-x
PMCID: PMC10067016
PMID: 37004571

Conflict of interest statement: The authors of this manuscript declare no relationships with any companies, whose products or services may be related to the subject matter of the article.

27. **Heteroresistance to rifampicin & isoniazid in clinical samples of patients with presumptive drug-resistant tuberculosis in Central India.**

Indian J Med Res. 2023 Apr 27. doi: 10.4103/ijmr.ijmr_607_22. Online ahead of print.

Desikan P(1), Panwalkar N(2), Punde RP(2), Khan Z(2), Pauranik A(2), Mirza SB(2), Chourey M(2), Anand S(3), Sachdeva KS(4).

BACKGROUND & OBJECTIVES: A combination of resistant and susceptible Mycobacterium tuberculosis (MTB) isolated from clinical specimens is referred to as heteroresistance. Heteroresistance leads to difficulties in drug resistance testing and may adversely affect treatment outcomes. The present study estimated the proportion of heteroresistance among MTB in clinical samples of presumptive drug-resistant tuberculosis (TB) patients in Central India.

METHODS: A retrospective analysis of data generated from line probe assay (LPA) at a tertiary care hospital in Central India between January 2013 and December 2018 was carried out. A heteroresistant MTB in a sample was indicated by the presence of both wild-type and mutant-type patterns on an LPA strip.
RESULTS: Data analysis was carried out on interpretable 11,788 LPA results. Heteroresistance in MTB was detected in 637 (5.4%) samples. Of these, heteroresistance in MTB was detected in 413 (64.8%), 163 (25.5%) and 61 (9.5%) samples with respect to rpoB, katG and inhA genes, respectively.
INTERPRETATION & CONCLUSIONS: Heteroresistance is considered a preliminary step in the development of drug resistance. Delayed or suboptimal anti-tubercular therapy in patients with heteroresistance of MTB may elicit full clinical resistance and negatively impact the National TB Elimination Programme. Further studies are, however, needed to determine the impact of heteroresistance on treatment outcomes in individual patients.

DOI: 10.4103/ijmr.ijmr_607_22
PMID: 37102517

Conflict of interest statement: None

28. **An integrated computational approach towards novel drugs discovery against polyketide synthase 13 thioesterase domain of Mycobacterium tuberculosis.**

Sci Rep. 2023 Apr 28;13(1):7014. doi: 10.1038/s41598-023-34222-8.

Altharawi A(1), Alossaimi MA(1), Alanazi MM(2), Alqahatani SM(1), Tahir Ul Qamar M(3).

The acquired drug resistance by Mycobacterium tuberculosis (M. tuberculosis) to antibiotics urges the need for developing novel anti-M. tuberculosis drugs that possess novel mechanism of action. Since traditional drug discovery is a labor-intensive and costly process, computer aided drug design is highly appreciated tool as it speeds up and lower the cost of drug development process. Herein, Asinex antibacterial compounds were virtually screened against thioesterase domain of Polyketide synthase 13, a unique enzyme that forms α-alkyl β-ketoesters as a direct precursor of mycolic acids which are essential components of the lipid-rich cell wall of M. tuberculosis. The study identified three drug-like compounds as the most promising leads; BBB_26582140, BBD_30878599 and BBC_29956160 with binding energy value of - 11.25 kcal/mol, -9.87 kcal/mol and - 9.33 kcal/mol, respectively. The control molecule binding energy score is -9.25 kcal/mol. Also, the docked complexes were dynamically stable with maximum root mean square deviation (RMSD) value of 3 Å. Similarly, the MM-GB\PBSA method revealed highly stable complexes with mean energy

values < - 75 kcal/mol for all three systems. The net binding energy scores are validated by WaterSwap and entropy energy analysis. Furthermore, The in silico druglike and pharmacokinetic investigation revealed that the compounds could be suitable candidates for additional experimentations. In summary, the study findings are significant, and the compounds may be used in experimental validation pipeline to develop potential drugs against drug-resistant tuberculosis.

DOI: 10.1038/s41598-023-34222-8
PMID: 37117557 [Indexed for MEDLINE]

29. **A Novel Tool to Identify Bactericidal Compounds against Vulnerable Targets in Drug-Tolerant M. tuberculosis found in Caseum.**

mBio. 2023 Apr 25;14(2):e0059823. doi: 10.1128/mbio.00598-23. Epub 2023 Apr 5.

Sarathy JP(1), Xie M(1), Jones RM(2), Chang A(3), Osiecki P(1), Weiner D(4)(5), Tsao WS(1), Dougher M(1), Blanc L(6), Fotouhi N(7), Via LE(4)(5), Barry CE 3rd(4)(8), De Vlaminck I(3), Sherman DR(2), Dartois VA(1)(9).

Caseous necrosis is a hallmark of tuberculosis (TB) pathology and creates a niche for drug-tolerant persisters within the host. Cavitary TB and high bacterial burden in caseum require longer treatment duration. An in vitro model that recapitulates the major features of Mycobacterium tuberculosis (Mtb) in caseum would accelerate the identification of compounds with treatment-shortening potential. We have developed a caseum surrogate model consisting of lysed and denatured foamy macrophages. Upon inoculation of Mtb from replicating cultures, the pathogen adapts to the lipid-rich matrix and gradually adopts a nonreplicating state. We determined that the lipid composition of ex vivo caseum and the surrogate matrix are similar. We also observed that Mtb in caseum surrogate accumulates intracellular lipophilic inclusions (ILI), a distinctive characteristic of quiescent and drug-tolerant Mtb. Expression profiling of a representative gene subset revealed common signatures between the models. Comparison of Mtb drug susceptibility in caseum and caseum surrogate revealed that both populations are similarly tolerant to a panel of TB drugs. By screening drug candidates in the surrogate model, we determined that the bedaquiline analogs TBAJ876 and TBAJ587, currently in clinical development, exhibit superior bactericidal against caseum-resident Mtb, both alone and as substitutions for bedaquiline in the bedaquiline-pretomanid-linezolid regimen approved for the treatment of multidrug-resistant TB. In summary, we have developed a physiologically relevant

nonreplicating persistence model that reflects the distinct metabolic and drug-tolerant state of Mtb in caseum. IMPORTANCE M. tuberculosis (Mtb) within the caseous core of necrotic granulomas and cavities is extremely drug tolerant and presents a significant hurdle to treatment success and relapse prevention. Many in vitro models of nonreplicating persistence have been developed to characterize the physiologic and metabolic adaptations of Mtb and identify compounds active against this treatment-recalcitrant population. However, there is little consensus on their relevance to in vivo infection. Using lipid-laden macrophage lysates, we have designed and validated a surrogate matrix that closely mimics caseum and in which Mtb develops a phenotype similar to that of nonreplicating bacilli in vivo. The assay is well suited to screen for bactericidal compounds against caseum-resident Mtb in a medium-throughput format, allowing for reduced reliance on resource intensive animal models that present large necrotic lesions and cavities. Importantly, this approach will aid the identification of vulnerable targets in caseum Mtb and can accelerate the development of novel TB drugs with treatment-shortening potential.

DOI: 10.1128/mbio.00598-23
PMCID: PMC10127596
PMID: 37017524 [Indexed for MEDLINE]

Conflict of interest statement: The authors declare no conflict of interest.

30. **Evaluation of Microsphere-based xMAP Test for gyrA Mutation Identification in Mycobacterium Tuberculosis.**

Biomed Environ Sci. 2023 Apr 20;36(4):384-387. doi: 10.3967/bes2023.046.

Ou XC(1), Zhao B(1), Song ZX(1), Pei SJ(2), Wang SF(1), He WC(1), Liu CF(1), Liu DX(1), Xing RD(1), Xia H(1), Zhao YL(1).

DOI: 10.3967/bes2023.046
PMID: 37105915 [Indexed for MEDLINE]

31. **Next-Generation Diarylquinolines Improve Sterilizing Activity of Regimens with Pretomanid and the Novel Oxazolidinone TBI-223 in a Mouse Tuberculosis Model.**

Antimicrob Agents Chemother. 2023 Apr 18;67(4):e0003523. doi: 10.1128/aac.00035-23. Epub 2023 Mar 15.

Li SY(1), Converse PJ(1), Betoudji F(1), Lee J(1), Mdluli K(2), Upton A(2)(3), Fotouhi N(2), Nuermberger EL(1).

A regimen comprised of bedaquiline (BDQ, or B), pretomanid, and linezolid (BPaL) is the first oral 6-month regimen approved by the U.S. Food and Drug Administration and recommended by the World Health Organization for the treatment of extensively drug-resistant tuberculosis. We used a well-established BALB/c mouse model of tuberculosis to evaluate the treatment-shortening potential of replacing bedaquiline with either of two new, more potent diarylquinolines, TBAJ-587 and TBAJ-876, in early clinical trials. We also evaluated the effect of replacing linezolid with a new oxazolidinone, TBI-223, exhibiting a larger safety margin with respect to mitochondrial toxicity in preclinical studies. Replacing bedaquiline with TBAJ-587 at the same 25-mg/kg dose significantly reduced the proportion of mice relapsing after 2 months of treatment, while replacing linezolid with TBI-223 at the same 100-mg/kg dose did not significantly change the proportion of mice relapsing. Replacing linezolid or TBI-223 with sutezolid in combination with TBAJ-587 and pretomanid significantly reduced the proportion of mice relapsing. In combination with pretomanid and TBI-223, TBAJ-876 at 6.25 mg/kg was equipotent to TBAJ-587 at 25 mg/kg. We conclude that replacement of bedaquiline with these more efficacious and potentially safer diarylquinolines and replacement of linezolid with potentially safer and at least as efficacious oxazolidinones in the clinically successful BPaL regimen may lead to superior regimens capable of treating both drug-susceptible and drug-resistant TB more effectively and safely.

DOI: 10.1128/aac.00035-23
PMCID: PMC10112056
PMID: 36920217 [Indexed for MEDLINE]

Conflict of interest statement: The authors declare a conflict of interest. Nader Fotouhi is a current employee of the TB Alliance. The remaining authors declare no conflict of interest.

33. **Diagnostic performance of the GenoType MTBDRplus VER 2.0 line probe assay for the detection of isoniazid resistant Mycobacterium tuberculosis in Ethiopia.**

PLoS One. 2023 Apr 26;18(4):e0284737. doi: 10.1371/journal.pone.0284737. eCollection 2023.

Moga S(1)(2), Bobosha K(3), Fikadu D(1), Zerihun B(1), Diriba G(1), Amare M(1), Kempker RR(4), Blumberg HM(4)(5), Abebe T(2).

BACKGROUND: Isoniazid (INH) resistant Mycobacterium tuberculosis (Hr-TB) is the most common type of drug resistant TB, and is defined as M tuberculosis complex

(MTBC) strains resistant to INH but susceptible to rifampicin (RIF). Resistance to INH precedes RIF resistance in almost all multidrug resistant TB (MDR-TB) cases, across all MTBC lineages and in all settings. Therefore, early detection of Hr-TB is critical to ensure rapid initiation of appropriate treatment, and to prevent progression to MDR-TB. We assessed the performance of the GenoType MTBDRplus VER 2.0 line probe assay (LPA) in detecting isoniazid resistance among MTBC clinical isolates.
METHODS: A retrospective study was conducted among M. tuberculosis complex (MTBC) clinical isolates obtained from the third-round Ethiopian national drug resistance survey (DRS) conducted between August 2017 and December 2019. The sensitivity, specificity, positive predictive value, and negative predictive value of the GenoType MTBDRplus VER 2.0 LPA in detecting INH resistance were assessed and compared to phenotypic drug susceptibility testing (DST) using the Mycobacteria Growth Indicator Tube (MGIT) system. Fisher's exact test was performed to compare the performance of LPA between Hr-TB and MDR-TB isolates.
RESULTS: A total of 137 MTBC isolates were included, of those 62 were Hr-TB, 35 were MDR-TB and 40 were INH susceptible. The sensitivity of the GenoType MTBDRplus VER 2.0 for detecting INH resistance was 77.4% (95% CI: 65.5-86.2) among Hr-TB isolates and 94.3% (95% CI: 80.4-99.4) among MDR-TB isolates (P = 0.04). The specificity of the GenoType MTBDRplus VER 2.0 for detecting INH resistance was 100% (95% CI: 89.6-100). The katG 315 mutation was observed in 71% (n = 44) of Hr-TB phenotypes and 94.3% (n = 33) of MDR-TB phenotypes. Mutation at position-15 of the inhA promoter region alone was detected in four (6.5%) Hr-TB isolates, and concomitantly with katG 315 mutation in one (2.9%) MDR-TB isolate.
CONCLUSIONS: GenoType MTBDRplus VER 2.0 LPA demonstrated improved performance in detecting INH resistance among MDR-TB cases compared to Hr-TB cases. The katG315 mutation is the most common INH resistance conferring gene among Hr-TB and MDR-TB isolates. Additional INH resistance conferring mutations should be evaluated to improve the sensitivity of the GenoType MTBDRplus VER 2.0 for the detection of INH resistance among Hr-TB cases.

DOI: 10.1371/journal.pone.0284737
PMCID: PMC10132600
PMID: 37099514 [Indexed for MEDLINE]

Conflict of interest statement: The authors have declared that no competing interests exist.

34. **MarR-Dependent Transcriptional Regulation of mmpSL5 Induces Ethionamide Resistance in Mycobacterium abscessus.**

Antimicrob Agents Chemother. 2023 Apr 18;67(4):e0135022. doi: 10.1128/aac.01350-22. Epub 2023 Mar 29.

Rodriguez R(1), Campbell-Kruger N(2), Gonzalez Camba J(2), Berude J(2), Fetterman R(2), Stanley S(2).

Mycobacterium abscessus (Mabs) is an emerging nontuberculosis mycobacterial (NTM) pathogen responsible for a wide variety of respiratory and cutaneous infections that are difficult to treat with standard antibacterial therapy. Mabs has a high degree of both innate and acquired antibiotic resistance to most clinically relevant drugs, including standard anti-mycobacterial agents. Ethionamide (ETH), an inhibitor of mycolic acid biosynthesis, is currently utilized as a second-line agent for treating multidrug-resistant tuberculosis infections. Here, we show that ETH displays activity against clinical strains of Mabs in vitro at concentrations that are >100× lower than other mycolic acid targeting drugs. Using transposon mutagenesis followed by transposon sequencing (Tn-Seq) and whole-genome sequencing of spontaneous ETH-resistant mutants, we identified MAB_2648c as a genetic determinant of ETH sensitivity in Mabs. MAB_2648c encodes a MarR family transcriptional regulator of the TetR class of regulators. We show that MAB_2648c represses expression of MAB_2649 (mmpS5) and MAB_2650 (mmpL5). Further, we show that derepression of these genes in MAB_2648c mutants confers resistance to ETH, but not other antibiotics. To identify determinants of resistance that may be shared across antibiotics with distinct mechanisms of action, we also performed Tn-Seq during treatment with amikacin and clarithromycin, drugs currently used clinically to treat Mabs. We found very little overlap in genes that modulate the sensitivity of Mabs to all three antibiotics, suggesting a high degree of specificity for resistance mechanisms in this emerging pathogen.

DOI: 10.1128/aac.01350-22
PMCID: PMC10112066
PMID: 36988462 [Indexed for MEDLINE]

Conflict of interest statement: The authors declare no conflict of interest.

35. **[Investigation of Efflux Pump Genes in Resistant Mycobacterium tuberculosis Complex Clinical Isolates Exposed to First Line Antituberculosis Drugs and Verapamil Combination].**

[Article in Turkish]

Mikrobiyol Bul. 2023 Apr;57(2):207-219. doi: 10.5578/mb.20239916.

Özgür D(1), Ersoy L(2), Ülger M(3), Tezcan Ülger S(2), Aslan G(2).

Tuberculosis (TB) is caused by Mycobacterium tuberculosis, still one of the most common life-threatening infectious diseases worldwide. Although drug resistance in M.tuberculosis is mainly due to spontaneous chromosomal mutations in genes encoding drug target or drug activating enzymes, the resistance cannot be explained only by these mutations. Low permeability of the cell wall, drug inactivating enzymes and especially efflux pumps (EPs) are other mechanisms of drug resistance in mycobacteria. Efflux pump inhibitors (EPIs) binding to M.tuberculosis EPs were shown to inhibit efflux of anti-TB drugs, to enhance M.tuberculosis killing, to reduce drug resistance and to produce synergistic effects with first line anti-TB drugs. In this study, we aimed to determine the minimum inhibitory concentration (MIC) of first-line anti-TB drugs in the presence of verapamil (VER) and the expression of 21 putative EP genes belonged to the ATP-binding cassette (ABC), major facilitator superfamily (MFS) and resistance-nodulation-division (RND) families which might have caused the resistance in nine M.tuberculosis complex clinical isolates resistant to all of the first line anti-TB drugs. MIC values of the isolates were determined in 96-well U-bottom plates by the resazurin microtiter test (REMA) method based on the color change principle. According to the determined MIC values of each isolate, freshly grown cultures in Middlebrook 7H9 broth were exposed to first-line anti-TB drugs and MIC of first-line anti-TB drugs in the presence of VER (½ MIC) at 37°C for 48 hours for RNA extraction. The non-drug exposed cultures were used as control. Total RNA was extracted using the RNeasy Mini Kit (Qiagen GmbH, Hilden, Germany) and then treated with DNase I (Thermo Fischer Scientific Inc., Waltham, MA). Complementary DNA (cDNA) from the extracted RNAs was synthesized with the "First strand cDNA synthesis kit" (Thermo Fischer Scientific Inc., Waltham, MA) using oligo primers. The expression levels of efflux pump genes by quantitative realtime polymerase chain reaction (qRt-PCR) were performed using the QuantiTect SYBR Green Rt-PCR Kit (Qiagen, Germany). The housekeeping sigma factor gene sigA (Rv2703) was used as internal control in qRt-PCR assays. Relative quantification of the clinical isolates was determined by the 2-ΔΔCt method by comparing the expression levels of efflux genes in cultures exposed to primary anti-TB drugs and VER with those of non-drug exposed cultures. MIC values of nine isolates by REMA method was determined between 32-512 µg/mL, 1-128 µg/mL, 2-32 µg/mL, 4-16 µg/mL and 15.62-250 µg/mL for streptomycin (SM), isoniazid (INH), rifampicin (RIF), ethambutol (EMB) and VER, respectively. In the presence of ½ MIC VER, it was determined that the MIC of SM decreased 2-32 fold in eight isolates, the MIC of INH decreased by 2-8 fold in nine isolates, the MIC of RIF decreased by 2-16 fold in eight isolates, and the

MIC of EMB decreased 2-4 fold in only five isolates. There was an increase in the expression of Rv1273c, Rv1456c, Rv1457 and Rv1819 efflux pump genes from the ABC family, Rv1634 and Rv0842 from the MFS family and Rv3823 efflux from the RND family in isolates exposed to ½ MIC of first-line anti-TB drugs stress. Rv1456c and Rv1819 were found to be associated with SM resistance, Rv1273c with EMB resistance, Rv1457, Rv0842 and Rv3823 with both RIF and EMB resistance, and Rv1634 with INH, RIF and EMB resistance. It was determined that there was a decrease in the expression levels of eight efflux pump genes from the ABC family (Rv1456c, Rv1457c, Rv1458c, Rv0194, Rv1272c, Rv1686c, Rv1687c, Rv1819c), six from MFS family (Rv0842, Rv0849, Rv1634, Rv2265, Rv2456c, Rv0876c) and two from RND family (Rv0507, Rv0676c) in isolates exposed to MIC of first-line anti-TB drugs in the presence of VER (½ MIC). Further studies with clinical isolates are needed to investigate the EPIs that can be used in alternative therapy and to determine the contribution of EPs to the development of resistance due to the increasing TB resistance.

DOI: 10.5578/mb.20239916
PMID: 37067206 [Indexed for MEDLINE]

36. **Optimal management of drug-resistant tuberculosis: Can India lead the way?**

Indian J Med Res. 2023 Apr 27. doi: 10.4103/ijmr.ijmr_300_23. Online ahead of print.

Masini T(1), Furin J(2), Udwadia Z(3), Guglielmetti L(4).

DOI: 10.4103/ijmr.ijmr_300_23
PMID: 37102512

Conflict of interest statement: None

37. **Liquid chromatography-tandem mass spectrometry analysis of delamanid and its metabolite in human cerebrospinal fluid using protein precipitation and on-line solid-phase extraction.**

J Pharm Biomed Anal. 2023 Apr 1;227:115281. doi: 10.1016/j.jpba.2023.115281. Epub 2023 Feb 3.

Mazanhanga MT(1), Joubert A(1), Castel SA(1), van der Merwe M(1), Maartens G(1), Dooley KE(2), Upton CM(3), Wiesner L(4).

The penetration of the antituberculosis drug delamanid into the central nervous

system is not established. The distribution of delamanid and its major metabolite, DM-6705, into the cerebrospinal fluid requires investigation. A liquid chromatography-tandem mass spectrometry method for the quantification of delamanid and DM-6705 in human cerebrospinal fluid was developed and validated. The calibration range for both analytes was 0.300 - 30.0 ng/mL. The deuterium-labelled analogue of delamanid (delamanid-d4) and OPC-14714 were used as internal standards for delamanid and DM-6705, respectively. Samples were processed by protein precipitation followed by on-line solid-phase extraction and high-performance liquid chromatography on an Agilent 1260 HPLC system. A Phenomenex Gemini-NX C18 (5.0 µm, 50 mm × 2.0 mm) analytical column was used for on-line solid-phase extraction, and a Waters Xterra MS C18 (5.0 µm, 100 mm × 2.1 mm) analytical column for chromatographic separation using gradient elution, at a flow rate of 300 µL/min. The total run time was 7.5 min. Analytes were detected by multiple reaction monitoring on an AB Sciex 5500 triple quadrupole mass spectrometer at unit mass resolution, with electrospray ionization in the positive mode. Accuracy and precision were assessed over three independent validation batches. Extraction recoveries were more than 98% and were consistent across the analytical range. Both analytes in CSF exhibited non-specific adsorption to polypropylene tubes. The method was used to analyse cerebrospinal fluid samples from patients with pulmonary tuberculosis in an exploratory pharmacokinetic study.

DOI: 10.1016/j.jpba.2023.115281
PMCID: PMC10023415
PMID: 36739721 [Indexed for MEDLINE]

Conflict of interest statement: 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.

Other Articles

38. **Literature Highlights.**

Int J Tuberc Lung Dis. 2023 Apr 1;27(4):245-247. doi: 10.5588/ijtld.23.9904.

Tiberi S(1), Graham S(2), Trauer JM(3), Blackbourn HD(4).

Literature Highlights is a digest of notable papers recently published in the leading respiratory journals. Coverage includes a 6-month, all-oral regimen for

rifampicin-resistant TB; phase 3 trials of two shorter regimen for drug-resistant TB; heterogeneity in M. tuberculosis transmission through whole-genome sequencing; vaping and pulmonary inflammation; impact of COVID-19 during pregnancy on mother and newborn; migrants and TB.

DOI: 10.5588/ijtld.23.9904
PMID: 37035977 [Indexed for MEDLINE]

39. **What's new in childhood tuberculosis.**

Curr Opin Pediatr. 2023 Apr 1;35(2):166-175. doi: 10.1097/MOP.0000000000001226. Epub 2023 Feb 7.

Finlayson H(1), Lishman J(1), Palmer M(2).

PURPOSE OF REVIEW: The current review identifies recent advances in the prevention, diagnosis, and treatment of childhood tuberculosis (TB) with a focus on the WHO's updated TB management guidelines released in 2022.
RECENT FINDINGS: The COVID-19 pandemic negatively affected global TB control due to the diversion of healthcare resources and decreased patient care-seeking behaviour. Despite this, key advances in childhood TB management have continued. The WHO now recommends shorter rifamycin-based regimens for TB preventive treatment as well as shorter regimens for the treatment of both drug-susceptible and drug-resistant TB. The Xpert Ultra assay is now recommended as the initial diagnostic test for TB in children with presumed TB and can also be used on stool samples. Point-of-care urinary lipoarabinomannan assays are promising as 'rule-in' tests for children with presumed TB living with HIV. Treatment decision algorithms can be used to diagnose TB in symptomatic children in settings with and without access to chest X-rays; bacteriological confirmation should always be attempted.
SUMMARY: Recent guideline updates are a key milestone in the management of childhood TB, and the paediatric TB community should now prioritize their efficient implementation in high TB burden countries while generating evidence to close current evidence gaps.

DOI: 10.1097/MOP.0000000000001226
PMID: 36749063 [Indexed for MEDLINE]

40. **Tuberculosis and malnutrition: The European perspective.**

Clin Nutr. 2023 Apr;42(4):486-492. doi: 10.1016/j.clnu.2023.01.016. Epub 2023 Feb 10.

Ockenga J(1), Fuhse K(2), Chatterjee S(3), Malykh R(4), Rippin H(5), Pirlich M(6), Yedilbayev A(7), Wickramasinghe K(8), Barazzoni R(9).

Tuberculosis (TB) is a leading infectious cause of death worldwide, despite ongoing efforts to limit its incidence and mortality. Although the European Region has made gains in TB incidence and mortality, it now contends with increasing numbers of multidrug- and rifampicin-resistant tuberculosis (MDR/RR-TB). Malnutrition is a major contributor to the burden of TB and may also be directly caused or enhanced by the onset of TB. The presence of malnutrition may worsen TB and MDR/RR-TB related treatment outcomes and contribute to growing TB drug-resistance. Preventing and treating all forms of malnutrition is an important tool to limit the spread of TB worldwide and improve TB outcomes and treatment efficacy. We carried out a scoping review of the existing evidence that addresses malnutrition in the context of TB. Our review found malnutrition increased the risk of developing TB in high-burden settings and increased the likelihood of developing unfavorable treatment outcomes, including treatment failure, loss to follow-up, and death. The potential impact of nutritional care and improved nutritional status on patient prognosis was more difficult to evaluate due to heterogeneity of patient populations, treatment protocols, and treatment durations and goals. High-quality trials that consider malnutrition as a major risk factor and relevant treatment target when designing effective strategies to limit TB spread and mortality are needed to inform evidence-based practice. In TB patients, we suggest that widespread and regular nutritional screening, assessment, and counselling, has the potential to increase effectiveness of TB management strategies and improve patient quality of life, overall outcomes, and survival.

DOI: 10.1016/j.clnu.2023.01.016
PMID: 36857957 [Indexed for MEDLINE]

41. **At Long Last: Short, All-Oral Regimens for Multidrug-Resistant Tuberculosis in the United States.**

Open Forum Infect Dis. 2023 Apr 3;10(4):ofad177. doi: 10.1093/ofid/ofad177. eCollection 2023 Apr.

Sinha P(1), Jacobson KR(1), Horsburgh CR Jr(1)(2)(3)(4), Acuña-Villaorduña C(1).

Author information:
(1)Section of Infectious Diseases, Department of Medicine, Boston University Chobanian & Avedisian School of Medicine, Boston, Massachusetts, USA.
(2)Department of Epidemiology, Boston University School of Public Health, Boston, Massachusetts, USA.
(3)Department of Biostatistics, Boston University School of Public Health, Boston, Massachusetts, USA.
(4)Department of Global Health, Boston University School of Public Health, Boston, Massachusetts, USA.

Multidrug-resistant tuberculosis (MDR-TB) has historically required longer treatment regimens that were associated with higher unfavorable outcomes and side effects rates compared with drug susceptible TB (DS-TB). During the last decade, several studies conducted mostly in high-incidence settings have shown that MDR-TB can be successfully treated using all-oral shorter regimens of 6- to 9-month duration. In this article, we review the evolution of MDR-TB treatment from the early long regimens with injectables agents (IAs), followed by the shorter regimens with IA, to the groundbreaking, all-oral, 6- to 9-month regimens. Finally, we propose a framework for implementation of the shorter all-oral regimens in the United States.

DOI: 10.1093/ofid/ofad177
PMCID: PMC10135426
PMID: 37125228

Conflict of interest statement: Potential conflicts of interest. All authors: No reported conflicts of interest.

42. **The chamber of secretome in Mycobacterium tuberculosis as a potential therapeutic target.**

Biotechnol Genet Eng Rev. 2023 Apr;39(1):1-44. doi: 10.1080/02648725.2022.2076031. Epub 2022 May 25.

Dwivedi M(1), Bajpai K(2).

Mycobacterium tuberculosis (MTB) causes one of the ancient diseases, Tuberculosis, affects people around the globe and its severity can be understood by its classification as a second infectious disease after COVID-19 and the 13th leading cause of death according to a WHO report. Despite having advanced

diagnostic approaches and therapeutic strategies, unfortunately, TB is still spreading across the population due to the emergence of drug-resistance MTB and Latent TB infection (LTBI). We are seeking for effective approaches to overcome these hindrances and efficient treatment for this perilous disease. Therefore, there is an urgent need to develop drugs based on operative targeting of the bacterial system that could result in both efficient treatment and lesser emergence of MDR-TB. One such promising target could be the secretory systems and especially the Type 7 secretory system (T7SS-ESX) of Mycobacterium tuberculosis, which is crucial for the secretion of effector proteins as well as in establishing host-pathogen interactions of the tubercle bacilli. The five paralogous ESX systems (ESX-1 to EXS-5) have been observed by in silico genome analysis of MTB, among which ESX-1 and ESX-5 are substantial for virulence and mediating host cellular inflammasome. The bacterium growth and virulence can be modulated by targeting the T7SS. In the present review, we demonstrate the current status of therapeutics against MTB and focus on the function and cruciality of T7SS along with other secretory systems as a promising therapeutic target against Tuberculosis.

DOI: 10.1080/02648725.2022.2076031
PMID: 35613080 [Indexed for MEDLINE]

43. **Update on drug treatments for multidrug resistant tuberculosis.**

Curr Opin Infect Dis. 2023 Apr 1;36(2):132-139. doi: 10.1097/QCO.0000000000000899. Epub 2023 Jan 18.

Davies GR(1), Aston S.

PURPOSE OF THE REVIEW: To describe important recent developments in the treatment of multidrug resistant tuberculosis (MDR-TB).
RECENT FINDINGS: In the last decade, novel and repurposed antituberculosis drugs have transformed MDR-TB treatment with improved rates of treatment success, better tolerability and safety and reduced duration. As recently as 2016, standard care relied on up to seven drugs for 24 months with treatment success no better than 70%. Seven drug shorter so-called "Bangladesh" style regimens subsequently achieved similar or better results at a duration of 9-12 months but concerns about first-line resistance additional to rifampicin hampered global uptake. After conditional approval in 2012, the novel agent bedaquiline was demonstrated to improve outcomes and reduce mortality when used in longer and shorter regimens, resulting in the replacement of injectable agents. In the last 2 years, clinical trials of all-oral 6-month three or four drug regimens containing bedaquiline, pretomanid and linezolid have shown superior efficacy against both longer and shorter traditional regimens, resulting in major changes

in WHO guidance.
SUMMARY: Although some concerns around safety and emergent bedaquiline resistance remain to be fully addressed, 6-month all oral regimens promise to transform the treatment of people with MDR-TB worldwide.

DOI: 10.1097/QCO.0000000000000899
PMID: 36718913 [Indexed for MEDLINE]

44. **Clinical implications of molecular drug resistance testing for Mycobacterium tuberculosis: a 2023 TBnet/RESIST-TB consensus statement.**

Lancet Infect Dis. 2023 Apr;23(4):e122-e137. doi: 10.1016/S1473-3099(22)00875-1. Epub 2023 Feb 28.

Domínguez J(1), Boeree MJ(2), Cambau E(3), Chesov D(4), Conradie F(5), Cox V(6), Dheda K(7), Dudnyk A(8), Farhat MR(9), Gagneux S(10), Grobusch MP(11), Gröschel MI(12), Guglielmetti L(13), Kontsevaya I(14), Lange B(15), van Leth F(16), Lienhardt C(17), Mandalakas AM(18), Maurer FP(19), Merker M(20), Miotto P(21), Molina-Moya B(22), Morel F(13), Niemann S(23), Veziris N(13), Whitelaw A(24), Horsburgh CR Jr(25), Lange C(18); TBnet and RESIST-TB networks.

Collaborators: Domínguez J, Boeree MJ, Cambau E, Chesov D, Conradie F, Cox V, Dheda K, Dudnyk A, Farhat MR, Gagneux S, Grobusch MP, Gröschel MI, Guglielmetti L, Kontsevaya I, Lange B, van Leth F, Lienhardt C, Mandalakas AM, Maurer F, Merker M, Miotto P, Molina-Moya B, Morel F, Niemann S, Veziris N, Whitelaw A, Horsburgh CR, Lange C.

Erratum in
Lancet Infect Dis. 2023 Mar 28;:

Drug-resistant tuberculosis is a substantial health-care concern worldwide. Despite culture-based methods being considered the gold standard for drug susceptibility testing, molecular methods provide rapid information about the Mycobacterium tuberculosis mutations associated with resistance to anti-tuberculosis drugs. This consensus document was developed on the basis of a comprehensive literature search, by the TBnet and RESIST-TB networks, about reporting standards for the clinical use of molecular drug susceptibility testing. Review and the search for evidence included hand-searching journals and searching electronic databases. The panel identified studies that linked mutations in genomic regions of M tuberculosis with treatment outcome data. Implementation of molecular testing for the prediction of drug resistance in M

tuberculosis is key. Detection of mutations in clinical isolates has implications for the clinical management of patients with multidrug-resistant or rifampicin-resistant tuberculosis, especially in situations when phenotypic drug susceptibility testing is not available. A multidisciplinary team including clinicians, microbiologists, and laboratory scientists reached a consensus on key questions relevant to molecular prediction of drug susceptibility or resistance to M tuberculosis, and their implications for clinical practice. This consensus document should help clinicians in the management of patients with tuberculosis, providing guidance for the design of treatment regimens and optimising outcomes.

DOI: 10.1016/S1473-3099(22)00875-1
PMID: 36868253 [Indexed for MEDLINE]

Conflict of interest statement: Declaration of interests JD reports a technology licence to GenID (Germany), and honoraria for lectures from Oxford Immunotec (UK). EC reports support for attendance, accommodation, and travel for ECCMID 2022, Lisboa from European Society of Clinical Microbiology and Infectious Diseases (ESCMID), for annual ERL-TB net meetings from ERL-TB net (Network of National Reference Centers for Tuberculosis in Europe), and for the annual congress 2022, Bologna, from the European Society for Mycobacteriology; is a member of the Executive committee of ESCMID; and is chair of the subcommittee for antimycobacterial agents of EUCAST (European Committee on Antimicrobial Susceptibility Testing). MRF reports grants or contracts from NIH/NIAID (5R01AI155765 and 5R21AI154089) and consulting fees (paid to them) from FIND. LG is a member (unpaid) of the data safety monitoring board for the XACT-19 clinical trial in University of Cape Town, Cape Town, South Africa, and is co-principal investigator of two phase 3 clinical trials on shorter treatment for MDR-TB (endTB and endTB-Q), funded by Unitaid. BL reports grants or contracts from European Union, German Ministry for Education and Research (BMBF), Kultusministerkonferenz, German Centre for Infection Research, and Helmholtz Association, and unpaid leadership or fiduciary roles for DZIF IAB, DZIF Steering Committee transplant Cohort, and TBnet chair Epidemiology. SN reports support for this manuscript (eg, funding, provision of study materials, medical writing, and article processing charges) from BMBF (German Center for Infection Research), DFG (Excellenz Cluster Precision Medicine in Chronic Inflammation EXC 2167), and Leibniz Science Campus Evolutionary Medicine of the LUNG (EvoLUNG), and consulting fees from Illumina advisory board in 2022. NV reports grants or contracts for a study on bedaquiline from Janssen. CRH reports grants or contracts from NIH/NIAID (R01AI134430, DAA3-19-65672, R01AI147316 U01AI152980, and R01AI146555), and Centers for Disease Control and Prevention (NU38PS004651); consulting fees from Otsuka Pharmaceuticals; participation on a

data safety monitoring board for SODUCU (PanACEA Sutezolid Dose-finding and Combination Evaluation), BEAT-Tuberculosis (Building Evidence for Advancing New treatment for tuberculosis), DECODE (PanACEA Delpazolid Dose-finding and Combination Development), and Médecins Sans Frontières; and a leadership or fiduciary role from the International Union Against Tuberculosis and Lung Diseases. CLa reports support for the present manuscript (eg, funding, provision of study materials, medical writing, and article processing charges) from DZIF (German Center of Infection Research); consulting fees from a consultation service to INSMED, a company that produced liposomal amikacin as an inhalative suspension for the treatment of non-tuberculous mycobacteria pulmonary disease (outside of the scope of this work); speakers' honoraria from Insmed, Gilead, and Janssen (all outside of the scope of this work); is a member of the data safety board of trials from Médecins Sans Frontières (outside of the scope of this work); is supported by the German Center for Infection Research (DZIF); and acknowledges funding from the European Commission (anTBiotic EU-H2020 733079, ClicTB EDCTP2 RIA2017T-2030, stool4TB EDCTP2 RIAD2018-2511, and UNITE4TB EU-IMI 101007873). All other authors declare no competing interests.

45. **Unfavorable treatment outcomes among patients with drug-resistant TB in Uganda.**

Int J Tuberc Lung Dis. 2023 Apr 1;27(4):291-297. doi: 10.5588/ijtld.22.0638.

Kintu TM(1), Mwanahamisi BS(1), Muwanguzi M(1), Kyagambiddwa T(1), Miiro E(1), Tishekwa N(2), Lodiong LJD(1), Timbiine AK(2), Tumukunde P(2), Baluku JB(3), Nuwagira E(4).

BACKGROUND: Drug-resistant TB (DR-TB) remains a significant public health burden and a threat to the progress made in TB control and prevention in sub-Saharan Africa.OBJECTIVE: To determine the risk-predictors of poor treatment outcomes in patients with DR-TB in Uganda.METHODS: We retrospectively reviewed medical records of adult Ugandans who had been treated for DR-TB at Mbarara Regional Referral Hospital (MRRH) in Uganda.RESULTS: Of the 385 files reviewed, 332 (86.2%) met the study inclusion criteria. Of these, 226 (68.1%) were men and 193 (58.1%) were HIV-positive. A total of 73 participants (22.7%) had unfavorable treatment outcomes (treatment failure, loss to follow-up or death). History of cigarette smoking (OR 5.10, 95% CI 2.4-11.4; P < 0.001), age >60 years (OR 6.32, 95% CI 2.2-18.6; P < 0.001), anemia (OR 2.38, 95% CI 1.1-5.3; P = 0.02) and thrombocytopenia (OR 3.60, 95% CI 1.6-8.1; P < 0.001) were independent predictors of unfavorable treatment outcomes.CONCLUSION: There is a high prevalence of unfavorable treatment outcomes among patients with DR-TB. Further research is required to design a prognostic model for DR-TB patients in a resource-limited setting.

DOI: 10.5588/ijtld.22.0638
PMID: 37035969 [Indexed for MEDLINE]

46. **A four-drug standardized short regimen for highly resistant TB in South-West Nigeria.**

Int Health. 2023 Apr 7:ihad023. doi: 10.1093/inthealth/ihad023. Online ahead of print.

Fadeyi MO(1), Decroo T(2), Ortuño-Gutiérrez N(3), Ahmed B(1), Jinadu A(1), El-Tayeb O(3), Adebola W(4), Kehinde A(5), Lynen L(2), Gils T(2).

BACKGROUND: Patients with TB resistant to rifampicin (Rr-TB), and those with additional resistance to fluoroquinolones (pre-XDR-TB), should be treated with bedaquiline-pretomanid-linezolid-moxifloxacin and bedaquiline-pretomanid-linezolid, respectively. However, pretomanid is not yet widely available.
METHODS: This is a pragmatic prospective single-arm study investigating the efficacy and safety of 9 mo of bedaquiline-delamanid-linezolid-clofazimine in patients with pre-XDR-TB or Rr-TB unresponsive to Rr-TB treatment in Nigeria.
RESULTS: From January 2020 to June 2022, 14 of 20 patients (70%) successfully completed treatment, five died and one was lost-to-follow-up. No one experienced a treatment-emergent grade three/four event. Treatment success was higher compared with global pre-XDR-TB treatment outcomes.
CONCLUSIONS: While pretomanid is unavailable, highly resistant TB can be treated with bedaquiline-delamanid-linezolid-clofazimine.

DOI: 10.1093/inthealth/ihad023
PMID: 37026448

47. **Myoinositol and methyl stearate increases rifampicin susceptibility among drug-resistant Mycobacterium tuberculosis expressing Rv1819c.**

Chem Biol Drug Des. 2023 Apr;101(4):883-895. doi: 10.1111/cbdd.14197. Epub 2023 Jan 5.

Nirmal CR(1), Rajadas SE(1), Balasubramanian M(1), Mohanvel SK(1), Aathi MS(2), Munishankar S(1), Chilamakuru NB(3), Thiruvenkadam K(1), Pandiya Raj AK(1),

Paraman R(4), Dusthackeer A(1).

The alarming increase in multidrug resistance, which includes Bedaquiline and Delamanid, stumbles success in Tuberculosis treatment outcome. Mycobacterium tuberculosis gains resistance to rifampicin, which is one of the less toxic and potent anti-TB drugs, through genetic mutations predominantly besides efflux pump mediated drug resistance. In recent decades, scientific interventions are being carried out to overcome this hurdle using novel approaches to save this drug by combining it with other drugs/molecules or by use of high dose rifampicin. This study reports five small molecules namely Ellagic acid, Methyl Stearate, Myoinositol, Rutin, and Shikimic acid that exhibit synergistic inhibitory activity with rifampicin against resistant TB isolates. In-silico examinations revealed possible blocking of Rv1819c-an ABC transporter efflux pump that was known to confer resistance in M. tuberculosis to rifampicin. The synergistic anti-TB activity was assessed using a drug combination checkerboard assay. Efflux pump inhibition activity of ellagic acid, myoinositol, and methyl stearate was observed through ethidium bromide accumulation assay in the drug-resistant M. tuberculosis clinical strains and recombinant Mycobacterium smegmatis expressing Rv1819c in coherence with the significant reduction in the minimum inhibitory concentration of rifampicin. Cytotoxicity of the active efflux inhibitors was tested using in silico and ex vivo methods. Myoinositol and methyl stearate were completely non-toxic to the hematological and epithelial cells of different organs under ex vivo conditions. Based on these findings, these molecules can be considered for adjunct TB therapy; however, their impact on other drugs of anti-TB regimen needs to be tested.

DOI: 10.1111/cbdd.14197
PMID: 36533863 [Indexed for MEDLINE]

48. **Diagnosing osteo-articular tuberculosis and multidrug resistance using real-time polymerase chain reaction and high-resolution melt-curve analysis.**

J Orthop Res. 2023 Apr;41(4):891-896. doi: 10.1002/jor.25410. Epub 2022 Jul 13.

Sharma K(1), Sharma M(2), Sharma A(3), Dhillon MS(4).

The study evaluated real-time quantitative polymerase chain reaction (qPCR) and high-resolution melt-curve analysis (HRM) for simultaneous diagnosis of osteo-articular tuberculosis (OATB) and drug resistance. Two hundred and fifty synovial fluid and pus specimens (20 confirmed OATB by culture, 130 suspected OATB, and 100 controls) processed in the Department of Medical Microbiology,

PGIMER were subjected to qPCR using rpoB, MPB64, and IS6110 genes. All OATB positive specimens were subjected to HRM for detecting resistance to rifampicin and isoniazid. qPCR detected 129/150 OATB cases with a sensitivity of 86% (95% for confirmed and 84.6% for suspected OATB cases) and specificity of 100%. rpoB and MPB64 genes had higher sensitivity than IS6110 (86% vs. 74.6%). HRM reported eight multidrug resistant (MDR), two mono-rifampicin, and five mono-isoniazid resistant cases, all were concordant with gene sequencing. qPCR followed by HRM analysis offer a simple, accurate, and rapid platform for simultaneous detection of OATB and MDR.

DOI: 10.1002/jor.25410
PMID: 35780389 [Indexed for MEDLINE]

49. **Recent developments of imidazo[1,2-a]pyridine analogues as antituberculosis agents.**

RSC Med Chem. 2023 Mar 3;14(4):644-657. doi: 10.1039/d3md00019b. eCollection 2023 Apr 26.

Samanta S(1), Kumar S(1), Aratikatla EK(1), Ghorpade SR(1), Singh V(1)(2).

Over the past 2000 years, tuberculosis (TB) has killed more people than any other infectious disease. In 2021, TB claimed 1.6 million lives worldwide, making it the second leading cause of death from an infectious disease after COVID-19. Unfortunately, TB drug discovery research was neglected in the last few decades of the twentieth century. Recently, the World Health Organization has taken the initiative to develop new TB drugs. Imidazopyridine, an important fused bicyclic 5,6 heterocycle has been recognized as a "drug prejudice" scaffold for its wide range of applications in medicinal chemistry. A few examples of imidazo[1,2-a]pyridine exhibit significant activity against multidrug-resistant TB (MDR-TB) and extensively drug-resistant TB (XDR-TB). Here, we critically review anti-TB compounds of the imidazo[1,2-a]pyridine class by discussing their development based on the structure-activity relationship, mode-of-action, and various scaffold hopping strategies over the last decade, which is identified as a renaissance era of TB drug discovery research.

DOI: 10.1039/d3md00019b
PMCID: PMC10131611
PMID: 37122538

Conflict of interest statement: There are no conflicts to declare.

50. **Synthesis and Anti-Mycobacterium tuberculosis Activity of Imidazo[2,1-b][1,3]oxazine Derivatives against Multidrug-Resistant Strains.**

ChemMedChem. 2023 Apr 1:e202300015. doi: 10.1002/cmdc.202300015. Online ahead of print.

Fernandes GFS(1)(2)(3), Manieri KF(1), Bonjorno AF(1), Campos DL(1), Ribeiro CM(1), Demarqui FM(1), Ruiz DAG(4), Nascimento-Junior NM(4), Denny WA(2), Thompson AM(2), Pavan FR(1), Dos Santos JL(1).

The emergence of multidrug-resistant strains of M. tuberculosis has raised concerns due to the greater difficulties in patient treatment and higher mortality rates. Herein, we revisited the 2-nitro-6,7-dihydro-5H-imidazo[2,1-b][1,3]oxazine scaffold and identified potent new carbamate derivatives having MIC90 values of 0.18-1.63 µM against Mtb H37Rv. Compounds 47-49, 51-53, and 55 exhibited remarkable activity against a panel of clinical isolates, displaying MIC90 values below 0.5 µM. In Mtb-infected macrophages, several compounds demonstrated a 1-log greater reduction in mycobacterial burden than rifampicin and pretomanid. The compounds tested did not exhibit significant cytotoxicity against three cell lines or any toxicity to Galleria mellonella. Furthermore, the imidazo[2,1-b][1,3]oxazine derivatives did not show substantial activity against other bacteria or fungi. Finally, molecular docking studies revealed that the new compounds could interact with the deazaflavin-dependent nitroreductase (Ddn) in a similar manner to pretomanid. Collectively, our findings highlight the chemical universe of imidazo[2,1-b][1,3]oxazines and their promising potential against MDR-TB.

DOI: 10.1002/cmdc.202300015
PMID: 37002895

51. **A host blood transcriptional signature differentiates multi-drug/rifampin-resistant tuberculosis (MDR/RR-TB) from drug susceptible tuberculosis: a pilot study.**

Mol Biol Rep. 2023 Apr;50(4):3935-3943. doi: 10.1007/s11033-023-08307-6. Epub 2023 Feb 7.

Madamarandawala P(1), Rajapakse S(2), Gunasena B(3), Madegedara D(4), Magana-Arachchi D(5).

BACKGROUND: Tuberculosis (TB), caused by the bacterium Mycobacterium tuberculosis is one of the top thirteen causes of death worldwide. The major challenge to control TB is the emergence of drug-resistant tuberculosis (DR-TB); specifically, multi-drug resistant TB which are resistant to the most potent drugs; rifampin and isoniazid. Owing to the inconsistencies of the current diagnostic methods, a single test cannot identify the whole spectrum of DR-TB associated mutations. Recently, host blood transcriptomics has gained attention as a promising technique that develops disease-specific RNA signatures/biomarkers. However, studies on host transcriptomics infected with DR-TB is limited. Herein, we intended to identify genes/pathways that are differentially expressed in multi-drug/rifampin resistant TB (MDR/RR-TB) in contrast to drug susceptible TB.
METHOD AND RESULTS: We conducted blood RNA sequencing of 10 pulmonary TB patients (4; drug susceptible and 6; DR-TB) and 55 genes that were differentially expressed in MDR/RR-TB from drug-susceptible/mono-resistant TB were identified. CD300LD, MYL9, VAMP5, CARD17, CLEC2B, GBP6, BATF2, ETV7, IFI27 and FCGR1CP were found to be upregulated in MDR/RR-TB in all comparisons, among which CLEC2B and CD300LD were not previously linked to TB. In comparison pathway analysis, interferon alpha/gamma response was upregulated while Wnt/beta catenin signaling, lysosome, microtubule nucleation and notch signaling were downregulated.
CONCLUSION: Up/down-regulation of immunity related genes/pathways speculate the collective effect of hosts' attempt to fight against continuously multiplying DR-TB bacteria and the bacterial factors to fight against the host defense. The identified genes/pathways could act as MDR/RR-TB biomarkers, hence, further research on their clinical use should be encouraged.

DOI: 10.1007/s11033-023-08307-6
PMID: 36749527 [Indexed for MEDLINE]

52. **Population Pharmacokinetics and Dose Evaluation of Cycloserine among Patients with Multidrug-Resistant Tuberculosis under Standardized Treatment Regimens.**

Antimicrob Agents Chemother. 2023 Apr 25:e0170022. doi: 10.1128/aac.01700-22. Online ahead of print.

Zhu Y(#)(1), Zhu L(#)(2), Davies Forsman L(#)(3)(4), Paues J(5)(6), Werngren J(7), Niward K(5)(6), Schön T(5)(6)(8), Bruchfeld J(3)(4), Xiong H(#)(1),

Alffenaar JW(#)(9)(10)(11), Hu Y(#)(1).

Although cycloserine is a recommended drug for the treatment of multidrug-resistant tuberculosis (MDR-TB) according to World Health Organization (WHO), few studies have reported on pharmacokinetics (PK) and/or pharmacodynamics (PD) data of cycloserine in patients with standardized MDR-TB treatment. This study aimed to estimate the population PK parameters for cycloserine and to identify clinically relevant PK/PD thresholds, as well as to evaluate the current recommended dosage. Data from a large cohort with full PK curves was used to develop a population PK model. This model was used to estimate drug exposure in patients with MDR-TB from a multicentre prospective study in China. The classification and regression tree was used to identify the clinically relevant PK/PD thresholds. Probability of target attainment was analyzed to evaluate the currently recommended dosing strategy. Cycloserine was best described by a two-compartment disposition model. A percentage of time concentration above MICs (T>MIC) of 30% and a ratio of area under drug concentration-time curve (AUC0-24h) over MIC of 36 were the valid predictors for 6-month sputum culture conversion and final treatment outcome. Simulations showed that with WHO-recommended doses (500 mg and 750 mg for patients weighing <45 kg and ≥45 kg), the probability of target attainment exceeded 90% at MIC ≤16 mg/L in MGIT for both T>MIC of 30% and AUC0-24h/MIC of 36. New clinically relevant PK/PD thresholds for cycloserine were identified in patients with standardized MDR-TB treatment. WHO-recommended doses were considered adequate for the MGIT MIC distribution in our cohort of Chinese patients with MDR-TB.

DOI: 10.1128/aac.01700-22
PMID: 37097151

53. **Comprehensive coverage on anti-mycobacterial endeavour reported during 2022.**

Eur J Med Chem. 2023 Apr 21;255:115409. doi: 10.1016/j.ejmech.2023.115409. Online ahead of print.

Dhameliya TM(1), Vekariya DD(2), Patel HY(2), Patel JT(2).

TB being one of the deadliest diseases and second most common infectious cause of deaths, poses the severe threat to global health. The extended duration of therapy owing to resistance and its upsurge in immune-compromised patients have been the driving force for the development of novel of anti-TB scaffolds. Recently, we have compiled the account of anti-mycobacterial scaffolds published during 2015-2020 and updated them in 2021. The present work involves the insights on the anti-mycobacterial scaffolds reported in 2022 with their mechanism of action, structure activity relationships, along with the key

perceptions for the design of newer anti-TB agents for the broader interests of medicinal chemists.

DOI: 10.1016/j.ejmech.2023.115409
PMID: 37120997

Conflict of interest statement: 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.

54. **Efficacy and Tolerability of Concomitant Use of Bedaquiline and Delamanid for Multidrug- and Extensively Drug-Resistant Tuberculosis: A Systematic Review and Meta-Analysis.**

2023 Apr 3;76(7):1328-1337. doi: 10.1093/cid/ciac876.

Holmgaard FB(1)(2), Guglielmetti L(3)(4), Lillebaek T(5)(6), Andersen ÅB(7), Wejse C(1)(2), Dahl VN(1)(2)(5).

The introduction of two novel drugs, bedaquiline and delamanid, has given hope for better and shorter treatments of drug-resistant tuberculosis. A systematic review was conducted to evaluate the efficacy and safety of concomitant bedaquiline and delamanid administration. Pooled estimates of World Health Organization-defined favorable treatment outcome and significant QTc-interval prolongation (QTc ≥500 ms or ≥60 ms increase from baseline) were calculated using a random-effects model. Thirteen studies including a total of 1031 individuals with multidrug-resistant/rifampicin-resistant tuberculosis who received bedaquiline and delamanid were included. The pooled estimate of favorable treatment outcome was 73.1% (95% confidence interval [CI]: 64.3-81.8%). Sputum culture conversion at 6 months ranged from 61% to 95%. Overall, the pooled proportion of QTc-prolongation was 7.8% (95% CI: 4.1-11.6%) and few cardiac events were reported (0.8%; n = 6/798). Rates of sputum culture conversion and favorable treatment outcome were high in patients treated concomitantly with bedaquiline and delamanid, and the treatment seemed tolerable with low rates of clinically significant cardiac toxicity.

DOI: 10.1093/cid/ciac876
PMID: 36331978 [Indexed for MEDLINE]

Conflict of interest statement: Potential conflicts of interest . L. G. is a principal investigator of 2 phase III randomized controlled trials, funded by Unitaid and sponsored by Médecins Sans Frontières, testing shorter regimens for MDR/RR-TB including bedaquiline and delamanid. C. W. is on an advisory board for Pfizer on COVID-19 treatments (2 meetings in 2022 for which an honorarium was paid) and was recently lecturing on future pandemics for pulmonology specialists for Novartis (payment or honoraria). These sponsors had no role in the study design, data collection, data analysis, data interpretation, or writing of the manuscript. All other authors report no potential conflicts. All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

55. **MTBDRplus and MTBDRsl for simultaneous diagnosis of gastrointestinal tuberculosis and detection of first-line and second-line drug resistance.**

J Gastroenterol Hepatol. 2023 Apr;38(4):619-624. doi: 10.1111/jgh.16124. Epub 2023 Jan 30.

Sharma K(1), Sharma M(1)(2), Sharma V(3), Sharma M(3), Parmar UPS(4), Samanta J(3), Sharma A(5), Kochhar R(3), Sinha SK(3).

BACKGROUND AND AIM: Emergence of drug resistance, especially to second-line drugs, hampers tuberculosis elimination efforts. The present study aimed to evaluate MTBDRplus and MTBDRsl assays for detecting first-line and second-line drug resistance, respectively, in gastrointestinal tuberculosis (GITB).
METHODS: Thirty ileocecal biopsy specimens, processed in the Department of Microbiology between 2012 and 2022, that showed growth of Mycobacterium tuberculosis on culture were included in the study. DNA, extracted from culture, was subjected to MTBDRplus and MTBDRsl (Hain Lifescience GmbH, Nehren, Germany), following manufacturer's instructions. Their performance was compared against phenotypic drug susceptibility testing (pDST) and gene sequencing.
RESULTS: Out of the 30 specimens, 4 (13.33%) were mono-isoniazid resistant, 4 (13.33%) were multidrug resistant (MDR), 2 (6.67%) were pre-extensively drug resistant (pre-XDR), and 2 (6.67%) were mono-fluoroquinolone resistant. The results were 100% concordant with pDST and gene sequencing.
CONCLUSIONS: In the wake of growing drug resistance in all forms of extrapulmonary tuberculosis, including GITB, MTBDRplus and MTBDRsl are reliable tools for screening of resistance to both first-line and second-line drugs.

DOI: 10.1111/jgh.16124
PMID: 36652396 [Indexed for MEDLINE]

56. **Bioisosteric modification of Linezolid identified the potential M. tuberculosis protein synthesis inhibitors to overcome the myelosuppression and serotonergic toxicity associated with Linezolid in the treatment of the multi-drug resistance tuberculosis (MDR-TB).**

J Biomol Struct Dyn. 2023 Apr 25:1-16. doi: 10.1080/07391102.2023.2203254. Online ahead of print.

Girase R(1), Ahmad I(1), Patel H(1).

Linezolid is the first and only oxazolidinone antibacterial drug was approved in the last 35 years. It exhibits bacteriostatic efficacy against M. tuberculosis and is a crucial constituent of the BPaL regimen (Bedaquiline, Pretomanid, and Linezolid), which was authorized by the FDA in 2019 for the treatment of XDR-TB or MDR-TB. Despite its unique mechanism of action, Linezolid carries a considerable risk of toxicity, including myelosuppression and serotonin syndrome (SS), which is caused by inhibition of mitochondrial protein synthesis (MPS) and monoamine oxidase (MAO), respectively. Based on the structure toxicity relationship (STR) of Linezolid, in this work, we used a bioisosteric replacement approach to optimize the structure of Linezolid at the C-ring and/or C-5 position for myelosuppression and serotogenic toxicity. Extensive hierarchical multistep docking, drug likeness prediction, molecular binding interactions analyses, and toxicity assessment identified three promising compounds (3071, 7549 and 9660) as less toxic potential modulators of Mtb EthR protein. Compounds 3071, 7549 and 9660 were having the significant docking score of -12.696 Kcal/mol, -12.681 Kcal/mol and -15.293 Kcal/mol towards the Mtb EthR protein with less MAO-A and B affinity [compound 3071: MAO A (-4.799 Kcal/mol) and MAO B (-6.552 Kcal/mol); compound 7549: MAO A (> -2.00 Kcal/mol) and MAO B (> -2.00 Kcal/mol) and compound 9660: MAO A (> -5.678 Kcal/mol) and MAO B (> -6.537Kcal/mol) and none of them shown the Leukopenia as a side effect due to the Myelosuppression. The MD simulation results and binding free energy estimations correspond well with docking analyses, indicating that the proposed compounds bind and inhibit the EthR protein more effectively than Linezolid. The quantum mechanical and electrical characteristics were evaluated using density functional theory (DFT), which also demonstrated that the proposed compounds are more reactive than Linezolid.Communicated by Ramaswamy H. Sarma.

DOI: 10.1080/07391102.2023.2203254
PMID: 37097976

57. **Comprehensive Drug Resistance Characterization of Pulmonary Tuberculosis in Algeria: Insights on Mycobacterium tuberculosis Strains by Whole-Genome Sequencing.**

Microb Drug Resist. 2023 Apr 28. doi: 10.1089/mdr.2022.0321. Online ahead of print.

Benremila D(1), Djoudi F(1), Gharout-Sait A(1), Kheloufi S(1), Spitaleri A(2), Battaglia S(2), Cabibbe AM(2), Cirillo DM(2).

In this study, we aimed to characterize drug-resistant strains by whole-genome sequencing (WGS), to describe the spreading lineages and the history of transmission. Drug susceptibility testing was performed by 96-well broth microdilution plates. The genomic DNA was extracted and purified; libraries were prepared and run on the Illumina NextSeq500 System. Among 82 isolates, 21 tuberculosis (TB) isolates (25.6%) were drug resistant, including 10 MDR and 4 pre-extensively drug-resistant (XDR)-TB. The mutation Ser315Thr in the katG gene was confirmed in 15 isolates. In rpoB, Ser450Leu and His445Asp mutations were the most common. Asp94Asn and Ala90Val mutations were reported in gyrA. The LAM family, the most TB drug resistant, was widely predominant in the north and the T sublineage in the south of the country. This study provides the first insight on TB drug resistance using WGS in Algeria and clearly describes the first pre-XDR-TB cases and lineage distribution across the country.

DOI: 10.1089/mdr.2022.0321
PMID: 37115530

58. **Exploration of the diversity of multi-drug resistant Mycobacterium tuberculosis complex in Lagos, Nigeria using WGS: Distribution of lineages, drug resistance patterns and genetic mutations.**

Tuberculosis (Edinb). 2023 Apr 16;140:102343. doi: 10.1016/j.tube.2023.102343. Online ahead of print.

Noorizhab MNF(1), Zainal Abidin N(2), Teh LK(1), Tang TH(3), Onyejepu N(4), Kunle-Ope C(4), Tochukwu NE(4), Sheshi MA(5), Nwafor T(6), Akinwale OP(7), Ismail AI(8), Nor NM(9), Salleh MZ(10).

Multidrug-resistant (MDR) or extensively drug-resistant (XDR) Tuberculosis (TB)

is a major challenge to global TB control. Therefore, accurate tracing of in-country MDR-TB transmission are crucial for the development of optimal TB management strategies. This study aimed to investigate the diversity of MTBC in Nigeria. The lineage and drug-resistance patterns of the clinical MTBC isolates of TB patients in Southwestern region of Nigeria were determined using the WGS approach. The phenotypic DST of the isolates was determined for nine anti-TB drugs. The sequencing achieved average genome coverage of 65.99X. The most represented lineages were L4 (n = 52, 83%), L1 (n = 8, 12%), L2 (n = 2, 3%) and L5 (n = 1, 2%), suggesting a diversified MTB population. In term of detection of M/XDR-TB, while mutations in katG and rpoB genes are the strong predictors for the presence of M/XDR-TB, the current study also found the lack of good genetic markers for drug resistance amongst the MTBC in Nigeria which may pose greater problems on local tuberculosis management efforts. This high-resolution molecular epidemiological data provides valuable insights into the mechanistic for M/XDR TB in Lagos, Nigeria.

DOI: 10.1016/j.tube.2023.102343
PMID: 37080082

Conflict of interest statement: Declaration of conflicts of interest This study has no conflicts of interest.

59. **Decentralized, Integrated Treatment of RR/MDR-TB and HIV Using a Bedaquiline-Based, Short-Course Regimen Is Effective and Associated With Improved HIV Disease Control.**

J Acquir Immune Defic Syndr. 2023 Apr 15;92(5):385-392. doi: 10.1097/QAI.0000000000003150.

Govender T(1), Jham MA(2), Zhang JC(3), Pillay S(4), Pak Y(5), Pillay P(4), Furin J(6), Malenfant J(7), Murphy RA(8)(9).

BACKGROUND: In decentralized sites, with fewer resources and a high prevalence of advanced HIV, the effectiveness of the new short-course, bedaquiline-based regimen for rifampicin-resistant and multidrug-resistant tuberculosis (RR/MDR-TB) is not well-described.
SETTING: Adults with pulmonary RR/MDR-TB initiating the short-course regimen in KwaZulu-Natal, South Africa were prospectively enrolled at a decentralized program that integrated person-centered TB care.
METHODS: In addition to standard of care monitoring, study visits occurred at enrollment and months 1, 2, 4, 6, and 9. Favorable RR/MDR-TB outcome was defined

as cure or treatment completion without loss to follow-up, death, or failure by treatment. In patients with HIV, we assessed antiretroviral therapy (ART) uptake, virologic and immunologic outcomes.
RESULTS: Among 57 patients, HIV was present in 73.7% (95% CI: 60.3-84.5), with a median CD4 count of 170 cells/mm 3 (intraquartile range 49-314). A favorable RR/MDR-TB outcome was achieved in 78.9% (CI: 67.1-87.9). Three deaths occurred, all in the setting of baseline advanced HIV and elevated viral load. Overall, 21.1% (95% CI: 12.1-32.9) experienced a severe or life-threatening adverse event, the most common of which was anemia. Among patients with HIV, enrollment resulted in increased ART uptake by 24% (95% CI: 12.1%-39.4%), a significant improvement from baseline (P = 0.004); virologic suppression during concomitant treatment was observed in 71.4% (n = 30, 95% CI: 55.4-84.3).
CONCLUSION: Decentralized, person-centered care for RR/MDR-TB in patients with HIV using the short-course, bedaquiline-based regimen is effective and safe. In patients with HIV, enrollment led to improved ART use and reassuring virologic outcomes.

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PMCID: PMC10006315
PMID: 36729541 [Indexed for MEDLINE]

Conflict of interest statement: All authors declare no conflicts of interest.

60. **Development of DNA Bio-chip for Detection of Mutations of rpoB, embB and inhA Genes in Drug-Resistant Mycobacterium Tuberculosis.**

Indian J Clin Biochem. 2023 Apr;38(2):242-250. doi: 10.1007/s12291-022-01044-w. Epub 2022 May 2.

Jain B(1)(2), Kulkarni S(1)(2).

Drug-resistant (DR) tuberculosis (TB) is a global threat to health security and TB control programs. Since conventional drug susceptibility testing (DST) takes several weeks, we have developed a molecular method for the rapid identification of DR strains of Mycobacterium Tuberculosis (M.tb) utilizing DNA bio-chips. DNA bio-chips were prepared by immobilizing oligonucleotides (probes) on highly microporous polycarbonate track-etched membranes (PC-TEM) as novel support. Bio-chip was designed to contain 15 specific probes to detect mutations in three genes (rpoB, embB, and inhA). A sensitive and specific chemiluminescence based bio-chip assay was developed based on multiplex PCR followed by hybridization on bio-chip. Fifty culture isolates were used to evaluate the ability of in-house

developed bio-chip to detect the mutations. Bio-chip analysis shows that 37.7% of samples show wild type sequences, 53.3% of samples were monoresistance showing resistance to either rifampicin (RMP), isoniazid (INH), or ethambutol (EMB). 4.4% of samples were polydrug resistant showing mutations in both the rpoB gene and embB gene while 4.4% of samples were multidrug-resistant (MDR), harboring mutations in the rpoB and inhA genes. The results were compared with DST and sequencing. Compared to sequencing, bio-chip assay shows a sensitivity of 96.5% and specificity of 100% for RMP resistance. For EMB and INH, the results were in complete agreement with sequencing. This study demonstrates the first-time use of PC-TEMs for developing DNA bio-chip for the detection of mutations associated with drug resistance in M.tb. Developed DNA bio-chip accurately detected different mutations present in culture isolates and thus provides detailed and reliable data for clinical diagnosis.

DOI: 10.1007/s12291-022-01044-w
PMCID: PMC10070561
PMID: 37025431

Conflict of interest statement: Conflict of interestAll the authors declare that they have no conflict of interest.

61. **Emergence of Canonical and Noncanonical Genomic Variants following In Vitro Exposure of Clinical Mycobacterium tuberculosis Strains to Bedaquiline or Clofazimine.**

Antimicrob Agents Chemother. 2023 Apr 18;67(4):e0136822. doi: 10.1128/aac.01368-22. Epub 2023 Mar 9.

Ismail N(1)(2), Dippenaar A(3), Warren RM(1), Peters RPH(2)(4), Omar SV(4)(5).

In Mycobacterium tuberculosis, bedaquiline and clofazimine resistance occurs primarily through Rv0678 variants, a gene encoding a repressor protein that regulates mmpS5/mmpL5 efflux pump gene expression. Despite the shared effect of both drugs on efflux, little else is known about other pathways affected. We hypothesized that in vitro generation of bedaquiline- or clofazimine-resistant mutants could provide insight into additional mechanisms of action. We performed whole-genome sequencing and determined phenotypic MICs for both drugs on progenitor and mutant progenies. Mutants were induced through serial passage on increasing concentrations of bedaquiline or clofazimine. Rv0678 variants were identified in both clofazimine- and bedaquiline-resistant mutants, with

concurrent atpE SNPs occurring in the latter. Of concern was the acquisition of variants in the F420 biosynthesis pathway in clofazimine-resistant mutants obtained from either a fully susceptible (fbiD: del555GCT) or rifampicin mono-resistant (fbiA: 283delTG and T862C) progenitor. The acquisition of these variants possibly implicates a shared pathway between clofazimine and nitroimidazoles. Pathways associated with drug tolerance and persistence, F420 biosynthesis, glycerol uptake and metabolism, efflux, and NADH homeostasis appear to be affected following exposure to these drugs. Shared genes affected by both drugs include Rv0678, glpK, nuoG, and uvrD1. Genes with variants in the bedaquiline resistant mutants included atpE, fadE28, truA, mmpL5, glnH, and pks8, while clofazimine-resistant mutants displayed ppsD, fbiA, fbiD, mutT3, fadE18, Rv0988, and Rv2082 variants. These results show the importance of epistatic mechanisms as a means of responding to drug pressure and highlight the complexity of resistance acquisition in M. tuberculosis.

DOI: 10.1128/aac.01368-22
PMCID: PMC10112258
PMID: 36892309 [Indexed for MEDLINE]

Conflict of interest statement: The authors declare no conflict of interest.

62. **Association of smoking and alcohol use with rifampin-resistant TB treatment outcomes.**

Int J Tuberc Lung Dis. 2023 Apr 1;27(4):338-340. doi: 10.5588/ijtld.22.0678.

Campbell JR(1), Chan ED(2), Anderson LF(3), Bonnet M(4), Brode SK(5), Cegielski JP(6), Guglielmetti L(7), Singla R(8), Fox GJ(9), Skrahina A(10), Rodrigues D(11), Kuksa L(12), Viiklepp P(13), Menzies D(14).

DOI: 10.5588/ijtld.22.0678
PMID: 37035974 [Indexed for MEDLINE]

63. **Assessing Pretomanid for Tuberculosis (APT), a Randomized Phase 2 Trial of Pretomanid-Containing Regimens for Drug-Sensitive Tuberculosis: 12-Week Results.**

Am J Respir Crit Care Med. 2023 Apr 1;207(7):929-935. doi: 10.1164/rccm.202208-1475OC.

Dooley KE(1), Hendricks B(2), Gupte N(3), Barnes G(4), Narunsky K(2), Whitelaw C(2), Smit T(2), Ignatius EH(4), Friedman A(2), Dorman SE(5), Dawson R(2); Assessing Pretomanid for Tuberculosis (APT) Study Team.

Comment in
Am J Respir Crit Care Med. 2023 Apr 1;207(7):816-818.

Rationale: Pretomanid is a new nitroimidazole with proven treatment-shortening efficacy in drug-resistant tuberculosis. Pretomanid-rifamycin-pyrazinamide combinations are potent in mice but have not been tested clinically. Rifampicin, but not rifabutin, reduces pretomanid exposures. Objectives: To evaluate the safety and efficacy of regimens containing pretomanid-rifamycin-pyrazinamide among participants with drug-sensitive pulmonary tuberculosis. Methods: A phase 2, 12-week, open-label randomized trial was conducted of isoniazid and pyrazinamide plus 1) pretomanid and rifampicin (arm 1), 2) pretomanid and rifabutin (arm 2), or 3) rifampicin and ethambutol (standard of care; arm 3). Laboratory values of safety and sputum cultures were collected at Weeks 1, 2, 3, 4, 6, 8, 10, and 12. Time to culture conversion on liquid medium was the primary outcome. Measurements and Main Results: Among 157 participants, 125 (80%) had cavitary disease. Median time to liquid culture negativity in the modified intention-to-treat population (n = 150) was 42 (arm 1), 28 (arm 2), and 56 (arm 3) days (P = 0.01) (adjusted hazard ratio for arm 1 vs. arm 3, 1.41 [95% confidence interval (CI), 0.93-2.12; P = 0.10]; adjusted hazard ratio for arm 2 vs. arm 3, 1.89 [95% CI, 1.24-2.87; P = 0.003]). Eight-week liquid culture conversion was 79%, 89%, and 69%, respectively. Grade ≥3 adverse events occurred in 3 of 56 (5%), 5 of 53 (9%), and 2 of 56 (4%) participants. Six participants were withdrawn because of elevated transaminase concentrations (five in arm 2, one in arm 1). There were three serious adverse events (arm 2) and no deaths. Conclusions: Pretomanid enhanced the microbiologic activity of regimens containing a rifamycin and pyrazinamide. Efficacy and hepatic adverse events appeared highest with the pretomanid and rifabutin-containing regimen. Whether this is due to higher pretomanid concentrations merits exploration. Clinical trial registered with www.clinicaltrials.gov (NCT02256696).

DOI: 10.1164/rccm.202208-1475OC
PMID: 36455068 [Indexed for MEDLINE]

64. **QTc prolongation with bedaquiline treatment for drug-resistant pulmonary TB in a programmatic setting.**

Int J Tuberc Lung Dis. 2023 Apr 1;27(4):329-331. doi: 10.5588/ijtld.22.0406.

Sachdeva KS(1), Bhatnagar AK(2), Bhaskar A(3), Singla N(4), Sridhar R(5), Ramraj B(3), Athawale A(6), Solanki R(7), Baruah SR(8), Patel Y(9), Ramachandran R(10), Padmapriyadarsini C(3).

DOI: 10.5588/ijtld.22.0406
PMID: 37035973 [Indexed for MEDLINE]

65. **Pharmacokinetics and Efficacy of the Benzothiazinone BTZ-043 against Tuberculous Mycobacteria inside Granulomas in the Guinea Pig Model.**

Antimicrob Agents Chemother. 2023 Apr 18;67(4):e0143822. doi: 10.1128/aac.01438-22. Epub 2023 Mar 28.

Eckhardt E(1), Li Y(2), Mamerow S(3), Schinköthe J(4), Sehl-Ewert J(1), Dreisbach J(5)(6), Corleis B(1), Dorhoi A(1), Teifke J(1), Menge C(3), Kloss F(2), Bastian M(1).

Tuberculosis (TB), caused by Mycobacterium tuberculosis, is the world's leading cause of mortality from a single bacterial pathogen. With increasing frequency, emergence of drug-resistant mycobacteria leads to failures of standard TB treatment regimens. Therefore, new anti-TB drugs are urgently required. BTZ-043 belongs to a novel class of nitrobenzothiazinones, which inhibit mycobacterial cell wall formation by covalent binding of an essential cysteine in the catalytic pocket of decaprenylphosphoryl-β-d-ribose oxidase (DprE1). Thus, the compound blocks the formation of decaprenylphosphoryl-β-d-arabinose, a precursor for the synthesis of arabinans. An excellent in vitro efficacy against M. tuberculosis has been demonstrated. Guinea pigs are an important small-animal model to study anti-TB drugs, as they are naturally susceptible to M. tuberculosis and develop human-like granulomas after infection. In the current study, dose-finding experiments were conducted to establish the appropriate oral dose of BTZ-043 for the guinea pig. Subsequently, it could be shown that the active compound was present at high concentrations in Mycobacterium bovis BCG-induced granulomas. To evaluate its therapeutic effect, guinea pigs were subcutaneously infected with virulent M. tuberculosis and treated with BTZ-043 for 4 weeks. BTZ-043-treated guinea pigs had reduced and less necrotic granulomas than vehicle-treated controls. In comparison to the vehicle controls a highly significant reduction of the bacterial burden was observed after BTZ-043 treatment at the site of infection and in the draining lymph node and spleen. Together, these findings indicate that BTZ-043 holds great promise as a new antimycobacterial drug.

DOI: 10.1128/aac.01438-22
PMCID: PMC10112198
PMID: 36975792 [Indexed for MEDLINE]

Conflict of interest statement: The authors declare no conflict of interest.

66. **Bedaquiline exposure in people with drug-resistant TB treated for diabetes: analysis of two phase 2 trials.**

Int J Tuberc Lung Dis. 2023 Apr 1;27(4):335-337. doi: 10.5588/ijtld.22.0581.

Bolhuis MS(1), Akkerman OW(2), Sturkenboom MGG(1), van Boven JFM(1), Alffenaar JC(3), Stevens J(1).

DOI: 10.5588/ijtld.22.0581
PMID: 37035978 [Indexed for MEDLINE]

67. **Next-Generation Diarylquinolines Improve Sterilizing Activity of Regimens with Pretomanid and the Novel Oxazolidinone TBI-223 in a Mouse Tuberculosis Model.**

Antimicrob Agents Chemother. 2023 Apr 18;67(4):e0003523. doi: 10.1128/aac.00035-23. Epub 2023 Mar 15.

Li SY(1), Converse PJ(1), Betoudji F(1), Lee J(1), Mdluli K(2), Upton A(2)(3), Fotouhi N(2), Nuermberger EL(1).

A regimen comprised of bedaquiline (BDQ, or B), pretomanid, and linezolid (BPaL) is the first oral 6-month regimen approved by the U.S. Food and Drug Administration and recommended by the World Health Organization for the treatment of extensively drug-resistant tuberculosis. We used a well-established BALB/c mouse model of tuberculosis to evaluate the treatment-shortening potential of replacing bedaquiline with either of two new, more potent diarylquinolines, TBAJ-587 and TBAJ-876, in early clinical trials. We also evaluated the effect of replacing linezolid with a new oxazolidinone, TBI-223, exhibiting a larger safety margin with respect to mitochondrial toxicity in preclinical studies. Replacing bedaquiline with TBAJ-587 at the same 25-mg/kg dose significantly reduced the proportion of mice relapsing after 2 months of treatment, while replacing linezolid with TBI-223 at the same 100-mg/kg dose did not significantly change the proportion of mice relapsing. Replacing linezolid or TBI-223 with sutezolid in combination with TBAJ-587 and pretomanid significantly reduced the proportion of mice relapsing. In combination with pretomanid and TBI-223, TBAJ-876 at 6.25 mg/kg was equipotent to TBAJ-587 at 25 mg/kg. We conclude that replacement of bedaquiline with these more efficacious and potentially safer diarylquinolines and replacement of linezolid with potentially safer and at least as efficacious oxazolidinones in the clinically successful BPaL regimen may lead to superior regimens capable of treating both drug-susceptible and drug-resistant TB more effectively and safely.

DOI: 10.1128/aac.00035-23
PMCID: PMC10112056
PMID: 36920217 [Indexed for MEDLINE]

Conflict of interest statement: The authors declare a conflict of interest. Nader Fotouhi is a current employee of the TB Alliance. The remaining authors declare no conflict of interest.

68. **Structure-directed identification of pyridine-2-methylamine derivatives as MmpL3 inhibitors for use as antitubercular agents.**

Eur J Med Chem. 2023 Apr 21;255:115351. doi: 10.1016/j.ejmech.2023.115351. Online ahead of print.

Wen Y(1), Lun S(2), Jiao Y(1), Zhang W(1), Liu T(1), Yang F(3), Tang J(4), Bishai WR(5), Yu LF(6).

Mycobacterial membrane protein Large 3 (MmpL3), an inner membrane protein, plays a crucial role in the transport of mycolic acids that are essential for the viability of M. tuberculosis and has been a promising therapeutic target for new anti-TB agents. Herein, we report the discovery of pyridine-2-methylamine antitubercular compounds using a structure-based drug design strategy. Compound 62 stands out as the most potent compound with high activity against M. tb strain H37Rv (MIC = 0.016 μg/mL) as well as the clinically isolated strains of MDR/XDR-TB (MIC = 0.0039-0.0625 μg/mL), low Vero cell toxicity (IC50 ≥ 16 μg/mL), and moderate liver microsomal stability (CLint = 28 μL/min/mg). Furthermore, the resistant mutant of S288T due to single nucleotide polymorphism in mmpL3 was resistant to pyridine-2-methylamine 62, demonstrating compound 62 is likely target to MmpL3.

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PMID: 37116266

Conflict of interest statement: 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.

69. **Unraveling the possible inhibitors for Chorismate synthase to combat**

tuberculosis using in silico approach.

J Biomol Struct Dyn. 2023 Apr;41(7):2823-2830. doi: 10.1080/07391102.2022.2039298. Epub 2022 Feb 15.

Hanif M(1), Khan S(2), Farooq U(2), Nouroz F(1), Sarwar R(2).

Tuberculosis antibiotic resistance is a huge concern to the global population. The goal of this study was to find new and effective compounds to treat multidrug-resistant tuberculosis by targeting Chorismate synthase (CS), a crucial enzyme for Mycobacterium tuberculosis survival (MbT). The potential of a library of compounds as selective anti - tuberculosis drugs was investigated. Docking was first conducted using MoE to determine the effectiveness of the compounds. Molecular docking studies followed by MD simulation studies (total of 500 ns) in combination with free energy calculations grade the ligands in terms of their binding affinities. In the ligand bound state of the CS, MD simulations revealed a change from stretched to bended motional shift in loop L19. The RMSF analysis also revealed this flexibility, which was confirmed by visual inspection of L19 at various time intervals during the experiment. It appears that ZF1(-25.43Kcal/mol) and ZF2 (-22.04Kcal/mol) form hbonds and have a high binding energy in the active region of protein. Residues wise distribution of binding energy reveals that Arg144, Trp4, Thr6, and L19 amino acid residues are engaged in binding of CS with inhibitors. In summary, the findings suggest that compounds ZF1 and ZF2 may be more effective and selective anti-TB agents than currently available drugs. Also the role of L19, mediated by αH9 and αH5 in the retention of ligand inside the active pocket, through the formation of lid was also revealed. This knowledge will aid in the discovery of drugs that are potent CS inhibitors. More experimental research and a better understanding of the structure-activity relationship could aid in the development of possible candidates with better CS inhibition.Communicated by Ramaswamy H. Sarma.

DOI: 10.1080/07391102.2022.2039298
PMID: 35168481 [Indexed for MEDLINE]

70. **Oral Linezolid Induced Early Onset Hepatic Encephalopathy- A Case Report of 65-year Old Diabetic Female.**

Curr Drug Saf. 2023 Apr 17. doi: 10.2174/1574886318666230417113910. Online ahead of print.

Upadhyay M(1), Purohit B(2), Pargi P(2).

INTRODUCTION/BACKGROUND: Linezolid is increasingly utilized to treat

gram-positive bacteria that are resistant to other antibiotics like vancomycin-resistant Staphylococcus aureus, methicillin-resistant Staphylococcus aureus as well as drug-resistant tuberculosis. It acts by inhibiting protein synthesis in bacteria. Although it is a relatively safe medicine, many reports of hepatotoxicity and neurotoxicity linked to long-term usage have been received but patients with pre-existing risk factors, such as diabetes and alcoholism, may have toxicity even after short-term use of linezolid.
CASE PRESENTATION: Here we are presenting a case of a 65-year-old female with diabetes who developed hepatic encephalopathy after one week of treatment with linezolid prescribed for nonhealing diabetic ulcer after a culture sensitivity test. After the use of linezolid 600mg BD for 8 days patient developed altered sensorium and breathlessness and had high bilirubin, SGOT, and SGPT. She was diagnosed with hepatic encephalopathy. Linezolid was withdrawn and after 10 days all laboratory parameters for liver function test were improved.
CONCLUSION: Care should be taken when prescribing linezolid in such patients with pre-existing risk factors as they are prone to develop hepatotoxic and neurotoxic adverse effects even after short-term use of linezolid.

DOI: 10.2174/1574886318666230417113910
PMID: 37070438

71. **Water network chemistry to exploit the nature of catalytic water molecules in Mtb DNA gyrase: a computational study to understand the binding mechanism of fluoroquinolones.**

J Biomol Struct Dyn. 2023 Apr 25:1-9. doi: 10.1080/07391102.2023.2199869. Online ahead of print.

Kumar GS(1), Sobhia ME(1).

The dynamics of DNA gyrase and mutants of DNA gyrA such as G88A, A90V, S91P, D94A, D94G, D94N, D94Y; and double-point mutant (S91P-D94G), are meticulously investigated using computational approaches. Molecular dynamics (MD) and hydration thermodynamics have shed light on the fundamental, mechanistic basis of mutations on the conformational stability of Quinolone Binding Pocket (QBP) of DNA gyrase. Analysis of MD results revealed the displacement of a single crystal water molecule (HOH201) from the catalytic site of wild-type (WT) and mutants of DNA gyrA. This prompted our research group to probe the five crystal water molecules present in the QBP of the enzyme using water thermodynamics. Hydration thermodynamics analysis revealed the displacement of HOH201 due to

unstable thermodynamic signatures. Further, the analysis highlighted significant changes in thermodynamic signatures and locations of five crystal water hydration sites upon mutation. Integrated MD simulations and water thermodynamics provided promising insights into the conformational changes and inaccessibility of the catalytic water molecule that can influence the design of DNA gyrase inhibitors.Communicated by Ramaswamy H. Sarma.

DOI: 10.1080/07391102.2023.2199869
PMID: 37121993