Pattern of Drug Resistance Among Patients Presenting with Relapse of Pulmonary Tuberculosis

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ABSTRACT

Objective: To assess the patterns of drug resistance (DR) among patients previously treated for pulmonary tuberculosis (TB).
Study Design: Cross-sectional study.
Place and Duration of Study: Department of Medicine, Pak-Emirates Military Hospital, Rawalpindi Pakistan, from Jul 2020 to Mar 2021.
Methodology: Patients with previously treated pulmonary TB were included, based on their clinical history and characteristic chest X-ray findings. Patients were evaluated through an examination of sputum and/or bronchoalveolar washings for acid-fast bacilli (AFB), Mycobacterium TB-GeneXpert/RIF (Rifampicin) assay, and mycobacterial culture with drug susceptibility testing (DST). Patients with resistance to any anti-TB drug were classified as cases of DR-TB, and a pattern of drug resistance was documented.
Results: One hundred twenty patients were identified as having been previously treated for TB. DST demonstrated DR-TB among 38(31.7%) cases, while isoniazid (INH) resistance was the most commonly reported problem in 29(24.2%) cases. Up to 26(21.6%) relapsing cases were resistant to Rifampicin (RFM). Moreover, 15(12.5%) instances of resistance against second-line anti-TB drugs (fluoroquinolones and aminoglycosides) were documented. Multidrug-resistant TB (MDR-TB) was the most prevalent resistance pattern in 20(16.7%) cases, followed by mono-drug-resistant TB in 7(5.8%) and extensively resistant TB (XDR-TB) in 2(1.6%) cases. No definitive resistance pattern was evident among 9(7.5%) cases.
Conclusion: MDR-TB constitutes a major barrier in the line of TB eradication in Pakistan. Strict compliance with anti-TB protocols, along with a universal application of DST, can minimize the overall prevalence of relapsing cases of TB.

Keywords: Antitubercular Agents, Drug Resistance, Drug Susceptibility Testing, Extensively Drug-resistant Tuberculosis, Multidrug-resistant Tuberculosis.

INTRODUCTION

Tuberculosis (TB), formerly known as consumption or phthisis (to wither away), is a multi-system disease that is classified as one of the top ten causes of mortality in developing countries. This highly contagious infection is caused by an Acid Fast bacillus, i.e., *Mycobacterium tuberculosis*, which spreads through direct person-to-person contact via droplet infection.[1,2]

Pakistan accounts for up to 60% of the cases of TB in the Eastern Mediterranean region of the World Health Organization (WHO), with an overall prevalence of 376 cases per 100,000 population.[3] With the rising trend of Drug susceptibility testing techniques, reports of drug-resistant cases of TB (DR-TB) have also become quite prevalent in the South Asian country.[4] In Pakistan, anti-tuberculosis therapy (ATT) has been made available to the general public

through the platform of the National Tuberculosis Control Programme (NTP). A few of the notorious factors which have substantially contributed to the contact spread of DR-TB include low socioeconomic status, poor hygiene, overcrowding, immuno-suppression due to HIV infection (human immuno-deficiency virus), diabetes, and inadequate social awareness regarding disease prevention and treatment.[5,6]

Epidemiological statistics pertinent to DR-TB have been evaluated worldwide. As per evidence, the global prevalence of MDR-TB approximates 5.6% among all TB cases, and the trend is rising. According to the Global Tuberculosis Report, India has the highest burden of TB (27%) and MDRTB (24%) in the world.[7] Samples from the Indian TB population show that nearly 20.4% of previously treated individuals had INH and RFM-resistant *Mycobacterium* in their sputum cultures. Meanwhile, 2% of cases also displayed resistance against Streptomycin. In China, drug-resistant TB is approximately ten times more prevalent among relapsing individuals as compared to

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newly diagnosed cases.[8] Recent epidemiological data from Pakistan have shown an alarmingly high percentage of MDR-TB (69%) among the patients previously treated with tuberculosis.[9] Pakistan has come to be included among seven countries that carry more than two-thirds of the disease burden of TB .Keeping in view the fact that the third-world population is in dire need of a robust TB control program, this study was designed to elaborate on the dynamic and complex patterns of drug resistance among TB patients in Pakistan.

METHODOLOGY

The cross-sectional study was conducted from July to March 2021 at the Medicine Department of Pak Emirates Military Hospital, Rawalpindi Pakistan, after permission from the Hospital Ethical Committee. The sample size was calculated using the WHO sample size calculator, keeping 4.36% as the reported prevalence of previously treated bacteriologically positive patients.[10]

Inclusion Criteria: Patients of either gender, aged above 12 years who presented with relapse of pulmonary TB suggested by clinical presentation, chest X-ray / High-Resolution CT chest findings and confirmed by Sputum or Bronchoalveolar acid-fast staining and / or Gene Xpert / RIF Assay, were included.

Exclusion Criteria: Patients with extrapulmonary or Miliary TB, patients reporting any new TB contact were excluded.

Thorough informed consent was taken from all the patients. Previously treated TB cases were defined as those individuals who had initially tested positive for tuberculosis via acid-fast staining or Gene Xpert / RIF assay done on sputum or bronchoalveolar lavage (BAL) and were administered standard anti-TB drug regimen comprising of Isoniazid, Rifampicin, Ethambutol and Pyrazinamide for six months with clinical improvement as indicated by weight gain and resolution of chest findings.

Two sputum specimens were collected for microbiological examination including one spot sample. They sent for MTB (Mycobacterium TB) Gene-Xpert/RIF (Rifampicin) assay, acid-fast bacilli (AFB) smear and mycobacterial culture with drug susceptibility testing. Patients unable to produce sputum were offered bronchoscopy, while bronchial washings/bronchoalveolar lavage and samples were sent for Gene-Xpert/RIF assay and culture with drug

susceptibility testing. The patients who were found to have resistance to any anti-TB drug were classified as having drug-resistant tuberculosis. The pattern of drug resistance was noted via drug susceptibility testing. The specialized data collection proforma was developed for this study, which included questions regarding drug sensitivity results and smear positivity.

Statistical Package for Social Sciences (SPSS) version 24.0 was used for the data analysis. Quantitative variables were expressed as Mean±SD and qualitative variables were expressed as frequency and percentages.

RESULTS

One hundred twenty patients of both genders previously treated for TB were sampled using consecutive sampling techniques. The mean age of these subjects was 32.59±13.12 years. 19(15.8%) had comorbid conditions. Ten individuals had diabetes mellitus, whereas one individual had rheumatoid arthritis, which was being treated with methotrexate and prednisolone. 27(22.5%) patients had been treated for TB with standard ATT for more than six months previously. Within this previously treated patient cohort, drug susceptibility testing demonstrated that a total of 38(31.7%) cases had DR-TB. Among these drugs, resistance against INH was the most commonly reported problem. 29(24.2%) patients were found to have it. The second most common resistance was towards Rifampicin, with 26(21.7%) patients with relapsing TB cases being found resistant to it. Pyrazinamide resistance ranked third with 14 cases (11.6%).

15(12.5%) cases of resistance against second-line ATT drugs were documented via drug susceptibility testing, most prominently against fluoroquinolones, 13(10.8%). Only 1.67% (n=2) cases were resistant against second-line Injectable drugs.

In terms of drug resistance patterns, MDR-TB was found to be the most prevalent form of resistance found in 20(16.7%) (Figure-1), followed by monodrug-resistant TB found in 7(5.8%) and XDR-TB found in 2(1.67%) cases (Table). Among 9(7.5%) cases of DR-TB, no identifiable pattern of drug resistance was evident. 9(7.5%) patients were found to have resistance to Fluoroquinolones. Among these nine patients, 4(3.3%) also resisted Rifampicin. Isolated resistance to Rifampicin was not documented (Figure-2).

The prevalence of MDR-TB has been evaluated

Table: Patterns of Drug Resistance (DR-TB) among the Previously Treated Cases (n=120) ATT: Anti-tuberculosis therapy; INH: Isoniazid; RFM: Rifampicin; FQL: Fluoroquinolones; AGSDs: Aminoglycosides; DR-TB: Drug-resistant tuberculosis; MDR-TB: Multidrug-resistant tuberculosis; XDR-TB: Extensive drug-resistant tuberculosis.

| Pattern of Drug Resistance | DR-TB cases = 38 (31.7%) | | | | | | |
| | First-line ATT drugs | | | | | Second-line ATT drugs | |
	INH (%)	RFM (%)	Ethambutol (%)	Pyrazinamide (%)	Streptomycin (%)	FQL (%)	Injectable AGSDs (Amikacin, Kanamycin, and Capreomycin) (%)
Mono DR-TB	7(5.8%)	Nil	Nil	Nil	Nil	Nil	Nil
MDR-TB	20(16.7%)	20(16.7%)	7(5.8%)	14(11.7%)	4(3.3%)	2(1.67%)	Nil
XDR-TB	2(1.67%)	2(1.67%)	Nil	Nil	Nil	2(1.67%)	2(1.67%)
Non-MDR, non-XDR	Nil	4(3.3%)	Nil	Nil	Nil	9(7.5%)	Nil
Total	29(24.2%)	26(21.7%)	7(5.8%)	14(11.7%)	4(3.3%)	13(10.8%)	2(1.67%)
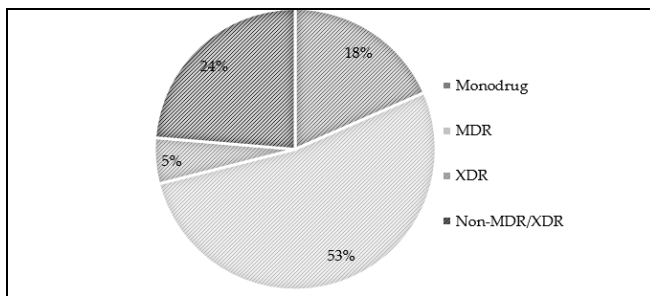
Figure-1: Drug Resistant Tuberculosis
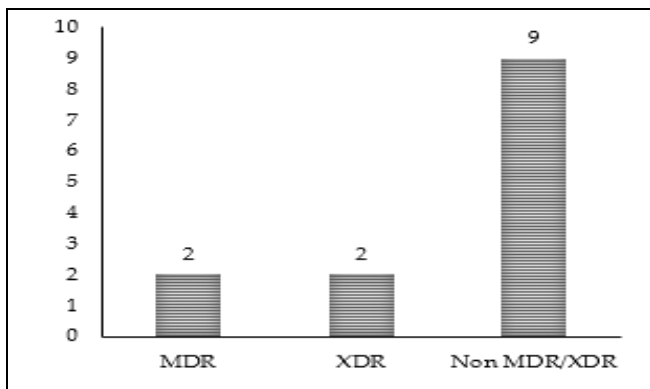
Figure-2: Fluoroquinolone Resistance Among Relapse TB Patients with DR TB on Drug Susceptibility Testing

DISCUSSION

MDR-TB is an emerging health crisis in Pakistan which is not only difficult to treat but requires expensive drugs for a prolonged period of time, leading to a decline in adherence to therapy on the part of the patient. Results of this study have shown that nearly 16.7% of the individuals who have been previously treated for TB develop resistance against two major first-line ATT drugs (INH and RFM).

throughout the globe. Hasan *et al*. have reported that out of an estimated 518,000 active cases of tuberculosis in Pakistan, 15,000 are MDR-TB.[11] In the neighbouring countries, among the previously treated cases of pulmonary TB, MDR-TB strains were isolated from nearly 22.2% of individuals in China.[12] The overall prevalence of DR-TB in various Chinese territories ranges from 27.5-67.5% among previously treated patients. Additionally, 39.2% of the previously cured cases were found to be INH-resistant in contrast to 24.2% cases of INH resistance determined by the authors. Gegia *et al*. have asserted that the treatment of such INH-resistant cases merely with the aid of other first-line agents poses a potential risk in the form of therapeutic failure and TB relapse.[13] In India, the prevalence of MDR-TB has been estimated at around 12.5%. In Bangladesh, the proportion of MDR-TB was 2.3% among new and 13.8% among previously treated TB patients, as reported in literature.[14,15] Moreover, treating these cases empirically is leading to a rise in the multidrug-resistant TB epidemic. Such cases need to be addressed cautiously by utilization of second-line ATT drugs.[16]

Regarding RFM resistance, the authors found an incidence of 21.7%, whereas monodrug resistance was not observed among patients with RFM resistance. Contrariwise, other researchers have found purely RFM resistance among as many as 1.1% cases of DR-TB, while a cumulative incidence of nearly 9.4% was recorded for RFM resistance.[17] This situation enhances the overall value of drug susceptibility testing and makes it even more important to enhance our laboratory capacity for drug susceptibility testing.18 Moreover, comparative analysis shows that these statistics reveal a more dreadful scenario of ATT

resistance in Pakistan, especially considering our resource limitations. Nevertheless, interregional demographic factors could be crucial in interpreting these results.

In contrast to MDR-TB, XDR-TB is much less widespread in the world. The authors found XDR-TB in 1.67% of the previously treated TB cases, which reveals that another threat is knocking at our doors if we do not nip it while it's in the bud. Refractoriness to ofloxacin was reported among 3.76% of TB patients in China.[16] Previous injudicious exposure to FQLs for empirical treatment of infections has been cited as one of the chief driving factors behind it.[17] Epidemiological statistics from Pakistan have shown that FQL resistance persists among as many as 52% of cases of MDR-TB.[18] Additionally, linezolid and newer agents such as bedaquiline have been utilized as potential candidates against the XDR strains of *M. tuberculosis*.[19,20]

LIMITATION OF STUDY

Cases of extrapulmonary TB were not considered by the authors, and this could undermine the statistical results of research as Extrapulmonary TB is even difficult to treat.

CONCLUSION

Drug-resistant tuberculosis represents a substantial health challenge in the path of TB eradication in Pakistan, posing a hefty expense to our slow-growing economy. With a sharply rising incidence of MDR-TB among previously treated individuals, only strict compliance to TB treatment protocols coupled with judicious use of anti-TB antibiotics can minimize the overall risk of TB relapse because of the potential for the disease to become drug-resistant. A routine application of drug susceptibility testing in patients of Tuberculosis who are at risk of having drug-resistant tuberculosis will form a cornerstone in the eradication of this disease from our country.

Conflict of Interest: None.

Authors' Contribution

Following authors have made substantial contributions to the manuscript as under:

MH & MS: Data acquisition, data analysis, data interpretation, critical review, approval of the final version to be published.

IF & AA: Study design, data interpretation, drafting the manuscript, critical review, approval of the final version to be published.

FS & MI: Conception, data acquisition, drafting the manuscript, approval of the final version to be published.

Authors agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

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