Effectiveness and safety of clofazimine in multidrug-resistant tuberculosis: a nationwide report from Brazil

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ABSTRACT Although clofazimine is used to treat multidrug-resistant tuberculosis (MDR-TB), there is scant information on its effectiveness and safety. The aim of this retrospective, observational study was to evaluate these factors as well as the tolerability of clofazimine in populations in Brazil, where it was administered at a daily dose of 100 mg·day^{-1} (body weight ⩾45 kg) as part of a standardised MDR-TB treatment regimen until 2006 (thereafter pyrazinamide was used).

All MDR-TB patients included in the Sistema de Informação de Tratamentos Especiais da Tuberculose (SITETB) individual electronic register were analysed. The effectiveness of clofazimine was assessed by comparing the treatment outcomes of patients undergoing clofazimine-containing regimens against those undergoing clofazimine-free regimens and its safety by describing clofazimine-attributed adverse events. A total of 1446 patients were treated with clofazimine-containing regimens and 1096 with pyrazinamide-containing regimens.

Although success rates were similar in patients treated with clofazimine *versus* those treated with pyrazinamide (880 out of 1446, 60.9%, *versus* 708 out of 1096, 64.6%; p=0.054), clofazimine-treated cases exhibited higher death rates due to tuberculosis than pyrazinamide-treated ones (314 out of 1446, 21.7%, *versus* 120 out of 1096, 10.9%) but fewer failures (78 out of 1446, 5.4%, *versus* 95 out of 1096, 8.7%) and less loss to follow-up (144 out of 1446, 10.0%, *versus* 151 out of 1096, 13.8%). No relevant differences were detected when comparing adverse events in patients treated with clofazimine-containing regimens to those treated with clofazimine-free regimens. However, the incidence of side-effects was less than previously reported (gastro-intestinal complaints: 10.5%; hyper-pigmentation: 50.2%; neurological disturbances: 9–13%).

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Introduction

The World Health Organization (WHO) has estimated that 480 000 cases of tuberculosis (TB) occurred in 2015 due to multidrug-resistant strains of *Mycobacterium tuberculosis* [1–5] (*i.e.* those strains resistant to at least rifampicin and isoniazid, the two most important anti-TB drugs) with 190 000 deaths, to which we need to add 100 000 rifampicin-resistant cases. Multidrug-resistant tuberculosis (MDR-TB) also includes extensively drug-resistant tuberculosis (XDR-TB), those strains resistant to any fluoroquinolone and to at least one of the second-line injectable drugs amikacin, capreomycin and kanamycin, and the most severe cases which have a drug-resistance pattern beyond XDR-TB [1–3]. Unfortunately, the success rate reported in treating MDR-TB cases ranges between 50 and 60% (52% globally according to the 2016 WHO report [1]), being lower in cases with a complicated drug-resistance pattern [1–5]. Treating MDR-TB and XDR-TB cases is lengthy (∼18–24 months), often toxic (with frequent adverse events) and costly [1–6].

While important efforts have been made in studying the potentiality of new drugs such as bedaquiline and delamanid [7, 8], research attention has recently been directed towards the so-called "repurposed drugs", *i.e.* those drugs originally designed for indications other than TB which have proved to be useful for treating MDR-TB and XDR-TB. Among these drugs, linezolid [9, 10] as well as various carbapenems [11–15] and macrolides [16] have recently raised scientific interest. Furthermore, clofazimine, a fat-soluble riminophenazine dye originally used to treat leprosy, has shown potentiality *in vitro* and *in vivo* as a sterilising drug to treat MDR-TB [17, 18].

The importance of clofazimine, originally part of WHO Group 5 (those drugs with unclear efficacy) [4], has increased after the WHO recommended the new "shorter regimen" [5], previously known as the Bangladesh regimen, which includes clofazimine [6, 19–26]. Although clofazimine is now part of WHO Group C [5], the evidence available on its safety and efficacy is relatively modest. A recently published trial [17] included only 53 cases treated with clofazimine, whereas the total number of patients in a systematic review on this drug [18] was 599 from eight studies (427 of which were from a single Bangladesh study). No large-scale study reporting individual safety and efficacy data on clofazimine is presently available. This current opportunity is presented by the information collected by the Brazilian National TB programme, where data on safety are collected on an individual basis [26] and effectiveness can be evaluated by comparing the outcomes of two standardised MDR-TB regimens (with and without clofazimine, respectively).

In Brazil the MDR-TB regimen is standardised in the majority of cases [27]. Clofazimine and ofloxacin were used until 2006 and, after this date, pyrazinamide replaced clofazimine. In 2010 levofloxacin replaced ofloxacin as the backbone fluoroquinolone (regimen: 1 month of amikacin or streptomycin five times a week, ethambutol, levofloxacin, pyrazinamide and terizidone, followed by 4 months of amikacin or streptomycin three times a week, ethambutol, levofloxacin, pyrazinamide and terizidone and 12 months of ethambutol, levofloxacin and terizidone) (table 1).

The shift from clofazimine to pyrazinamide followed a request by the WHO, as the global availability of this drug did not allow for treatment of all MDR-TB cases managed in Brazil, and priority use was in the management of leprosy. The new regimen, which is still used in the country, has been designed by the Brazilian National TB programme in agreement with the WHO. The aim of the present study is to evaluate the effectiveness, safety and tolerability of clofazimine administered within a standardised MDR-TB regimen in Brazil.

Methods

This retrospective, observational study is part of an ongoing collaboration involving the European Respiratory Society (ERS), the Brazilian Society of Respiratory Medicine (SBPT) and the Brazilian National TB Programme. With 0.9% of the global TB burden in 2015 (33% of that in the Americas) Brazil notified 30.9 new cases per 100 000 population (63 189 cases) and 2.2 deaths per 100 000 population. According to the Brazilian Ministry of Health, in the same year there were 1027 drug-resistant cases among this number, of which 442 were MDR-TB cases (43.0%) and eight (0.8%) which met the criteria for XDR-TB [1].

All Brazilian patients needing a treatment different to the standard 6-month regimen for drug-susceptible TB are notified in the national register (SITETB). The SITETB system is an individual, electronic register designed as an upgraded version of the previously used software for MDR-TB cases (the TBMR register). It includes demographic and clinical information (age, sex, body weight, education, job information, risk factors and co-morbidities, chest radiography, previous TB treatment, drug-resistance profile and

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TABLE 1 Dosage and drug administration details that comprised the treatment regimens for multidrug-resistant tuberculosis in Brazil, 2000–2010

Drug	Daily dose (mg·kg^{-1})	
	Body weight ⩽45 kg	Body weight >45 kg
Clofazimine-containing regimen (2000–2006)		
Amikacin	500	750–1000[#]
Ofloxacin	400	800
Clofazimine	50	100
Terizidone	500	750
Ethambutol	800	1200
Streptomycin	500	750–1000[#]
Pyrazinamide-containing regimen (2006–2010)		
Amikacin	500	750–1000[#]
Levofloxacin[¶]	750	1000
Pyrazinamide	1000	1500
Terizidone	500	750
Ethambutol	800	1200
Streptomycin	500	750–1000[#]

[#]: at a body weight of ⩾60 kg a dosage of 1000 mg·kg^{-1} was used; [¶]: levofloxacin was introduced in 2010.

TB-treatment duration) as well as treatment outcomes. The information provided by SITETB was analysed and, if not otherwise specified, the WHO definitions for treatment outcomes were used as follows. Treatment completed as recommended by the national policy with no evidence of failure and with three or more consecutive negative cultures taken at least 30 days apart after the intensive phase. Treatment completed: treatment completed as recommended by the national policy with no evidence of failure but with no record that three or more consecutive cultures taken at least 30 days apart after the intensive phase were negative. Treatment failed: treatment terminated or permanent regimen change required in respect of at least two TB drugs due to lack of conversion by the end of the intensive phase, bacteriological reversion in the continuation phase after conversion to negative, evidence of additional acquired resistance to fluoroquinolones or second-line injectable drugs, or adverse drug reactions. Died: patient dies for any reason during the course of treatment. Lost to follow-up: treatment interrupted for two or more consecutive months. Treatment success: the sum of the cured and treatment completed categories [1]. Patients were considered to be smokers when smoking ⩾10 cigarettes·day^{-1} and to be alcohol consumers based on pre-defined daily amounts (*e.g.* 1 dose·day^{-1} of "cachaça", a local alcoholic drink, or ⩾3 beers·day^{-1}).

Drug-susceptibility testing (DST) is usually performed in Brazil for ethambutol and pyrazinamide (this last using Canetti's proportions method with a pyrazinamide concentration of 100 µg·mL^{-1} in previously acidified media). Before 2010, ethionamide and pyrazinamide were tested frequently, while fluoroquinolones and injectable drugs were tested in a minority of patients. After 2011, DST for fluoroquinolones and injectable drugs was performed more frequently, however, DST was not performed for clofazimine. In practice, DST was performed using solid media until 2010 (Löwenstein–Jensen medium, Middlebrook 7H10 agar or Middlebrook 7H11 agar) and then liquid media (Mycobacterial Growth Indicator Tubes; MGIT™; BD Diagnostic, Sparks, MD, USA) was used in quality-assured laboratories belonging to the WHO network [26, 27].

Clofazimine was administered at a dose of 100 mg·day^{-1} or 50 mg·day^{-1} when body weight was ⩾ or <45 kg, respectively, as per Brazilian guidelines. Based on the DST results recorded in the register, the cohort of MDR-TB cases treated from 2000 to 2010 was selected, removing 65 poly-resistant TB cases (those with resistance to two or more drugs including isoniazid but not rifampicin) and 22 mono-resistant TB cases (all were resistant to rifampicin and were removed as having belonged to the pre-Xpert era and, consequently, had not been managed as MDR-TB). The effectiveness of clofazimine was assessed by comparing the treatment outcomes of patients undergoing clofazimine-containing regimens *versus* those undergoing clofazimine-free regimens. Clofazimine safety was assessed by describing adverse events attributed to clofazimine and to each single drug comprising the regimen. Absolute frequencies (percentages) and median values (with interquartile ranges (IQR)) were used to describe qualitative and quantitative variables. When appropriate, Chi-squared, Fisher exact or Mann–Whitney tests were performed to assess statistically significant differences for qualitative and quantitative variables. A multivariable logistic regression analysis was carried out to assess the role of demographic, epidemiological, bacteriological and clinical variables on treatment success. A two-tailed, p-value of less than 0.05 was considered statistically

significant. All statistical analyses were performed using STATA version 14 (StataCorp LP 2015, College Station, TX, USA).

Results

A total of 2542 cases had evidence from DST of resistance to both isoniazid and rifampicin (*i.e.* involved MDR-TB). The comparison of demographic and clinical data in MDR-TB patients, including the drug-resistance profile, is summarised in table 2. A total of 1446 patients were treated with a clofazimine-containing regimen while 1096 patients were treated with a pyrazinamide-containing regimen. The patients treated with clofazimine were significantly less exposed to TB-related risk factors, such as drug abuse, heavy alcohol consumption or a tobacco smoking habit, than those treated with pyrazinamide ($p<0.0001$). Interestingly, clofazimine-treated cases had a lower prevalence of diabetes mellitus than those undergoing the pyrazinamide-containing regimen (57 out of 1445, 3.9%, *versus* 132 out of 1096, 12.1%; $p<0.0001$). The proportion of MDR-TB cases, excluding XDR-TB and pre-XDR-TB cases, was similar (98.6% amongst clofazimine-treated patients *versus* 99.2% amongst pyrazinamide-treated patients), as was the proportion of pre-XDR-TB cases (*i.e.* those resistant to fluoroquinolones or to a second-line injectable drug but not both). Clofazimine-treated patients experienced a median of three (IQR: 2–3) previous anti-TB treatments compared to two (IQR: 2–3; $p<0.0001$) in the pyrazinamide-treated group. No major differences in clinical presentation were detected in patients undergoing clofazimine-containing treatment *versus* those exposed to a clofazimine-free regimen, although the group treated with clofazimine had a slightly greater proportion of pulmonary cases and a slightly smaller proportion of pulmonary and extra-pulmonary cases. Treatment duration for MDR-TB cases was similar under both regimens (18 months). Directly-observed therapy (DOT) was performed to a significantly higher degree amongst those patients receiving the pyrazinamide regimen (67.3%) than amongst those treated with clofazimine (34.6%; $p<0.0001$).

The comparison of treatment outcomes between the standardised regimens is described in table 3. Although treatment success was similar in patients given a clofazimine regimen compared to those on a pyrazinamide regimen (880 out of 1446, 60.9%, *versus* 708 out of 1096, 64.6%; p-value=0.054), the cases treated with clofazimine experienced more deaths due to TB (314 out of 1446, 21.7%, *versus* 120 out of 1096, 11%) but fewer treatment failures (78 out of 1446, 5.4%, *versus* 95 out of 1096, 8.7%) and less loss to follow-up (144 out of 1446, 10.0%, *versus* 151 out of 1096, 13.8%). The proportion of deaths remained significantly higher in the clofazimine-treated group also stratifying cases according to the number of resistances (*i.e.*, three). By the end of treatment, patients on a clofazimine regimen had gained a median of 3 kg of body weight compared with their pre-treatment baseline weight (from 50 kg (IQR: 48.0–58.0) to 53 kg (IQR: 48.0–63.5); table 2).

The multivariable logistic regression analysis showed that treatment success significantly increased or decreased for the role played by the following variables: male gender (odds ratio (OR): 0.8; p=0.009), age (OR: 1.0; p=0.001), employment status (OR: 1.5; p=0.004), advanced educational level (OR: 2.4; $p<0.0001$), high weight before treatment (OR: 1.0; $p<0.0001$), lack of previous exposure to TB therapies (OR: 0.8; $p<0.0001$), longer treatment duration (OR: 1.0; p=0.04), DOT (OR: 1.3; p=0.03) and resistance to ethambutol (OR: 0.7; p=0.002). No relevant differences were detected in the comparison of adverse events in patients treated with a clofazimine-containing regimen compared to a clofazimine-free regimen (table 4).

Discussion

The aim of this study is to evaluate the effectiveness, safety and tolerability of clofazimine used within a standardised MDR-TB regimen in a large cohort of Brazilian TB patients. The study results suggest that both regimens are rather effective in achieving treatment success, as the rates achieved are higher than those reported in the largest MDR-TB cohort ever published [2, 3]. However, both regimens used in Brazil can be considered rather weak as they include three active, or likely to be active, second-line TB drugs. Furthermore, no major differences are detected when comparing adverse events in patients treated with a clofazimine-containing regimen *versus* a clofazimine-free regimen. Interestingly, the cases treated with clofazimine were less exposed to important TB-related risk factors, such as drug abuse and alcohol and tobacco use, while having a lower prevalence of diabetes mellitus. These differences are likely to be due to the economic and demographic changes which occurred over time in Brazil, although information bias (improvement of the surveillance system over time) cannot be excluded. From a clinical point of view, the prevalence of MDR-TB, pre-XDR-TB and XDR-TB cases was similar in both groups, although the patients treated with clofazimine had experienced more previous TB treatments than those treated with pyrazinamide. DOT was performed to a significantly higher degree amongst patients on a pyrazinamide regimen than amongst those receiving clofazimine, as a result of programmatic improvements over time in Brazil and the decision, which was taken in 2005, to make it a strategic component of the anti-TB strategy.

TABLE 2 Demographic, epidemiological and clinical characteristics of the multidrug-resistant (MDR) tuberculosis (TB) cohort in Brazil, 2000–2010

Variable	Clofazimine regimen	Pyrazinamide regimen	p-value
Male	952/1446 (65.8)	714/1096 (65.2)	0.72
Age years	38 (29–47)	39 (28–49)	0.45
Job			
Housewife	33/1446 (2.3)	146/1096 (13.3)	<0.0001
Unemployed	38/1446 (2.6)	200/1096 (18.3)	
Retired	7/1446 (0.5)	58/1096 (5.3)	
Self-employed	66/1446 (4.6)	277/1096 (25.3)	
Other	1302/1446 (90.0)	415/1096 (37.9)	
Period of education years			
0	116/1130 (10.3)	78/1019 (7.7)	<0.0001
1–3	313/1130 (27.7)	235/1019 (23.1)	
4–7	496/1130 (43.9)	374/1019 (36.7)	
8–11	126/1130 (11.2)	251/1019 (24.6)	
>11	79/1130 (7.0)	81/1019 (7.9)	
Resistance pattern			
MDR-TB	1425/1446 (98.6)	1087/1096 (99.2)	0.16
pre-XDR-TB	18/1446 (1.2)	8/1096 (0.7)	0.21
XDR-TB	3/1446 (0.2)	1/1096 (0.1)	0.53
Clinical presentation			
Pulmonary	1429/1446 (98.8)	1058/1096 (96.5)	<0.0001
Extra-pulmonary	14/1446 (1.0)	7/1096 (0.6)	
Pulmonary and extra-pulmonary	3/1446 (0.2)	31/1096 (2.8)	
Pulmonary involvement			
Normal	3/1435 (0.2)	0/1089 (0.0)	<0.0001
Unilateral, cavitary	191/1435 (13.3)	214/1089 (19.7)	
Unilateral, not cavitary	92/1435 (6.4)	85/1089 (7.8)	
Bilateral, cavitary	961/1435 (67.0)	683/1089 (62.7)	
Bilateral, not cavitary	188/1435 (13.1)	107/1089 (9.8)	
Extra-pulmonary involvement			
Lymph nodes	8/16 (50.0)	10/25 (40.0)	0.27
Bones	4/16 (25.0)	3/25 (12.0)	
Pleurae	2/16 (12.5)	2/25 (8.0)	
Other	2/16 (12.5)	10/25 (40.0)	
Weight at baseline kg	50.0 (48.0–58.0)	54.0 (47.0–62.0)	<0.0001
Weight at end of treatment kg	53.0 (48.0–63.5)	58.5 (50.0–68.0)	<0.0001
Number of previous TB therapies	3 (2–3)	2 (2–3)	<0.0001
Acquired resistance	1293/1446 (89.4)	837/1096 (76.4)	<0.0001
Drug use			
Amikacin	1365/1446 (94.4)	810/1096 (73.9)	<0.0001
Ofloxacin	1426/1446 (98.6)	824/1096 (75.2)	<0.0001
Levofloxacin	0/1446 (0.0)	265/1096 (24.2)	<0.0001
Terizidone	1438/1446 (99.5)	1079/1096 (98.5)	0.01
Streptomycin	58/1446 (4.0)	289/1096 (26.4)	<0.0001
Ethambutol	1441/1446 (99.7)	1081/1096 (98.6)	0.005
Ciprofloxacin	0/1446 (0.0)	0/1096 (0.0)	
Metronidazole	2/1446 (0.1)	3/1096 (0.3)	0.66
Clarithromycin	0/1446 (0.0)	2/1096 (0.2)	0.19
Ethionamide	5/1446 (0.4)	39/1096 (3.6)	<0.0001
Moxifloxacin	2/1446 (0.1)	7/1096 (0.6)	0.045
Isoniazid	2/1446 (0.1)	2/1096 (0.2)	1.0
Rifampicin	3/1446 (0.2)	4/1096 (0.4)	0.47
Fluoroquinolones	1426/1446 (98.6)	1082/1096 (98.7)	0.82
Aminoglycosides	1423/1446 (98.4)	1092/1096 (99.6)	0.003
Treatment duration months	18.0 (16.0–20.0)	18.0 (16.0–19.0)	0.03
N of resistance <3	18.0 (15.5–21.0)	18.0 (15.5–19.0)	0.03
N of resistance ⩾3	18.0 (17.0–20.0)	18.0 (17.0–20.0)	0.45
MDR-TB	18.0 (16.0–20.0)	18.0 (16.0–19.0)	0.04
pre-XDR-TB/XDR-TB	23.0 (18.0–33.0)	18.0 (18.0–24.0)	0.73

Continued

TABLE 2 Continued

Variable	Clofazimine regimen	Pyrazinamide regimen	p-value
DOT	500/1446 (34.6)	738/1096 (67.3)	<0.0001
HIV testing	86/1444 (6.0)	84/ 1085(7.7)	0.08
Risk factors			
Exposure to corticosteroids	3/1445 (0.2)	3/1096 (0.3)	1.0
Transplantation	0/1445 (0.0)	0/1096 (0.0)	
Drug abuse	24/1445 (1.7)	107/1096 (9.8)	<0.0001
Alcohol user	52/1445 (3.6)	210/1096 (19.2)	<0.0001
Tobacco user	3/1445 (0.2)	35/1096 (3.2)	<0.0001
Exposure to TNF-α	0/1445 (0.0)	0/1096 (0.0)	
Diabetes mellitus	57/1445 (3.9)	132/1096 (12.1)	<0.0001
Silicosis	3/1445 (0.2)	2/1096 (0.2)	1.0
Co-morbidities			
Neoplasia	6/1445 (0.4)	9/1096 (0.8)	0.19
Renal failure	3/1445 (0.2)	5/1096 (0.5)	0.30
Hepatitis	5/1445 (0.4)	9/1096 (0.8)	0.17
Mental disorder	16/1445 (1.1)	30/1096 (2.8)	0.002
AIDS	79/1445 (5.5)	76/1096 (7.0)	0.13
Other diseases	42/1445 (2.9)	128/1096 (11.7)	<0.0001
Seizures	0/1445 (0.0)	0/1096 (0.0)	
Drug-resistance			
Pyrazinamide	614/1123 (54.7)	139/466 (29.8)	<0.0001
Ethambutol	586/1368 (42.8)	341/1047 (32.6)	<0.0001
Aminoglycosides	639/1384 (46.2)	398/1031 (38.6)	<0.0001
Amikacin	6/39 (15.4)	3/31 (9.7)	0.72
Kanamycin	1/6 (16.7)	2/16 (12.5)	1.0
Streptomycin	638/1381 (46.2)	397/1030 (38.5)	<0.0001
Fluoroquinolones	18/46 (39.1)	8/35 (22.9)	0.12
Ofloxacin	16/44 (36.4)	6/32 (18.8)	0.10
Levofloxacin	2/3 (66.7)	0/4 (0.0)	0.14
Moxifloxacin		1/1 (100.0)	
Ciprofloxacin	1/6 (16.7)	1/15 (6.7)	0.50
Ethionamide	342/1015 (33.7)	72/323 (22.3)	<0.0001
Clofazimine			
Terizidone	1/1 (100.0)		
n of resistance	3 (3–4)	3 (2–3)	<0.0001
N of resistance ⩾3	1238/1446 (85.6)	676/1096 (61.7)	<0.0001

Data are presented as n/n (%) or median (interquartile range), unless otherwise stated. XDR: extensively drug-resistant; DOT: directly-observed therapy; TNF-α: tumour necrosis factor-α.

The outcome in the clofazimine-treated group (60.9% treatment success) was slightly less positive than that reported in a recent systematic review (65% treatment success in 599 cases treated with clofazimine) [18]. This systematic review included a single large study, the Bangladesh study [20], and several small-scale studies which enrolled between 5 and 46 patients [18]. Unfortunately, several details important for the comparison of our study with the systematic review are not available. One study quoted in the systematic review reports the duration of treatment with clofazimine as being 608 days (*i.e.* 20 months) as opposed to the 18 months in our study, while information on the daily dose of clofazimine is missing in five out of the eight studies in the review. However, 84% of the cases analysed in the systematic review were prescribed clofazimine at a dose of 100 mg·day^{-1}, the same as administered in our study. Treatment outcome definitions varied across studies in the review [18] and demographic data were also different. The proportion of females ranged between 20 and 85.7%, the median being 34.2% in our study, while the mean age ranged between 26 and 50 years (the median age in our study was 38 years). The HIV prevalence, when tested, was very low (one case per series only was HIV-positive, this being 6% in our cohort).

In a recent randomised open trial performed in China, 53 cases treated with clofazimine (administered for 21 months at a dose of 100 mg·day^{-1}) were compared with 52 controls. Treatment success was significantly higher in the treatment group when compared to the control group (73.6% *versus* 53.8%; p=0.035) and culture conversion was significantly more rapid, as was cavity closure (evaluated by chest radiography) [17]. Importantly, the background regimen used in the Chinese trial was stronger than in our study, including prothionamide, pyrazinamide and a fluoroquinolone (moxifloxacin, gatifloxacin or levofloxacin) (100% of

TABLE 3 Treatment outcomes for the multidrug-resistant tuberculosis (TB) cohort in Brazil, 2000–2010

Treatment outcome	Clofazimine regimen	Pyrazinamide regimen	p-value
Outcome			
Cured	421/1446 (29.1)	384/1096 (35.0)	<0.0001
Treatment completed	459/1446 (31.7)	324/1096 (29.6)	
Died	314/1446 (21.7)	120/1096 (11.0)	
Died (non-TB cause)	29/1446 (2.0)	22/1096 (2.0)	
Lost to follow-up	144/1446 (10.0)	151/1096 (13.8)	
Failed	78/1446 (5.4)	95/1096(8.7)	
Treatment success	880/1446 (60.9)	708/1096 (64.6)	0.054
n of resistance <3			
Cured	46/208 (22.1)	141/420 (33.6)	0.003
Treatment completed	75/208 (36.1)	141/420 (33.6)	0.54
Died	51/208 (24.5)	36/420 (8.6)	<0.0001
Died (non-TB cause)	4/208 (1.9)	13/420 (3.1)	0.38
Lost to follow-up	17/208 (8.2)	55/420 (13.1)	0.07
Failed	15/208 (7.2)	34/420 (8.1)	0.69
n of resistance ⩾3			
Cured	375/1238 (30.3)	243/676 (36.0)	0.01
Treatment completed	384/1238 (31.0)	183/676 (27.1)	0.07
Died	263/1238 (21.2)	84/676 (12.4)	<0.0001
Died (non-TB cause)	25/1238 (2.0)	9/676 (1.3)	0.27
Lost to follow-up	127/1238 (10.3)	96/676 (14.2)	0.01
Failed	63/1238 (5.1)	61/676 (9.0)	0.001

Data are displayed as n/n (%) unless otherwise stated.

cases), capreomycin or amikacin (79.2% of cases), ethambutol (39.6% of cases), and clarithromycin (49.1% of cases) based on DST results. All the available data suggests that clofazimine is active in MDR-TB cases. In our study, the slightly better results of the pyrazinamide-containing regimen are explained by the higher resistance profile initially observed in Brazil, particularly to fluoroquinolones (>30%) [26], by the higher number of previous treatments, and by the higher DOT coverage in this group. While a delayed diagnosis might have played a role we cannot study this based on the available data. However, patients treated with

TABLE 4 Adverse events notified in the multidrug-resistant tuberculosis cohort in Brazil, 2000–2010

Adverse event	Clofazimine regimen	Pyrazinamide regimen	p-value
Hyperpigmentation	725/1445 (50.2)	63/1096 (5.8)	<0.0001
Arthralgia	194/1445 (13.4)	231/1096 (21.1)	<0.0001
Gastrointestinal intolerance	151/1445 (10.5)	102/1096 (9.3)	0.34
Hearing impairment	133/1445 (9.2)	86/1096 (7.9)	0.23
Insomnia	104/ 1445 (7.2)	73/1096 (6.7)	0.60
Headache	88/1445 (6.1)	70/1096 (6.4)	0.76
Mental disorder	85/1445 (5.9)	51/1096 (4.7)	0.17
Visual impairment	56/1445 (3.9)	38/1096 (3.5)	0.59
Renal impairment	27/1445 (1.9)	13/1096 (1.2)	0.17
Neuropathy	0/1445 (0.0)	0/1096 (0.0)	
Haematologic abnormality	0/1445 (0.0)	0/1096 (0.0)	
Allergic reaction	0/1445 (0.0)	2/1096 (0.2)	0.19
Electrolyte disorder	0/1445 (0.0)	0/1096 (0.0)	
Hypothyroidism	0/1445 (0.0)	0/1096 (0.0)	
Hyperuricemia	0/1445 (0.0)	0/1096 (0.0)	
Haematuria	0/1445 (0.0)	0/1096 (0.0)	
Nystagmus	2/1445 (0.1)	1/1096 (0.1)	1.0
Seizure	0/1445 (0.0)	1/1096 (0.1)	0.43
Other	0/1445 (0.0)	2/1096 (0.2)	0.19

Data are displayed as n/n (%) unless otherwise stated.

clofazimine had significantly fewer risk factors or co-morbidities (drug or alcohol abuse, a tobacco smoking habit, or diabetes mellitus).

The initial annual mortality rate decreased over time in the clofazimine group (data not shown) and we believe that while this cannot be attributed to cardiological complications (prolonging of the Q-T interval) it can be attributed to a higher prevalence of resistance (of which we have evidence) and delayed diagnosis (which, as stated above, we cannot investigate). Increased MDR-TB mortality rates are commonly observed in the early stages of implementation of large-scale MDR-TB programmes due to improved notification (deaths which, although previously occurring, were not notified) and programmatic factors such as delayed diagnosis, delayed treatment initiation and insufficient drug availability (leading to the use of drugs generally available at hospital level such as ciprofloxacin/ofloxacin and/or amikacin). Therefore, given that systematic DST for all drugs was not possible, some of the MDR-TB cases might have in fact been pre-XDR-TB or XDR-TB cases, further contributing to the increase in mortality. The use of a weaker fluoroquinolone (ofloxacin) in the clofazimine regimen compared to that used in the pyrazinamide regimen (levofloxacin) might also have contributed to increased mortality.

Recent discussions have focussed on the mechanism of action of clofazimine, which is not yet well known [28, 29]. In fact, clofazimine reaches high concentration in macrophages and is able to tackle the inhibitory effect of *M. tuberculosis* derived factors on phagocyte intracellular killing. The potential synergistic effect of clofazimine with γ-interferon, pyrazinamide and clarithromycin deserves to be studied further [18]. In addition, clofazimine has a long half-life, which can also contribute to the final effect of the drug although its effect is evident after the second week of treatment [28–33]. Furthermore, a recent study suggests that clofazimine has excellent sterilising activity in the murine model [29]. Recently, the possibility of developing a rapid test to detect susceptibility to clofazimine has been demonstrated, allowing the correct identification of eligible individuals in the near future [31]. Furthermore, recent evidence indicates that the use of clofazimine does not favour the development of resistance against bedaquiline [32, 33]. In fact, cross-resistance with bedaquiline has been attributed to the transcriptional regulator Rv0678 with concomitant upregulation of the multi-substrate efflux pump MmpI.5. As such, the mutation in Rv0678 initially considered responsible for the cross-resistance should be considered a confounding factor [32, 33].

In terms of safety, the global evidence available before our study consisted of the previously mentioned systematic review and a clinical trial [17, 18]. The systematic review [18] concluded that although the optimal clofazimine dose is not yet known (being 100 mg·day^{-1} in the majority of available studies), adverse events are in general minor and rarely life-threatening. Although important, the review is affected by relevant between-study heterogeneity and by the epidemiological observational nature, with potential selection biases. Gastro-intestinal intolerance was observed in 40–50% of cases, with 75–100% reporting brownish skin pigmentation while ichthyosis and skin darkness were notified in 8–20% of patients. In the Chinese trial [17], 94.3% of cases reported adverse events of the skin and 47.2% reported ichthyosis, while 11.3% had gastro-intestinal disturbances and, overall, <4% had neurological complaints. In our study, adverse events were even lower: gastro-intestinal complaints were recorded in 10.5% of cases and hyper-pigmentation in 50.2% of patients. Neurological disturbances were reported in 9–13% of cases.

Our study has several limitations, being based on programmatic information in a setting where it is not possible to perform a complete DST for each patient, and where DST, despite being performed in quality-controlled laboratories belonging to the WHO network, cannot be performed with a complete panel in all cases. As a consequence, no additional information can be provided as to whether any of the MDR-TB cases were, in reality, pre-XDR-TB or XDR-TB cases. This shortcoming can hinder the interpretation of the results and higher proportions of difficult-to-treat cases (*i.e.*, pre-XDR-TB or XDR-TB) could, in fact, affect the effectiveness indicators. In terms of effectiveness, the difference in the prevalence of risk factors discussed above is probably biasing the comparison in favour of the pyrazinamide-containing regimen, which was introduced in a mature phase of the Brazilian MDR-TB programme. Safety was evaluated using programmatic data which could be associated with a high rate of under-reporting. Therefore, a complete safety assessment (*e.g.* an ECG analysis) was not performed. The risk of underestimating adverse events (those occurring but not being recorded) is potentially high and should be kept under consideration when safety data from routine surveillance programmes are generalised to a population context. Furthermore, it was not possible to homogenise for the main confounding variables of the two treatment arms. If the population-based dataset is an important scientific resource (showing a real-world scenario), the role played by multiple confounding variables and their interactions could hinder the reliability and comparison of the findings with other settings. An epidemiological design, based on inclusion and exclusion criteria, can reduce the background noise related to the confounding covariates. The logistic regression model cannot significantly preserve the inferentiality of the findings when the sampling source is represented by a population whose statistical units are

consecutively recruited. Moreover, important variables such as individual follow-up of the enrolled patients were not collected. However, the database is quality-controlled (being used to authorise the prescription of second-line TB drugs) and contains a large, nationwide comprehensive record (over 6000 cases) from Brazil, one of the high TB-burden countries.

Conclusions

To our knowledge, this is the largest ever cohort treated with clofazimine and the first study reporting all cases from a major country. The results of the therapeutic performance of clofazimine within a standardised regimen indicate that the drug is effective at the programmatic level (ensured success rates above 60%), safe [17, 18] and, as discussed elsewhere [13], does not increase the prevalence of drug resistance. The take-home message for the clinician is that: 1) clofazimine can be added to an optimised background regimen, designed as per WHO recommendations, within both individualised or standardised regimens (which include the newly recommended "shorter" regimen) [5, 34]; and 2) the tolerability of the drug seems to be confirmed. Furthermore, the study results confirm that the 100 mg·day^{-1} dose of clofazimine is probably adequate, as recently demonstrated both *in vitro* and *in vivo* in a murine model [28]. In conclusion, clofazimine seems to have the potential to further contribute to the successful treatment of more TB cases affected by multidrug-resistance, although results from randomised, controlled clinical trials are necessary to provide a definite answer [30].

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