May Literature

1. Multidrug-resistant tuberculosis during pregnancy and adverse birth outcomes: a systematic review and meta-analysis.

BJOG. 2021 Jun;128(7):1125-1133. doi: 10.1111/1471-0528.16573. Epub 2020 Nov 23.

Alene KA(1)(2)(3), Jegnie A(4), Adane AA(3)(5).

BACKGROUND: Multidrug-resistant tuberculosis (MDR-TB) is a major global public health concern. However, there is a dearth of literature on whether MDR-TB and its medications impact maternal and perinatal outcomes, and when such evidence exists the findings are conflicting.
OBJECTIVES: This systematic review and meta-analysis aimed to examine the impact of MDR-TB and its medications during pregnancy on maternal and perinatal outcomes.
SEARCH STRATEGY: PubMed, Scopus and Web of Science databases were searched from earliest to February 2020.
SELECTION CRITERIA: Records were screened based on pre-defined selection criteria and assessed for quality by two independent reviewers.
DATA COLLECTION AND ANALYSIS: A meta-analysis was performed using the random effects model to calculate pooled prevalence for each outcome.
MAIN RESULTS: Of the 72 records identified, 12 were included in the systematic review and meta-analysis, consisting of 174 pregnant women with MDR-TB and 110 adverse outcomes. Maternal death, pregnancy loss, preterm birth and low birthweight were the most common maternal and perinatal adverse outcomes reported in the studies. The overall pooled prevalence was 7.5% (95% CI 3.2-12.8) for maternal death, 10.6% (95% CI 6.0-16.3) for pregnancy loss, 12.9% (95% CI 0.0-38.0) for preterm birth and 23.7% (95% CI 17.0-31.0) for low birthweight.
CONCLUSIONS: The findings suggest that MDR-TB is associated with a high risk of adverse maternal and perinatal outcomes, but these should be interpreted cautiously because the evidence is largely preliminary. Adequately powered prospective cohort studies are urgently required to corroborate these findings.
TWEETABLE ABSTRACT: Multidrug-resistant tuberculosis may increase the risk of adverse maternal and perinatal outcomes.

DOI: 10.1111/1471-0528.16573
PMID: 33068306

2. Whole genome sequencing analysis of multidrug-resistant tuberculosis in Singapore, 2006-2018.

Eur J Clin Microbiol Infect Dis. 2021 May;40(5):1079-1083. doi: 10.1007/s10096-020-04100-6. Epub 2020 Nov 15.

Chee CBE(1), Lim LKY(2), Ong RTH(3), Sng LH(4), Hsu LY(3), Lee VJM(5), Wang YT(2).

There were 290 multidrug-resistant (MDR)-TB cases diagnosed in Singapore from 2006 to 2018. Eighty-one percent were foreign-born. Spoligotyping and MIRU-VNTR methods identified 108 patients in 24 clusters. The Beijing spoligotype accounted for 22 clusters. Whole genome sequencing (WGS) analysis reduced the number of clustered patients and clusters to 43 and nine respectively. One MIRU cluster was redefined into three WGS clusters. All the clusters had foreign-born source cases. Forty percent of local-born, versus 9% of foreign-born, MDR-TB cases belonged to WGS clusters. WGS more accurately elucidated potential MDR-TB transmission which was overestimated by conventional genotyping methods in Singapore.

DOI: 10.1007/s10096-020-04100-6
PMID: 33190171

3. Spectrum of Bacterial Colonization in Patients Hospitalized for Treatment of Multidrug-Resistant Tuberculosis.

Microb Drug Resist. 2021 May;27(5):691-697. doi: 10.1089/mdr.2020.0073. Epub 2020 Oct 19.

Annear D(1), Gaida R(2), Myburg K(1), Black J(2)(3), Truter I(2), Bamford C(4)(5), Govender S(1).

This study investigated the bacterial colonization in patients admitted for treatment of drug-resistant tuberculosis in a specialized TB hospital. Identification and antimicrobial susceptibility testing of bacterial isolates (n = 62) from nasal, groin, and rectal swabs [patient cohort (n = 37)] were determined by the VITEK-MS system. Resistance gene analysis was by PCR and DNA sequencing. Molecular typing of Klebsiella pneumoniae isolates was by Multilocus Sequencing Typing (MLST). Patients (n = 13/37; 35%) were colonized by multidrug-resistant (MDR) bacteria (ESBL and MRSA) on admission. Of the 24 patients who were not colonized by MDR bacteria on admission, 46% (17/37) became colonized by MDR bacteria within 1 month of admission, mostly with ESBL-producing Enterobacteriales and resistance to aminoglycosides and

fluoroquinolones. ESBL Escherichia coli (41/62; 66%) and K. pneumoniae (14/62; 23%) predominated. Genes encoding for ESBLs (blaCTX-M-14, blaCTX-M-15, blaSHV-28, blaOXA-1, and blaOXY-2) and plasmid-mediated quinolone resistant genes (qnrB1, qnrB4, and qnrB10) were detected. MLST revealed genetic diversity among the K. pneumoniae isolates from hospitalized patients. This study provides insight into bacterial pathogen colonization in hospitalized TB patients with the first occurrence of the qnrB4 and qnrB10 genes and co-expression of genes: qnrB4+aac(6')-lb-cr, qnrB10+aac(6')-lb-cr, qnrB4+qnrS1, and qnrB10+qnrS1 in fluoroquinolone-resistant E. coli isolates within South Africa. However, the source and colonization routes of these isolates could not be determined.

DOI: 10.1089/mdr.2020.0073
PMID: 33074767

4. Emergence of Specific gyrA Mutations Associated High-Level Fluoroquinolone-Resistant Mycobacterium tuberculosis among Multidrug-Resistant Tuberculosis Cases in North India.

Microb Drug Resist. 2021 May;27(5):647-651. doi: 10.1089/mdr.2020.0240. Epub 2020 Sep 29.

Singh PK(1), Singh U(1), Jain A(1).

Aim: This study aims to determine the frequency and pattern of gyrA/B mutations in multidrug-resistant (MDR) Mycobacterium tuberculosis (MTB) strains and also to assess the association between different gyrA/B mutations with phenotypic resistance to moxifloxacin (MOX) at clinical breakpoint (CB) drug concentration. Method: A total of 106 clinical MTB isolates carrying gyrA/B mutations were included consecutively. Culture-based MOX susceptibility was tested at CB (1.0 µg/mL) followed by its correlation with gyrA/B mutations using Genotype MTBDRsl assay. The mutations associated with phenotypic resistance were further analyzed on a large dataset of 1,825 MDR tuberculosis (TB) patients. Result: D94G and A90V mutations within gyrA were significantly associated with resistance and susceptible phenotype ($p < 0.001$), respectively. Of 1,825 MDR patients, gyrA/B mutations were found in 58.8% cases, of which fluoroquinolone (FQ) resistance was concluded among 97.9%, 0.8%, and 1.3% patients due to mutation in gyrA, gyrB, and in both the genes, respectively. D94G alone (45.9%) followed by A90V (21.2%) mutations in gyrA gene was most frequent. Conclusion: Our study showed that MDR-TB has emerged in northern India with additional FQ resistance. Different selection pressure and transmission may result in prevailing accumulation of specific gyrA mutations causing high-level FQ resistance, therefore, current control measures need to be strengthened.

DOI: 10.1089/mdr.2020.0240
PMID: 32991238

5. Anti-tuberculosis site-specific oral delivery system that enhances rifampicin bioavailability in a fixed-dose combination with isoniazid.

Drug Deliv Transl Res. 2021 Jun;11(3):894-908. doi: 10.1007/s13346-020-00847-9. Epub 2020 Sep 8.

Luciani-Giacobbe LC(1), Lorenzutti AM(2), Litterio NJ(2), Ramírez-Rigo MV(3)(4), Olivera ME(5).

The in vivo release segregation of rifampicin (RIF) and isoniazid (INH) has been proposed as a strategy to avoid RIF acid degradation, which is known as one of the main factors for reduced RIF bioavailability and can result in drug-resistant tuberculosis. So far, this strategy has been scarcely explored. The aims of this study were to investigate the stability and bioavailability of RIF after combination of a very fast release matrix of RIF with a sustained delivery system of INH. A series of INH-alginic acid complexes (AA-INH) was obtained and characterized. Independent and sequential release profile of AA-INH at biorrelevant media of pH 1.20 and 6.80 was explored. In addition, AA-INH was combined with a RIF-carboxymethylcellulose very fast release complex (CMC-RIF) obtained previously and subjected to acid dissolution assays to evaluate RIF acid stability and determine RIF and INH dissolution efficiencies. Finally, a pharmacokinetic study in dogs was carried out. The AA-INH was easily obtained in solid-state. Their characterization revealed its ionic nature, with a loading capacity of around 30%. The dissolution efficiencies (15 min) confirmed release segregation in acid media with 7.8 and 65.6% for AA-INH and CMC-RIF, respectively. INH release rate from the AA-INH system was slow in acid media and increased in simulated intestinal media. The complete release of INH was achieved after 2 h in simulated intestinal media in the sequential release experiments. The acid degradation of RIF was significantly reduced (36.7%) when both systems were combined and oral administration to dogs revealed a 42% increase in RIF bioavailability. In conclusion, CMC-RIF and AA-INH may be useful for the formulation of a site-specific solid dosage form to overcome some of the main obstacles in tuberculosis treatment. Graphical abstract.

DOI: 10.1007/s13346-020-00847-9
PMID: 32901368

6. Treating Drug-resistant Tuberculosis Infection: No More Excuses.

Clin Infect Dis. 2021 May 18;72(10):1716-1718. doi: 10.1093/cid/ciaa328.

Reuter A(1), Furin J(2).

Comment on
Clin Infect Dis. 2021 May 18;72(10):1709-1715.

DOI: 10.1093/cid/ciaa328
PMID: 32266941

7. Therapeutic strategies and outcomes of MDR and pre-XDR-TB in Italy: a nationwide study.

Int J Tuberc Lung Dis. 2021 May 1;25(5):395-399. doi: 10.5588/ijtld.21.0036.

Riccardi N(1), Saderi L(2), Borroni E(3), Tagliani E(3), Cirillo DM(4), Marchese V(5), Matteelli A(5), Piana A(6), Castellotti P(7), Ferrarese M(7), Gualano G(8), Palmieri F(9), Girardi E(10), Codecasa L(7), Sotgiu G(2).

BACKGROUND: Treatment outcomes in multidrug-resistant TB (MDR-TB) patients are suboptimal in several low-incidence countries.METHODS: The primary outcome measure was the proportion of successfully treated patients in Italy during an 18-year period. Secondary outcomes were treatment outcomes in certain drug-containing regimens and the possibility for the WHO shorter MDR-TB regimen.RESULTS: In the 191 patients included (median age at admission: 33 years; 67.5% male, following drug-resistance patterns were found: MDR-TB in 68.6%, pre-extensively drug-resistant TB (pre-XDR-TB) in 30.4% and XDR-TB in 1.1% patients. The most frequently prescribed drugs were fluoroquinolones in 84.6% cases, amikacin in 48.7%, linezolid in 34.6% and meropenem/clavulanic acid in 29.5%. The median duration of treatment was 18 months. Treatment success was achieved in 71.2% patients, of whom, 44% were cured and 27.2% completed treatment. Treatment success rates did not statistically differ between the MDR- (68.8%) and pre-XDR-TB (77.6%) groups (P = 0.26). Treatment success rates had large variability between North and South of Italy (81.3% vs. 53.3%). Only 22.5% of the cases would have been eligible for shorter MDR-TB regimensCONCLUSION: Our study highlights variability in treatment outcomes in MDR- and pre-XDR-TB patients. Study findings confirmed the potential utility of linezolid and, for patients with limited oral options, meropenem/clavulanic acid and amikacin.

DOI: 10.5588/ijtld.21.0036
PMID: 33977908

8. Recent Progress and Challenges for Drug-Resistant Tuberculosis Treatment.

Pharmaceutics. 2021 Apr 21;13(5):592. doi: 10.3390/pharmaceutics13050592.

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Control of Mycobacterium tuberculosis infection continues to be an issue, particularly in countries with a high tuberculosis (TB) burden in the tropical and sub-tropical regions. The effort to reduce the catastrophic cost of TB with the WHO's End TB Strategy in 2035 is still obstructed by the emergence of drug-resistant TB (DR-TB) cases as result of various mutations of the MTB strain. In the approach to combat DR-TB, several potential antitubercular agents were discovered as inhibitors for various existing and novel targets. Host-directed therapy and immunotherapy also gained attention as the drug-susceptibility level of the pathogen can be reduced due to the pathogen's evolutionary dynamics. This review is focused on the current progress and challenges in DR-TB treatment. We briefly summarized antitubercular compounds that are under development and trials for both DR-TB drug candidates and host-directed therapy. We also highlighted several problems in DR-TB diagnosis, the treatment regimen, and drug discovery that have an impact on treatment adherence and treatment failure.

DOI: 10.3390/pharmaceutics13050592
PMID: 33919204

9. Multidrug-resistant tuberculosis imported into low-incidence countries-a GeoSentinel analysis, 2008-2020.

J Travel Med. 2021 May 12:taab069. doi: 10.1093/jtm/taab069. Online ahead of print.

Eimer J(1), Patimeteeporn C(2), Jensenius M(3), Gkrania-Klotsas E(4), Duvignaud A(5), Barnett ED(6), Hochberg NS(7), Chen LH(8), Trigo-Esteban E(9), Gertler M(10), Greenaway C(11), Grobusch MP(12)(13), Angelo KM(2), Hamer DH(7)(14), Caumes E(15)(16), Asgeirsson H(1)(17).

BACKGROUND: Early detection of imported multidrug-resistant tuberculosis (MDR-TB) is crucial, but knowledge gaps remain about migration- and travel-associated MDR-TB epidemiology. The aim was to describe epidemiologic characteristics among international travelers and migrants with MDR-TB. METHODS: Clinician-determined and microbiologically confirmed MDR-TB diagnoses deemed to be related to travel or migration were extracted from GeoSentinel, a global surveillance network of travel and tropical medicine clinics, from

January 2008 through December 2020. MDR-TB was defined as resistance to both isoniazid and rifampicin. Additional resistance to either a fluoroquinolone or a second-line injectable drug was categorized as pre-extensively drug-resistant (pre-XDR) TB, and as extensively drug-resistant (XDR) TB when resistance was detected for both. Sub-analyses were performed based on degree of resistance and country of origin.
RESULTS: Of 201 patients, 136 had MDR-TB (67.7%), 25 had XDR-TB (12.4%), 23 had pre-XDR TB (11.4%), and 17 had unspecified MDR- or XDR-TB (8.5%); 196 (97.5%) were immigrants, of which 92 (45.8%) originated from the former Soviet Union. The median interval from arrival to presentation was 154 days (interquartile range [IQR]: 10-751 days); 34.3% of patients presented within 1 month after immigration, 30.9% between 1 and 12 months, and 34.9% after ≥1 year. Pre-XDR- and XDR-TB patients from the former Soviet Union other than Georgia presented earlier than those with MDR-TB (26 days [IQR: 8-522] vs. 369 days [IQR: 84-827]) while patients from Georgia presented very early, irrespectively of the level of resistance (8 days [IQR: 2-18] vs. 2 days [IQR: 1-17]).
CONCLUSIONS: MDR-TB is uncommon in traditional travellers. Purposeful medical migration may partly explain differences in time to presentation among different groups. Public health resources are needed to better understand factors contributing to cross-border MDR-TB spread and to develop strategies to optimize care of TB-infected patients in their home countries before migration.

DOI: 10.1093/jtm/taab069
PMID: 33987682

10. Treatment Outcomes Among Pediatric Patients With Highly Drug-Resistant Tuberculosis: The Role of New and Repurposed Second-Line Tuberculosis Drugs.

J Pediatric Infect Dis Soc. 2021 Apr 30;10(4):457-467. doi: 10.1093/jpids/piaa139.

Madzgharashvili T(1), Salindri AD(2), Magee MJ(3)(4), Tukvadze N(1), Avaliani Z(1), Blumberg HM(3)(4)(5), Kempker RR(5), Lomtadze N(1).

BACKGROUND: Among pediatric patients with multidrug-resistant tuberculosis (MDR-TB), limited data exist regarding treatment outcomes in the context of the new and repurposed second-line TB drugs (SLDs). We aimed to describe the treatment outcomes among pediatric MDR-TB patients receiving new and repurposed SLDs including the proportion who achieved favorable outcomes.

METHODS: We conducted a retrospective cohort study among pediatric patients (age ≤18 years) treated for MDR-TB in the country of Georgia from 2009 to 2016. A "new and repurposed" SLD regimen was defined as a regimen that included linezolid, bedaquiline, and/or delamanid. Favorable treatment outcome was defined by treatment completion or documented microbial "cure" status at the end of treatment. We assessed the association between the use of the new and repurposed SLDs with MDR-TB treatment outcomes using bivariate analyses and log-binomial regression.
RESULTS: There were 124 pediatric MDR-TB patients (median age: 13.7; interquartile range: 4.6-16.0) initiating treatment; 119 (96.0%) had a treatment outcome recorded and were included in our analyses. Eighteen (15.1%) patients received new and repurposed SLDs from 2015 or later. After adjusting for potential confounders, the proportion achieving favorable MDR-TB treatment outcomes was higher among patients treated with SLD regimens that included new and/or repurposed drugs when compared with those treated without (adjusted risk ratio: 1.17; 95% confidence interval: 0.51-2.72).
CONCLUSIONS: We observed a high proportion of favorable treatment outcomes among pediatric patients with MDR-TB receiving the new and repurposed SLDs. Further studies to evaluate the efficacy and children's tolerability of the new and repurposed SLDs are still warranted.

DOI: 10.1093/jpids/piaa139
PMCID: PMC8087132
PMID: 33347564

11. Multi-drug resistant tuberculosis, ten years later.

Med Clin (Barc). 2021 Apr 23;156(8):393-401. doi: 10.1016/j.medcli.2020.08.018. Epub 2021 Jan 31.

[Article in English, Spanish]

Caminero Luna JA(1), Pérez Mendoza G(2), Rodríguez de Castro F(2).

Drug-resistant tuberculosis, especially those with resistance to rifampicin (RR-TB), has become one of the main obstacles to achieving the dream of eradicating tuberculosis. Furthermore, it is necessary to combine three or four different drugs in the attempt to cure TB, however, unfortunately, there are few available that can be considered genuinely effective. Fortunately, the notable

worldwide increase in RR-TB in recent years has led to the investment of resources in the development of new drugs for TB, and other drugs investigated for other diseases have been successfully tested on TB. This has resulted in a clear change in the clinical management of these patients over the last 3-4 years, and it is now easier to design therapeutic regimens and achieve higher success rates. All these changes are updated in this review.

DOI: 10.1016/j.medcli.2020.08.018
PMID: 33531151 [Indexed for MEDLINE]

12. Treatment and outcomes of Multidrug-resistant tuberculosis in Auckland 1995-2018.

Intern Med J. 2021 May 7. doi: 10.1111/imj.15341. Online ahead of print.

Cutfield T(1)(2), Mowlem L(2), Paynter J(2), Christmas T(2), Harrison A(2), Lewis C(2), Newton S(3), Nisbet M(1)(2).

INTRODUCTION: New Zealand has a low burden of tuberculosis, however multidrug-resistant tuberculosis (MDR-TB) still represents a challenge for clinicians. This is the first description of clinical aspects of MDR-TB in New Zealand.
METHODS: Clinical data were obtained for patients treated for MDR-TB at Auckland District Health Board (ADHB).
RESULTS: There were 60 patients nationally with MDR-TB between 1989 and 2018; 41/60 (69%) received care at ADHB. Pulmonary infection was present in 36/41 (88%) of patients, with 19/41 (46%) with smear-positive sputum (smear 1-2+ in 6/41, 15%; smear 3-4+ in 13/41, 32%). The median duration of treatment was 22 months (range 7.5-26) for 18/41 (44%) patients who completed MDR-TB treatment by August 2018. The median duration of amikacin treatment was 6 months (range 2-12) for the 23/38 (61%) in whom these data were available. All 38 patients who received treatment for MDR-TB experienced adverse effects, most commonly gastrointestinal (66%), neurologic (50%), ototoxicity (47%) and psychiatric (37%). Complications of intravenous access were experienced by 10/37 (27%) patients. Of the 19/41 (46%) patients who completed treatment, 18/19 (95%) achieved cure. There was one case who had recurrence who had inadequate treatment, and one case who had spontaneous resolution without treatment. Seventeen (41%) patients left Auckland prior to completion of treatment, mostly to return to their country of origin (15/17, 88%).
CONCLUSION: MDR-TB is uncommon in New Zealand. Treatment is frequently associated with adverse events, however rates of cure for people completing

treatment in New Zealand are high.

DOI: 10.1111/imj.15341
PMID: 33961727

13. Compassionate use of delamanid in adults and children for drug-resistant tuberculosis: 5-year update.

Eur Respir J. 2021 May 20;57(5):2002483. doi: 10.1183/13993003.02483-2020. Print 2021 May.

Ghosh S(1), Breitscheidel L(2), Lazarevic N(1), Martin A(1), Hafkin J(3), Hittel N(1).

BACKGROUND: Although delamanid has been approved for the treatment of multidrug-resistant TB (MDR-TB) in numerous regions, in areas where it is not yet registered it can be accessed as part of salvage therapy (in particular for those patients with limited treatment options) via the Otsuka compassionate use programme. Here we present the analysis of interim treatment outcomes by 24 weeks of more than 200 MDR-TB patients globally who received delamanid under this programme.
METHODS: We evaluated treatment efficacy with respect to culture negativity at 24 weeks, as well as the safety profile of delamanid, in an MDR-TB patient cohort treated under compassionate use between 2014 and 2019.
RESULTS: Among patients who received delamanid as part of a multidrug regimen, 123 (61%) out of 202 had extensively drug-resistant TB (XDR-TB), 66 (33%) out of 202 had HIV co-infection and 34 (17%) out of 202 were children aged between 6 and 17 years. Of those patients who were culture positive at delamanid treatment initiation and who completed 24 weeks of delamanid treatment in combination with other anti-tuberculosis (TB) drugs, culture negativity was achieved in 116 (79%) out of 147 cases. The corresponding rates of culture negativity for patients with XDR-TB and HIV co-infection, as well as the paediatric subgroup were 69 (77%) out of 90, 44 (92%) out of 48 and 20 (80%) out of 25, respectively. QT interval prolongation was the most frequently observed serious adverse event and was reported in 8% of patients receiving delamanid. Overall, treatment safety outcomes did not reveal any new or unidentified risks.
CONCLUSIONS: The use of delamanid combined with other active drugs has the potential to achieve high rates of culture negativity in difficult-to-treat drug-resistant TB cases, with a favourable safety profile.

DOI: 10.1183/13993003.02483-2020
PMID: 33243846

14. Emergence of additional drug resistance during treatment of multidrug-resistant tuberculosis in China: a prospective cohort study.

Clin Microbiol Infect. 2021 Apr 23:S1198-743X(21)00169-5. doi: 10.1016/j.cmi.2021.04.001. Online ahead of print.

Hu Y(1), Zheng X(1), Davies Forsman L(2), Ning Z(3), Chen C(4), Gao Y(1), Zhang Z(3), Lu W(4), Werngren J(5), Bruchfeld J(2), Hoffner S(6), Xu B(7).

OBJECTIVES: Little is known about how additional second-line drug resistance emerges during multidrug-resistant tuberculosis (MDR-TB) treatment. The present study aimed to investigate the influence of microevolution, exogenous reinfection and mixed infection on second-line drug resistance during the recommended 2-year MDR-TB treatment.
METHODS: Individuals with MDR-TB were enrolled between 2013 and 2016 in a multicentre prospective observational cohort study and were followed up for 2 years until treatment completion. Whole-genome sequencing (WGS) was applied for serial Mycobacterium tuberculosis isolates from study participants throughout the treatment, to study the role of microevolution, exogenous reinfection and mixed infection in the development of second-line drug resistance.
RESULTS: Of the 286 enrolled patients with MDR-TB, 63 (22.0%) M. tuberculosis isolates developed additional drug resistance during the MDR-TB treatment, including 5 that fulfilled the criteria of extensively drug-resistant TB. By comparing WGS data of serial isolates retrieved from the patients throughout treatment, 41 (65.1%) of the cases of additional second-line drug resistance were the result of exogenous reinfection, 18 (28.6%) were caused by acquired drug resistance, i.e. microevolution, while the remaining 4 (6.3%) were caused by mixed infections with drug-resistant and drug-susceptible strains. In multivariate analysis, previous TB treatment (adjusted hazard ratio (aHR) 2.51, 95% CI 1.51-4.18), extensive disease on chest X-ray (aHR 3.39, 95% CI 2.03-5.66) and type 2 diabetes mellitus (aHR 4.00, 95% CI 2.22-7.21) were independent risk factors associated with the development of additional second-line drug resistance.
CONCLUSIONS: A large proportion of additional second-line drug resistance emerging during MDR-TB treatment was attributed to exogenous reinfection, indicating the urgency of infection control in health facilities as well as the need for repeated drug susceptibility testing throughout MDR-TB treatment.

DOI: 10.1016/j.cmi.2021.04.001
PMID: 33895338

15. Trends, Characteristics and Treatment Outcomes of Patients with Drug-Resistant Tuberculosis in Uzbekistan: 2013-2018.

Int J Environ Res Public Health. 2021 Apr 27;18(9):4663. doi: 10.3390/ijerph18094663.

Safaev K(1), Parpieva N(1), Liverko I(1), Yuldashev S(1), Dumchev K(2), Gadoev J(3), Korotych O(4), Harries AD(5)(6).

Uzbekistan has a high burden of drug-resistant tuberculosis (TB). Although conventional treatment for multidrug-resistant TB (MDR-TB) and extensively drug-resistant TB (XDR-TB) has been available since 2013, there has been no systematic documentation about its use and effectiveness. We therefore documented at national level the trends, characteristics, and outcomes of patients with drug-resistant TB enrolled for treatment from 2013-2018 and assessed risk factors for unfavorable treatment outcomes (death, failure, loss to follow-up, treatment continuation, change to XDR-TB regimen) in patients treated in Tashkent city from 2016-2017. This was a cohort study using secondary aggregate and individual patient data. Between 2013 and 2018, MDR-TB numbers were stable between 2347 and 2653 per annum, while XDR-TB numbers increased from 33 to 433 per annum. At national level, treatment success (cured and treatment completed) for MDR-TB decreased annually from 63% to 57%, while treatment success for XDR-TB increased annually from 24% to 57%. On multivariable analysis, risk factors for unfavorable outcomes, death, and loss to follow-up in drug-resistant TB patients treated in Tashkent city included XDR-TB, male sex, increasing age, previous TB treatment, alcohol abuse, and associated comorbidities (cardiovascular and liver disease, diabetes, and HIV/AIDS). Reasons for these findings and programmatic implications are discussed.

DOI: 10.3390/ijerph18094663
PMCID: PMC8124452
PMID: 33925705

16. Design of multidrug-resistant tuberculosis treatment regimens based on DNA sequencing.

Clin Infect Dis. 2021 Apr 26:ciab359. doi: 10.1093/cid/ciab359. Online ahead of print.

Grobbel HP(1)(2)(3), Merker M(2)(4), Köhler N(1)(2)(3), Andres S(5), Hoffmann H(6)(7), Heyckendorf J(1)(2)(3), Reimann M(1)(2)(3), Barilar I(4), Dreyer V(4), Hillemann D(5), Kalsdorf B(1)(2)(3), Kohl TA(4), Sanchez-Carballo P(1)(2)(3), Schaub D(1)(2)(3), Todt K(6)(7), Utpatel C(4), Maurer FP(5)(8), Lange C(1)(2)(3)(9), Niemann S(2)(4)(5).

BACKGROUND: Comprehensive and reliable drug susceptibility testing (DST) is urgently needed to provide adequate treatment regimens for patients with multidrug-resistant/rifampicin-resistant tuberculosis (MDR/RR-TB). We investigated if next generation sequencing (NGS) analysis of Mycobacterium tuberculosis complex isolates and genes implicated in drug resistance can guide the design of effective MDR/RR-TB treatment regimens.
METHODS: NGS-based genomic DST predictions of M. tuberculosis complex isolates from MDR/RR-TB patients admitted to a TB reference center in Germany between 01/01/2015 and 04/30/2019 were compared with phenotypic DST results of Mycobacteria growth indicator tubes (MGIT). Standardized treatment algorithms were applied to design individualized therapies based on either genomic or phenotypic DST results, and discrepancies were further evaluated by determination of minimum inhibitory drug concentrations (MIC) using Sensititre MYCOTBI and UKMYC microtiter plates.
RESULTS: In 70 patients with MDR/RR-TB, agreement among 1048 pairwise comparisons of genomic and phenotypic DST was 86.3%; 76 (7.2%) results were discordant, and 68 (6.5%) could not be evaluated due to presence of polymorphisms with yet unknown implications for drug resistance. Importantly, 549/561 (97.9%) predictions of drug susceptibility were phenotypically confirmed in MGIT, and 27/64 (42.2%) false positive results were linked to previously described mutations mediating a low or moderate MIC increase. Virtually all drugs (99.0%) used in combination therapies that were inferred from genomic DST, were confirmed to be susceptible by pDST.
CONCLUSIONS: NGS-based genomic DST can reliably guide the design of effective MDR/RR-TB treatment regimens.

DOI: 10.1093/cid/ciab359
PMID: 33900387

17. Strategies to Combat Multi-Drug Resistance in Tuberculosis.

Acc Chem Res. 2021 May 18;54(10):2361-2376. doi: 10.1021/acs.accounts.0c00878. Epub 2021 Apr 22.

Singh V(1)(2), Chibale K(1)(2).

Conspectus"Drug resistance is an unavoidable consequence of the use of drugs; however, the emergence of multi-drug resistance can be managed by accurate diagnosis and tailor-made regimens."Antimicrobial resistance (AMR), is one of the most paramount health perils that has emerged in the 21st century. The global increase in drug-resistant strains of various bacterial pathogens prompted the World Health Organization (WHO) to develop a priority list of AMR pathogens. Mycobacterium tuberculosis (Mtb), an acid-fast bacillus that causes tuberculosis (TB), merits being one of the highest priority pathogens on this list since drug-resistant TB (DR-TB) accounts for ∼29% of deaths attributable to AMR. In recent years, funded collaborative efforts of researchers from academia, not-for-profit virtual R&D organizations and industry have resulted in the continuous growth of the TB drug discovery and development pipeline. This has so far led to the accelerated regulatory approval of bedaquiline and delamanid for the treatment of DR-TB. However, despite the availability of drug regimes, the current cure rate for multi-drug-resistant TB (MDR-TB) and extensively drug-resistant TB (XDR-TB) treatment regimens is 50% and 30%, respectively. It is to be noted that these regimens are administered over a long duration and have a serious side effect profile. Coupled with poor patient adherence, this has led to further acquisition of drug resistance and treatment failure. There is therefore an urgent need to develop new TB drugs with novel mechanism of actions (MoAs) and associated regimens.This Account recapitulates drug resistance in TB, existing challenges in addressing DR-TB, new drugs and regimens in development, and potential ways to treat DR-TB. We highlight our research aimed at identifying novel small molecule leads and associated targets against TB toward contributing to the global TB drug discovery and development pipeline. Our work mainly involves screening of various small molecule chemical libraries in phenotypic whole-cell based assays to identify hits for medicinal chemistry optimization, with attendant deconvolution of the MoA. We discuss the identification of small molecule chemotypes active against Mtb and subsequent structure-activity relationships (SAR) and MoA deconvolution studies. This is followed by a discussion on a chemical series identified by whole-cell cross-screening against Mtb, for which MoA deconvolution studies revealed a pathway that explained the lack of in vivo efficacy in a mouse model of TB and reiterated the importance of selecting an appropriate growth medium during phenotypic screening. We also discuss our efforts on drug repositioning toward addressing DR-TB. In the concluding section, we preview some promising future directions and the challenges inherent in advancing the drug pipeline to address

DR-TB.

DOI: 10.1021/acs.accounts.0c00878
PMID: 33886255

18. Genetic variability in multidrug-resistant Mycobacterium tuberculosis isolates from patients with pulmonary tuberculosis in North India.

BMC Microbiol. 2021 Apr 21;21(1):123. doi: 10.1186/s12866-021-02174-6.

Singh AV(1), Singh S(2), Yadav A(2), Kushwah S(2), Yadav R(2), Sai DK(2), Chauhan DS(2).

BACKGROUND: Information on the genetic variability of drug resistant isolates of Mycobacterium tuberculosis is of paramount importance to understand transmission dynamics of disease and to improve TB control strategies. Despite of largest number of multidrug-resistant (MDR) tuberculosis cases (1, 30,000; 27% of the global burden), strains responsible for the expansion or development of drug-resistant Mycobacterium tuberculosis infections have been poorly characterized in India. Present study was aimed to investigate the genetic diversity in MDR isolates of Mycobacterium tuberculosis in North India.
RESULTS: Spacer oligonucleotide typing (spoligotyping) was performed on 293 clinical MDR isolates of Mycobacterium tuberculosis recovered from cases of pulmonary tuberculosis from North India. Spoligotyping identified 74 distinct spoligotype patterns. Comparison with an international spoligotype database (spoldb4 database) showed that 240 (81.91%) and 32 (10.92%) strains displayed known and shared type patterns, while 21 (7.16%) strains displayed unique spoligotype patterns. Among the phylogeographic lineages, lineage 3 (East African-Indian) was found most predominant lineage (n = 159, 66.25%), followed by lineage 2 (East Asian; n = 34, 14.16%), lineage 1 (Indo-Oceanic; n = 30, 12.50%) and lineage 4 (Euro American; n = 17, 7.08%). Overall, CAS1_DEL (60.41%; SITs 2585, 26, 2694, 309, 381, 428, 1401, 141, 25, 1327) was found most pre-dominant spoligotype pattern followed by Beijing (14.16%; SITs255, 260, 1941, 269) and EAI3_IND (5.00%; SITs 298, 338, 11). The demographic and clinical characteristics were not found significantly associated with genotypic lineages of MDR-M.tuberculosis isolates recovered from pulmonary TB patients of North India.
CONCLUSIONS: Present study reveals high genetic diversity among the Mycobacterium tuberculosis isolates and highlights that SIT141/CAS1_Del followed by SIT26/ Beijing lineage is the most common spoligotype responsible for the development and transmission of MDR-TB in North India. The high presence of shared type and unique spoligotype patterns of MDR strains indicates epidemiological significance of locally evolved strains in ongoing transmission

of MDR-TB within this community which needs to be further monitored using robust molecular tools with high discriminatory power.

DOI: 10.1186/s12866-021-02174-6
PMCID: PMC8059304
PMID: 33879047

19. Whole genome analysis of extensively drug resistant Mycobacterium tuberculosis strains in Peru.

Sci Rep. 2021 May 4;11(1):9493. doi: 10.1038/s41598-021-88603-y.

Santos-Lazaro D(1), Gavilan RG(1)(2), Solari L(1), Vigo AN(1), Puyen ZM(3)(4).

Peru has the highest burden of multidrug-resistant tuberculosis in the Americas region. Since 1999, the annual number of extensively drug-resistant tuberculosis (XDR-TB) Peruvian cases has been increasing, becoming a public health challenge. The objective of this study was to perform genomic characterization of Mycobacterium tuberculosis strains obtained from Peruvian patients with XDR-TB diagnosed from 2011 to 2015 in Peru. Whole genome sequencing (WGS) was performed on 68 XDR-TB strains from different regions of Peru. 58 (85.3%) strains came from the most populated districts of Lima and Callao. Concerning the lineages, 62 (91.2%) strains belonged to the Euro-American Lineage, while the remaining 6 (8.8%) strains belonged to the East-Asian Lineage. Most strains (90%) had high-confidence resistance mutations according to pre-established WHO-confident grading system. Discordant results between microbiological and molecular methodologies were caused by mutations outside the hotspot regions analysed by commercial molecular assays (rpoB I491F and inhA S94A). Cluster analysis using a cut-off ≤ 10 SNPs revealed that only 23 (34%) strains evidenced recent transmission links. This study highlights the relevance and utility of WGS as a high-resolution approach to predict drug resistance, analyse transmission of strains between groups, and determine evolutionary patterns of circulating XDR-TB strains in the country.

DOI: 10.1038/s41598-021-88603-y
PMCID: PMC8097007
PMID: 33947918

20. Tuberculosis, COVID-19 and hospital admission: Consensus on pros and cons based on a review of the evidence.

Pulmonology. 2021 May-Jun;27(3):248-256. doi: 10.1016/j.pulmoe.2020.12.016. Epub

2021 Jan 28.

Migliori GB(1), Visca D(2), van den Boom M(3), Tiberi S(4), Silva DR(5), Centis R(6), D'Ambrosio L(7), Thomas T(8), Pontali E(9), Saderi L(10), Schaaf HS(11), Sotgiu G(10); contributing members of the Global Tuberculosis Network.

The scientific debate on the criteria guiding hospitalization of tuberculosis (TB) and COVID-19 patients is ongoing. The aim of this review is to present the available evidence on admission for TB and TB/COVID-19 patients and discuss the criteria guiding hospitalization. Furthermore, recommendations are made as derived from recently published World Health Organization documents, based on Global Tuberculosis Network (GTN) expert opinion. The core published documents and guidelines on the topic have been reviewed. The proportion of new TB cases admitted to hospital ranges between 50% and 100% while for multidrug-resistant (MDR) TB patients it ranges between 85 and 100% globally. For TB patients with COVID-19 the proportion of cases admitted is 58%, probably reflecting different scenarios related to the diagnosis of COVID-19 before, after or at the same time of the active TB episode. The hospital length of stay for drug-susceptible TB ranges from 20 to 60 days in most of countries, ranging from a mean of 10 days (USA) to around 90 days in the Russian Federation. Hospitalization is longer for MDR-TB (50-180 days). The most frequently stated reasons for recommending hospital admission include: severe TB, infection control concerns, co-morbidities and drug adverse events which cannot be managed at out-patient level. The review also provides suggestions on hospital requirements for safe admissions as well as patient discharge criteria, while underlining the relevance of patient-centred care through community/home-based care.

DOI: 10.1016/j.pulmoe.2020.12.016
PMCID: PMC7843149
PMID: 33547028 [Indexed for MEDLINE]

21. Prevalence of aminoglycoside-induced hearing loss in drug-resistant tuberculosis patients: A systematic review: Ototoxic hearing loss in drug-resistant tuberculosis.

J Infect. 2021 May 17:S0163-4453(21)00252-8. doi: 10.1016/j.jinf.2021.05.010. Online ahead of print.

Dillard LK(1), Martinez RX(2), Perez LL(2), Fullerton AM(3), Chadha S(2), McMahon CM(3).

OBJECTIVES: Estimate the prevalence of ototoxic hearing loss in drug-resistant tuberculosis (DR-TB) patients treated with aminoglycoside antibiotics via a systematic review and meta-analysis. Estimate the annual preventable cases of hearing loss in DR-TB patients and leverage findings to discuss primary, secondary and tertiary prevention.
METHODS: Studies published between 2005 and 2018 that reported prevalence of post-treatment hearing loss in DR-TB patients were included. We performed a random effects meta-analysis to determine pooled prevalence of ototoxic hearing loss overall and by medication type. Preventable hearing loss cases were estimated using World Health Organization (WHO) data on DR-TB treatment and prevalence determined by the meta-analysis.
RESULTS: Eighteen studies from 10 countries were included. Pooled prevalence of ototoxic hearing loss and corresponding 95% confidence interval (CI) was 40.62% CI [32.77% - 66.61%] for all drugs (kanamycin: 49.65% CI [32.77% - 66.61%], amikacin: 38.93% CI [26.44% - 53.07%], capreomycin: 10.21% CI [4.33% - 22.21%]). Non-use of aminoglycosides may result in prevention of approximately 50,000 hearing loss cases annually.
CONCLUSIONS: Aminoglycoside use results in high prevalence of ototoxic hearing loss. Widespread prevention of hearing loss can be achieved by following updated WHO guidelines for DR-TB treatment. When hearing loss cannot be avoided, secondary and tertiary prevention should be prioritized.

DOI: 10.1016/j.jinf.2021.05.010
PMID: 34015383

22. Predictors of mortality and loss to follow-up among drug resistant tuberculosis patients in Oromia Hospitals, Ethiopia: A retrospective follow-up study.

PLoS One. 2021 May 6;16(5):e0250804. doi: 10.1371/journal.pone.0250804. eCollection 2021.

Woldeyohannes D(1), Tekalegn Y(2), Sahiledengle B(2), Assefa T(3), Aman R(2), Hailemariam Z(4), Mwanri L(5), Girma A(6).

BACKGROUND: Drug resistance tuberculosis (DR-TB) patients' mortality and loss to follow-up (LTF) from treatment and care is a growing worry in Ethiopia. However, little is known about predictors of mortality and LTF among drug-resistant tuberculosis patients in Oromia region, Ethiopia. The current study aimed to identify predictors of mortality and loss to follow-up among drug resistance tuberculosis patients in Oromia Hospitals, Ethiopia.

METHODS: A retrospective follow up study was carried out from 01 November 2012 to 31 December 2017 among DR-TB patients after calculating sample size using single proportion population formula. Mean, median, Frequency tables and bar charts were used to describe patients' characteristics in the cohort. The Kaplan-Meier curve was used to estimate the probability of death and LTF after the treatment was initiated. The log-rank test was used to compare time to death and time to LTF. The Cox proportional hazard model was used to determine predictors of mortality and LTF after DR-TB diagnosis. The Crude and adjusted Cox proportional hazard ratio was used to measure the strength of association whereas p-value less than 0.05 were used to declare statistically significant predictors.
RESULT: A total of 406 DR-TB patients were followed for 7084 person-months observations. Among the patients, 71 (17.5%) died and 32 (7.9%) were lost to follow up (LTF). The incidence density of death and LTF in the cohort was 9.8 and 4.5 per 1000 person-months, respectively. The median age of the study participants was 28 years (IQR: 27.1, 29.1). The overall cumulative survival probability of patients at the end of 24 months was 77.5% and 84.5% for the mortality and LTF, respectively. The independent predictors of death was chest radiographic findings (AHR = 0.37, 95% CI: 0.17-0.79) and HIV serostatus 2.98 (95% CI: 1.72-5.19). Drug adverse effect (AHR = 6.1; 95% CI: 2.5, 14.34) and culture test result (AHR = 0.1; 95% CI: 0.1, 0.3) were independent predictors of LTF.
CONCLUSION: This study concluded that drug-resistant tuberculosis mortality and LTF remains high in the study area. Continual support of the integration of TB/HIV service with emphasis and work to identified predictors may help in reducing drug-resistant tuberculosis mortality and LTF.

DOI: 10.1371/journal.pone.0250804
PMCID: PMC8101723
PMID: 33956812

23. Assessment of training and mentoring for DR-TB care decentralization in Tanzania.

Hum Resour Health. 2021 Apr 26;19(1):56. doi: 10.1186/s12960-021-00600-4.

Lyakurwa D(1), Lyimo J(2), Mulder C(3)(4), Pelzer PT(3), Koppelaar I(3), Heus M(3).

INTRODUCTION: Drug-resistant TB (DR-TB) care shifted from centralized to decentralized care in Tanzania in 2015. This study explored whether DR-TB training and mentoring supported healthcare workers' (HCWs) DR-TB care performance.

METHODS: This mixed study assessed HCWs' DR-TB care knowledge, the training quality, and the mentoring around 454 HCWs who were trained across 55 DR-TB sites between January 2016 and December 2017. Pre- and post-training tests, end-of-training evaluation, supervisor's interviews, DR-TB team self-assessment and team focus group discussion were conducted among trained HCWs. Interim and final treatment results of the national central site and the decentralized sites were compared.
RESULTS: HCW's knowledge increased for 15-20% between pre-training and post-training. HCWs and supervisors perceived mentoring as most appropriate to further develop their DR-TB competencies. Culture negativity after 6 months of treatment was similar for the decentralized sites compared to the national central site, 81% vs 79%, respectively, whereas decentralized sites had less loss to follow-up (0% versus 3%) and fewer deaths (3% versus 12%). Delays in laboratory results, stigma, and HCWs shortage were reported the main challenges of decentralized care.
CONCLUSIONS: Training and mentoring to provide DR-TB care at decentralized sites in Tanzania improved HCWs' knowledge and skills in DR-TB care and supported observed good interim and final patient treatment outcomes despite health system challenges.

DOI: 10.1186/s12960-021-00600-4
PMCID: PMC8077954
PMID: 33902587

24. Leukocytes from Patients with Drug-Sensitive and Multidrug-Resistant Tuberculosis Exhibit Distinctive Profiles of Chemokine Receptor Expression and Migration Capacity.

J Immunol Res. 2021 Apr 21;2021:6654220. doi: 10.1155/2021/6654220. eCollection 2021.

Ocaña-Guzmán R(1), Téllez-Navarrete NA(1), Ramón-Luing LA(1), Herrera I(2), De Ita M(3), Carrillo-Alduenda JL(4), Choreño-Parra JA(5), Medina-Quero K(6), Zúñiga J(5)(7), Chávez-Galán L(1).

Tuberculosis (TB), caused by Mycobacterium tuberculosis (Mtb), remains as a leading infectious cause of death worldwide. The increasing number of multidrug-resistant TB (MDR-TB) cases contributes to the poor control of the TB epidemic. Currently, little is known about the immunological requirements of protective responses against MDR-TB. This is of major relevance to identify immune markers for treatment monitoring and targets for adjuvant immunotherapies. Here, we hypothesized that MDR-TB patients display unique immunophenotypical features and immune cell migration dynamics compared to

drug-sensitive TB (DS-TB). Hence, we prospectively conducted an extensive characterization of the immune profile of MDR-TB patients at different time points before and after pharmacological therapy. For this purpose, we focused on the leukocyte expression of chemokine receptors, distribution of different monocyte and lymphocyte subsets, plasma levels of chemotactic factors, and in vitro migration capacity of immune cells. Our comparative cohort consisted of DS-TB patients and healthy volunteer donors (HD). Our results demonstrate some unique features of leukocyte migration dynamics during MDR-TB. These include increased and prolonged circulation of CD3+ monocytes, CCR4+ monocytes, EM CD4+ T cells, EM/CM CD8+ T cells, and CXCR1+CXCR3+ T cells that is sustained even after the administration of anti-TB drugs. We also observed shared characteristics of both MDR-TB and DS-TB that include CCR2+ monocyte depletion in the blood; high plasma levels of MPC-1, CCL-7, and IP-10; and increased responsiveness of leukocytes to chemotactic signals in vitro. Our study contributes to a better understanding of the MDR-TB pathobiology and uncovers immunological readouts of treatment efficacy.

DOI: 10.1155/2021/6654220
PMCID: PMC8084684
PMID: 33977111

25. Survival analysis of patients with tuberculosis and risk factors for multidrug-resistant tuberculosis in Monrovia, Liberia.

PLoS One. 2021 Apr 23;16(4):e0249474. doi: 10.1371/journal.pone.0249474. eCollection 2021.

Carter BB(1), Zhang Y(1), Zou H(1), Zhang C(1), Zhang X(1), Sheng R(1), Qi Y(1), Kou C(1), Li Y(1).

We reviewed the records of 337 confirmed cases of tuberculosis patients in Monrovia, the capital of Liberia, 2015. The risk factors affecting the survival and multidrug-resistance of tuberculosis patients were examined. Kaplan-Meier analysis and the log-rank test were used to assess the differences in survival among the patients, while Cox regression model was used for multivariate analysis. The qualitative data was tested with chi-square test in the single factor analysis of multidrug-resistant TB. Multivariate analysis was performed using binary logistic regression analysis. The significance level for all the tests were set at 0.05. The mean period of the follow-up of patients was 10 months. In the 337 patients, 33 (9.8%) died, the 21-month survival rate was 90.2%. The results of multivariate Cox regression analysis show that

overcrowding (HR = 7.942, 95% CI 3.258-19.356), former smoking (HR = 3.773, 95% CI 1.601-8.889), current smoking (HR = 3.546, 95% CI 1.195-10.521), multidrug-resistance tuberculosis (HR = 4.632, 95% CI 1.913-11.217) were risk factors for death during anti-tuberculosis treatment in TB patients in Liberia. The results of binary logistic regression analysis show that extra-pulmonary (OR = 2.032, 95% CI 1.133-3.644), family history of TB (OR = 2.387, 95% CI 1.186-4.807) and current smoking (OR = 3.436, 95% CI 1.681-7.027) were risk factors for multidrug-resistant tuberculosis. These results can provide insights on local tuberculosis early intervention, increase public health awareness, and strengthen the control of factors that may affect the survival and multidrug-resistance of tuberculosis patients.

DOI: 10.1371/journal.pone.0249474
PMCID: PMC8064579
PMID: 33891596

26. Tuberculosis: An Overview of the Immunogenic Response, Disease Progression, and Medicinal Chemistry Efforts in the Last Decade toward the Development of Potential Drugs for Extensively Drug-Resistant Tuberculosis Strains.

J Med Chem. 2021 Apr 22;64(8):4359-4395. doi: 10.1021/acs.jmedchem.0c01833. Epub 2021 Apr 7.

Sharma A(1)(2), De Rosa M(3), Singla N(1), Singh G(2), Barnwal RP(1), Pandey A(4).

Tuberculosis (TB) is a slow growing, potentially debilitating disease that has plagued humanity for centuries and has claimed numerous lives across the globe. Concerted efforts by researchers have culminated in the development of various strategies to combat this malady. This review aims to raise awareness of the rapidly increasing incidences of multidrug-resistant (MDR) and extensively drug-resistant (XDR) tuberculosis, highlighting the significant modifications that were introduced in the TB treatment regimen over the past decade. A description of the role of pathogen-host immune mechanisms together with strategies for prevention of the disease is discussed. The struggle to develop novel drug therapies has continued in an effort to reduce the treatment duration, improve patient compliance and outcomes, and circumvent TB resistance mechanisms. Herein, we give an overview of the extensive medicinal chemistry efforts made during the past decade toward the discovery of new chemotypes, which are potentially active against TB-resistant strains.

DOI: 10.1021/acs.jmedchem.0c01833
PMID: 33826327

27. Impacts of a comprehensive tuberculosis control model on the quality of clinical services and the financial burden of treatment for patients with drug-resistant tuberculosis in China: a mixed-methods evaluation.

Infect Dis Poverty. 2021 Apr 21;10(1):54. doi: 10.1186/s40249-021-00832-5.

Jiang WX(1), Li ZP(2), Zhao Q(2), Gao MQ(3), Long Q(1), Wang WB(2), Huang F(4), Wang N(5), Tang SL(6).

BACKGROUND: The China National Health Commission-Gates TB Project Phase III implemented a comprehensive TB control model including multiple interventions to address the burden of drug-resistant TB (DRTB). This study aims to evaluate the quality of DRTB clinical services and assess the financial burden of DRTB patients during the intervention period.
METHODS: A mixed-methods approach was used to evaluate the effectiveness of interventions in the three project provinces: Zhejiang, Jilin and Ningxia Hui Autonomous Region. The quantitative data included de-identified DRTB registry data during 2015-2018 in project provinces from China CDC, medical records of DRTB patients registered in 2018 (n = 106) from designated hospitals, and a structured DRTB patient survey in six sample prefectures in 2019. The quality of clinical services was evaluated using seven indicators across patient screening, diagnosis and treatment. Logistic regression was conducted to explore factors associated with the extremely high financial burden. Semi-structured in-depth interviews with policymakers and focus group discussions with physicians and DRTB patients were conducted to understand the interventions implemented and their impacts.
RESULTS: The percentage of bacterially confirmed patients taking a drug susceptibility test (DST) increased significantly between 2015 and 2018: from 57.4 to 93.6% in Zhejiang, 12.5 to 86.5% in Jilin, and 29.7 to 91.4% in Ningxia. The treatment enrollment rate among diagnosed DRTB patients also increased significantly and varied from 73 to 82% in the three provinces in 2018. Over 90% of patients in Zhejiang and Jilin and 75% in Ningxia remained in treatment by the end of the first six months' treatment. Among all survey respondents 77.5% incurred extremely high financial burden of treatment. Qualitative results showed that interventions on promoting rapid DST technologies and patient referral were successfully implemented, but the new financing policies for reducing patients' financial burden were not implemented as planned.
CONCLUSIONS: The quality of DRTB related clinical services has been significantly improved following the comprehensive interventions, while the financial burden of DRTB patients remains high due to the delay in implementing financing policies. Stronger political commitment and leadership are required for multi-channel financing to provide additional financial support to DRTB

patients.

DOI: 10.1186/s40249-021-00832-5
PMCID: PMC8059277
PMID: 33883030

28. Scaling Up Molecular Diagnostic Tests for Drug-Resistant Tuberculosis in Uzbekistan from 2012-2019: Are We on the Right Track?

Int J Environ Res Public Health. 2021 Apr 28;18(9):4685. doi: 10.3390/ijerph18094685.

Yuldashev S(1), Parpieva N(1), Alimov S(1), Turaev L(1), Safaev K(1), Dumchev K(2), Gadoev J(3), Korotych O(4), Harries AD(5)(6).

Uzbekistan has a large burden of drug-resistant tuberculosis (TB). To deal with this public health threat, the National TB Program introduced rapid molecular diagnostic tests such as Xpert MTB/RIF (Xpert) and line probe assays (LPAs) for first-line and second-line drugs. We documented the scale-up of Xpert and LPAs from 2012-2019 and assessed whether this led to an increase in patients with laboratory-confirmed multidrug-resistant/rifampicin-resistant TB (MDR/RR-TB) and extensively drug-resistant TB (XDR-TB). This was a descriptive study using secondary program data. The numbers of GeneXpert instruments cumulatively increased from six to sixty-seven, resulting in annual assays increasing from 5574 to 107,330. A broader use of the technology resulted in a lower proportion of tests detecting Mycobacterium tuberculosis with half of the positive results showing rifampicin resistance. LPA instruments cumulatively increased from two to thirteen; the annual first-line assays for MDR-TB increased from 2582 to 6607 while second-line assays increased from 1435 in 2016 to 6815 in 2019 with about one quarter to one third of diagnosed patients showing second-line drug resistance. Patient numbers with laboratory-confirmed MDR-TB remained stable (from 1728 to 2060) but there was a large increase in patients with laboratory-confirmed XDR-TB (from 31 to 696). Programmatic implications and ways forward are discussed.

DOI: 10.3390/ijerph18094685
PMCID: PMC8124440
PMID: 33924862

29. Comparative Utility of Genetic Determinants of Drug Resistance and Phenotypic Drug Susceptibility Profiling in Predicting Clinical Outcomes in Patients With Multidrug-Resistant Mycobacterium tuberculosis.

Front Public Health. 2021 Apr 22;9:663974. doi: 10.3389/fpubh.2021.663974. eCollection 2021.

Che Y(1), Yang T(1), Lin L(1), Xiao Y(2), Jiang F(2), Chen Y(2), Chen T(1), Zhou J(2).

Setting: Programmatic management of drug-resistant tuberculosis in Ningbo, China. Objective: To assess whether data-driven genetic determinants of drug resistance patterns could outperform phenotypic drug susceptibility testing in predicting clinical meaningful outcomes among patients with multidrug-resistant tuberculosis (MDR-TB). Design: We conducted a prospective cohort study of 104 MDR-TB patients. All MDR-TB isolates underwent drug susceptibility testing and genotyping for mutations that could cause drug resistance. Study outcomes were time to sputum smear conversion and probability of treatment success, as well as time to culture conversion within 6 months. Data were analyzed using latent class analysis, Kaplan-Meier curves, and Cox regression models. Results: We report that latent class analysis of data identified two latent classes that predicted sputum smear conversion with P = 0.001 and area under receiver-operating characteristic curve of 0.73. The predicted latent class memberships were associated with superior capability in predicting sputum culture conversion at 6 months and overall treatment success compared to phenotypic drug susceptibility profiling using boosted logistic regression models. Conclusion: These results suggest that genetic determinants of drug resistance in combination with phenotypic drug-resistant tests could serve as useful biomarkers in predicting treatment prognosis in MDR-TB.

DOI: 10.3389/fpubh.2021.663974
PMCID: PMC8100237
PMID: 33968888

30. Drug Resistance Pattern of M. tuberculosis Complex in Oromia Region of Ethiopia.

Infect Drug Resist. 2021 May 4;14:1679-1689. doi: 10.2147/IDR.S294559. eCollection 2021.

Bedru H(1), Fikru M(2), Niguse W(2), Jemal A(2), Getinet G(2), Gobena A(3)(4), Hailu A(5), Peter S(6).

PURPOSE: Multidrug resistant tuberculosis is an emerging problem in many parts of the world. The aim of this study was to determine the drug resistance pattern

of Mycobacterium tuberculosis complex in Oromia Region of Ethiopia.
PATIENTS AND METHODS: A cross-sectional study was conducted from Jan 2017 to June 2018 on 450 pulmonary tuberculosis patients who visited health facilities in nine administrative zones of Oromia Region. Socio-demographic characteristics and relevant clinical information were obtained using a structured questionnaire. Line Probe Assay for first and second line drugs was used to assess the pattern of drug resistance. SPSS version 20 was used for statistical analysis.
RESULTS: Median age was 26 years and 240 (53.3%) patients were males. About 24% of them were previously treated for tuberculosis. Thirty-four (7.6%) were HIV co-infected. Line Probe Assay interpretable results were obtained for 387 isolates. Thirty (7.8%) were resistant to rifampicin and isoniazid and thus were multidrug resistant isolates. Among the multidrug resistant samples, three were found to be extensively drug resistant and one was pre-extensively drug resistant. Previous treatment history (AOR 9.94 (95% CI 3.73-26.51), P < 0.001) and nutritional status below normal (AOR 3.15 (95% CI 1.13-8.81), P < 0.029) were found to be associated with multidrug resistance. The chi-square tests have shown that there was a significant difference between the BCG vaccinated and the non-vaccinated in developing multidrug resistant tuberculosis at P = 0.027.
CONCLUSION: The proportion of multidrug resistance is above the WHO estimate for the country, Ethiopia, and the fact that some zones were at risk of transmission of extensively drug resistant tuberculosis warrant great attention of the control program holders even though it has to be verified through the conventional method.

DOI: 10.2147/IDR.S294559
PMCID: PMC8106478
PMID: 33976556

31. Greenness exposure and all-cause mortality during multi-drug resistant tuberculosis treatment: A population-based cohort study.

Sci Total Environ. 2021 Jun 1;771:145422. doi: 10.1016/j.scitotenv.2021.145422. Epub 2021 Jan 26.

Greenness exposure and all-cause mortality during multi-drug resistant tuberculosis treatment: A population-based cohort study.

Ge E(1), Gao J(2), Ren Z(3), Liu X(4), Luo M(5), Zhong J(6), Fei F(7), Chen B(8), Wang X(9), Wei X(10), Peng Y(11).

BACKGROUND: Living closer to greenness were thought to benefit various health outcomes. We aimed to assess the association between residential greenness and mortality among patients undergoing multidrug resistant tuberculosis (MDR-TB) treatment.
METHODS: We enrolled all local MDR-TB patients reported in Zhejiang, China from 2009 to 2017 and followed them throughout the treatment. We calculated the contemporaneous normalized difference vegetation index (NDVI) in the 250 and 500 m radius around patient's residence. Cox proportional hazards regression models with time-varying NDVI were used to assess the impact of greenness exposure on all-cause mortality during MDR-TB treatment, adjusting for potential individual and contextual covariates.
RESULTS: We ascertained 1,621 active MDR-TB cases, which contributed 3036 person-years at risk with an average follow-up of 684 days (s.d. 149 days) per patient. Among them, there were 163 deaths during follow-up, representing a crude mortality rate of 537 deaths per 10,000 person-years. Patients exposed to the second quintile (Q2) of greenness within the 500 m buffer had around 64% reduced mortality risk over the lowest quintile of greenness with hazard ratio (HR) = 0.364 (95% CI: 0.109-1.22). In lower nighttime light (NTL) areas, the hazard ratios (HR) per quintile increase in NDVI within the 500 m buffer were Q2: 0.35 (95% CI: 0.10-1.18), Q3: 0.24 (95% CI: 0.09-0.66), Q4: 0.26 (95% CI: 0.10-0.69), and Q5: 0.26 (95% CI: 0.10-0.71) relevant to the lowest quintile Q1, with a trend of p-value ≤0.01. Patients who were female, younger (<60 years), resided in urban areas, or had high PM2.5 (i.e. particles with diagram ≤2.5 μm) exposure were more likely to benefit from greenness exposure. Associations were neither observed with NDVI in the 250 m buffer nor for patients living in higher NTL areas. There was a non-linear exposure-response relationship between greenness and deaths with p-value ≤0.05.
CONCLUSION: Increasing greenness exposure along with medical treatment reduces all-cause mortality among patients living in lower NTL areas.

DOI: 10.1016/j.scitotenv.2021.145422
PMID: 33548711 [Indexed for MEDLINE]

32. Effectiveness and cardiac safety of bedaquiline-based therapy for drug-resistant tuberculosis: a prospective cohort study.

Clin Infect Dis. 2021 Apr 21:ciab335. doi: 10.1093/cid/ciab335. Online ahead of print.

Brust JCM(1), Gandhi NR(2)(3), Wasserman S(4), Maartens G(4)(5), Omar SV(6)(7), Ismail NA(6)(7), Campbell A(2), Joseph L(1), Hahn A(1), Allana S(2),

Hernandez-Romieu AC(3), Zhang C(1), Mlisana K(8), Viljoen CA(9), Zalta B(10), Ebrahim I(4), Franczek M(2), Master I(11), Ramangoaela L(12), Te Riele J(13), Meintjes G(4); PROBeX Study Team.

BACKGROUND: Bedaquiline improves treatment outcomes in patients with rifampin-resistant TB (RR-TB) but prolongs the QT-interval and carries a black-box warning by the U.S. Food and Drug Administration. The World Health Organization recommends that all patients with RR-TB receive a regimen containing bedaquiline, yet a phase 3 clinical trial demonstrating its cardiac safety has not been published.
METHODS: We conducted an observational cohort study of RR-TB patients from 3 provinces in South Africa who received regimens containing bedaquiline. We performed rigorous cardiac monitoring, including electrocardiograms (ECGs) performed in triplicate at four time points during bedaquiline therapy. Participants were followed until the end of therapy or 24 months. Outcomes included final tuberculosis treatment outcome and QT-prolongation, defined as any QTcF>500 ms or an absolute change from baseline (△ QTcF) >60 ms.
RESULTS: We enrolled 195 eligible participants, of whom 40% had extensively drug-resistant (XDR) TB. Most participants (97%) received concurrent clofazimine. 74% of participants were cured or successfully completed treatment, and outcomes did not differ by HIV status. QTcF continued to increase throughout bedaquiline therapy, with a mean increase of 23.7 (SD 22.7) ms from baseline to month 6. Four participants experienced a QTcF>500 ms and 19 experienced a △QTcF>60 ms. Older age was independently associated with QT-prolongation. QT-prolongation was neither more common nor severe in participants receiving concurrent lopinavir-ritonavir.
CONCLUSIONS: Severe QT-prolongation was uncommon and did not require permanent discontinuation of either bedaquiline or clofazimine. Close QT-monitoring may be advisable in older patients.

DOI: 10.1093/cid/ciab335
PMID: 33882121

33. Risk Factors for Adverse Events in Household Contacts Prescribed Preventive Treatment for Drug-resistant Tuberculosis Exposure.

Clin Infect Dis. 2021 May 18;72(10):1709-1715. doi: 10.1093/cid/ciaa327.

Malik AA(1)(2)(3), Becerra MC(4)(5)(6), Lash TL(1), Cranmer LM(7), Omer SB(8)(9)(10), Fuad J(2), Siddiqui S(2), Amanullah F(11), Jaswal M(2), Salahuddin

N(11), Keshavjee S(4)(5)(6), Hussain H(3), Gandhi NR(1).

Comment in
Clin Infect Dis. 2021 May 18;72(10):1716-1718.

BACKGROUND: Completion of tuberculosis (TB) preventive treatment is important to optimize efficacy; treatment-related adverse events (AEs) sometimes result in discontinuation. This study describes the occurrence of AEs and their risk factors during a 6-month, 2-drug, fluoroquinolone-based preventive treatment for household contacts of patients with drug-resistant TB in Karachi, Pakistan.
METHODS: The primary outcome was development of any clinical AE during preventive treatment. Adverse events were categorized using the AE grading tables of the National Institutes of Health. Time-to-event analysis with Kaplan-Meier curves and Cox proportional hazards models accounting for recurrence were used to analyze associated risk factors.
RESULTS: Of the 172 household contacts on preventive treatment, 36 (21%) developed 64 AEs during 813 months of treatment. The incidence of AEs over 6 months of treatment was 7.9 per 100 person-months; 16 per 100 person-months with a fluoroquinolone and ethionamide, and 4.4 per 100 person-months with a fluoroquinolone and ethambutol. There were 53 (83%) grade 1 and 11 grade 2 AEs, with no grade 3 or 4 AEs. In multivariable analysis, the risk of AEs was higher in contacts prescribed ethionamide as compared to ethambutol adjusting for age, sex, and body mass index (adjusted hazard ratio, 2.1 [95% confidence interval {CI}, 1.2-3.6]). Overall, there was no notable difference in treatment completion among the contacts who experienced an AE and those who did not (crude odds ratio, 1.1 [95% CI, .52-2.5]).
CONCLUSIONS: A fluoroquinolone-based preventive treatment regimen for drug-resistant TB exposure is well tolerated. Regimens with ethionamide are more likely to result in AEs.

DOI: 10.1093/cid/ciaa327
PMID: 32266942

34. Comprehensive review on mechanism of action, resistance and evolution of antimycobacterial drugs.

Life Sci. 2021 Jun 1;274:119301. doi: 10.1016/j.lfs.2021.119301. Epub 2021 Mar 3.

Chauhan A(1), Kumar M(2), Kumar A(3), Kanchan K(4).

Tuberculosis is one of the deadliest infectious diseases existing in the world since ancient times and still possesses serious threat across the globe. Each year the number of cases increases due to high drug resistance shown by Mycobacterium tuberculosis (Mtb). Available antimycobacterial drugs have been classified as First line, Second line and Third line antibiotics depending on the time of their discoveries and their effectiveness in the treatment. These antibiotics have a broad range of targets ranging from cell wall to metabolic processes and their non-judicious and uncontrolled usage in the treatment for years has created a significant problem called multi-drug resistant (MDR) tuberculosis. In this review, we have summarized the mechanism of action of all the classified antibiotics currently in use along with the resistance mechanisms acquired by Mtb. We have focused on the new drug candidates/repurposed drugs, and drug in combinations, which are in clinical trials for either treating the MDR tuberculosis more effectively or involved in reducing the time required for the chemotherapy of drug sensitive TB. This information is not discussed very adequately on a single platform. Additionally, we have discussed the recent technologies that are being used to discover novel resistance mechanisms acquired by Mtb and for exploring novel drugs. The story of intrinsic resistance mechanisms and evolution in Mtb is far from complete. Therefore, we have also discussed intrinsic resistance mechanisms of Mtb and their evolution with time, emphasizing the hope for the development of novel antimycobacterial drugs for effective therapy of tuberculosis.

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PMID: 33675895 [Indexed for MEDLINE]

35. Toward a Phage Cocktail for Tuberculosis: Susceptibility and Tuberculocidal Action of Mycobacteriophages against Diverse Mycobacterium tuberculosis Strains.

mBio. 2021 May 20;12(3):e00973-21. doi: 10.1128/mBio.00973-21.

Guerrero-Bustamante CA(1), Dedrick RM(1), Garlena RA(1), Russell DA(1), Hatfull GF(2).

The global health burden of human tuberculosis (TB) and the widespread antibiotic resistance of its causative agent Mycobacterium tuberculosis warrant new strategies for TB control. The successful use of a bacteriophage cocktail to treat a Mycobacterium abscessus infection suggests that phages could play a role in tuberculosis therapy. To assemble a phage cocktail with optimal therapeutic

potential for tuberculosis, we have explored mycobacteriophage diversity to identify phages that demonstrate tuberculocidal activity and determined the phage infection profiles for a diverse set of strains spanning the major lineages of human-adapted strains of the Mycobacterium tuberculosis complex. Using a combination of genome engineering and bacteriophage genetics, we have assembled a five-phage cocktail that minimizes the emergence of phage resistance and cross-resistance to multiple phages, and which efficiently kills the M. tuberculosis strains tested. Furthermore, these phages function without antagonizing antibiotic effectiveness, and infect both isoniazid-resistant and -sensitive strains.IMPORTANCE Tuberculosis kills 1.5 million people each year, and resistance to commonly used antibiotics contributes to treatment failures. The therapeutic potential of bacteriophages against Mycobacterium tuberculosis offers prospects for shortening antibiotic regimens, provides new tools for treating multiple drug-resistant (MDR)-TB and extensively drug-resistant (XDR)-TB infections, and protects newly developed antibiotics against rapidly emerging resistance to them. Identifying a suitable suite of phages active against diverse M. tuberculosis isolates circumvents many of the barriers to initiating clinical evaluation of phages as part of the arsenal of antituberculosis therapeutics.

DOI: 10.1128/mBio.00973-21
PMID: 34016711

36. Epidemiological profile of patients with rifampicin-resistant tuberculosis: an analysis of the Uganda National Tuberculosis Reference Laboratory Surveillance Data, 2014-2018.

Antimicrob Resist Infect Control. 2021 May 8;10(1):76. doi: 10.1186/s13756-021-00947-2.

Bahizi G(1)(2), Majwala RK(3)(4), Kisaka S(5), Nyombi A(3)(6), Musisi K(3)(6), Kwesiga B(7), Bulage L(7), Ario AR(7)(8), Turyahabwe S(3)(8).

BACKGROUND: Drug-resistant tuberculosis (DR-TB), including rifampicin-resistant tuberculosis (RR-TB) and multidrug-resistant tuberculosis (MDR-TB, or RR-TB with additional isoniazid resistance), presents challenges to TB control. In Uganda, the GeneXpert test provides point-of-care testing for TB and rifampicin resistance. Patients identified with RR-TB receive culture-based drug susceptibility testing (DST) to identify additional resistance, if any. There are few data on the epidemiological profiles of current DR-TB patients in Uganda. We described patients with RR-TB in Uganda and assessed the trends of

RR-TB to inform TB control interventions.
METHODS: We identified patients with RR-TB whose samples were referred for culture and DST during 2014-2018 from routinely-generated laboratory surveillance data at the Uganda National Tuberculosis Reference Laboratory. Data on patient demographics and drug sensitivity profile of Mycobacterium tuberculosis isolates were abstracted. Population data were obtained from the Uganda Bureau of Statistics to calculate incidence. Descriptive epidemiology was performed, and logistic regression used to assess trends.
RESULTS: We identified 1474 patients whose mean age was 36 ± 17 years. Overall incidence was 3.8/100,000 population. Males were more affected by RR-TB than females (4.9 vs. 2.7/100,000, $p \leq 0.01$). Geographically, Northern Uganda was the most affected region (IR = 6.9/100,000) followed by the Central region (IR = 5.01/100,000). The overall population incidence of RR-TB increased by 20% over the evaluation period (OR = 1.2; 95% CI 1.15-1.23); RR-TB in new TB cases increased by 35% (OR = 1.35; 95% CI 1.3-1.4) and by 7% in previously-treated cases (OR = 1.07; 95% CI 1.0-1.1). Of the 1474 patients with RR-TB, 923 (63%) were culture-positive of whom 670 (72%) had full DST available. Based on the DST results, 522/670 (78%) had MDR-TB.
CONCLUSION: Between 2014 and 2018, the incidence of RR-TB increased especially among newly-diagnosed TB patients. We recommend intensified efforts and screening for early diagnosis especially among previously treated patients. Mechanisms should be in put to ensure that all patients with RR-TB obtain DST.

DOI: 10.1186/s13756-021-00947-2
PMCID: PMC8106164
PMID: 33964986

37. 'We had to manage what we had on hand, in whatever way we could': adaptive responses in policy for decentralized drug-resistant tuberculosis care in South Africa.

Health Policy Plan. 2021 Apr 21;36(3):249-259. doi: 10.1093/heapol/czaa147.

Kielmann K(1), Dickson-Hall L(2), Jassat W(3), Le Roux S(2), Moshabela M(4), Cox H(5), Grant AD(4)(6)(7), Loveday M(8), Hill J(2)(6), Nicol MP(2)(9), Mlisana K(10), Black J(11).

In 2011, the South African National TB Programme launched a policy of decentralized management of drug-resistant tuberculosis (DR-TB) in order to expand the capacity of facilities to treat patients with DR-TB, minimize delays to access care and improve patient outcomes. This policy directive was implemented to varying degrees within a rapidly evolving diagnostic and treatment landscape for DR-TB, placing new demands on already-stressed health

systems. The variable readiness of district-level systems to implement the policy prompted questions not only about differences in health systems resources but also front-line actors' capacity to implement change in resource-constrained facilities. Using a grounded theory approach, we analysed data from in-depth interviews and small group discussions conducted between 2016 and 2018 with managers (n = 9), co-ordinators (n = 15), doctors (n = 7) and nurses (n = 18) providing DR-TB care. Data were collected over two phases in district-level decentralized sites of three South African provinces. While health systems readiness assessments conventionally map the availability of 'hardware', i.e. resources and skills to deliver an intervention, a notable absence of systems 'hardware' meant that systems 'software', i.e. health care workers (HCWs) agency, behaviours and interactions provided the basis of locally relevant strategies for decentralized DR-TB care. 'Software readiness' was manifest in four areas of DR-TB care: re-organization of service delivery, redressal of resource shortages, creation of treatment adherence support systems and extension of care parameters for vulnerable patients. These strategies demonstrate adaptive capacity and everyday resilience among HCW to withstand the demands of policy change and innovation in stressed systems. Our work suggests that a useful extension of health systems 'readiness' assessments would include definition and evaluation of HCW 'software' and adaptive capacities in the face of systems hardware gaps.

DOI: 10.1093/heapol/czaa147
PMCID: PMC8059133
PMID: 33582787

38. Inhibitors of aminoacyl-tRNA synthetases as antimycobacterial compounds: An up-to-date review.

Bioorg Chem. 2021 May;110:104806. doi: 10.1016/j.bioorg.2021.104806. Epub 2021 Mar 6.

Bouz G(1), Zitko J(2).

Aminoacyl-tRNA synthetases (aaRSs) are crucial for the correct assembly of amino acids to cognate tRNA to maintain the fidelity of proteosynthesis. AaRSs have become a hot target in antimicrobial research. Three aaRS inhibitors are already in clinical practice; antibacterial mupirocin inhibits the synthetic site of isoleucyl-tRNA synthetase, antifungal tavaborole inhibits the editing site of leucyl-tRNA synthetase, and antiprotozoal halofuginone inhibits proline-tRNA

synthetase. According to the World Health Organization, tuberculosis globally remains the leading cause of death from a single infectious agent. The rising incidence of multidrug-resistant tuberculosis is alarming and urges the search for new antimycobacterial compounds, preferably with yet unexploited mechanism of action. In this literature review, we have covered the up-to-date state in the field of inhibitors of mycobacterial aaRSs. The most studied aaRS in mycobacteria is LeuRS with at least four structural types of inhibitors, followed by TyrRS and AspRS. Inhibitors of MetRS, LysRS, and PheRS were addressed in a single significant study each. In many cases, the enzyme inhibition activity translated into micromolar or submicromolar inhibition of growth of mycobacteria. The most promising aaRS inhibitor as an antimycobacterial compound is GSK656 (compound 8), the only aaRS inhibitor in clinical trials (Phase IIa) for systemic use against tuberculosis. GSK656 is orally available and shares the oxaborole tRNA-trapping mechanism of action with antifungal tavaborole.

DOI: 10.1016/j.bioorg.2021.104806
PMID: 33799176

39. Malnutrition is Associated with Delayed Sputum Culture Conversion Among Patients Treated for MDR-TB.

Infect Drug Resist. 2021 Apr 28;14:1659-1667. doi: 10.2147/IDR.S293461.
eCollection 2021.

Bade AB(1), Mega TA(2), Negera GZ(3).

BACKGROUND: Clinicians use sputum culture conversion as an interim indicator of the efficacy of multi-drug resistant tuberculosis (MDR-TB) treatment and to determine treatment duration. Yet, limited studies have been published in Ethiopia.
OBJECTIVE: The objective of this study was to determine the predictors of delayed culture conversion among patients receiving MDR-TB treatment at selected treatment centers in Ethiopia.
PATIENTS AND METHODS: A multi-center observation study was conducted among MDR-TB patients in South and Southwestern Ethiopia from April 14 to May 14, 2019. The data of patients treated from January 2013 to July 2019 were reviewed using a data abstraction tool. The data were analyzed. Descriptive statistics was computed using SPSS version 21 software program. Cox regression was used to identify predictors of delayed culture conversion. Hazard ratios with a two-sided p-value <0.05 were considered statistically significant.

RESULTS: Of 200 included MDR-TB patients, 108 (54%) were males. Majority,159 (79.5%) of the patients had a culture conversion time of less than two months, while 15 (7.5%) had delayed culture conversion (greater than 120 days). Patient's registration group (after loss to follow- (adjusted hazard ratio (AHR)=16.215, 95% CI [3.839, 68.498]), after treatment failure (AHR=12.161, 95% CI [2.516, 58.793]), history of previous TB treatment (AHR=4.007, 95% CI [3.115, 62.990])) and low BMI (AHR= 1.257; 95% CI [0.725,1.547] were identified as a risk factors for delayed culture conversion.
CONCLUSION: Our finding showed that nearly 80% of the patients achieve sputum culture conversion by the second month of treatment. Delayed culture conversion was more likely among patients with malnutrition (BMI<18.5kg/m2), after treatment failure, previous TB treatment, and after lost to follow-up.

DOI: 10.2147/IDR.S293461
PMCID: PMC8089472
PMID: 33953577

40. Nanoparticle-mediated macrophage targeting-a new inhalation therapy tackling tuberculosis.

Drug Deliv Transl Res. 2021 Jun;11(3):1037-1055. doi: 10.1007/s13346-020-00815-3.

Makled S(1), Boraie N(1), Nafee N(2)(3).

Despite the potent clinical efficacy of linezolid (LNZ) against drug-resistant tuberculosis, its safety and tolerability remain of major concern. Our objective is to develop antitubercular inhalable LNZ nano-embedded microparticles. In this context, LNZ incorporated in non-structured lipid carriers (NLCs) was characterized in terms of colloidal, morphological, thermal, and release profiles. The potential of LNZ-NLCs to cross mucosal barriers and invade alveolar macrophages (AM, MH-S cells) was appraised. In vivo proof of concept was accomplished via orotracheal administration to mice. Respirable microparticles prepared by spray drying NLCs with diluents were assessed for their size, shape, flowability, aerosolization performance, and lung deposition pattern. NLCs (809-827 nm in size, zeta potential - 37.4 to - 58.9 mV) ensued 19% LNZ loading and pH-independent sustained release. Penetration studies revealed 73% LNZ crossing mucus within 1 h. Meanwhile, viability assay on A549 cells ensured an IC50 of 1.2 and 0.32 mg/mL for plain and LNZ-NLCs, respectively. CLSM confirmed phagocytosis of NLCs by MH-S macrophages, while H&E staining demonstrated NLC accumulation in murine AM in vivo with no signs of

histopathological/biochemical changes. Bronchoalveolar lavage showed significantly low levels of LDH and total proteins (TP) for LNZ-NLCs highlighting their superior safety. Respirable microparticles embedding LNZ-NLCs ensured excellent aerosolization (MMAD 2 μm, FPF 93%) denoting perfect alveolar deposition. The developed inhalation therapy provided sustained LNZ release, mucus penetrability, potential safety in therapeutic doses, in vitro and in vivo macrophage targetability, and preferential deposition in the deep lung. Overall positive outcomes rely on reduced dose, dosing frequency, and per se superior safety circumventing systemic-associated life-threatening side effects. Graphical abstract.

DOI: 10.1007/s13346-020-00815-3
PMID: 32617866

41. Genomic epidemiology of tuberculosis in eastern Malaysia: insights for strengthening public health responses.

Microb Genom. 2021 May;7(5). doi: 10.1099/mgen.0.000573.

Bainomugisa A(1), Meumann EM(2)(3), Rajahram GS(4)(5)(6), Ong RT(7), Coin L(8), Paul DC(9), William T(10)(4)(6), Coulter C(1), Ralph AP(1)(2)(3).

Tuberculosis is a leading public health priority in eastern Malaysia. Knowledge of the genomic epidemiology of tuberculosis can help tailor public health interventions. Our aims were to determine tuberculosis genomic epidemiology and characterize resistance mutations in the ethnically diverse city of Kota Kinabalu, Sabah, located at the nexus of Malaysia, Indonesia, Philippines and Brunei. We used an archive of prospectively collected Mycobacterium tuberculosis samples paired with epidemiological data. We collected sputum and demographic data from consecutive consenting outpatients with pulmonary tuberculosis at the largest tuberculosis clinic from 2012 to 2014, and selected samples from tuberculosis inpatients from the tertiary referral centre during 2012-2014 and 2016-2017. Two hundred and eight M. tuberculosis sequences were available for analysis, representing 8% of cases notified during the study periods. Whole-genome phylogenetic analysis demonstrated that most strains were lineage 1 (195/208, 93.8%), with the remainder being lineages 2 (8/208, 3.8%) or 4 (5/208, 2.4%). Lineages or sub-lineages were not associated with patient ethnicity. The lineage 1 strains were diverse, with sub-lineage 1.2.1 being dominant (192, 98%). Lineage 1.2.1.3 isolates were geographically most widely distributed. The greatest diversity occurred in a border town sub-district. The time to the most recent common ancestor for the three major lineage 1.2.1 clades was estimated to be the year 1966 (95% HPD 1948-1976). An association was found between failure of culture conversion by week 8 of treatment and infection with

lineage 2 (4/6, 67%) compared with lineage 1 strains (4/83, 5%) (P<0.001), supporting evidence of greater virulence of lineage 2 strains. Eleven potential transmission clusters (SNP difference ≤12) were identified; at least five included people living in different sub-districts. Some linked cases spanned the whole 4-year study period. One cluster involved a multidrug-resistant tuberculosis strain matching a drug-susceptible strain from 3 years earlier. Drug resistance mutations were uncommon, but revealed one phenotype-genotype mismatch in a genotypically multidrug-resistant isolate, and rare nonsense mutations within the katG gene in two isolates. Consistent with the regionally mobile population, M. tuberculosis strains in Kota Kinabalu were diverse, although several lineage 1 strains dominated and were locally well established. Transmission clusters - uncommonly identified, likely attributable to incomplete sampling - showed clustering occurring across the community, not confined to households or sub-districts. The findings indicate that public health priorities should include active case finding and early institution of tuberculosis management in mobile populations, while there is a need to upscale effective contact investigation beyond households to include other contacts within social networks.

DOI: 10.1099/mgen.0.000573
PMID: 33945455

42. Low Rate of Acquired Linezolid Resistance in Multidrug-Resistant Tuberculosis Treated With Bedaquiline-Linezolid Combination.

Front Microbiol. 2021 May 3;12:655653. doi: 10.3389/fmicb.2021.655653. eCollection 2021.

Du J(1), Gao J(1), Yu Y(2), Li Q(3), Bai G(4), Shu W(1), Gao M(5), Liu Y(1), Wang L(6), Wang Y(7), Xue Z(7), Huo F(6), Li L(1), Pang Y(6).

In this retrospective study in China, we aimed to: (1) determine the prevalence of linezolid (LZD) resistance among multidrug-resistant tuberculosis (MDR-TB)-infected patients; (2) monitor for dynamic LZD susceptibility changes during anti-TB treatment; and (3) explore molecular mechanisms conferring LZD resistance. A total of 277 MDR-TB patients receiving bedaquiline (BDQ)-containing regimens in 13 TB specialized hospitals across China were enrolled in the study. LZD and BDQ susceptibility rates were determined using the minimum inhibitory concentration (MIC) method, then DNA sequences of patient isolates were analyzed using Sanger sequencing to detect mutations conferring LZD resistance. Of 277 patients in our cohort, 115 (115/277, 41.5%) with prior LZD exposure yielded 19 (19/277, 6.9%) isolates exhibiting LZD resistance. The LZD resistance rate of LZD-exposed group isolates significantly exceeded the

corresponding rate for non-exposed group isolates (P = 0.047). Genetic mutations were observed in 10 (52.6%, 10/19) LZD-resistant isolates, of which a Cys154Arg (36.8%, 7/19) substitution within ribosomal protein L3 was most prevalent. Analysis of sequential positive cultures obtained from 81 LZD-treated patients indicated that cultured organisms obtained from most patients (85.2%, 69/81) retained original LZD MIC values; however, organisms cultured later from two patients exhibited significantly increased MIC values that were attributed to the rplC substitution T460C. Overall, LZD resistance was detected in 6.9% of patients of an MDR-TB cohort in China. Low rate of acquired LZD resistance was noted in MDR-TB treated with BDQ-LZD combination.

DOI: 10.3389/fmicb.2021.655653
PMCID: PMC8126624
PMID: 34012425

43. Diagnostic accuracy of the FluoroType MTB and MTBDR VER 2.0 assays for the centralised high throughput detection of Mycobacterium tuberculosis complex DNA and isoniazid and rifampicin resistance.

Clin Microbiol Infect. 2021 Apr 29:S1198-743X(21)00209-3. doi: 10.1016/j.cmi.2021.04.022. Online ahead of print.

Dippenaar A(1), Derendinger B(1), Dolby T(2), Beylis N(3), van Helden PD(1), Theron G(1), Warren RM(1), de Vos M(4).

OBJECTIVES: To evaluate the accuracy of two new molecular diagnostic tests for the detection of drug resistant tuberculosis, the FluoroType MTB and MTBDR VER 2.0 assays, in combination with manual and automated DNA extraction methods.
METHODS: Sputa from 360 Xpert Ultra Mycobacterium tuberculosis complex (MTBC)-positive patients and 250 Xpert Ultra MTBC-negative patients were tested. GenoType MTBDRplus served as reference for MTBC and drug resistance detection. Sanger sequencing was used to resolve discrepancies.
RESULTS: FluoroType MTB VER 2.0 showed similar MTBC sensitivity compared to FluoroType MTBDR VER 2.0 [manual DNA extraction: 91.6% (294/321) vs. 89.8% (291/324) (p=0.4); automated DNA extraction: 92.1% (305/331) vs 87.7% (291/332) (p=0.05)]. FluoroType MTBDR VER2.0 showed comparable diagnostic accuracy to FluoroType MTBDR VER1.0 as previously reported for the detection of MTBC and rifampicin and isoniazid resistance.
CONCLUSIONS: The FluoroType MTB and MTBDR VER 2.0 assays together with an automated DNA extraction and PCR set-up platform may improve laboratory

operational efficiency for the diagnosis of MTBC and resistance to rifampicin and isoniazid and show promise for the implementation in a centralised molecular drug susceptibility testing model.

DOI: 10.1016/j.cmi.2021.04.022
PMID: 33933566

44. Rapid Tuberculosis Diagnostics Including Molecular First- and Second-Line Resistance Testing Based on a Novel Microfluidic DNA Extraction Cartridge.

J Mol Diagn. 2021 May;23(5):643-650. doi: 10.1016/j.jmoldx.2021.02.004. Epub 2021 Feb 23.

Beutler M(1), Homann AR(2), Mihalic M(3), Plesnik S(3), Niebling L(2), Eckart M(4), Allerheiligen V(4), Czurratis D(5), Maharjan B(6), Shrestha B(6), Parpieva N(7), Turaev L(7), Sayfutdinov Z(7), Hofmann-Thiel S(8), Grasse W(9), Metzger-Boddien C(9), Paust N(2), Hoffmann H(10).

Xpert MTB/RIF testing has improved tuberculosis (TB) diagnostics and rifampicin (Rif) resistance testing worldwide. However, it has weaknesses, such as its restriction to Rif resistance testing and the inability to use extracted DNA for further testing. Herein, a holistic diagnostic workflow, including TB detection and resistance testing toward Rif, isoniazid, and important second-line drugs (SLDs), based on a novel microfluidic DNA extraction cartridge (TB-Disk), is presented. DNA from 73 precharacterized sputum samples was extracted with TB-Disk, including 45 clinical and bacteriologically confirmed TB samples, nine TB-negative samples, and 19 sputum samples spiked with twofold dilutions of TB bacteria. The extracted DNA was subjected to further testing with FluoroType MTB (FT-MTB), GenoType MTBDRplus (GT-plus), and GenoType MTBDRsl. A total of 100% (20/20) and 72% (18/25) of smear-positive and smear-negative TB samples were identified as Mycobacterium tuberculosis complex positive. A total of 79% (33/42) of subsequently GT-plus tested samples yielded a valid result. Eight samples were identified as multidrug-resistant TB by GT-plus and further tested for resistance toward SLDs using GenoType MTBDRsl, yielding 75% (6/8) valid results. FT-MTB with cartridge-based DNA extraction (Disk-DNA) and DNA extracted with FluoroLyse yielded similar analytical sensitivities. FT-MTB with Disk-DNA was 100% specific. TB-Disk in combination with FT-MTB enables sensitive TB detection. The Disk-DNA can be further used for screening resistance toward first-line drugs and SLDs.

DOI: 10.1016/j.jmoldx.2021.02.004
PMID: 33636391

45. A systematic review of pharmacoeconomic evaluations on oral diarylquinoline-based treatment for drug-resistant tuberculosis: from high to low burden countries.

Expert Rev Pharmacoecon Outcomes Res. 2021 Apr 30. doi: 10.1080/14737167.2021.1925111. Online ahead of print.

Fekadu G(1), Yao J(1), You JHS.

INTRODUCTION: There is a rising global interest in the pharmacoeconomic evaluations of bedaquiline (BDQ), a novel oral diarylquinoline, for treatment of drug-resistant tuberculosis (DR-TB).
AREAS COVERED: This article systematically reviewed publications retrieved from Medline, American Psychological Association-Psychology information, Web of Science, Embase, Scopus, Science direct, Centre for Reviews and Dissemination, and CINAHL Complete during 2010-2020 on pharmacoeconomic studies on BDQ for DR-TB treatment. Ten Markov model-based cost-effectiveness analyses identified were conducted in high (n=4), intermediate (n=2), and low (n=4) TB burden countries.
EXPERT OPINION: The paucity of model-based health economic analyses on BDQ-containing regimens for DR-TB indicated that further pharmacoeconomic research of BDQ-based regimens, on the aspects of duration of BDQ treatment, types of DR-TB indicated, and settings of regions and health-systems, is highly warranted to inform global cost-effective use of BDQ-based regimens for DR-TB treatment.

DOI: 10.1080/14737167.2021.1925111
PMID: 33931005

46. Evaluation of the 2016-2020 regional tuberculosis response framework, WHO Western Pacific Region.

Bull World Health Organ. 2021 May 1;99(5):330-341A. doi: 10.2471/BLT.20.268060. Epub 2021 Mar 2.

Viney K(1), Lowbridge C(2), Morishita F(3), Rahevar K(3), Oh KH(3), Islam T(3), Marais BJ(4).

OBJECTIVE: To assess the implementation of the Regional framework for action on implementation of the End TB Strategy in the Western Pacific, 2016-2020 in countries and areas in the World Health Organization Western Pacific Region.
METHODS: We used a mixed methods approach to assess the framework's measurable and perceived impact. We conducted an analysis of national tuberculosis strategic plans, a cross-sectional survey of senior staff of tuberculosis programmes, key informant interviews and some country case studies.
FINDINGS: Of the 37 countries and areas of the Western Pacific Region, 14 had a national tuberculosis strategic plan, including all countries and areas with a high incidence of tuberculosis. Most senior tuberculosis programme staff who responded to the survey (16/23) found the regional framework useful when developing their national targets and grant applications. Programmatic challenges identified included financing, human resources, public-private mix, active case finding, and paediatric and drug-resistant tuberculosis. Most of the 17 key informants thought that the regional framework's categorization of actions (for all settings, for specific settings and for pre-elimination settings) was useful, but that the added value of the regional framework over other relevant documents was not obvious because of overlap in content.
CONCLUSION: The regional framework influenced national level tuberculosis control planning and implementation in a positive way. A future regional framework should provide a longer-term strategic horizon and specifically address emerging trends and persistent problems faced by countries or areas of the region.

DOI: 10.2471/BLT.20.268060
PMCID: PMC8061668
PMID: 33958821

47. Compatibility of Evolutionary Responses to Constituent Antibiotics Drive Resistance Evolution to Drug Pairs.

Mol Biol Evol. 2021 May 4;38(5):2057-2069. doi: 10.1093/molbev/msab006.

Jahn LJ(1), Simon D(1), Jensen M(1), Bradshaw C(1), Ellabaan MMH(1), Sommer MOA(1).

Antibiotic combinations are considered a relevant strategy to tackle the global antibiotic resistance crisis since they are believed to increase treatment efficacy and reduce resistance evolution (WHO treatment guidelines for drug-resistant tuberculosis: 2016 update.). However, studies of the evolution of

bacterial resistance to combination therapy have focused on a limited number of drugs and have provided contradictory results (Lipsitch, Levin BR. 1997; Hegreness et al. 2008; Munck et al. 2014). To address this gap in our understanding, we performed a large-scale laboratory evolution experiment, adapting eight replicate lineages of Escherichia coli to a diverse set of 22 different antibiotics and 33 antibiotic pairs. We found that combination therapy significantly limits the evolution of de novode novo resistance in E. coli, yet different drug combinations vary substantially in their propensity to select for resistance. In contrast to current theories, the phenotypic features of drug pairs are weak predictors of resistance evolution. Instead, the resistance evolution is driven by the relationship between the evolutionary trajectories that lead to resistance to a drug combination and those that lead to resistance to the component drugs. Drug combinations requiring a novel genetic response from target bacteria compared with the individual component drugs significantly reduce resistance evolution. These data support combination therapy as a treatment option to decelerate resistance evolution and provide a novel framework for selecting optimized drug combinations based on bacterial evolutionary responses.

DOI: 10.1093/molbev/msab006
PMCID: PMC8097295
PMID: 33480997

48. Does Ghana's National Health Insurance Scheme provide financial protection to tuberculosis patients and their households?

Soc Sci Med. 2021 May;277:113875. doi: 10.1016/j.socscimed.2021.113875. Epub 2021 Mar 27.

Pedrazzoli D(1), Carter DJ(2), Borghi J(3), Laokri S(4), Boccia D(2), Houben RM(5).

Financial barriers are a key limitation to accessing health services, such as tuberculosis (TB) care in resource-poor settings. In Ghana, the National Health Insurance Scheme (NHIS), established in 2003, officially offers free TB care to those enrolled. Using data from the first Ghana's national TB patient cost survey, we address two key questions 1) what are the key determinants of costs and affordability for TB-affected households, and 2) what would be the impact on costs for TB-affected households of expanding NHIS to all TB patients? We reported the level of direct and indirect costs, the proportion of TB-affected

households experiencing catastrophic costs (defined as total TB-related costs, i.e., direct and indirect, exceeding 20% of their estimated pre-diagnosis annual household income), and potential determinants of costs, stratified by insurance status. Regression models were used to determine drivers of costs and affordability. The effect of enrolment into NHIS on costs was investigated through Inverse Probability of Treatment Weighting Analysis. Higher levels of education and income, a bigger household size and an multi-drug resistant TB diagnosis were associated with higher direct costs. Being in a low wealth quintile, living in an urban setting, losing one's job and having MDR-TB increased the odds of experiencing catastrophic costs. There was no evidence to suggest that enrolment in NHIS defrayed medical, non-medical, or total costs, nor mitigated income loss. Even if we expanded NHIS to all TB patients, the analyses suggest no evidence for any impact of insurance on medical cost, income loss, or total cost. An expansion of the NHIS programme will not relieve the financial burden for TB-affected households. Social protection schemes require enhancement if they are to protect TB patients from financial catastrophe.

DOI: 10.1016/j.socscimed.2021.113875
PMID: 33848718

49. Extracellular matrix-inspired inhalable aerogels for rapid clearance of pulmonary tuberculosis.

Biomaterials. 2021 Jun;273:120848. doi: 10.1016/j.biomaterials.2021.120848. Epub 2021 Apr 22.

Simonson AW(1), Umstead TM(2), Lawanprasert A(1), Klein B(1), Almarzooqi S(1), Halstead ES(2), Medina SH(3).

Tuberculosis (TB) remains a leading cause of death from a single infectious agent, and limiting the spread of multidrug-resistant TB (MDR-TB) is now an urgent global health priority. Essential to the persistence of this disease is the ability of Mycobacterium tuberculosis (Mtb) to circumvent host defenses by infecting lung macrophages to create a cellular niche for its survival and proliferation. This has urged the development of new therapeutic strategies that act through mechanisms distinct from conventional antibiotics, and thus are effective against MDR bacteria, while being able to efficiently kill persister Mtb cells in infected host macrophages. Here, we report a new class of gel-like microparticle aerosols, or 'aerogels', designed to exploit metabolic vulnerabilities of Mtb pathogens and TB-infected macrophages to enable preferential delivery of synergistic peptide-antibiotic combinations for potent

and rapid antitubercular therapy. This is achieved by formulating aerogels through the supramolecular assembly of a de novo designed anti-TB peptide and the extracellular matrix (ECM)-derived polysaccharide, hyaluronic acid (HA). Importantly, HA serves as a nutrient source for Mtb cells during tissue invasion and proliferation, and is recognized by CD44 receptors highly expressed on lung macrophages during TB infection. By exploiting this metabolic substrate for pathogen targeting, HA aerogels are shown to avidly bind and kill both drug-sensitive and drug-resistant mycobacteria, while being efficiently internalized into macrophage host cells in vitro and in vivo to clear Mtb persisters. This multifaceted bioactivity suggests aerogels may serve as a versatile inhalable platform upon which novel biomaterials-enabled therapeutics can be developed to rapidly clear pulmonary MDR-TB.

DOI: 10.1016/j.biomaterials.2021.120848
PMID: 33915409

50. The lipolytic activity of LipJ, a stress-induced enzyme, is regulated by its C-terminal adenylate cyclase domain.

Future Microbiol. 2021 May;16:487-507. doi: 10.2217/fmb-2020-0223. Epub 2021 May 7.

Kumari B(1), Kaur J(1), Maan P(1)(2), Kumar A(3), Kaur J(1).

Aim: The confirmation of lipolytic activity and role of Rv1900c in the Mycobacterium physiology Methods: rv1900c/N-terminus domain (rv1900NT) were cloned in pET28a/Escherichia coli, purified by affinity chromatography and characterized. Results: A zone of clearance on tributyrin-agar and activity with pNP-decanoate confirmed the lipolytic activity of Rv1900c. The Rv1900NT demonstrated higher enzyme specific activity, Vmax and kcat, but Rv1900c was more thermostable. The lipolytic activity of Rv1900c decreased in presence of ATP. Mycobacterium smegmatis expressed rv1900c/rv1900NT-altered colony morphology, growth, cell surface properties and survival under stress conditions. The effect was more prominent with Rv1900NT as compared with Rv1900c. Conclusion: The study confirmed the lipolytic activity of Rv1900c and suggested its regulation by the adenylate cyclase domain and role in the intracellular survival of bacteria.

Plain Language Summary: Lay abstract Tuberculosis (TB) remains the top contagious/infectious killer in the world. It is caused by the bacteria Mycobacterium tuberculosis. The bacteria resides/replicates in the immune cell

that normally has to eradicate infectious microorganisms. Though the treatment of TB is available, the emergence of drug-resistant bacteria is of major concern. The treatment of drug-resistant TB has been reported to be more difficult due to lengthy and complex treatment regimens. Therefore, there is an urgent need for new and better drugs to treat TB/drug-resistant TB. For this purpose understanding the role of each protein in the physiology of mycobacteria is required. Lipids play a critical role in the intracellular survival of this pathogen in the host. Our study demonstrated that LipJ supported the intracellular survival of bacteria. Therefore, it could be a potential drug target.

DOI: 10.2217/fmb-2020-0223
PMID: 33960821

51. Microevolution of Mycobacterium tuberculosis hetero-resistance subpopulations in a patient receiving 27 years of tuberculosis treatment in Germany.

Antimicrob Agents Chemother. 2021 Apr 26:AAC.02520-20. doi: 10.1128/AAC.02520-20. Online ahead of print.

Sonnenkalb L(1), Strohe G(2), Dreyer V(1), Andres S(3), Hillemann D(3), Maurer FP(3)(4), Niemann S(1)(5), Merker M(6).

Pre-existing and newly emerging resistant pathogen subpopulations (hetero-resistance) are potential risk factors for treatment failure of multi/extensively drug resistant (MDR/XDR) tuberculosis (TB). Intra-patient evolutionary dynamics of Mycobacterium tuberculosis complex (Mtbc) strains and their implications on treatment outcomes are still not completely understood.MethodsTo elucidate how Mtbc strains escape therapy, we analysed 13 serial isolates by whole genome sequencing from a German patient. Sequencing data was compared to phenotypic drug susceptibility profiles, and the patient's collective 27-year treatment history, to further elucidate factors fostering intra-patient resistance evolution.ResultsThe patient endured five distinct TB episodes, ending in resistances to 16 drugs and a nearly untreatable XDR-TB infection. The first isolate obtained, during the patient's 5th TB episode, presented fixed resistance mutations to seven anti-TB drugs including isoniazid, rifampicin, streptomycin, pyrazinamide, prothionamide, para-aminosalicyclic acid and cycloserin/terizidone. Over the next 13 years a dynamic evolution with co-existing, heterogeneous subpopulations was observed in six out of 13 sequential bacterial isolates. The emergence of drug-resistant subpopulations coincided with frequent changes in treatment regimens, which often included two or less active compounds. This evolutionary arms race between competing sub-populations, ultimately resulted in the fixation of a single XDR

variant.ConclusionOur data demonstrates the complex intra-patient microevolution of Mtbc subpopulations during failing MDR/XDR-TB treatment. Designing effective treatment regimens based on rapid detection of (hetero-) resistance is key to avoid resistance development and treatment failure.

DOI: 10.1128/AAC.02520-20
PMID: 33903103

52. Impact of the revised definition of extensively drug resistant tuberculosis.

Eur Respir J. 2021 May 6:2100641. doi: 10.1183/13993003.00641-2021. Online ahead of print.

Veziris N(1), Bonnet I(2), Morel F(2), Guglielmetti L(2), Maitre T(3), Fournier Le Ray L(3), Sougakoff W(2), Robert J(2), Aubry A(2); CNR MyRMA.

DOI: 10.1183/13993003.00641-2021
PMID: 33926973

53. Five-year microevolution of a multidrug-resistant Mycobacterium tuberculosis strain within a patient with inadequate compliance to treatment.

BMC Infect Dis. 2021 Apr 29;21(1):394. doi: 10.1186/s12879-021-06069-9.

Fernandez Do Porto DA(#)(1)(2), Monteserin J(#)(3)(4), Campos J(3), Sosa EJ(5), Matteo M(6), Serral F(1), Yokobori N(3)(4), Benevento AF(5), Poklepovich T(3), Pardo A(1)(5), Wainmayer I(3), Simboli N(3), Castello F(1), Paul R(3), Martí M(2)(5), López B(3), Turjanski A(7)(8), Ritacco V(9)(10).

BACKGROUND: Whole-genome sequencing has shown that the Mycobacterium tuberculosis infection process can be more heterogeneous than previously thought. Compartmentalized infections, exogenous reinfections, and microevolution are manifestations of this clonal complexity. The analysis of the mechanisms causing the microevolution -the genetic variability of M. tuberculosis at short time scales- of a parental strain into clonal variants with a patient is a relevant issue that has not been yet completely addressed. To our knowledge, a whole genome sequence microevolution analysis in a single patient with inadequate adherence to treatment has not been previously reported. CASE PRESENTATION: In this work, we applied whole genome sequencing analysis for a more in-depth analysis of the microevolution of a parental Mycobacterium

tuberculosis strain into clonal variants within a patient with poor treatment compliance in Argentina. We analyzed the whole-genome sequence of 8 consecutive Mycobacterium tuberculosis isolates obtained from a patient within 57-months of intermittent therapy. Nineteen mutations (9 short-term, 10 fixed variants) emerged, most of them associated with drug resistance. The first isolate was already resistant to isoniazid, rifampicin, and streptomycin, thereafter the strain developed resistance to fluoroquinolones and pyrazinamide. Surprisingly, isolates remained susceptible to the pro-drug ethionamide after acquiring a frameshift mutation in ethA, a gene required for its activation. We also found a novel variant, (T-54G), in the 5' untranslated region of whiB7 (T-54G), a region allegedly related to kanamycin resistance. Notably, discrepancies between canonical and phage-based susceptibility testing to kanamycin were previously found for the isolate harboring this mutation. In our patient, microevolution was mainly driven by drug selective pressure. Rare short-term mutations fixed together with resistance-conferring mutations during therapy.
CONCLUSIONS: This report highlights the relevance of whole-genome sequencing analysis in the clinic for characterization of pre-XDR and MDR resistance profile, particularly in patients with incomplete and/or intermittent treatment.

DOI: 10.1186/s12879-021-06069-9
PMCID: PMC8082761
PMID: 33926375 [Indexed for MEDLINE]

54. Identification and validation of potent Mycobacterial proteasome inhibitor from Enamine library.

J Biomol Struct Dyn. 2021 May 6:1-11. doi: 10.1080/07391102.2021.1914173. Online ahead of print.

Tyagi R(1), Srivastava M(2), Singh B(3), Sharma S(1)(4), Pandey RP(1), Asthana S(2), Kumar D(5), Raj VS(1).

As a consequence of present status of tuberculosis (TB) it is the obligation to develop novel targets and potential drugs so that rate of drug resistant TB can be declined. Mycobacterium proteasome is considered to be significant target for the purpose of drug designing as it is responsible for resisting the effect of NO (nitric oxide) immune system defence mechanism against the bacterial cells. Small compounds library from Enamine database has already been tested using virtual screening and molecular docking studies. Further a reanalysis with two picked out significant compounds Z1020863610, Z106766984 was carried out using molecular dynamic simulation studies and in vitro validations (in vitro susceptibility assay, enzyme inhibition assay and MTT assay). In silico outcome that two inhibiters were interacting at the active site pocket of receptor with

high stability, was found to be very consistent with in vitro results. So it was conferred that compounds (Z1020863610, Z106766984) are potential lead for future process of drug development (in vivo testing and clinical trials).Communicated by Ramaswamy H. Sarma.

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PMID: 33955331

55. Clinical and epidemiological features of tuberculosis isolated from critically ill patients.

Rev Argent Microbiol. 2021 May 14:S0325-7541(21)00046-8. doi: 10.1016/j.ram.2021.02.011. Online ahead of print.

Hurtado J(1), Coitinho C(2), Nin N(3), Buroni M(3), Hurtado FJ(4), Robello C(5), Greif G(6).

Human tuberculosis is still a major world health concern. In Uruguay, contrary to the world trend, an increase in cases has been observed since 2006. Although the incidence of MDR-resistant strains is low and no cases of XDR-TB were registered, an increase in the number of patients with severe tuberculosis requiring critical care admission was observed. As a first aim, we performed the analysis of the genetic structure of strains isolated from patients with severe tuberculosis admitted to an intensive care unit. We compared these results with those corresponding to the general population observing a statistically significant increase in the Haarlem genotypes among ICU patients (53.3% vs 34.7%; $p<0.05$). In addition, we investigated the association of clinical outcomes with the genotype observing a major incidence of hepatic dysfunctions among patients infected with the Haarlem strain ($p<0.05$). The cohort presented is one of the largest studied series of critically ill patients with tuberculosis.

DOI: 10.1016/j.ram.2021.02.011
PMID: 34001412

56. Identification of Potential Binders of Mtb Universal Stress Protein (Rv1636) Through an in silico Approach and Insights Into Compound Selection for Experimental Validation.

Front Mol Biosci. 2021 May 3;8:599221. doi: 10.3389/fmolb.2021.599221.
eCollection 2021.

Chakraborti S(1), Chakraborty M(2), Bose A(2), Srinivasan N(1), Visweswariah
SS(2).

Millions of deaths caused by Mycobacterium tuberculosis (Mtb) are reported worldwide every year. Treatment of tuberculosis (TB) involves the use of multiple antibiotics over a prolonged period. However, the emergence of resistance leading to multidrug-resistant TB (MDR-TB) and extensively drug-resistant TB (XDR-TB) is the most challenging aspect of TB treatment. Therefore, there is a constant need to search for novel therapeutic strategies that could tackle the growing problem of drug resistance. One such strategy could be perturbing the functions of novel targets in Mtb, such as universal stress protein (USP, Rv1636), which binds to cAMP with a higher affinity than ATP. Orthologs of these proteins are conserved in all mycobacteria and act as "sink" for cAMP, facilitating the availability of this second messenger for signaling when required. Here, we have used the cAMP-bound crystal structure of USP from Mycobacterium smegmatis, a closely related homolog of Mtb, to conduct a structure-guided hunt for potential binders of Rv1636, primarily employing molecular docking approach. A library of 1.9 million compounds was subjected to virtual screening to obtain an initial set of ~2,000 hits. An integrative strategy that uses the available experimental data and consensus indications from other computational analyses has been employed to prioritize 22 potential binders of Rv1636 for experimental validations. Binding affinities of a few compounds among the 22 prioritized compounds were tested through microscale thermophoresis assays, and two compounds of natural origin showed promising binding affinities with Rv1636. We believe that this study provides an important initial guidance to medicinal chemists and biochemists to synthesize and test an enriched set of compounds that have the potential to inhibit Mtb USP (Rv1636), thereby aiding the development of novel antitubercular lead candidates.

DOI: 10.3389/fmolb.2021.599221
PMCID: PMC8126637
PMID: 34012976

57. Levofloxacin pharmacokinetics in saliva as measured by a mobile microvolume UV spectrophotometer among people treated for rifampicin-resistant TB in Tanzania.

J Antimicrob Chemother. 2021 May 12;76(6):1547-1552. doi: 10.1093/jac/dkab057.

Mohamed S(1), Mvungi HC(2), Sariko M(3), Rao P(1), Mbelele P(2), Jongedijk EM(4), van Winkel CAJ(4), Touw DJ(4), Stroup S(1), Alffenaar JC(5)(6)(7), Mpagama S(3), Heysell SK(1).

BACKGROUND: Early detection and correction of low fluoroquinolone exposure may improve treatment of MDR-TB.
OBJECTIVES: To explore a recently developed portable, battery-powered, UV spectrophotometer for measuring levofloxacin in saliva of people treated for MDR-TB.
METHODS: Patients treated with levofloxacin as part of a regimen for MDR-TB in Northern Tanzania had serum and saliva collected concurrently at 1 and 4 h after 2 weeks of observed levofloxacin administration. Saliva levofloxacin concentrations were quantified in the field via spectrophotometry, while serum was analysed at a regional laboratory using HPLC. A Bayesian population pharmacokinetics model was used to estimate the area under the concentration-time curve (AUC0-24). Subtarget exposures of levofloxacin were defined by serum AUC0-24 <80 mg·h/L. The study was registered at Clinicaltrials.gov with clinical trial identifier NCT04124055.
RESULTS: Among 45 patients, 11 (25.6%) were women and 16 (37.2%) were living with HIV. Median AUC0-24 in serum was 140 (IQR = 102.4-179.09) mg·h/L and median AUC0-24 in saliva was 97.10 (IQR = 74.80-121.10) mg·h/L. A positive linear correlation was observed with serum and saliva AUC0-24, and a receiver operating characteristic curve constructed to detect serum AUC0-24 below 80 mg·h/L demonstrated excellent prediction [AUC 0.80 (95% CI = 0.62-0.94)]. Utilizing a saliva AUC0-24 cut-off of 91.6 mg·h/L, the assay was 88.9% sensitive and 69.4% specific in detecting subtarget serum AUC0-24 values, including identifying eight of nine patients below target.
CONCLUSIONS: Portable UV spectrophotometry as a point-of-care screen for subtarget levofloxacin exposure was feasible. Use for triage to other investigation or personalized dosing strategy should be tested in a randomized study.

DOI: 10.1093/jac/dkab057
PMCID: PMC8120342
PMID: 33675664

58. The importance of pharmacokinetics/pharmacodynamics assessment in Phase IIB/III trials for MDR-TB treatment.

Int J Tuberc Lung Dis. 2021 May 1;25(5):336-339. doi: 10.5588/ijtld.21.0072.

Märtson AG(1), Kim HY(2), Marais B(3), Alffenaar JW(2).

DOI: 10.5588/ijtld.21.0072
PMID: 33977900

59. Inhibitors of F(1)F(0)-ATP synthase enzymes for the treatment of tuberculosis and cancer.

Future Med Chem. 2021 May;13(10):911-926. doi: 10.4155/fmc-2021-0010. Epub 2021 Apr 13.

Denny WA(1)(2).

The spectacular success of the mycobacterial F1F0-ATP synthase inhibitor bedaquiline for the treatment of drug-resistant tuberculosis has generated wide interest in the development of other inhibitors of this enzyme. Work in this realm has included close analogues of bedaquiline with better safety profiles and 'bedaquiline-like' compounds, some of which show potent antibacterial activity in vitro although none have yet progressed to clinical trials. The search has lately extended to a range of new scaffolds as potential inhibitors, including squaramides, diaminoquinazolines, chloroquinolines, dihydropyrazolo[1,5-a]pyrazin-4-ones, thiazolidinediones, diaminopyrimidines and tetrahydroquinolines. Because of the ubiquitous expression of ATP synthase enzymes, there has also been interest in inhibitors of other bacterial ATP synthases, as well as inhibitors of human mitochondrial ATP synthase for cancer therapy. The latter encompass both complex natural products and simpler small molecules. The review seeks to demonstrate the breadth of the structural types of molecules able to effectively inhibit the function of variants of this intriguing enzyme.

DOI: 10.4155/fmc-2021-0010
PMID: 33845594

60. Outcomes of Children Born to Pregnant Women With Drug-resistant Tuberculosis Treated With Novel Drugs in Khayelitsha, South Africa: A Report of Five Patients.

Pediatr Infect Dis J. 2021 May 1;40(5):e191-e192. doi: 10.1097/INF.0000000000003069.

Acquah R(1), Mohr-Holland E(1), Daniels J(1), Furin J(2), Loveday M(3), Mudaly

V(4), Reuter A(1).

This brief report presents a series of 5 pregnant women treated for rifampicin-resistant tuberculosis with the novel drugs bedaquiline, delamanid, and linezolid as part of an optimized backbone regimen and reviews the outcomes of the children born to them. Although the case series is small, all children had excellent birth outcomes suggesting pregnant women should not be denied access to novel therapies for RR-TB.

DOI: 10.1097/INF.0000000000003069
PMCID: PMC8043512
PMID: 33847295

61. Semisynthetic modifications of antitubercular lanostane triterpenoids from Ganoderma.

J Antibiot (Tokyo). 2021 May 12:1-8. doi: 10.1038/s41429-021-00422-5. Online ahead of print.

Chinthanom P(1), Vichai V(1), Dokladda K(1), Sappan M(1), Thongpanchang C(1), Isaka M(2).

Antitubercular lanostane triterpenoids isolated from mycelial cultures of the basidiomycete Ganoderma australe were structurally modified by semisynthesis. One of the synthetic compounds, named GA003 (9), showed more potent activity against Mycobacterium tuberculosis H37Ra than the lead natural lanostane (1). GA003 was also significantly active against the virulent strain (H37Rv) as well as extensively drug-resistant tuberculosis strains.

DOI: 10.1038/s41429-021-00422-5
PMCID: PMC8113785
PMID: 33981028