Potential antimicrobial agents for the treatment of MDR-TB

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Abstract:

Treatment of multidrug-resistant tuberculosis (MDR-TB) is challenging because of the high toxicity and poor efficacy of second-line drugs. Therefore new anti-TB drugs are urgently needed. Unfortunately, development of new drugs takes time, is difficult and expensive. In order to speed up novel treatments for MDR-TB, we suggest considering expanding the indications of already available drugs. Six drugs with antimicrobial activity (phenothiazines, metronidazole, doxycycline, disulfiram, tigecycline and co-trimoxazole) are not listed in WHO guidelines on MDR-TB treatment but could be potential candidates for evaluation against *M. tuberculosis.*

We reviewed *in vitro*, *in vivo* and clinical anti-TB activity of these drugs in addition to the pharmacokinetics (PK) and side effects. We discussed the potential role of these drugs for treatment of MDR-TB. Of the drugs effective against active replicating TB, co-trimoxazole seems the most promising one because of its consistent pharmacokinetic profile, easy penetration into epithelial lining fluid (ELF) and its safety profile. For the more challenging dormant state of TB, thioridazine may play a potential role as an adjuvant for treatment of MDR-TB.

A strategy consisting of PK/PD studies, dose finding and phase III studies is needed to explore these drugs further for their potential role in the treatment of MDR-TB.

IntroductionMultidrug-resistant (MDR) tuberculosis (TB) is an infectious disease caused by *Mycobacterium tuberculosis (M. tuberculosis)* that is resistant to the two most powerful drugs: isoniazid and rifampin as well as fluoroquinolone and at least one of the injectable drugs (amikacin, capreomycin and kanamycin) [1-4]. The problem with the loss of rifampin is particularly important because although many drugs have potential to kill rapidly dividing metabolically active organisms, few drugs are active when the persistent population of *M. tuberculosis* has switched their genetic program to a quiescent, dormant phenotype. These persisters provide the major challenge for the immune system as well as for drug treatment to achieve sterility [5].

WHO estimated that 2.5% of all TB patients and 3% of all new cases are infected with MDR-TB [6, 7]. According to WHO an estimated 650,000 prevalent cases of MDR-TB occurred globally in 2010 [8].

The proportions of new TB cases with MDR-TB at country level in Eastern European countries range at an alarming level between 19.4% –32.3% [9]. One recent report from Belarus even indicates that almost half of newly diagnosed treatment-naïve patients with TB actually have MDR-TB [10]. Further, the fact that patients < 35 years of age showed two folds higher odds of MDR-TB than those aged ≥35 years suggests that in Belarus, emergence of MDR-TB is rampant and recent[11].

Most of the drug resistance is caused by mutations in the genome of *M. tuberculosis* coding for drug targets, but there is also evidence for mutations resulting in upregulation of bacterial efflux pumps potentially reducing susceptibility for several drug groups [12, 13]. An important feature of *M. tuberculosis* is the fact that under stress conditions, including drug therapy, the organism is able to change its genetic program.

Clearly, new antituberculous drugs are urgently needed. Unfortunately development of new drugs takes time, is difficult and expensive and low and middle income countries that constitute 95% of TB cases around the world have limited resources for health expenditure as well as lack of support for research [14]. In order to speed up novel treatments for MDR-TB, we explore the possibility of expanding the indications of already available drugs in the market.

The aim of this review is therefore to describe antimicrobial drugs that are not listed in WHO guidelines on MDR-TB but have *in vitro* and/or *in vivo* or clinical antituberculous activity and are available on the market labeled for other diseases. These drugs are phenothiazines, metronidazole, doxycycline, disulfiram, tigecycline and co-trimoxazole. In addition suggestions are made which drug is likely to be eligible for further evaluation against TB.

Methods

PubMed and Google scholar searches of published literature in English language were done manually in ^{31}December 2012 using a combination of the name of each drug and *M. tuberculosis*. The names of the drugs were combined with more specific search terms: *in vitro, in vivo* and clinical studies, pharmacokinetics and side effects. No exclusion criterion based on study methodology was applied. Further cross-references were obtained from bibliographies of identified and relevant papers. The results were divided per drug and within each case the subheading for *in vitro, in vivo* and clinical data were used. Based on these

data, suggestions are made which drug is the most promising for further evaluation for the treatment of MDR-TB.

Results

In total 85 references according to the search criteria were retrieved. The results for each drug are discussed in details below. A summary of the results, based on *in vitro*, *in vivo* and clinical data is provided in table 1.

Phenothiazine

Phenothiazines as thioridazine, chlorpromazine and promethazine belong to the group of anti-psychotic drugs [15]. The phenothiazine derivative; thioridazine is a neuroleptic compound that has been used for over four decades [16].

They have the ability to inhibit the bacterial efflux pump which protects the bacterial cell against harmful substances to which the cell is exposed [15]. These compounds have been shown to inhibit the *in vitro* growth of MDR-TB. Phenothiazines may be concentrated more than 10-folds in macrophages which phagocyte tuberculosis bacilli [17].

In vitro studies

Phenothiazine family compounds including chlorpromazine and thioridazine are known to possess appreciable levels of antimicrobial activity against MTB organisms. The anti-MDR-MTB activity was similar in thioridazine and chlorpromazine [18]. Thioridazine inhibits the growth of clinical isolates of *M. tuberculosis* that are resistant to streptomycin, rifampin, isoniazid, ethambutol and pyrazinamide (first line anti-TB drugs). Thioridazine is bactericidal at a concentration ≥32 mg/L. MDRTB shows similar susceptibility to chlorpromazine and thioridazine [19].

Minimum inhibitory concentration of thioridazine required for 50% inhibition of *M. tuberculosis* H37Rv clinical isolates is 2.5 mg/L [20].

The macrophage plays an important role in concentrating these drugs in the phagolysosome of macrophages. Phenothiazines like thioridazine interfere with calcium and potassium distribution, lowering the pH in the macrophage lysosomal apparatus thereby disrupting the integrity of the cell wall of the bacterium and facilitating the penetration of these drugs into *M. tuberculosis* [21].

Because thioridazine does not appear toxic to the macrophage in vitro, it could be used in the treatment of intracellular *M. tuberculosis* infections [21].

In vivo studies

Groups of five female BALB/C mice were infected intraperitoneally with10^6 colony forming units (CFU) /mL of *M. tuberculosis* and treated with thioridazine in the dose range 0.05-0.5 mg/day which is equivalent to that used for psychosis in humans (1200 mg/day). There was a more than five log reduction in the number of CFU/kg derived from the lungs of infected mice compared to the control group within one month [22].

Clinical studies

When thioridazine is administered orally, it is rapidly absorbed with peak plasma concentrations occurring within 2-3 hours. Thioridazine is widely distributed in tissues like liver, blood and kidney [23, 24]. However, it is distributed less favorably to the brain [25].

Thioridazine is not useful for treatment of cavitary pulmonary *M. tuberculosis* because the concentration of thioridazine required for killing or inhibiting *M. tuberculosis* outside the macrophage exceeded the concentration that could be achieved in patients receiving standard dosages [21].

Thioridazine undergoes 5-sulfoxidation and *N*-demethylation by CYP1A2 and CYP3A4 while CYP2D6 catalyzes mono-2- and di-2-sulfoxidation of thioridazine in the human liver. CYP2D6 and CYP3A4 catalyze thioridazine mono-2-sulfoxidation [26].

Thioridazine is almost entirely bound to serum proteins 99.85% while its metabolites are bound to serum proteins to a lesser extent. This could be a problem because a small change in the binding capacity of proteins can change the unbound concentration of thioridazine that has clinical activity. The unbound concentrations of thioridazine metabolites including thioridazine side-chain sulfoxide and thioridazine side–chain sulfone are higher (2 and 9 times respectively) than the unbound concentration of thioridazine [27]**.**

Thioridazine can be used as adjuvant for regimens of four to five drugs for treatment of MDRTB. It has been administered in drug resistant TB until the susceptibility of strains is known [17].

Of 17 XDR-TB patients, 14 patients were treated with thioridazine in combination with linezolid and/or moxifloxacin in a daily dose of only 25 mg for 2 weeks after which the dose was increased by 25 mg weekly until dosing reached 200 mg/day. The combined therapy of thioridazine, moxifloxacin and linezolid cured 61% of patients and 22% of patients who were still on treatment, showed beneficial response. Thioridazine was discontinued in two patients because of pancytopenia in one patient and allergic dermatitis in other. In this study, the authors speculate that thioridazine could have contributed to an earlier bacteriological sputum conversion. No prolongation of the QT interval or other heart complication was observed in the patients that received thioridazine. Combined therapy including linezolid, moxifloxacin and thioridazine was associated with a relapse-free cure in most cases [28].

In another study, thioridazine cured10 out of 12 patients, the other 2 patients responded as well but they dropped out of the program [29].

The side effects of thioridazine like for all phenothiazines are dose dependent and include QTc prolongation, thereby increasing the risk of Torsade Pointes with subsequent risk of sudden death [30]. To avoid the risk of sudden death in patients whose QTc is significantly increased, the patients should be screened by ECG before and during treatment with thioridazine [29]. Because chlorpromazine causes frequent and serious side effects when administered chronically, it is not a good candidate drug for the treatment of MDRTB [31].

Metronidazole

Metronidazole is currently licensed for the oral treatment of infections like protozoa (trichomoniasis, amaebiasis) and anaerobic bacteria [32].

The mechanism of action of metronidazole includes severe damage to the bacillary DNA by exposing the bacilli to unstable products that result from reduction of the nitro group in metronidazole interfering with enzymes involved in DNA repair [33].

Metronidazole has only been evaluated to a limited extent for TB showing good bactericidal action in anaerobic conditions against dormant tubercle bacilli. Unfortunately it has no effect on aerobic cultures of *M. tuberculosis* [32, 34].

In vitro studies

Metronidazole showed no effect when added to bone marrow–derived macrophages infected with *M. tuberculosis* and does not decrease the bacterial load although a high concentration of metronidazole was used [35].

Adding metronidazole to a regimen of rifampin, moxifloxacin and amikacin and/or capreomycin significantly improved killing of dormant (anaerobic and drug-tolerant) *M. tuberculosis* in an adipocyte model [36].

An acidic culture model of *M. tuberculosis* was used to test metronidazole alone and in combination with other drugs against aerobic, 5-day-old (A5) and hypoxic, 5-, 12-, 19-day-old

(H5, H12, H19, respectively) bacilli after 7, 14, 21 days of exposure. Metronidazole was inactive against A5 and H5 but it has efficacy against H12 and H19 cells increased with hypoxia and exposure time with 4 and 3.8 log reductions in the number of CFU respectively on day 21. A combination of metronidazole with potent anti-A5 and –H5 drugs (rifampin, moxifloxacin, amikacin after 14 days of exposure killed H19 cells only [37].

In vivo studies

Although metronidazole had no effect on the growth of *M*. *tuberculosis* in lungs of aerosol-infected mice in a dose of 15mg/kg, there was a relatively small but statistically significant reduction in bacterial counts when the mice established a chronic disease state, when a proportion of bacilli were in a dormant state. The possible explanation for the low activity of metronidazole is that the *M. tuberculosis* is not in a state of anaerobic metabolism in which it is susceptible to metronidazole [35].

In another in vivo study using a granuloma model of *M. tuberculosis* dormancy (mouse hollow fiber model) metronidazole (100mg/kg) failed to show a beneficial effect against bacilli although there was immunohistochemical and mutant survival based evidence of tissue hypoxia. The possible explanations of this result could be due to non-optimal concentration of metronidazole or poor penetration into granulomatosis lesions or non-sufficient hypoxia to permit reductive activation of metronidazole [38].

Activity of metronidazole in guinea-pigs was also not promising, metronidazole at 50 and 100 mg/kg combined with standard regimens showed no significant activity for the treatment of guinea-pigs infected with *M*. *tuberculosis* perhaps because of poor penetration of metronidazole into the necrotic core of the hypoxic granulomas or to the concentrations of metronidazole effective against *M. tuberculosis* under completely anaerobic and microaerophilic conditions [39]. These results were comparable to the *in vivo* study in which

C3HeB/FeJ (Kramnik) and BALB/c mice were infected by aerosol with *M. tuberculosis* and treated for 7 to 8 weeks with 200 mg/kg metronidazole. Although necrotic lesions in the Kramnik model showed evidence of hypoxia, metronidazole had no bactericidal activity against Erdman strain of *M. tuberculosis* in this model and also in BALB/c mice [40]

Clinical studies

The oral bioavailability of metronidazole is almost complete (98.9%)[41] . Peak serum levels are reached within 1-3 hours [42]. Metronidazole is bound 10-20% to plasma proteins [42]. The half-life of metronidazole is about 8 h [41]. Metronidazole is distributed in different tissues and it has good penetration into the cerebral spinal fluid (CSF) and central nervous system (CNS) [44]. It is metabolized in the liver resulting in the formation of two oxidation products: alcohol or hydroxyl metabolite, 1-(2-hydroxyethyl)-2-hydroxymethyl-5-nitroimidazole which has an antimicrobial activity of 30-75% compared to metronidazole. The acidic metabolite, 2-methyl-5-nitroimidazole-1-acetic acid which has 5% activity of the metronidazole is only detected in patients with renal dysfunction [45, 46]. Metronidazole and its metabolites are mainly excreted in urine [43].

There are few clinical studies of metronidazole for TB. In a single blinded study, metronidazole (400 mg three times daily) was administered for eight weeks in addition to an anti-TB regimen. Addition of metronidazole to standard treatment with streptomycin, isoniazid and rifampicin resulted in a significant improvement in clinical response, with reduction of sputum quantity, a high radiographic improvement and an improvement in sensitivity of anti-TB drugs in comparison with placebo group. This study confirmed that metronidazole has a beneficial adjuvant role in the treatment of TB [47].

In an ongoing clinical trial, metronidazole is used in a dose of 500 mg three times daily in combination with standard second line antituberculous drugs in MDR-TB patients. The results of this study are to be expected soon [NCT00425113].

Metronidazole is generally well tolerated. Adverse reactions include reversible neutropenia, minor gastrointestinal side effects, metallic taste, vaginal and urethral burning and darkening of the urine. Central nervous system side effects include ataxia, vertigo, peripheral neuropathy and headache [48]. Metronidazole can produce a reaction similar to that of disulfiram when administered to patients using alcohol because the interaction between metronidazole and ethanol leads to accumulation of acetaldehyde in the blood which is toxic [49]. As alcohol abuse is a risk factor for getting infected with TB, care should be taken in use of this drug in patients with alcohol abuse.

Tetracyclines

Doxycycline and tigecycline belong to the tetracycline group of antimicrobials which exhibit a broad-spectrum of activity against different pathogens including Gram-positive and Gram-negative bacteria [50].

Tetracyclines are bacteriostatic and act by binding to the bacterial 30S ribosomal subunit and inhibiting protein synthesis. The tetracyclines are effective for the treatment of a wide range of infectious diseases like community-acquired respiratory tract infection, sexually transmitted disease and skin conditions [50].

Doxycycline is an antibiotic with broad spectrum matrix metalloproteinase (MMP) inhibitory activity [51]. It reduces the expression of MMPs thereby reducing tissue damage in TB and suppressing mycobacterial growth. Because doxycycline is safe, cheap and available, it may represent a new adjunctive therapy to improve outcomes and reduce mortality in TB [52].

Tigecycline (GAR-936) is a new semisynthetic glycylcycline (tetracyclines analogue) and is effective against intra-abdominal and skin and soft tissue infections caused by staphylococci, enterococci or streptococci as well as most enterobacteriaceae and anaerobic pathogens [50][53]. Tigecycline is effective against rapidly growing mycobacteria (*M. fortuitum, M.*

chelonae and M. abscessus) but showed no activity against the more slowly growing mycobacteria (*M. tuberculosis)* [53, 54].

In vitro studies

Doxycycline suppressed MMP1 and MMP3 and TNF–α secretion from primary human macrophages infected with *M. tuberculosis* at 72 hour in a dose dependent manner. Doxycycline is bacteriostatic to *M. tuberculosis* clinical isolates with a MIC of 2.5µg/ml [52].

In 69 MDR-TB isolates obtained from patients in the Samara region of Russia, 5 (7.4%) isolates were resistant to doxycycline [55]. In *vitro,* MIC values of tigecycline against clinical isolates of *M. tuberculosis* were as high as 8-64 µg/ml [56].

In vivo studies

Doxycycline decreased mycobacterial replication in infected guinea pigs but showed no effect on MMP activity. Doxycycline in a dose of 5 or 20 mg/kg suppresses lung CFU at 10 weeks in a dose dependent manner. The results showed that doxycycline improved the outcome in TB by acting directly on mycobacterial proliferation rather than on MMP activity [52].

Clinical studies

Doxycycline is available in the oral form unlike tigecycline that is only available in an injectable formulation [50]. Doxycycline is well absorbed from the gastrointestinal tract and bioavailability ranging between 75-100% [57, 50]. It is absorbed quickly and reaches its maximum serum concentration (Cmax) within 4 h [50]. Cmax and area under the plasma concentration time curve (AUC) of tigecycline are proportional with the dose [50, 53]. In

epithelial lining fluid the area under the curve of tigecycline was 2.28 µg. h/ml which is higher than that of serum1.73±0.64 µg. h/ml [58]. Doxycycline is more lipophilic so that it exhibits greater penetration into the tissues especially the brain, eye, prostate and intestinal epithelia [50]. Higher concentrations of doxycycline are found also in kidney, liver and bowel [57]. Tigecycline has a large volume of distribution 7-10L/kg [59]. It shows good penetration into tissues like bones, skin, liver and lung [50]. The protein binding of doxycycline is between 60-95% and of tigecycline ranges from 71 -89% [60, 50].

Tigecycline has a half-life of 15-36 hours which is greater than the 12h half-life of the doxycycline [50, 58]. No more than 15% of tigecycline is excreted in the urine in unchanged form [50, 61]. About 30-40% of doxycycline is excreted unchanged in the urine [50].

Doxycycline is safe in humans and may suppress immunopathologic MMPs thereby reducing tissue damage in TB patients using a low dose (20 mg twice daily). It may achieve sufficient concentration in the lung interstitium to decrease the growth of *M. tuberculosis* and modulate MMP activity and expression [52]. Unfortunately no clinical data are available for the potential use of tigecycline for the treatment of MDR-TB in humans.

The most common side effects of doxycycline are gastrointestinal side effects and skin reactions [62]. In a study with healthy subjects, tigecycline had no serious side effects except nausea and vomiting which were dose-related [61].

Disulfiram

Disulfiram (tetraethylthiuram disulfide, DSF) has been used orally in the clinical treatment of alcoholism since 1949. DSF is a prodrug and is enzymatically metabolized in the blood to metabolites primarily diethyldithiocarbamate (DDC or DETC) within 4 min [63, 64].

DSF and DDC exhibit growth-inhibitory activity against bacteria, fungi, protozoa, and viruses. DSF is effective against MDR/XDR-TB and exhibits bactericidal activity *in vivo* and *in vitro* [63]. The mechanism of action of DDC against *M. tuberculosis* has been reported as

inhibition of beta-class carbonic anhydrases (beta-CAs) from *M. tuberculosis* [65]. However the mechanism of action of DSF against *M. tuberculosis* is not completely understood [52].

In vitro studies

Peripheral blood mononuclear cells (PBMC) of healthy and HIV subjects were preincubated with 100-1000 ng /ml of DDC and then infected with *M. tuberculosis* H37Rv. DDC reduced CFU of these intracellular growing mycobacteria. DDC enhances the antimycobacterial activity of monocyte–derived macrophages from healthy volunteers injected with 5mg/kg body weight DDC exvivo. DDC can enhance macrophage maturation by the induction of 1, 25-$(OH)_2$ cholecalciferol (vitamin D3) [66]. Vitamin D3 may play an important role in the pathological process in tuberculosis by downregulating the levels of matrix metalloproteinases (MMPs) and upregulating the levels of tissue inhibitor of metalloproteinase (TIMPs) [67].

An in vitro study showed that DETC was highly active against tubercle bacilli with MIC of 8 µg /ml [64].

DSF and DDC showed antitubercular activity against more than 40 clinical isolates of *M. tuberculosis* including MDR/XDR-TB strains. The MIC90s of DSF and DDC against clinical isolates were 1.56 and 3.13 µg/ml respectively. They show also bactericidal activity against intracellular *M. tuberculosis* in human monocyte leukemia cell line (THP-1) at 6-30 µg/ml and 10-30 µg/ml respectively [63].

Since no cross resistance of DSF and DDC with other anti-TB drugs was observed, these compounds may be of potential value for future regimens against MDR/XDR-TB [63].

In vivo studies

DSF kills *M. tuberculosis* at (80-160) µg/kg in a mouse model with chronic TB [63]. DETC enhances the activity of pyrazinamide and rifampin two-fold when co-administered in mice at 100 mg/kg showing activity against persisters of *M. tuberculosis* [64].

Clinical studies

DSF is rapidly and completely absorbed following oral administration and is quickly reduced to DDC. DDC is metabolized to diethylamine, carbon disulfide (CS_2), DDC methyl ester, DDC glucuronide and DDC sulfate, a small amount of DDC is reoxidized to DSF. DSF, DDC and CS_2 are widely distributed throughout the body in lipids of various tissues and highest levels of these compounds are found in skeletal muscle [68]. In humans more than 90% of orally administered DSF is eliminated within 3 days by renal clearance as DDC and DDC glucuronide and to a small extent as DDC sulfate and via the breath as CS_2 [68]. Few publications showed the activity of DSF or its metabolites against TB. Only one study found that DDC reduces the incidence of infections in HIV co-infected TB patients probably by stimulating the antimicrobial activity of mononuclear phagocytes [66].

The side effects of disulfiram are mainly on the CNS including psychosis or confusional state that occurs in the early period of DSF therapy with higher dosages of DSF (500 mg/day). Another serious side effect is peripheral neuropathy. All these side effects are reversible [69]. Rarely, DSF can cause fatal hepatitis [69].

Co-trimoxazole

Co-trimoxazole (SXT) is a synergistic combination of two antimicrobial agents; trimethoprim (TMP) and sulfamethoxazole (SMX). Its mechanism of action is interference with folic acid synthesis of bacteria. SXT is predominantly used for the treatment of urinary tract infections and prophylaxis and treatment of *Pneumocystis jiroveci pneumonia* (PCP) in HIV patients [70, 71]. There is ongoing debate about the use of SXT for the treatment of TB. Some studies mentioned that only SMX was effective against *M. tuberculosis* while TMP is not [72, 73].

In vitro studies

M. tuberculosis strains appeared to be susceptible to SXT in 43 of 44 (98%) isolates tested. These isolates were sensitive to TMP-SMX at MIC ≤1/19 µg/ml [74]. Another study showed that SMX inhibits 80% and 99% growth of all 117 clinical isolates at MIC 19 mg/l and 38 mg/l respectively [75]. Drug susceptibility testing in a recent study mentioned that MICs values of SMX for *M. tuberculosis* ranged from 4.75-25 µg/ml [76]. In 7H9 broth, *M. tuberculosis* was susceptible to SMX; MIC90 was 8 µg/ ml. In this study SMX achieved an excellent activity against *M. tuberculosis* [77]. One *in vitro* study showed that *M. tuberculosis* strain H37Rv was susceptible to SMX and not to TMP at MIC of 8.5µg/ml. When SXT was added to an isoniazid or rifampin treated *M. tuberculosis* culture isolate, SMX with and without TMP was efficient at killing and preventing its growth and thereby preventing the emergence of drug resistance [78].

In one *in vitro* study, *M. tuberculosis strains* were exposed to either TMP/SMX combination, SMX and TMP alone or SMX in combination with the first line tuberculosis drugs (isoniazid, rifampicin and ethambutol). The results showed that TMP had a negligible effect on the growth of *M. tuberculosis* while SMX inhibited 80% of the growth of *M. tuberculosis at 4.75* mg/L. There was no synergistic activity between TMP and SMX combination but an additive effect was observed. In combination with isoniazid, rifampicin and ethambutol, SMX had a synergistic effect with rifampicin, an additive effect with ethambutol and no effect with isoniazid [11].

Clinical studies

The total absorption of SMX is 85-90%. The concentrations of the non-protein-bound fraction of SMX to TMP in the plasma varied between subjects and ranged from 1:5 to 1:40 [79]. The

explanation could be that SMX has a smaller volume of distribution (10-20L) than TMP (69-133L). In the blood, SMX is bound to plasma proteins to the extent of 58-66% (34-42% free) [79, 80]. SMX is well distributed in most body fluids and also in cerebrospinal fluid so that it may be a good candidate for the treatment of TB meningitis [79]. Concentrations of SMX in sputum, middle ear fluid and sinus fluid are about 20-27% of serum and in bronchial secretion 60-100% of those in serum [81]. SXT penetrates epithelial lining fluid (ELF) easily because it is lipophilic and inflammation independent [82, 83]. Following distribution of SMX, it is partially acetylated and glucuronide –conjugated in the liver [84]. SMX is excreted as acetyl derivative for 60-65% and 15% appears as glucuronides. These metabolites constitute 70% of total SMX in urine and 20-35% in plasma [79]. The plasma half–life of SMX is 9 hours [79].

Limited data are available on the pharmacokinetic parameters of SXT in MDR-TB patients. Only one study described the PK parameters after receiving 480 mg and 960 mg of co-trimoxazole. PK parameters of SMX in TB patients including area under the curve, clearance and volume of distribution are lower than the values observed in patients with other indications. These patients seem to display a consistent PK profile for SMX. SXT was safe and well tolerated except for one patient who had gastrointestinal side effects after receiving 960 mg of SXT [76].

The adherence and tolerance to the drug was good when SXT was used in a daily dose of 960 mg as prophylaxis to reduce the mortality in adults with HIV infection and TB, to prevent or treat opportunistic infections [85]. The occurrence of one or several side effects that could be due to SXT was approximately similar in the placebo group (n=372) and in the co-trimoxazole group (n=371). Thus, it can be concluded that the side effects of SXT even in HIV –infected individuals is rather mild [85]. A dose of 960 mg/day of SXT improved the survival of HIV-positive TB patients dramatically so that SXT prophylaxis should be added to the routine care of HIV-positive TB patients [86].

In five clinical studies summarized in [71], co-trimoxazole was safe, feasible and effective in the prophylactic treatment of HIV patients with TB; it not only reduced PCP, but also malaria and other bacterial (pneumococcal) airway infections, The impact on infections with *M. tuberculosis* was not considered in these studies.

In general TMP-SMX is a safe medication and well tolerated. The most common side effects include gastrointestinal intolerance, nausea, vomiting, anorexia and diarrhea [87, 84]. Possible side effects in the blood are hyperkalemia, slight increase in the serum creatinine level (not representing loss of glomerular filtration but rather, reversible decrease in tubular excretion of the creatinine molecule) and hypernatremia, they occur especially in patients with renal dysfunction [84, 87 88]. Hematological abnormalities include leucopenia, agranulocytosis, thrombocytopenia, hemolytic and aplastic anemia [89].

Discussion:

The need for new drugs to improve the treatment of patients with MDR-TB has received a great deal of attention. These patients are currently treated with a combination of second-line drugs that are more expensive, more toxic, and less effective than the drugs used in standard therapy, often resulting in poor outcomes. There are numerous obstacles like the lack of commercial interest of new anti-TB drugs including perceived lack of need and profit opportunity in addition to the difficulties of identifying potentially active new compounds and of clinical development of promising new compounds. Consequently, exploring the antimicrobial activity of drugs that are already available on the market would be a tremendous asset. Therefore this review identifies the pharmacokinetics, pharmacodynamics, *and in vitro, in vivo* and clinical data of these drugs that can provide an advantage to explore further in the treatment of MDR-TB.

Based on the *in vitro, in vivo* and clinical data of the drugs discussed in this review, some of these drugs could be promising candidates for treatment of MDR-TB in the future like thioridazine, doxycycline, metronidazole and co-trimoxazole. They are effective either against rapid or non- replicating stage of *M. tuberculosis.* These drugs have also favorable pharmacokinetics like CNS and CSF penetration that may be useful for the treatment of TB meningitis.

However, thioridazine has no activity against rapidly replicating metabolically active *M. tuberculosis* such as in active cavitary pulmonary TB because of poor penetration of this drug into the cavities and therefore, lacks efficacy against this stage of TB.

The same is true for metronidazole that is predominantly active under anaerobic conditions; this drug however seems to have activity against dormant *M. tuberculosis.* Compounds of similar structure of metronidazole have been shown to have potent antitubercular activity *in vitro* and *in vivo* like CG17341 and PA 824 [90]. Thus, metronidazole could be used as a lead compound for the synthesis of new drugs against MDR-TB. Doxycycline has bacteriostatic

no bactericidal effect against *M. tuberculosis* and is therefore not the first choice to develop further.

Co-trimoxazole could be the promising drug for the treatment of MDR-TB because of its consistent pharmacokinetic profile in combination with other anti-TB drugs. It is an easy to administer, cheap, well tolerated and safe antibiotic. It has also encouraging activity in vitro.

The rest has some limitations and limited data that may restrict their uses for the treatment of MDR-TB. Because of the fatal outcome of a disulfiram-alcohol reaction; this drug should not be administered to TB patients that are alcohol abusers. Tigecycline is administered only parenterally and is not effective against the more slowly growing mycobacteria such as *M. tuberculosis*.

There is only limited antimicrobial pharmacokinetic-pharmacodynamic (PK/PD) information available for the drugs discussed here. This could be an obstacle to find the suitable doses and to predict their efficacy in the treatment of MDR-TB patients. Therefore antimicrobial PK/PD derived in preclinical PK/PD models like the hollow fiber infection model and the mouse models and additional clinical trials offer the possibility to study toxicity and also to determine the duration of therapy. These will be important steps in the further development of these drugs for use in MDR-TB.

A clinical prospective study should be performed to evaluate the pharmacokinetic parameters and tolerance in order to find the suitable dose for the treatment of MDR-TB patients in phase II study. In a phase III study, the clinical efficacy and long term safety of the drug should be investigated by comparing the sterilizing activities of the candidate alone and when substituted for one of standard anti-TB drugs. These approaches are essential for the efficient clinical development of the potential antimicrobial drugs for TB treatment.

Table1. Summary of *in vitro, in vivo* and clinical activity of the drugs against *M. tuberculosis*

Drug/formulation	***in vitrol* activity (MIC)**	***in vivol* activity**	**Clinical/activity**	**Pharmacokinetics**	**Recommendations**
Phenothiazine :					
Thioridazine (oral)	(+) 2.5 µg/ml [20]	(+)	(+)	Wide distribution into tissues [23] [24]	Clinical trials are needed to investigate the suitable dose for treatment of cavitary pulmonary *M. tuberculosis*
Metronidazole (oral)	(-) ,(+) in combination with anti-TB regimen MIC of metronidazole against *M. tuberculosis* is unknown	(-)	(+) in combination with anti-TB regimen	Good penetration into CNS and CSF [44]	Further clinical studies are required. Treatment of TB meningitis.
Tetracyclines :					
Tigycycline (parenteral)	( ) 8-64 µg/ml[56]	( )	( )	High AUC in ELF and good penetration in the lung [50] [58]	*In vitro, in vivo* and clinical studies against slowly growing *M. tuberculosis*
Doxycycline (oral)	(+), (-) 2.5 µg/ml[52]	(+)	(+)	Good penetration in the brain And sufficient concentration in lung interstitium [50]	Treatment of TB meningitis
Disulfiram (oral)	(+) 8 µg/ml [64] [64]	(+)		( )	Further clinical studies especially pharmacokinetics
Co-trimoxazole (TMP /SMX)	(+) 1/19 µg/ml [74] [74]	( )	(+)	Distributed mostly in CSF [79]	Good candidate for treatment of TB meningitis

(oral)					*In vivo* studies are required

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