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Review article

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# Drug-resistant tuberculosis: a persistent global health concern

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## Abstract

Drug-resistant tuberculosis (TB) is estimated to cause 13% of all antimicrobial resistance-attributable deaths worldwide and is driven by both ongoing resistance acquisition and person-to-person transmission. Poor outcomes are exacerbated by late diagnosis and inadequate access to effective treatment. Advances in rapid molecular testing have recently improved the diagnosis of TB and drug resistance. Next-generation sequencing of *Mycobacterium tuberculosis* has increased our understanding of genetic resistance mechanisms and can now detect mutations associated with resistance phenotypes. All-oral, shorter drug regimens that can achieve high cure rates of drug-resistant TB within 6–9 months are now available and recommended but have yet to be scaled to global clinical use. Promising regimens for the prevention of drug-resistant TB among high-risk contacts are supported by early clinical trial data but final results are pending. A person-centred approach is crucial in managing drug-resistant TB to reduce the risk of poor treatment outcomes, side effects, stigma and mental health burden associated with the diagnosis. In this Review, we describe current surveillance of drug-resistant TB and the causes, risk factors and determinants of drug resistance as well as the stigma and mental health considerations associated with it. We discuss recent advances in diagnostics and drug-susceptibility testing and outline the progress in developing better treatment and preventive therapies.

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## Introduction

Tuberculosis (TB), caused by the aerophilic intracellular obligate pathogen *Mycobacterium tuberculosis*, is a globally endemic bacterial infection that transmits person-to-person via the airborne route. Although pulmonary disease is the most common form of TB, it can also affect other organs, most commonly lymph nodes but also the pleura, central nervous system, musculoskeletal system and other organ systems[1]. TB treatment can range from as low as 4 months to as high as 24 months of multi-antibiotic regimens depending on the drug resistance profile, the initial clinical picture and progression during the therapeutic course[2,3]. Microbiological confirmation of disease is an important component of treatment delivery and monitoring, but it can be difficult to achieve due to the slow-growing nature of *M. tuberculosis* and its fastidiousness in culture. It is further challenged by the ability of *M. tuberculosis* to cause disease with a very low bacterial load (paucibacillary disease), which is hard to detect with currently available methods[4]. The interplay between exposure to anti-TB drugs during treatment, person-to-person transmission, global travel and poor quality TB care has led to the emergence and establishment of different drug-resistant strains of *M. tuberculosis* in geographically distinct regions across the globe[5,6].

Drug-resistant TB (DR-TB) is classified by the extent of resistance to key agents in anti-TB drug regimens. The most important of these agents is rifampicin (Rif), a bactericidal and sterilizing drug that allowed shortening of treatment duration[7,8]. Multidrug-resistant TB (MDR-TB) is defined as *M. tuberculosis* resistant to both Rif and isoniazid (Inh)[9]. Rif-resistant TB (RR-TB) is considered the entry point into second-line treatment given that individuals with RR-TB are treated with similar regimens to those with MDR-TB, regardless of Inh resistance[10]. Nucleic acid amplification tests (NAATs) for Rif resistance, especially the widely adopted Xpert MTB/Rif assay (Cepheid Inc., California, USA), have enabled improved diagnosis and surveillance of RR-TB since the early 2010s[11].

Diagnostic testing for Inh resistance continues to be more limited than for Rif due to costs and the lower sensitivity of molecular assays for Inh[12]. However, in many parts of the world, Inh-resistant and Rif-susceptible TB (Hr-TB) is the most common form of DR-TB, with MDR-TB being the second most common[13]. Inh is an important first-line drug for TB, and it is both potent and well tolerated[14–16]. However, evidence on the impact of resistance to Inh on the rate of treatment failure suggests a lesser effect than for MDR/RR-TB[17,18]. The recommended regimen for Hr-TB is similar to that of drug-susceptible disease replacing Inh with a fluoroquinolone[2,19,20].

The past decade has been marked by the introduction of new drugs for the treatment of MDR/RR-TB, including bedaquiline (Bdq) and the nitroimidazoles pretomanid and delamanid (Dlm), and by increased recognition of the efficacy of the late-generation fluoroquinolones and linezolid (Lzd). In 2021, the WHO announced a revision to the definition of extensively drug-resistant TB (XDR-TB) and suggested a standard definition for pre-XDR-TB for the first time. Pre-XDR-TB is now defined as MDR-TB additionally resistant to a late-generation fluoroquinolone, such as moxifloxacin (Mfx) or levofloxacin (Lfx), and XDR-TB is defined as MDR-TB with resistance to Lfx or Mfx and at least one additional group A drug (Bdq or Lzd)[10,21]. These revisions highlight the clinical impact of these advanced forms of drug resistance and de-emphasize the role of toxic, second-line injectable agents, of which only amikacin and streptomycin are still recommended in settings when all-oral compositions cannot be used or when rescue treatment is necessary[2,10].

In this Review, we examine the current epidemiological trends for DR-TB, gaps in the care cascade, and key factors and determinants associated with drug resistance. We explore current and developing approaches to diagnosing DR-TB and provide an updated overview of the latest antibiotic treatment regimens and preventive strategies. We also explore the stigma and mental health implications of being diagnosed with DR-TB.

## Global epidemiology and care cascade

### DR-TB burden

The WHO estimates that 410,000 (95% uncertainty interval (UI) 370,000–450,000) people developed MDR/RR-TB in 2022, accounting for 3.9% (95% UI 3.7–4.1%) of the 10.6 million estimated incident TB cases for that year[22]. Of these, 160,000 people died[22]. MDR-TB, excluding XDR-TB, is estimated to cause 13% (95% UI 10–19%) of all antimicrobial resistance-attributable deaths worldwide[23]. The proportion of people exposed to MDR-TB is unknown. Mathematical modelling suggests that ~19 million people have a latent MDR-TB infection and are at risk of activation[24].

The burden of MDR/RR-TB varies substantially between regions and across countries. Of the 177,853 reported MDR/RR-TB cases, the highest-burden regions were Southeast Asia (47%), Europe (19%), Western Pacific (15%) and Africa (13%), followed by the Eastern Mediterranean (3%) and the Americas (3%)[22]. The WHO lists 30 high DR-TB burden countries[22] (Fig. 1a), defined based on both estimated absolute numbers of cases with MDR/RR-TB and population incidence rates. The WHO estimates that India, the Philippines and the Russian Federation are the highest-burden countries and account for 26.8%, 7.6% and 7.6% of all MDR/RR-TB cases, respectively[22]. Pakistan was considered one of the three highest-burden countries for RR-TB until new methods to estimate RR-TB were developed in 2022 (ref. 25), leading to a major downward revision for RR-TB incidence estimates[22].

Rif mono-resistant TB (RMR-TB) is estimated to be increasing in prevalence, from 12% of all MDR/RR-TB cases in 2014 to 22% in 2019 (refs. 9,26). The relative importance of RMR-TB as a contributor to the RR-TB burden varies substantially across high MDR/RR-TB burden countries, ranging from less than 5% of all RR-TB in countries as diverse as Ethiopia and Bangladesh to around 40% in Tajikistan and South Africa[27]. In some settings, RMR-TB may be associated with HIV infection[28,29]. There is also some evidence that RMR-TB is driving the increase in RR-TB[30].

In 2022, the incidence rate of Inh-resistant TB was estimated at 1.3 million cases (95% UI 0.39–2.3 million), including people with both Hr-TB and MDR-TB[22]. A review of survey data from 156 different settings in 2003–2017 suggests an Hr-TB prevalence of 7.4% and 11.4% among never-treated and previously treated persons with TB, respectively[31]. Most of these cases are unlikely to be diagnosed, given that Rif resistance is the usual entry point into further drug susceptibility testing (DST).

### The global care cascade for DR-TB

The TB cascade of care assesses the continuum of disease from diagnosis to treatment outcome to identify steps where persons are lost to follow-up or do not progress to a favourable outcome[32–35] (Fig. 1b). A larger proportion of persons with MDR/RR-TB initiated second-line treatment from 2015 to 2019 (23% to 41% of all estimated MDR/RR-TB cases)[22]. However, this proportion declined to 36% in 2020 due to setbacks in MDR/RR-TB diagnosis and care during the COVID-19 pandemic[10,22,36] (Fig. 1c). This setback partially reversed in 2022, when 175,650 (43%, 95% UI 39–48%) of the estimated 410,000

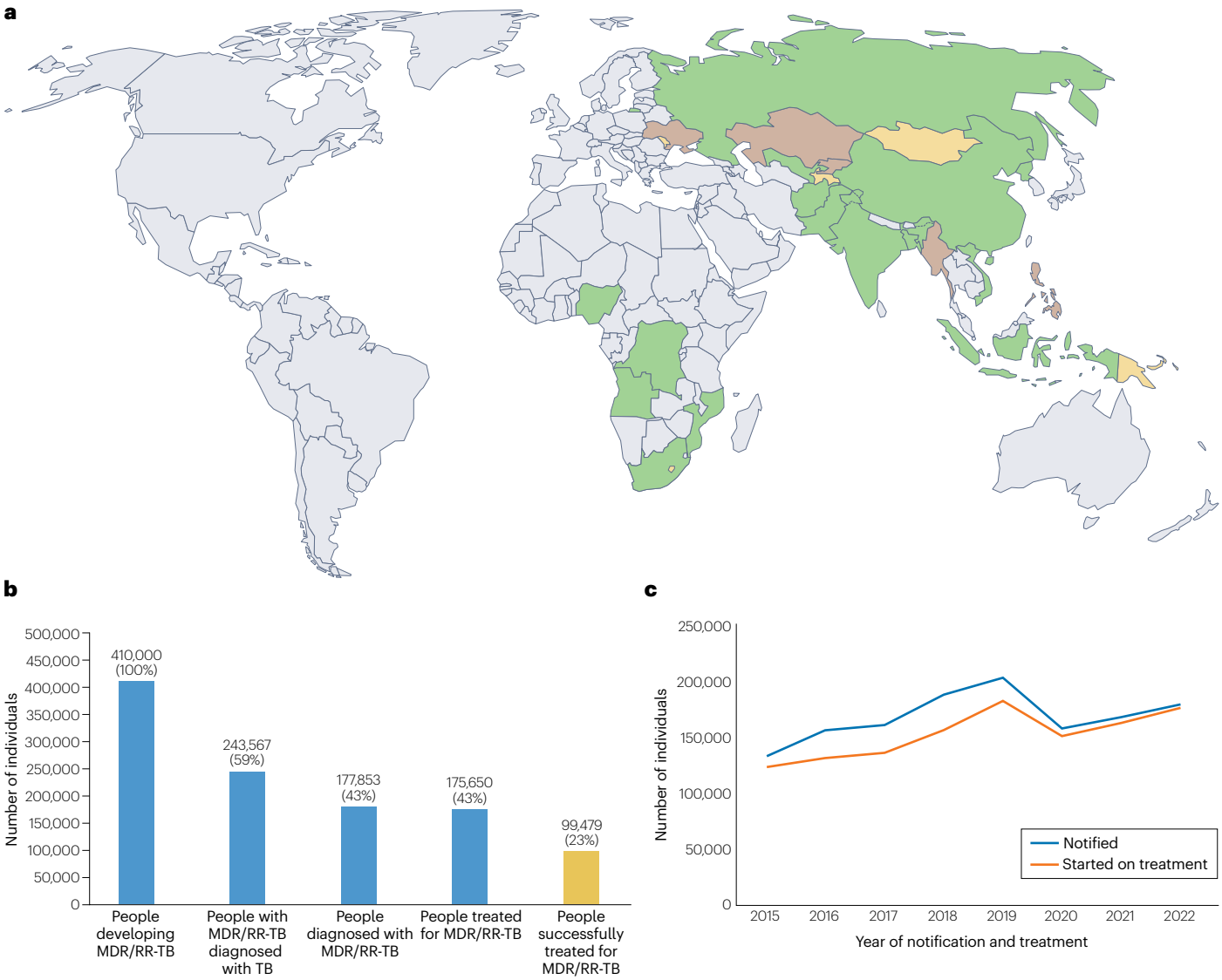


**Fig. 1 | Global epidemiology and cascade of care for MDR/RR-TB. a**, The map represents the top 30 countries with the highest burden of multidrug-resistant or rifampicin-resistant tuberculosis (MDR/RR-TB), which include countries in Asia, Africa, Europe and Oceania. Countries in green correspond to those ranking among the top 20 with the highest burden based on cases and include India, Vietnam, Bangladesh, South Africa, Nigeria, the Democratic Republic of Congo, Mozambique and Russia. Countries in yellow are among the top 10 with the highest adjusted burden per 100,000 individuals and include Papua New Guinea, Lesotho, Mongolia, Tajikistan and Moldova. Countries in brown fall into both categories and include Kazakhstan, Myanmar, Kyrgyzstan, Ukraine and the Philippines[22]. **b**, The graph shows the cascade of care for MDR/RR-TB based on estimated numbers, reported notifications of TB and MDR/RR-TB, number of individuals started on treatment for MDR/RR-TB, and number of treatment success cases[9,22]. The cascade of care refers to the sequential stages that include diagnosis, treatment, and management of persons with TB and its analysis can inform and guide efforts to improve TB control. Of approximately 410,000 individuals actively developing MDR/RR-TB, 59% were diagnosed with TB, while 43% were diagnosed with MDR/RR-TB and received treatment for it. Of the 437,000 estimated MDR/RR-TB cases in 2020, only 23% had a successful MDR/RR-TB treatment outcome (yellow, most recent available data). **c**, The graph shows the numbers of individuals with notified MDR/RR-TB (blue) and individuals who were started on treatment (orange) by year. Data from ref. 22. In 2015, the WHO estimated, for the first time, the total number of MDR/RR-TB cases[22].

(95% UI 370,000–450,000) individuals with MDR/RR-TB were started on second-line treatment[22] (Fig. 1b,c). Despite these improvements, only 825,000 individuals with MDR/RR-TB were enroled on treatment between 2018 and 2022, reaching 55% of the 1.5 million United Nations treatment target for that time period[22]. After treatment initiation, only 23% of the estimated 437,000 people with MDR/RR-TB completed treatment in 2020 (the last year for which treatment success data is available)[10,22] (Fig. 1b). Globally, treatment success for MDR/RR-TB has slowly improved to approximately 63% of people started on treatment from approximately 50% of treated cases in 2012 (ref. 22) (Supplementary Fig. 1), a number that is expected to increase with the widespread implementation of newer and better all-oral regimens.

This leaky global cascade of care was further corroborated by standardized patient surveys[37–40] in the high-burden countries of India and South Africa. Standardized patients are coached to portray symptoms of TB in a standardized manner to evaluate provider expertise in case assessment and treatment[37]. In the interactions where standardized patients presented with recurrent TB and possible MDR/RR-TB, only 3% were recommended any DST in India[41]. In South Africa, nearly 70% of the standardized patient interactions were correctly managed in the private sector when the standardized patients presented with a history of previous TB[39]. Timely resistance testing can bridge a gap in the cascade, increase treatment success and decrease transmission networks for DR-TB.

Access to TB diagnostics for bacteriological confirmation of disease and testing for drug resistance is still limited[2,22,42]. In 2022, 7.5 million people were officially diagnosed with pulmonary TB and notified as TB cases[22]. Only 53% of these diagnoses were bacteriologically confirmed as having TB, of which 73% were tested for Rif resistance[22]. Access to DST for second-line TB drugs to guide treatment is even more limited[42]. Data from 2021 shows that only 50% of individuals diagnosed with MDR/RR-TB were tested for fluoroquinolone resistance that year[9]. According to a country-level report in 2020 (ref. 43), only 29% of 35 countries that use Bdq, Dlm, Lzd and/or clofazimine (Cfz) had DST available for these drugs despite an increasing number of reports of the emergence of resistance to these agents[44–48].

The diagnostic gap in the care cascade is particularly relevant for children with MDR/RR-TB. Globally, between 25,000 and 32,000 children aged 0–14 years are estimated to develop MDR/RR-TB annually[49]. In 2018–2022, only 21,600 children were started on second-line therapy for MDR/RR-TB (corresponding to 19% of the 2018–2022 target)[9]. The lack of effective diagnostics for TB and DR-TB that can detect paucibacillary disease in children[50] and the lack of contact investigation for adults with MDR/RR-TB through which children can be identified and treated render diagnosis and treatment a challenging task in this population group. Recent modelling suggests that implementation of effective household contact management for MDR/RR-TB could have prevented MDR/RR-TB development in more than 5,600 children and could have averted 3,600 infant deaths due to TB in 2019 alone[51].

## Causes and risk factors of DR-TB

DR-TB can be acquired de novo during an episode of active TB (acquired resistance) or can be transmitted directly from an individual with DR-TB to a new host (transmitted resistance)[52] (Fig. 2). The proportion of acquired versus transmitted resistance varies by country and public health system[53–57]. Empirical and modelling data suggest that, in high-burden settings, more than 90% of all incident MDR/RR-TB is due to transmitted resistance[53,58]. Transmitted resistance explains at least 46% of all MDR/RR-TB in one pooled estimate across 20 countries, ranging from more than 28% of Inh resistance in the United Kingdom to more than 61% of Inh resistance in South Africa[56,59]. In household contact studies that have employed genotyping to more accurately infer recent transmission, DR-TB was measured to transmit at similar rates to drug-susceptible TB[60]. Several studies have reported longer delays in diagnosis and treatment initiation for MDR/RR-TB compared with drug-susceptible TB[61], suggesting that persons with MDR/RR-TB may remain infectious for longer, further potentiating transmission[62]. This emphasizes that early diagnosis and effective treatment as well as other public health interventions to reduce exposure to DR-TB in hospitals and congregate environments are important for controlling DR-TB.

Resistance acquisition within 2–5 years of initial TB treatment is more common in lower-resource health systems compared with well-resourced health systems, where acquired resistance rarely occurs[56,63]. Directly observed therapy became the standard of care for TB in the early 1990s with the initial goal of decreasing the risk of acquired resistance and improving overall treatment outcomes[64]. Since then, directly observed therapy has been shown to have limited to no effect on resistance acquisition or treatment outcome, and data supports between-patient pharmacokinetic variability as a more important factor[65–67]. The order of drug resistance acquisition in TB is

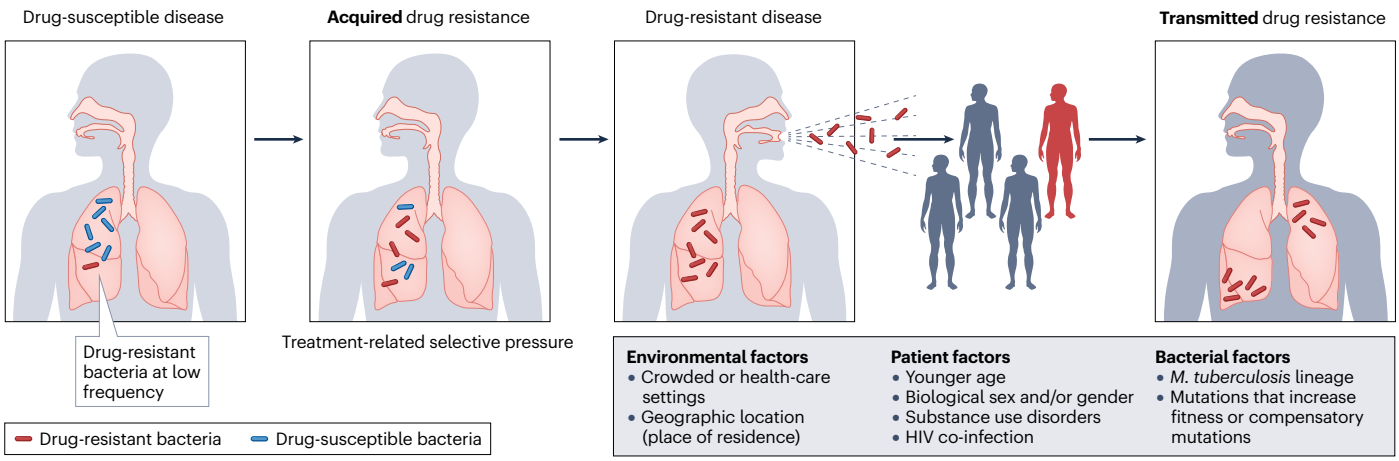


**Fig. 2 | Acquisition and transmission of DR-TB.** Drug-resistant tuberculosis (DR-TB) can be acquired over the course of an episode of active TB undergoing ineffective treatment, where drug-resistant subpopulations (red) can emerge at low frequency and be selected for over drug-susceptible populations (blue) during treatment, which is known as 'acquired resistance'. Persons with active DR-TB can subsequently transmit the disease directly to their contacts through the air (for example, when coughing, sneezing or speaking). Drug-resistant *Mycobacterium tuberculosis* cells released into the air can be inhaled by a susceptible individual, who can develop DR-TB with no prior history of TB treatment (transmitted resistance) (rightmost two panels). Diverse environmental (geographic location, crowded spaces), patient (person's age, biological sex and/or gender, substance use, HIV co-infection) and bacterial (genetic mutations, lineage) factors can contribute to the transmission of DR-TB among individuals.

predictable, starting with Inh, followed by Rif and streptomycin, then ethambutol and pyrazinamide, and lastly fluoroquinolones and injectable agents[54,56,68]. This order relates to the relative bacterial fitness cost of the different resistance mutations and to the usage pattern of the drugs[54,56,68], for example, resistance amplification to second-line drugs occurs largely among isolates already resistant to first-line agents. Molecular dating has demonstrated that streptomycin resistance was commonly acquired around Rif-resistance acquisition, probably amplified by the historical addition of streptomycin when first-line treatment is failing[56]. However, the existence of fluoroquinolone mono-resistant TB, acquired at low rates in South Asia[13,69], and the increasing prevalence of RMR-TB demonstrate that this is not the only possible sequence of resistance acquisition. RMR-TB may be acquired preferentially in specific populations, for example, in persons with HIV co-infection due, possibly, to poor drug absorption or drug–drug interactions[70].

Risk factors for DR-TB can be broadly classified into environmental factors, patient factors and bacterial factors (Fig. 2). Some consistent environmental factors include geographic location and residence or prolonged stay in crowded localities[61,71–77]. Living in areas with a higher prevalence of MDR-TB increases the likelihood of exposure to drug-resistant strains of *M. tuberculosis*. Further, it is recognized that DR-TB does not distribute evenly in high-prevalence geographies; significant spatial heterogeneity within these geographies has been reported, including concentration in correctional facilities[76–78], slums[61,79], migrant or refugee camps, and other settings[71–73,80–82]. Hospitals and other health-care settings can also pose a risk for exposure to DR-TB, especially if there is limited or no use of personal protective equipment[74,75,83–85].

Patient-level risk factors include the affected person's history of prior TB treatment, younger age, biological sex and/or gender, HIV, and alcohol or other substance use disorders[61,73,86–90]. Prior TB treatment associates with higher rates of resistance at 17% (95% UI 11–23%) compared with 3.3% (95% UI 2.6–4.0%) among individuals without prior TB history[22]. This is thought to relate to either acquired resistance during the prior TB treatment episode or to missed primary or transmitted resistance before the prior TB treatment[73,88,89]. The association between younger age and DR-TB is not well understood but it could be attributed to a higher proportion of recently transmitted TB disease among younger individuals possibly compounded by additional risk factors correlated with age such as substance use[61,91]. Reactivation of disease acquired in the past when resistance may have been less prevalent may be more common among older individuals, but recent work casts doubt regarding the role of reactivation in driving the TB epidemic[92]. Although active TB disease is more common in men than in women, several reports suggest higher than expected rates of drug resistance among women, especially young women[61,90,93–95]. Reasons for this are not well understood and raise concerns about biological sex and/or gender disparities in disease risk and/or access to high-quality TB care. Co-infection with HIV has been variably associated with DR-TB infection, possibly mediated by different immune pressures on pathogen populations in host and/or pharmacodynamic factors resulting from drug–drug interactions[89,96]. Substance use disorders, especially of illicit drugs, correlate with gathering in crowded or poorly ventilated spaces[97] and with acquisition of resistance through decreased treatment adherence[98–100].

Bacterial factors include infection with specific *M. tuberculosis* lineages, including modern lineage 2, also known as the Beijing lineage[56,69,101,102]. The association between the *M. tuberculosis* Beijing lineage and drug resistance may relate to intrinsic biological differences between the lineages that facilitate transmission or resistance acquisition due to higher bacterial fitness, gene–gene interactions or ease of acquisition of mutations that compensate for fitness cost related to drug-resistance mutations[69,101,103,104]. In some parts of the world, for example, in the region of the former Union of Soviet Socialist Republics, the association between the Beijing lineage and resistance may relate to founder effects that have both potentiated transmission and resistance acquisition. For instance, limited testing for resistance in congregated settings and nosocomial spread in health-care facilities could have contributed to this association[103]. Ongoing research aims to disentangle the environmental, patient and bacterial factors that have potentiated resistance acquisition and transmission.

## Biological and genetic determinants of drug resistance

### Intrinsic resistance

*M. tuberculosis* has varying degrees of intrinsic resistance to several antibiotic classes, including penicillins, cephalosporins and macrolides[105–107]. Mycobacteria are closely related to *Streptomyces* species and have a similarly diverse set of mechanisms to self-protect from antibiotics[105,108] such as a hydrophobic waxy cell envelope, a low number of water-filled porins[109] and low lipid fluidity of the cell membrane[110], making it impenetrable to a wide range of compounds[105]. Other important intrinsic mechanisms include drug target modification, like methyltransferase modification of ribosomal RNA and intrinsic resistance to ribosome-targeting macrolide antibiotics[111], drug efflux[108], and enzymatic drug inactivation such as the extended-spectrum β-lactamase BlaC[112] (Fig. 3). Clavulanate has been described to inhibit BlaC and restore susceptibility to amoxicillin in vitro, although the combination has been shown to have poor efficacy in clinical trials[113,114]. Carbapenems, including meropenem, are poor substrates for BlaC and retain activity against DR-TB[115] but their efficacy in salvage treatment of XDR-TB is yet to be confirmed[105]. Although there is interest in developing compounds that can reverse or inhibit intrinsic mechanisms of antibiotic resistance in *M. tuberculosis*, no such agent is currently used for the clinical treatment of TB.

### Acquired resistance

The more clinically relevant form of resistance is acquired resistance to drugs in current use. Acquired resistance is mediated by the acquisition of new core genomic DNA changes through mutation or other mechanisms[116]. Acquired resistance in *M. tuberculosis* is thought to be a largely irreversible process, in which reversion back to a wild-type genotype through the loss of the specific acquired mutation is extremely rare. This is thought to relate to the high fitness of the commonly observed drug-resistance variants, with the exception of kanamycin resistance, where reversion of *eis* promoter variants was observed in clinical isolates[117].

The most common acquired resistance mechanism in *M. tuberculosis* is genetic alteration in the drug targets, which have been shown to confer resistance to Rif, Inh, ethambutol, streptomycin, aminoglycosides, fluoroquinolones and Lzd (Fig. 3). Mutations in drug target genes (Table 1) impair drug binding and/or drug action. Another common mechanism is through alteration of drug-activating enzymes that encode resistance to Inh, pyrazinamide, ethionamide, pretomanid and Dlm[118–121] (Table 1). Less common resistance mechanisms include the upregulation of a drug-inactivating enzyme or drug efflux. Enzymes that inactivate the drug, such as Eis acetyltransferase for amikacin and kanamycin, can be upregulated through mutations in the *eis* promoter or an upstream regulator of *eis* such as Whib6 and Whib7, resulting in

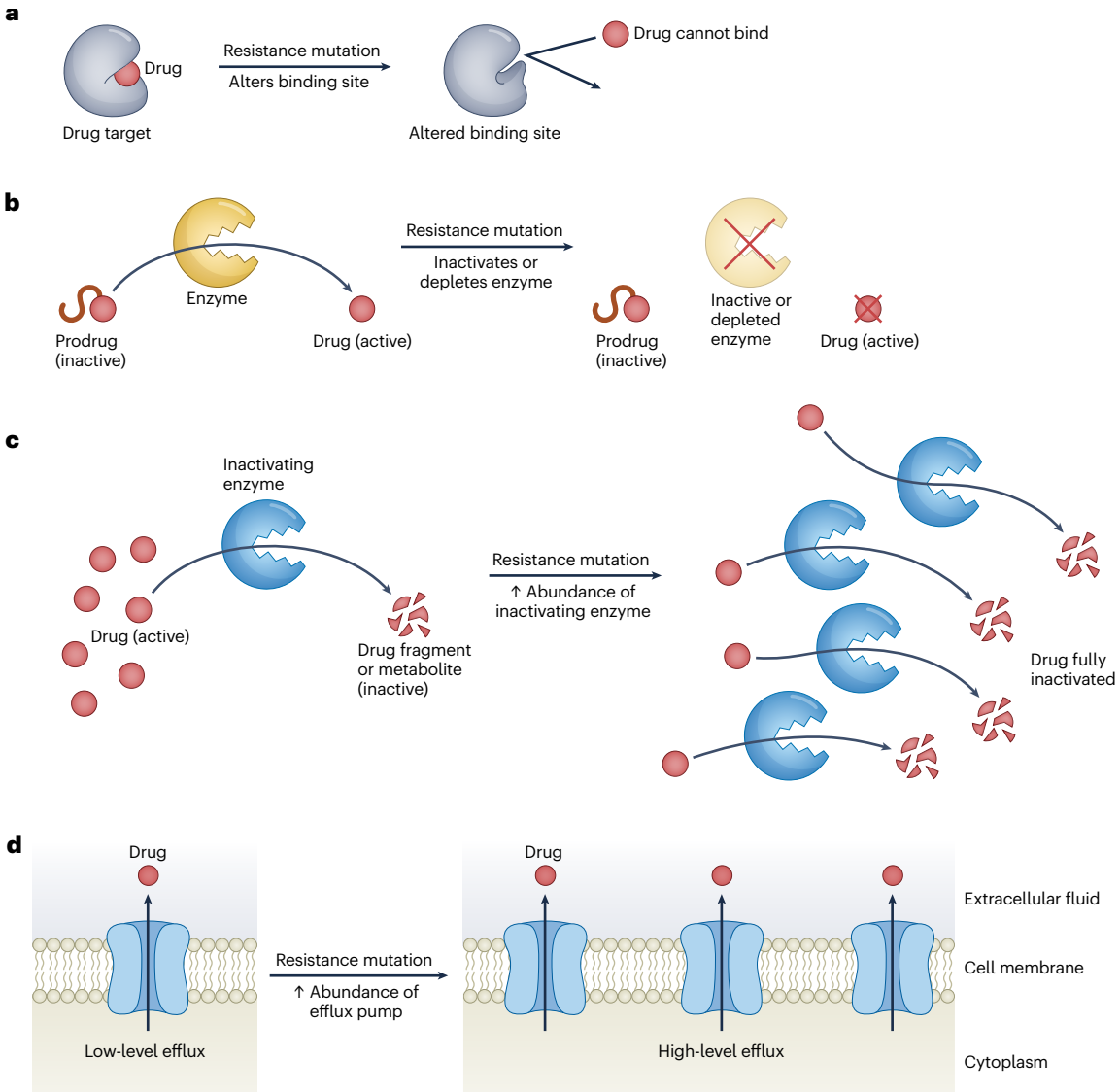


**Fig. 3 | Common mechanisms of drug resistance in *Mycobacterium tuberculosis*.** The diagram outlines the most common mechanisms of acquired drug resistance in *M. tuberculosis*. **a**, A mutation-induced alteration of the bacterial drug target prevents the drug from interacting with its binding site, leading to a loss of drug activity (for example, resistance to rifampicin, pyrazinamide, linezolid and ethambutol). **b**, Inactivation or depletion of the bacterial enzyme responsible for converting an inactive prodrug into its active form, resulting in the drug no longer being activated (for example, resistance to isoniazid, pyrazinamide and ethionamide). **c**, A mutation results in increased abundance of a bacterial enzyme that inactivates the drug, resulting in loss of drug activity (for example, resistance to kanamycin and amikacin). **d**, Enhanced drug efflux, mediated through regulatory variation as a result of frameshifts in a transcriptional repressor (for example, resistance to bedaquiline and clofazimine).

drug inactivation and resistance[122]. The Bdq target is ATP synthetase subunit E (encoded by *atpE*) but mutations in this gene are rare, likely due to the high fitness cost[120]. The most common mechanism of Bdq resistance involves mutations in the *mmpR5* gene (Rv0678), which encodes a transcriptional repressor that downregulates the transmembrane transporter MmpL5-S5 (Rv0676c-Rv0677c). The upregulation of MmpL5-S5 confers low-level resistance to Bdq likely through increased Bdq transport or efflux across the membrane[117,120,123]. Bdq resistance is the only mechanism of acquired resistance that is currently attributable to efflux in *M. tuberculosis*. Cross-resistance between Bdq and Cfz is expected with *mmpR5* mutations[120]. There is evidence that prior Cfz use for TB treatment has increased the population frequency of *mmpR5* mutations[45].

Non-coding variants can also mediate resistance either through downregulating enzymes that mediate prodrug-to-drug activation or upregulating target proteins or inactivating enzymes as in Eis. In general, for the same protein that can be affected by both non-coding variation and coding variation, non-coding resistance mutations result in lower-level antibiotic resistance as seen in the case of *inhA* coding region versus promoter variants for Inh resistance[124].

**Table 1 | Summary of mechanisms of drug resistance in *Mycobacterium tuberculosis***

| Drug | Tier[a] | Gene | Putative drug-resistance mechanism | Assayed by tNGS[b] | Refs. |
|---|---|---|---|---|---|
| Amikacin, capreomycin | 2 | *ccsA* | Unknown | No | 124 |
| Amikacin, capreomycin | 2 | *fprA* | Unknown | No | 124 |
| Amikacin, capreomycin | 2 | *aftB* | Unknown | No | 124 |
| Amikacin, kanamycin | 1 | *eis* | Drug inactivation | Yes | 254 |
| Amikacin, streptomycin, capreomycin | 2 | *whiB6* | Regulation of drug inactivator | No | 124 |
| Amikacin, streptomycin, capreomycin, kanamycin | 1 | *rrs* | Drug target alteration | Yes | 255 |
| Amikacin, streptomycin, kanamycin | 1 | *whiB7* | Regulation of drug inactivator | No | 255 |
| Bedaquiline | 1 | *atpE* | Drug target alteration | No | 120 |
| Bedaquiline, clofazimine | 1 | *pepQ* | Unknown | No | 120 |
| Bedaquiline, clofazimine | 1 | *mmpLS5-Rv0678* (*mmpR5*) | Drug efflux | Yes | 120 |
| Bedaquiline, clofazimine | 2 | *Rv1979c* | Unknown | No | 120 |
| Capreomycin | 1 | *tlyA* | Altered drug target modification required for drug effect | Yes | 255 |
| Delamanid, pretomanid | 1 | *ddn* | Loss of prodrug activation | No | 256 |
| Delamanid, pretomanid | 1 | *fbiABC* | Loss of prodrug activation | No | 256 |
| Delamanid, pretomanid | 1 | *Rv2983* (*fbiD*) | Loss of prodrug activation | No | 256 |
| Delamanid, pretomanid | 1 | *fgd1* | Loss of prodrug activation | No | 256 |
| Ethambutol | 1 | *embCAB* | Drug target alteration | Yes: *embB* only | 257,258 |
| Ethambutol | 2 | *embR* | Regulation of drug target | No | 259 |
| Ethambutol | 2 | *ubiA* | Competition with drug for target binding | No | 258 |
| Ethionamide | 2 | *Rv3083* (*mymA*) | Loss of prodrug activation | No | 260 |
| Ethionamide | 1 | *ethA* | Loss of prodrug activation | Yes | 260,261 |
| Ethionamide | 2 | *ethR* | Regulation of prodrug activator | No | 260,261 |
| Ethionamide | 2 | *mshA* | Loss of prodrug activation | No | 262 |
| Isoniazid | 1 | *ahpC*[d] | Compensatory | Yes | 263 |
| Isoniazid | 1 | *furA–katG* | Loss of prodrug activation | Yes: *katG* only | 255 |
| Isoniazid, ethionamide | 1 | *inhA* | Drug target alteration | Yes | 255,264 |
| Isoniazid, ethionamide | 1 | *fabG1*, *inhA* promoter | Regulation of drug target | Yes | 255,264 |
| Isoniazid, ethionamide | 2 | *ndh* | Loss of prodrug activation | No | 265 |
| Isoniazid, pyrazinamide, streptomycin | 2 | *Rv1258c* | Unknown | No | 266,267 |
| Isoniazid, rifampicin | 2 | *Rv2752c* (*RNase J*) | Unknown[c] | No | 124,137 |
| Linezolid | 1 | *rplC* | Drug target alteration | Yes | 120 |
| Linezolid | 1 | *rrl* | Drug target alteration | Yes | 120 |
| Moxifloxacin, levofloxacin | 1 | *gyrB-gyrA* | Drug target alteration | Yes | 255 |
| Pyrazinamide | 1 | *clpC1* | Drug target degradation | No | 268 |
| Pyrazinamide | 1 | *panD* | Drug target alteration | No | 268 |
| Pyrazinamide | 1 | *pncA* | Loss of prodrug activation | Yes | 269 |
| Pyrazinamide | 2 | *PPE35* | Unknown | No | 124 |
| Pyrazinamide | 2 | *Rv3236c* (*kefB*) | Unknown | No | 124 |
| Rifampicin | 1 | *rpoB*[d] | Drug target alteration | Yes | 255 |
| Streptomycin | 1 | *rpsL* | Drug target alteration | Yes | 255 |
| Streptomycin | 1 | *gid* | Altered drug target modification required for drug effect | Yes | 255,270 |

Table is based on the Catalogue of mutations in *Mycobacterium tuberculosis* Complex and Their Associations with Drug Resistance, 1st edition[271]. tNGS, targeted next-generation sequencing. [a]Based on the WHO mutation catalogue expert review[125,271]. [b]Deeplex-MycTB assay (GenoScreen). [c]Associated with drug tolerance in ref. 137 but role in clinical resistance not known. [d]*rpoC* and *rpoA* have been shown to be associated with resistance compensation for fitness cost without evidence of a direct effect on resistance[272].

Resistance implicates more than 40 genetic loci, with the mechanism not being understood for some of them[125] (Table 1 and Supplementary Table 1). There are several open questions about *M. tuberculosis* resistance mechanisms, including the limited sensitivity of resistance prediction based on known resistance mutations, especially for second-line or recently introduced drugs[125]. Additionally, phenotypic DST is prone to suboptimal thresholding due to uncertainties in critical concentrations, making it a less-than-optimal benchmark for genotypic tests for all drugs[126,127].

Better quantification of interactions between mutations (epistasis) in the same or different loci[117,122,128] is needed to improve resistance prediction[118,129–132]. For example, MmpR5 loss-of-function mutations lead to Bdq resistance by themselves but, if a loss-of-function mutation is present in the efflux pump encoded by *mmpL5-S5*, the isolate is Bdq hyper-susceptible irrespective of MmpR5 function[117].

Other newly recognized phenomena are genotypic drug tolerance and resilience, in which mutations can have measurable changes in antibiotic effect without affecting the minimal inhibitory concentration[133–137]. For example, a recent transposon mutagenesis screen identified *cinA* as a mediator for multidrug tolerance to first-line and second-line antitubercular drugs[136]. These effects manifested in differential growth or metabolism rates in the presence of drugs, as observed with mutations in *glpK* and *prpR*. Another effect was a quicker recovery after transient antibiotic stress. However, the clinical significance of these experimental observations remains unclear.

Mixed populations of wild-type and resistance-associated variants in a single clonal TB infection, also known as hetero-resistance, have been increasingly described in the literature[138]. The detection of hetero-resistance has been enabled by the ability of high-throughput sequencing to capture low-frequency populations in pathogen isolates, and its frequency may depend on the drug and its usage in the clinic. These subpopulations, especially when present at very low frequencies, are thought to be transient but their frequency can increase due to positive selection, resulting in fixation of resistance variants in the right environment[139–141]. According to current literature, a variant frequency exceeding 20% is considered likely to be clinically relevant and predictive of subsequent fixation. However, the significance of variants with lower frequencies and the management of hetero-resistance observed before or during treatment are not yet clear[139–141].

## Diagnosis of DR-TB

In 2023, the WHO published a standard[142] that recommends universal access to WHO-recommended rapid molecular diagnostics for testing individuals with presumptive TB as well as universal DST at least for Rif for all cases of microbiologically confirmed TB and for fluoroquinolone for persons with proven RR-TB[143]. However, 60% of DR-TB remains undetected globally[22]. Universal access to testing that can accurately and comprehensively diagnose drug resistance is an important step to improve treatment outcomes and control of DR-TB[144,145]. Resource and infrastructure limitations and scant practice implementation are key challenges to universal access to DST[146].

Culture-based DST has been the gold standard for decades but is limited by availability and long turnaround times ranging from 2 to 8 weeks[147]. The only recommended method for solid culture-based DST defines resistance as ≥1% of growth observed at the lab-defined critical concentration of the drug in comparison to an inoculum on a control plate; this process can take up to 42 days[148]. Liquid culture systems like BACTEC MGIT can shorten this time frame to approximately 10 days and are globally standardized but fail to detect clinically significant resistance in certain cases of RR-TB, which has major implications on treatment decisions[148–151].

In the past decade, rapid molecular tests have emerged, and several NAATs are currently endorsed by the WHO (Table 2). Low-complexity NAATs are recommended by the WHO as an initial

**Table 2 | WHO-endorsed and emerging tests for tuberculosis and drug-resistant tuberculosis**

| Methodology | Drug detection | Time | Sample |
|---|---|---|---|
| **WHO-endorsed tests** | | | |
| Xpert, GeneXpert Edge | Rifampicin, isoniazid, second-line injectables, fluoroquinolones | Less than 2 h | Sputum and more than 10 other specimen types |
| Line probe assay | Rifampicin, isoniazid, second-line injectables, fluoroquinolones, pyrazinamide, ethambutol | 5 h to 1 day (GenoScholar PZA-TB[a]) | Sputum and culture |
| Loop-mediated isothermal amplification | Possibility for resistance detection | Less than 1 h | Sputum |
| Truelab | Rifampicin (isoniazid, second-line injectables, fluoroquinolones and linezolid are under development) | Less than 1 h | Sputum |
| Centralized drug-sensitivity testing (Roche or Abbott) | Rifampicin, isoniazid, second-line injectables, fluoroquinolones | 8 h or less | Sputum, culture, bronchial alveolar lavage, sediment |
| Targeted sequencing | Rifampicin, isoniazid, second-line injectables, fluoroquinolones, pyrazinamide, ethambutol, linezolid, bedaquiline, clofazimine | 2–3 days | Sputum or early positive culture |
| **Emerging technologies[b]** | | | |
| Point-of-care nucleic acid amplification test | Rifampicin, isoniazid, second-line injectables, fluoroquinolones | Less than 2 h | Sputum |
| Whole-genome sequencing | Rifampicin, isoniazid, second-line injectables, fluoroquinolones, pyrazinamide, ethambutol, linezolid, bedaquiline, clofazimine, ethionamide | 2–3 days | Early positive culture |

[a]The specified time is specific for the GenoScholar PZA-TB that detects pyrazinamide resistance. [b]Tests that are not yet endorsed by the WHO but are under development or evaluation.

diagnostic test for detection of TB and RR-TB in sputum over smear microscopy or culture and phenotypic DST[4]. Globally, the most widely used test is the Xpert MTB/Rif (Cepheid, Sunnyvale, USA, first recommended in 2010)[152,153]. A follow-on development that improved TB diagnostic sensitivity is the Xpert MTB/Rif Ultra, particularly in the settings of paucibacillary disease or HIV infection but at the expense of lower specificity[154] (Supplementary Table 2). Broader use of Xpert or Xpert Ultra has been limited by the requirements for stable electrical power supply, temperature control, cartridge supply, cost, lack of quality assurance and maintenance support[154–156]. The real-time PCR Truenat assay (Molbio Diagnostics, Goa, India) is a WHO-recommended alternative to Xpert or Xpert Ultra, with the battery-operated platform enabling testing closer to the person affected by TB[4,157,158]. The platform is currently primarily used in India but, with its expansion to additional drugs including Inh[159], slightly lower operating costs, and ability to reflex into sequencing from the extracted eluate, it is also slowly getting uptake worldwide. The use of Xpert has improved RR-TB diagnosis[155]. However, its effect on overall TB mortality has been modest[160–162]. The lack of impact is potentially due to empiric treatment of suspected cases and ineffective linkage to care for diagnosed persons. There is a current lack of implementation research aimed at designing solutions to deal with these challenges[163,164].

The Xpert MTB/XDR (Cepheid, Sunnyvale, USA) is a 90-min follow-on test that can differentiate high-level and low-level resistance to Inh and fluoroquinolones as well as resistance to second-line injectable drugs (amikacin, kanamycin and capreomycin) with, on average, moderate-to-high diagnostic accuracy in comparison to other laboratory-based molecular tests[165,166].

Another group of tests are the moderate-complexity automated NAATs that operate on laboratory-based instruments, which require more infrastructure and technical skill. The limited infrastructure for sample transportation and result reporting has led many countries with a high TB burden to limit the use of these centralized tests for diagnosis. They have been recommended by the WHO based on comparable performance to Xpert[4]. In comparison to laboratory requirements for phenotypic culture-based DST, these assays require a lower biosafety level (2 versus 3) and offer test results in a few hours as well as high throughput. Furthermore, the platforms can multiplex and leverage economies of scale, making them attractive where sample transportation to a central laboratory is feasible[167]. The four recommended tests may be used for the detection of *M. tuberculosis* as well as resistance to both first-line TB drugs Rif and Inh (all targeting the Rif resistance-determining region of the *rpoB* gene for Rif, and the *katG* and *inhA* regions for Inh, with slightly varying exact targets)[4,167] (Supplementary Table 2).

Line probe assays have lower complexity than the centralized assays described above and can be performed in intermediate laboratories (biosafety level 2 to 3)[4,168,169]. Results can be read in 5 h (ref. 4) (Supplementary Table 2). In addition to the tests available for Rif, Inh, fluoroquinolones, ethambutol and injectables, the WHO also recently recommended the use of the LPA Genoscholar PZA-TB II assay (Nipro, Osaka, Japan) for the detection of resistance to pyrazinamide[4,170,171].

Integrated molecular assays for critical DR-TB drugs such as Bdq and Lzd are not yet available but are in development as understanding of the genotypic correlates for drug resistance matures[120,172–175]. Several expert groups and the WHO provided a consensus mutation catalogue that should form the basis of genotype-based assays[118,125,172,176,177]. The guidelines were recently updated to recommend specific tNGS technologies based on a WHO-commissioned systematic review of published and unpublished data. However, the current accuracy for prediction of resistance to critical DR-TB drugs Bdq and Lzd is suboptimal, with sensitivity being 68% and 69%, respectively[178]. Such levels of accuracy are inadequate to substitute culture-based DST until further advances are made. To avoid overcalling drug-resistance-associated mutations, generalizable association methods that balance the sensitivity and specificity of the final genotypic prediction are vital to advance genotypic DST for all drugs[125].

Next-generation sequencing (NGS) is an adaptable approach for rapid and comprehensive resistance diagnosis by detecting known mutations in either targeted genes or the whole genome. The workflow conceptually includes four steps: DNA extraction with quality control, library preparation, sequencing and data analysis[179]. The commercially available NGS platforms suitable for clinical use for DR-TB diagnosis include the Illumina MiSeq, the ThermoFisher Scientific Ion Torrent Personal Genome, the Qiagen GeneReader NGS system and the Oxford Nanopore MinION, with the latter having the advantage of a minimal equipment footprint, which, however, comes with a higher cost of consumables[179]. Due to the lack of consensus for interpretation and the still limited evidence of its performance in real-world settings, the implementation of Nanopore in clinical practice is yet to be established[180].

Whole-genome sequencing (WGS) can detect all known genetic determinants of drug resistance (including for new and repurposed drugs) and is easily adaptable to detect and discover new resistance mutations such as the I491F mutation in *rpoB*, which is not detectable by NAAT assays[181,182]. It also provides additional information on compensatory mutations, lineage, hetero-resistance, epistasis and transmission relatedness[118,125,183–185] (Table 1). The use of WGS for the detection of *M. tuberculosis* drug resistance may resolve discordance in phenotypic and target-specific genotypic tests[186,187]. WGS is currently performed from cultured isolates of *M. tuberculosis* as it needs a relatively high quantity of good-quality DNA to generate full WGS data, which may preclude widespread adoption due to the delay associated with culture. Nonetheless, WGS results can be generated a median of 9 days faster than final reference laboratory reports and at a cost 7% cheaper than present diagnostic workflows[177]. Attempts to perform direct WGS from uncultured sputum have had limited success in sputum with low bacterial load[188–191]. Sputa with smaller amounts of bacteria have been successfully sequenced by adding a bait enrichment step[191], which remains prohibitively expensive to scale up for clinical use. Improved sample processing is needed to replace the entire phenotypic DST workflow process with WGS, but a phase IV trial is currently studying the feasibility of using WGS approaches to guide treatment for RR-TB[192].

Sequencing directly from uncultured clinical samples, such as sputum or stool, has a faster turnaround time; however, low bacillary samples remain problematic[193]. tNGS can be implemented through either an amplicon-based assay or a hybridization or capture-based assay to sequence full-length genes with high depth, allowing for the detection of low-frequency resistance mutations. tNGS provides rapid sequence information for a greater number of loci than existing molecular tests (Supplementary Table 2). Currently in the TB diagnostic pipeline for DR-TB detection directly from sputum samples is the Deeplex-MycTB assay (GenoScreen) used for identification of mycobacterial species and prediction of drug resistance for species within the *M. tuberculosis* complex (detection of 18 resistance-associated gene targets). Other promising platforms include Oxford Nanopore Technology ONT EPI 2ME (detection of 16 resistance gene targets) and Deepcheck ABL (detection of 13 gene targets). tNGS seems to be a highly appealing

**Table 3 | Key second-line medications used to treat drug-resistant tuberculosis**

| WHO drug group | Drug | Recommended dosing | Primary adverse events | Recommended monitoring | Comments |
|---|---|---|---|---|---|
| Group A | Bedaquiline | Adults: 400 mg daily for 14 days, then 200 mg thrice weekly OR 200 mg daily for 8 weeks, then 100 mg daily[222] | QTcF prolongation (occasional), elevated transaminases (uncommon)[222] | ECG at baseline and at least at 2, 12 and 24 weeks for QTcF prolongation, liver function tests at baseline, then monthly[222] | Can be safely extended beyond 24 weeks[1]; alternative dosing strategies are being assessed; cannot be given with efavirenz[2]; paediatric formulation of 20-mg tablet available[2,50] |
| | Linezolid | 600 mg daily[222] | Bone marrow suppression (early in treatment), optic neuritis and peripheral neuropathy (later in treatment, usually after 8 weeks), lactic acidosis (occasional), diarrhoea and nausea (common)[222] | Baseline, weekly then monthly complete blood count, visual acuity and peripheral neuropathy screening every 2 months[222] | Doses above 600 mg daily are associated with higher rate of toxicity; alternative dosing strategy of lowering dose to 300 mg daily or 600 mg thrice weekly was assessed in TB-PRACTECAL study[2]; paediatric formulation of 150-mg tablet available[222] |
| | Levofloxacin or moxifloxacin | 750–1,000 mg daily for levofloxacin[222]; 400–800 mg daily for moxifloxacin[222] | QTcF prolongation (occasional), arthralgia (common), Achilles tendon rupture (occasional), endovascular toxicity, especially in the elderly (uncommon)[222] | ECG at baseline and at least at 2, 12 and 24 weeks for QTcF prolongation, especially if used with other QTcF-prolonging agents[222] | Levofloxacin less likely to cause QTcF prolongation[222]; paediatric formulations of both are available[222] |
| Group B | Clofazimine | 100 mg daily[222] | Skin hyperpigmentation (common), QTcF prolongation (frequent), vomiting or gastrointestinal intolerance (uncommon)[222] | Counselling about skin pigmentation, baseline and monthly ECG to assess for QTcF prolongation if used with other QTcF-prolonging agents[222] | Skin changes may lead to inadvertent disclosure and be distressing to people receiving treatment[2]; can have cross-resistance with bedaquiline[2]; paediatric formulation of 50-mg tablet available[222] |
| | Cycloserine or terizidone | 10–15 mg/kg/day[222] | Neuropsychiatric effects (common), psychosis (frequent), seizures (occasional)[222] | Peak concentrations should be monitored throughout treatment, baseline and monthly depression screening[222]; counselling and emotional support provided on an ongoing basis | May lead to increased neuropsychiatric effects when used with delamanid or efavirenz[222] |
| Group C | Ethambutol | 15–25 mg/kg/day[222] | Optic neuropathy (occasional), liver toxicity (uncommon)[222] | Baseline and monthly visual acuity and colour discrimination screening; counselling about vision[222] | Should only be used if susceptibility is confirmed or as part of a standardized regimen; paediatric formulation of 100-mg tablet available[222] |
| | Delamanid | 100 mg twice a day[222] | Neuropsychiatric effects (occasional), mild QTcF prolongation (occasional)[222] | Albumin levels before treatment initiation; ECG at baseline; baseline and monthly depression screening and counselling important if taken with cycloserine or efavirenz[222] | May lead to increased neuropsychiatric effects when used with cycloserine, terizidone or efavirenz[222]; can be safely extended beyond 24 weeks[2]; nitroimidazole agent of choice for people less than 14 years of age or pregnant people[2]; paediatric formulation of 25 mg available[50,222] |
| | Pyrazinamide | 20–30 mg/kg/day[222] | Hyperuricaemia (common) potentially leading to gout (occasional), arthralgia (frequent), hepatotoxicity (occasional)[222] | Liver function tests at baseline and monthly[222] | Should only be used if susceptibility is confirmed or as part of a standardized regimen[2]; paediatric formulation of 150-mg tablet available[222] |
| | Meropenem or imipenem-cilastatin (both plus clavulanic acid) | 1 g thrice daily or 2 g twice daily for meropenem[222]; 1 g twice a day for imipenem-cilastatin[222] | Gastrointestinal upset (common), thrush (common), pseudomembranous colitis (occasional), seizures (uncommon)[222] | No routine assessment needed[222] | Must be given intravenously; must be given 30–60 min after oral 500 mg of amoxicillin plus 125 mg of clavulanic acid (orally)[222] |

**Table 3 (continued) | Key second-line medications used to treat drug-resistant tuberculosis**

| WHO drug group | Drug | Recommended dosing | Primary adverse events | Recommended monitoring | Comments |
|---|---|---|---|---|---|
| Group C (continued) | Amikacin or streptomycin | 10–20 mg/kg/day up to 1,000 mg/day[222] | Proteinuria (common), nephrotoxicity (occasional), ototoxicity (occasional), cranial nerve VIII toxicity (occasional)[222] | Must have baseline and monthly audiometry; baseline and monthly renal function testing[222] | Should only be used in rescue regimens and if susceptibility is confirmed[2]; should not be routinely used for drug-resistant tuberculosis treatment[2] |
| | Ethionamide or protionamide | 15–20 mg/kg/day up to 1,000 mg/day[222] | Gastrointestinal intolerance (common), hepatotoxicity (occasional), hypothyroidism (occasional), gynaecomastia (uncommon)[222] | Baseline TSH and then every 3 months thereafter[222] | Should only be used when no other options[2] as it was associated with worse treatment outcomes in some studies[273] |
| | Para-aminosalicylic acid | 8–12 g per day divided into 2 or 3 doses[222] | Gastrointestinal upset (common), hypothyroidism (frequent), hepatotoxicity (uncommon), coagulopathy (uncommon)[222] | Baseline and monthly liver function testing; baseline TSH and then every 3 months thereafter[222] | Should only be used when no other options[2] as it was associated with worse treatment outcomes in some studies[273]; must be administered with acidic food[222] |
| Ungrouped drugs | Pretomanid | 200 mg daily[222] | Headache (common), gastrointestinal upset (common), hepatotoxicity, myelosuppression, testicular or reproductive toxicity (animal studies)[222] | Liver function tests at baseline, 2 weeks and monthly after; ECG and baseline electrolytes[222] | Has only been tested in combination with other medications and should not be used or added to regimen combination in which it was not tested[2]; should not be given to children under age 14 years or pregnant people[2] |
| | High-dose isoniazid | 10–15 mg/kg/day up to 600 mg (ref. 222) | Peripheral neuropathy (frequent), gastrointestinal upset (frequent), abnormal liver function tests (frequent), hepatitis (occasional), arthralgia (occasional), severe hypersensitivity (uncommon), drug-induced lupus (uncommon)[222] | Baseline liver function testing and monitoring as required[222] | Must be given in combination with vitamin $B_6$ (ref. 222) |

The table describes the recommended dosing and monitoring for 14 drugs or classes in current use for the treatment of drug-resistant tuberculosis. ECG, electrocardiogram; QTcF, corrected QT interval; TSH, thyroid-stimulating hormone.

approach, providing a per-sample turnaround time of 3 days directly from clinical samples to confirm drug resistance. It offers economy of scale in the short term, which makes it affordable and accessible in resource-limited settings. However, the need for batching currently limits the turnaround time of this method to at least 24 days[194].

Although data suggest considerable benefits could accrue from routine access to WGS-derived resistance prediction in high-TB-burden settings[195], uptake in such settings is hindered by integration into existing workflows, technical training and expert guidance regarding interpretation of sequencing data[196]. Currently, routine WGS or tNGS are not seen as affordable for high-burden countries. In July 2023, the WHO issued a rapid guidance document[178] on the use of tNGS for DR-TB and recommended that tNGS can be considered an alternative for prioritized target populations requiring comprehensive DST with faster results compared with phenotypic DST or where access to phenotypic DST is limited. This was followed by a document in October 2023 that provided guidance on implementation to accelerate scale-up[179].

## Treatment of DR-TB

Recently, the treatment of MDR/RR-TB has been radically transformed by the development of all-oral shorter regimens — the result of several trials and studies showing that different combinations of Bdq-containing all-oral regimens improve treatment success and/or reduce the risk of in-treatment mortality compared with injectable-containing regimens[18,197–207]. In the context of clinical trial conditions, it is possible to achieve treatment outcomes comparable to those observed in cases of drug-susceptible TB[198]. The WHO issued a conditional recommendation for a standardized regimen to treat DR-TB as well as strategies for designing personalized regimens based on a hierarchical categorization of second-line medications according to their efficacy and safety[2,208]. Group A drugs are associated with improved treatment outcomes and decreased mortality, group B drugs are associated with improved outcomes with a less clear impact on mortality, and group C drugs are associated with varying outcomes[18]. A minimum of four to five drugs should be used in combination to design a treatment regimen (Table 3).

The WHO-recommended standard 6-month regimen for MDR/RR-TB consists of four drugs based on the results of the phase II–III TB-PRACTECAL trial (Clinicaltrials.gov, NCT02589782)[2,209]. The recommended regimen —abbreviated as the 'BPaLM' regimen — is Bdq (400 mg daily for 14 days followed by 200 mg three times a week for 22 weeks), pretomanid (200 mg daily for 24 weeks), Mfx (400 mg daily for 24 weeks) and Lzd (600 mg daily for 16 weeks followed by either 300 mg daily or 600 mg thrice weekly for the remaining 8 weeks)[198]. The trial was halted early by the data safety monitoring board after results showed that 89% of people in the study arm were successfully treated

## Glossary

**Directly observed therapy**
Refers to the delivery of anti-tuberculosis drug treatment under direct observation of health workers, community workers or family members with the goal of improving adherence.

**Extensively drug-resistant TB**
(XDR-TB). Defined as multidrug-resistant or rifampicin-resistant tuberculosis with further resistance to fluoroquinolones and to either bedaquiline or linezolid or both (key second-line drugs).

**Inh-resistant and Rif-susceptible TB**
(Hr-TB). Defined as resistance to isoniazid and susceptibility to rifampicin.

**Multidrug-resistant TB**
(MDR-TB). Defined as resistance to rifampicin and isoniazid, the two most important first-line drugs used to treat tuberculosis (TB), regardless of resistance to other TB drugs.

**Pre-XDR-TB**
Defined as multidrug-resistant or rifampicin-resistant tuberculosis with resistance to fluoroquinolones.

**Rif mono-resistant TB**
(RMR-TB). Defined as resistance to rifampicin (Rif), with susceptibility to isoniazid.

**Rif-resistant TB**
(RR-TB). Defined as resistance to rifampicin (Rif), regardless of resistance to other tuberculosis (TB) drugs. Individuals with RR-TB are treated with regimens similar to those for multidrug-resistant TB (MDR-TB) and are therefore grouped with MDR-TB as MDR/RR-TB.

compared with only 52% in the standard-of-care arm[210]. The BPaLM regimen is recommended for people over the age of 14 years who have not been previously treated with Bdq, Lzd or a nitroimidazole agent (Dlm or pretomanid), and for people whose TB strains have known or likely susceptibility to fluoroquinolones. At the 72-week trial time point, individuals who received at least one dose of BPaLM regimen exhibited a sharp decrease in severe adverse events compared with standard care[198].

The WHO also recommended another 6–9-month regimen consisting of Bdq, pretomanid and Lzd (600 mg daily) for persons whose strains of TB have known or likely resistance to fluoroquinolones[2]. This recommendation is based, in part, on the PRACTECAL study (which showed similar outcomes among people whose strains of TB had fluoroquinolone resistance and those whose strains did not, although the numbers were small) and on the non-randomized, non-controlled regimens tested in the Nix-TB and ZeNix-TB studies (NCT02333799 and NCT03086486 (refs. 211,212), which showed treatment success rates of approximately 90% even when a lower dose of Lzd was used; 600 mg daily as opposed to 1,200 mg daily)[197]. The WHO also made a conditional recommendation for a standardized 9-month regimen for people whose strains of TB have known or likely susceptibility to fluoroquinolones. This regimen is based on programmatic data from South Africa[203] and consists of Bdq (400 mg daily for 14 days followed by 200 mg three times a week for 22 weeks), Lfx (15–20 mg/kg daily for 9 months), Cfz (100 mg daily for 9 months), Lzd (600 mg daily for 8 weeks), pyrazinamide (20–30 mg/kg daily for 9 months), ethambutol (15–25 mg/kg daily for 9 months) and high-dose Inh (10–15 mg/kg per day for 6 months). The regimen is recommended for people above the age of 6 years with non-severe disease.

A number of other shorter regimens for DR-TB treatment are currently being assessed either in operational research cohorts or in randomized trials[213] and are outlined in Supplementary Table 3. The MDR-END randomized controlled trial (NCT02619994)[214] conducted in South Korea assessed a shorter, Bdq-sparing regimen consisting of Dlm (100 mg twice daily), Lzd (600 mg daily for 8 weeks, followed by 300 mg daily or 600 mg every other day), Lfx (750–1,000 mg daily) and pyrazinamide (1,000–2,000 mg daily) given for 9–12 months. Preliminary data show that 75.0% of the 72 people who received the shorter regimen had a successful outcome compared with 70.6% of the 85 people who received the longer, standard-of-care regimen, showing the non-inferiority of yet another shorter regimen for DR-TB[215]. A preliminary report for the BEAT-TB regimen (NCT04062201)[216] in which Dlm was given in combination with Bdq, Lzd and Cfz showed a, thus far, 91% treatment success rate and offers a fluoroquinolone-free regimen for individuals unable to receive this category of medication (due to resistance or intolerance)[199]. The endTB clinical trial (NCT02754765)[217] is a phase III, multi-site randomized controlled trial testing multiple 6–9-month regimens for DR-TB with various combinations of Bdq, Dlm, Lfx, Lzd and/or Cfz[218]. The endTB-Q trial is a phase III randomized controlled trial assessing a regimen of Bdq, Dlm, Lzd and Cfz for people whose strains of TB have known or likely resistance to fluoroquinolones[219]. These two randomized trials will provide the highest-quality evidence for the use of all-oral shorter regimens for DR-TB, and published results are expected in 2024. To complement these more formal studies, multiple countries are conducting operational research to assess a 6–9-month regimen of group A and B drugs (Bdq, Lfx, Lzd, Cfz, and cycloserine or terizidone)[220] (Supplementary Table 3). Ongoing trials that utilize novel methodologies to assess the combination of newer compounds also hold great promise for continued improvements in treatment[221].

Individuals with previous exposure to or whose strains have known resistance to Bdq, Lzd, and/or the nitroimidazole agents (that is, Dlm or pretomanid) require the use of individualized rescue regimens. Longer (for example, 18–24 months) individualized regimens should also be given to people with severe forms of DR-TB, including disseminated disease, osteoarticular DR-TB and DR-TB meningitis[222].

Figure 4 summarizes regimens for DR-TB by target population. These recommendations consist of the updated WHO recommendations when such recommendations exist[2,222]. Recommendations for children and pregnant individuals are based on expert consensus for groups not covered under the WHO guidelines[223,224]. The WHO now advises that Bdq and Dlm can be given to children of all ages[50]. Pretomanid is only recommended for non-pregnant persons over the age of 14 years as animal studies suggest reproductive toxicity and safety data in humans is still pending. Although they were not included in the clinical trials supporting the 6-month BPaLM regimen, children and pregnant people should be offered other all-oral shorter regimens. For children, the duration of the regimen can be based on the extent of disease. For children with non-severe disease, such as isolated lymphadenitis or unilateral, non-cavitary pulmonary disease, regimens of 6–9 months may be considered. This is based on data from the SHINE trial, which focused on drug-susceptible TB in children with non-severe disease[225] extrapolated to children with DR-TB[226]. Child-friendly formulations of second-line drugs are available and should be given to children under the age of 6 years[227]. People living with HIV can also be treated with the recommended shorter regimens. They should be prioritized to receive dolutegravir-based antiretroviral therapy since Bdq cannot be given with efavirenz, and they should be monitored closely for overlapping toxicity[222].

A recent meta-analysis showed that individuals with Hr-TB who are treated with standard first-line TB regimens have a higher risk of

developing additional Rif resistance compared with individuals with drug-susceptible TB, with subsequent poor treatment outcomes[228,229]. This supports the more recent recommendations to treat Hr-TB with a modified first-line regimen that includes fluoroquinolones[20].

Ultimately, regimen composition and length of treatment for DR-TB are guided by knowledge of the resistance pattern of the *M. tuberculosis* strain of an individual[230]. Given the high risk of poor treatment outcomes, high risk of side effects and prevalence of comorbidities, offering person-centred care for this population is an essential part of DR-TB treatment and control. This may include additional time spent with the provider for educating the patient about the disease and its causes, providing other forms of treatment counselling and adherence support throughout care as well as connecting patients to survivor support networks, managing expectations of adverse events, and addressing socioeconomic needs and barriers to treatment success.

## Preventive therapy for DR-TB

Identified contacts of known RMR-TB cases can be prescribed Inh preventive monotherapy over a period of 6 months or more[231]. The WHO conditionally advises preventive treatment of household contacts of known MDR-TB cases based on individualized risk assessment[231]. There is uncertainty around the optimal regimen for TB prevention among contacts of persons with MDR-TB and its efficacy. Some country guidelines recommend a fluoroquinolone-based preventive regimen for contacts of patients with MDR-TB. The WHO provides a conditional recommendation on preventive treatment based on evidence for fluoroquinolone-based preventive and tailored treatment approaches[232,233]. Most tested regimens to date range from 6 to 9 months in duration and include a fluoroquinolone[234]. Pyrazinamide-containing regimens have substantial adverse effects and are, hence, less favoured than a fluoroquinolone-containing regimen[234]. The use of first-line

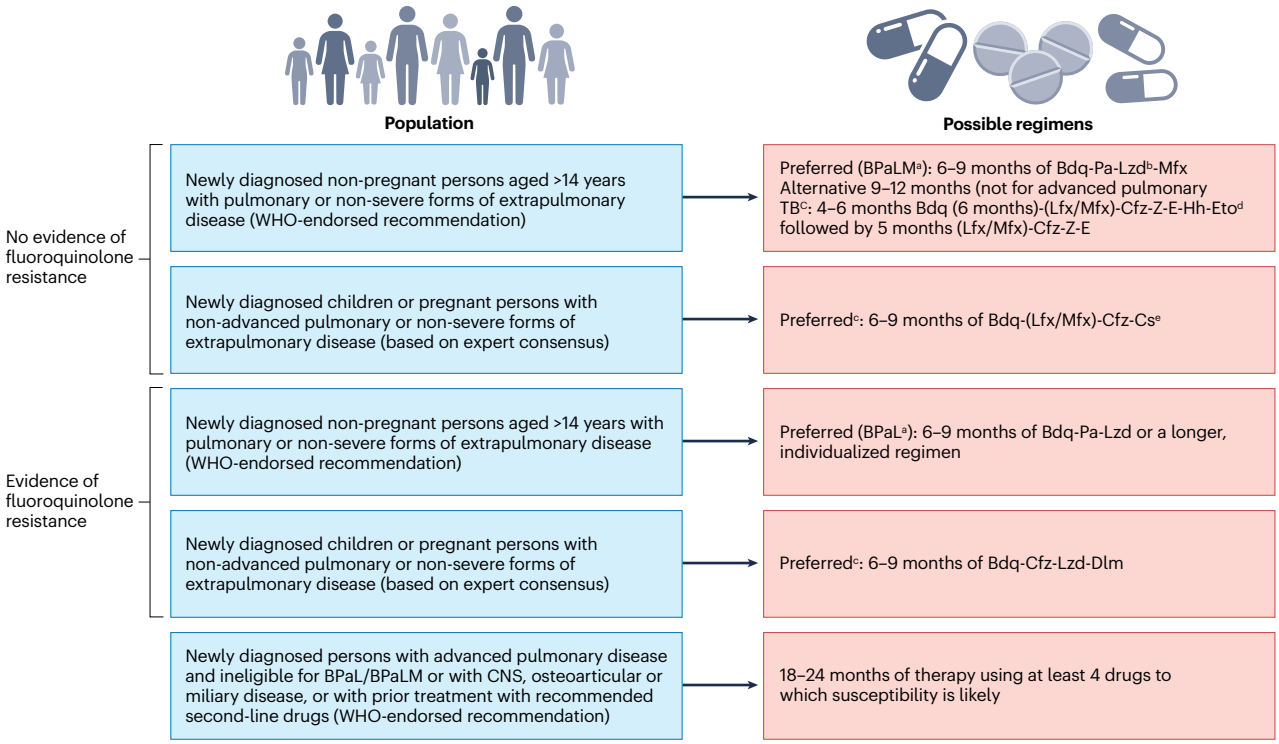


**Fig. 4 | Possible regimens for populations living with drug-resistant tuberculosis.** The schematic outlines target populations defined by prior treatment, age, pregnancy status, and infecting *Mycobacterium tuberculosis* resistance profile and the suggested drug treatment regimen based on these characteristics. Recommendations are endorsed by the WHO when available[2], and recommendations for children and pregnant individuals are based on expert consensus for groups not covered under the WHO recommendations[222–224]. Bdq, bedaquiline; Cfz, clofazimine; CNS, central nervous system; Cs, cycloserine; Dlm, delamanid; E, ethambutol; Eto, ethionamide; Hh, high-dose isoniazid; Lfx, levofloxacin; Lzd, linezolid; Mfx, moxifloxacine; Pa, pretomanid; TB, tuberculosis; Z, pyrazinamide. [a]BPaL (Bdq-Pa-Lzd) or BPaLM (Bdq-Pa-Lzd-Mfx) regimens are not recommended for individuals younger than 15 years old, pregnant individuals or those with severe forms of extrapulmonary disease, including those affecting the CNS, or osteoarticular and military disease. [b]For persons with a low likelihood of fluoroquinolone resistance, Lzd can be given for shorter periods of time (that is, 8 weeks or until fluoroquinolone results are back). [c]Non-BPaL or BPaLM all-oral 9-month regimens are inferior to BPaL or BPaLM and are not recommended for advanced pulmonary disease (that is, in individuals over 15 years old with evidence of bilateral cavitation or bilateral disease on imaging). These regimens are not recommended for severe forms of extrapulmonary disease (that is, in individuals over 15 years old with CNS diseases, osteoarticular, pericardial or military disease, or in individuals younger than 15 years with extrapulmonary disease other than peripheral lymphadenopathy, isolated mediastinal lymphadenopathy without evidence of compression or uncomplicated pleural effusion). [d]Eto can be replaced with a 2-month course of Lzd. [e]At least one of Cs, Dlm or Lzd should be administered to complete the four-drug regimen and the choice should be assessed on an individual basis with proper follow-up. In younger children, Cs can be challenging to administer. A limited but growing body of evidence supports the replacement of Cs with Dlm in both children and pregnant persons. In children with non-severe disease with no evidence of resistance to other agents, Lzd is often not favoured given the need for frequent monitoring (including blood draws) or given as a short 8-week dose. In children with severe disease, Lzd should be given in addition to Cs or Dlm.

preventive therapy for contacts of MDR-TB cases is not recommended although there is some evidence that Inh-based preventive therapy may protect household contacts of persons with MDR-TB against the development of secondary TB[235]. Three ongoing phase III clinical trials are evaluating the efficacy of MDR-TB preventive treatment; specifically, Lfx monotherapy versus placebo (TB-CHAMP ISRCTN92634082 (ref. 236) and V-QUIN ACTRN12616000215426 (ref. 237); with promising early results[238]) and delamanid versus Inh (PHOENIx NCT03568383 (ref. 239), results expected in 2026). In February 2024, the WHO issued a rapid communication based on results from the TB-CHAMP and V-QUIN trials stating Lfx should be offered to all household contacts of persons with DR-TB, and full guidelines are expected in July of 2024 (ref. 240).

## Stigma and mental health

A diagnosis of TB is a substantial source of stigma and social isolation[241,242]. Current evidence suggests that people with MDR-TB are more likely to experience stigma and social isolation than people with susceptible TB[243–245]. Focus-group studies conducted in South Africa showed that being diagnosed with MDR-TB is subject to more stigma than testing positive for HIV. The stigma associated with MDR-TB was linked to lower adherence to treatment compared with the adherence observed in antiretroviral therapy despite the stigma associated with HIV[245]. Current estimates for depression and/or anxiety in people living with MDR-TB range from 15% to 80%[246,247]. MDR-TB treatment initiation has historically been led by medically trained physicians in most countries, with limited focus on mental health or other allied support for care. Although several efforts to address stigma and mental health burden across all TB have focused on education and/or training additional community stakeholders[248], there is a need for MDR/RR-TB-specific interventions[249,250]. Historically, a substantial proportion of stigma or mental health challenges specific to MDR-TB treatment are related to the use of injectable anti-TB drugs and the drug Cs, which can lead to psychiatric side effects and require frequent encounters with medical providers[251]. Current recommendations limit the use of these medications in favour of shorter and safer oral regimens, thus addressing some DR-TB-specific stigma and mental health difficulties. The oral regimens also provide an opportunity to introduce decentralized, nurse-led MDR-TB care that may further reduce stigma.

## Conclusions and future directions

DR-TB continues to be a significant public health problem and poses a threat to the efforts to control TB[22]. Serious delays in providing adequate care, together with limited access to DR-TB diagnostics and treatment, are contributing to a modest cure rate in relation to the total estimated MDR/RR-TB burden[22,42]. Multiple environmental, patient and bacterial risk factors have all been associated with MDR/RR-TB, although it is not yet clear how some of the risk groups should be targeted or what interventions are most effective for DR-TB prevention. MDR-TB rates are fuelled by both transmission and drug resistance acquisition, with the former predominating among persons with MDR-TB without prior TB[56]. Recent drug resistance acquisition is more common in lower-income countries[56]. Put together, these aspects highlight a need for more research to guide public health interventions for the prevention of DR-TB and the need to strengthen health systems to diagnose, treat, support and monitor persons with DR-TB.

The past few years have witnessed significant advances in both the diagnosis and treatment of DR-TB. NAATs directly applied to sputum are now more widely used, improving diagnostic accuracy in comparison to sputum smear microscopy, which should be replaced by molecular testing[252]. NAATs have improved RR-TB detection globally albeit with a modest impact on mortality, partially attributed to the empirical treatment of suspected cases[160–162]. This could be avoided if comprehensive drug-resistance profiles were available. Towards this goal, there is expanding research on the use of tNGS and WGS for diagnostic purposes. Large efforts to perform WGS of *M. tuberculosis* clinical isolates around the globe have increased our knowledge of the genetic determinants of drug resistance in this pathogen. Sequencing offers a favourable cost profile and turnaround time given the number of drugs assayed, and the WHO has recently endorsed the use of tNGS[178].

An important global campaign called '1/4/6 × 24'[253] is now under way to scale up the 6-month, all-oral BPaL and BPaLM treatment regimen for DR-TB as well as shorter regimens for drug-susceptible TB and preventive treatment. The better-performing all-oral regimens could significantly reduce poor outcomes from MDR/RR-TB but their worldwide implementation into clinical practice is lagging globally due to inertia in regulatory, political and health systems[2,22]. Household contact management of MDR-TB is estimated to prevent, on average, many secondary cases, including in children. Ongoing clinical trials aim to identify the optimal regimen for MDR-TB chemoprophylaxis and early results support the use of fluoroquinolone monotherapy[236–239].

Although the burden of stigma, social isolation, anxiety and mood disorders in MDR-TB is estimated to be high, there is very limited study of MDR-specific interventions for prevention and management[249,250]. Addressing stigma and mental health issues in persons with MDR-TB is an identified research priority. Observed adherence to treatment has been shown to have a limited effect on resistance acquisition and treatment outcomes[65–67]. It is essential for the health-care community to acknowledge this observation to reduce the stigma and blame placed on persons with MDR-TB for their illness. Lastly, use of the all-oral novel MDR-TB treatment regimens promises to lessen side effects and decentralize care. This, in turn, promises to improve cure rates and decrease the burden of stigma and mental health challenges.

Further research into the risk factors, drivers, and genetic and biological mechanisms of DR-TB is essential. Simultaneously, efforts should focus on improving the quality of DR-TB care by refining diagnostic methods, treatment regimens and non-medical interventions (such as food supplementation and social benefits) to improve treatment outcomes.

Such efforts in both research and clinical care will require stronger policy attention on DR-TB, especially considering the WHO goals to end the global TB epidemic by 2035. Eradication of DR-TB will require collaborative initiatives across various sectors, including personal, societal and health system interventions. Substantial funding investments and inclusion of DR-TB into pandemic preparedness and universal health coverage agendas are crucial.

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## Author contributions

M.P., M.F., H.C., C.M.D., J.F. and M.G. wrote the article and, together with C.R. and M.S.A.E.A. researched data for the article. All authors contributed substantially to the discussion of the content and reviewed and/or edited the manuscript before submission.

## Competing interests

The authors declare no competing interests.

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