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SUSCEPTIBILITY

In Vitro Activity of Tedizolid against *Mycobacterium tuberculosis*

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ABSTRACT Tedizolid is a novel oxazolidinone with activities against Gram-positive microorganisms, including mycobacteria. We studied the *in vitro* activity of tedizolid against 120 *Mycobacterium tuberculosis* strains, including susceptible, first-line-resistant, and multidrug-resistant isolates. MIC was tested using the Bactec 960 MGIT system. MIC_{90} and MIC_{50} were 0.5 and 0.25 μg/ml, respectively, in susceptible and resistant strains. Tedizolid may be an alternative in the treatment of resistant *M. tuberculosis*.

KEYWORDS *Mycobacterium tuberculosis*, resistance, tedizolid

Tuberculosis (TB) is the ninth leading cause of death worldwide. In 2017, 6.4 million new cases of TB were reported. Drug-resistant TB is a continuing threat. An estimated 558,000 new cases were rifampin-resistant TB, of which 460,000 were multidrug-resistant TB (MDR-TB). The percentage of extremely resistant TB was 8.5%. Due to the emergence of MDR-TB, the evaluation of new compounds is necessary (1). Oxazolidinones are in development for possible use in TB therapy (2). Tedizolid phosphate is a novel oxazolidinone prodrug (TR-701) that is transformed in the serum into the active drug tedizolid TR-700 (formerly DA-7157), which has a broad range of activities against Gram-positive microorganisms, including mycobacteria. Initial experience with tedizolid has shown advantages in antimicrobial potency against key organisms, including those with reduced susceptibility to linezolid, lower incidence of adverse effects, and favorable pharmacokinetics (3). Its pharmacokinetic/pharmacodynamic properties allow for tedizolid to be administered orally once daily, making it potentially useful for indications requiring prolonged treatment. Although oxazolidinones are generally well tolerated, reversible myelosuppression, particularly thrombocytopenia, is well documented with linezolid, primarily in treatment courses of >14 days (4–7). Tedizolid has proved to be a viable alternative in these cases (8). Tedizolid inhibits protein by binding to the 23S rRNA of the 50S subunit, thereby preventing the formation of the 70S initiation complex (9). Tedizolid has shown activity against acid-fast bacilli. Some studies showed *in vitro* activities of tedizolid against species of nontuberculous mycobacteria (10–12). Reports of *in vitro* and *in vivo* (intracellular) activities against *Mycobacterium tuberculosis*, including MDR strains (4), and *Nocardia brasiliensis* have been published (13, 14). The aim of the present study was to determine the *in vitro* activity of tedizolid-susceptible and -resistant *M. tuberculosis* isolates.

The MICs of tedizolid against 120 *M. tuberculosis* strains selected from the Mycobacteria Reference Center, Faculty of Medicine, Córdoba, Spain, were determined. A total of 84 strains were identified as drug resistant (59 singly drug resistant, 25 MDR), and 36 strains were pan-susceptible. The activity of tedizolid against *M tuberculosis* strains was tested with the Bactec 960 MGIT system (Becton, Dickinson, Sparks, MD) (15). The MICs of tedizolid were determined to be between 0.015 and 16 μg/ml. All strains were stored at −70°C and subcultured on Lowenstein-Jensen (LJ) medium before testing. The

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TABLE 1 Tedizolid MIC ranges for 120 strains of *M. tuberculosis* tested

Strain type	No. of strains	MIC range (μg/ml)	No. (%C[a]) of isolates with MIC (μg/ml) of:				MIC_{50} (μg/ml)	MIC_{90} (μg/ml)	*P*
			0.006	0.125	0.25	0.5			
Susceptible	36	0.125–0.5	0	11 (30.5)	21(88.8)	4 (100)	0.25	0.5	
Resistant	59	0.06–0.5	1 (1.6)	21 (37.2)	31(89.8)	6 (100)	0.25	0.5	0.8171
MDR	25	0.125–0.5	0	10 (40.0)	12(88.0)	3 (100)	0.25	0.5	0.7109

[a]%C, cumulative percentage.

tedizolid compound was solubilized and diluted 2-fold in dimethyl sulfoxide, and 0.1 ml volume was added to the corresponding MGIT culture tube. Bacterial suspensions were prepared by dispensing two or three loops of bacteria from fresh LJ slopes into 3 ml of phosphate-buffered saline (PBS), and the mixture was homogenized by sonication in an ultrasound water bath. The suspensions were allowed to sediment for 20 min, and the upper phase was transferred to a new tube and allowed to sediment for another 15 min before adjustment to a McFarland standard of 0.5 and dilution (1:5) in PBS (mixture A). Half a milliliter of mixture A was used to inoculate the MGIT culture tubes containing the drug and an undiluted (drug-free) growth control. Additionally, a 1:100-diluted bacterial suspension was made from mixture A to prepare the proportional-growth control of the MGIT test. Briefly, 0.8 ml of an oleic acid-albumin-dextrose-catalase enrichment was added to each MGIT culture tube. Before all tubes were placed into the Bactec MGIT 960 instrument for analysis, they were shaken gently for homogenization. The MICs were determined with the Bactec MGIT 960 system. MIC was defined as the lowest drug concentration at which the growth index (GI) value was 100 at the point that the proportional growth control reached a GI of 400, as recommended by the manufacturer. Also, the MICs of susceptible and resistant MTB isolates were determined. MICs of strains were compared with the activity of linezolid.

As drug stability control, strain ATCC 29212 of *Enterococcus faecalis* was tested. *E. faecalis* had a tedizolid MIC of 0.25 μg/ml, within the acceptable concentration range (0.025 to 1 μg/ml) indicated by manufacture.

Statistical differences between antimicrobial resistance frequencies were determined using the χ^2 test. The level of significance used was $P < 0.05$.

The results are shown in Table 1. The MIC ranges for susceptible and resistant strains were 0.125 to 0.5and 0.06 to 0.5 μg/ml, respectively. The MIC_{50} and MIC_{90} values were 0.25 and 0.5 μg/ml for susceptible and resistant isolates, respectively. All 120 *M. tuberculosis* strains studied were inhibited at tedizolid concentrations of $\leq$0.5 μg/ml. Tedizolid showed 1- to 3-fold greater potency than linezolid. The MIC_{50} and MIC_{90} values of linezolid were 1 μg/ml for both. One strain with a linezolid MIC of 2 μg/ml exhibited an MIC for tedizolid of 0.25 μg/ml. In a previous *in vitro* study, tedizolid was tested against 95 strains of *M. tuberculosis*, 34 of which were resistant to isoniazid or rifampin and 30 to both; the MIC_{50} and MIC_{90} values were 0.25 and 0.5 mg/ml, respectively, for the evaluated isolates, including strains that were resistant to both isoniazid and rifampin (14). Tedizolid was also tested against *M. tuberculosis*-infected THP-1 macrophages and showed good intracellular killing activity comparable with that of rifampin or moxifloxacin (13). All MICs were lower than the breakpoint established for linezolid by the Clinical and Laboratory Standards Institute (10). No breakpoints for acid-fast bacilli have been proposed.

Tedizolid is presented as a drug with high activity against *M. tuberculosis*. In this study, tedizolid exhibited greater potency than linezolid. Larger studies are needed to provide definitive data on the efficacy of tedizolid in resistant isolates of *M. tuberculosis*. It may be an alternative treatment for resistant *M. tuberculosis*.

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