Original Article

Drug resistance patterns in pulmonary tuberculosis

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Abstract

Objective: To determine the resistance patterns of mycobacterium tuberculosis (MTB) isolates among category I and II patients of pulmonary tuberculosis.

Methods: This cross sectional study was conducted at the Department of Medicine, Liaquat University of Medical and Health Sciences Jamshoro, from November 2008 to September 2009. Patients were divided into category I and II. The sputa were collected, stained with Ziehl-Nielsen (Z-N) staining and ultimately inoculated on Lowenstein-Jensen (L-J) media for six weeks. Out of 890 pulmonary tuberculosis (PTB) patients, the growth was obtained in 285 cases. The Drug sensitivity testing (DST) for Isoniazid (INH), Rifampicin (RIF), Ethambutol (EMB) Pyrazinamide (PZA) and Streptomycin (SM) were performed. The data was analyzed on SPSS 10.0. A p-value of <0.05 was taken as significant.

Result: Out of 285 cases, 176 (61.75%) were male and 109 (38.24%) female. The mean age was 37 ± 19.90 years. The DST showed drug sensitive and drug resistant isolates in 80 (28.05%) and 205 (71.92%) cases respectively (p=0.001). The drug resistant tuberculosis (DR-TB) rates for individual drugs; INH, RIF, EMB, PZA and SM were 51,22%, 15.4%, 13.33%, 9%12, and 3.85% respectively (p=0.03). The MDR-TB isolates were detected in 120 (42.10%) cases, including 5 (5.88%) in category I and 115 (57.50%) in category II patients (p=0.0001).

Conclusion: Drug resistant and multidrug resistant tuberculosis was observed mainly in category II patients. However, primary MDR was also observed in category I patients and reflects dissemination of MDR cases within the community.

Keywords: Drug resistant tuberculosis, Multidrug resistance, MDR-TB. (JPMA 61:229; 2011).

Introduction

Tuberculosis (TB) is a major cause of illness and death worldwide, especially in Asia and Africa. Globally, 9.2 million new cases (139 per million populations), including 4.1 million new smear-positive cases (44% of the total) and 1.7 million deaths from TB occurred in 2006. This is an increase from 9.1 million cases in 2005, due to population growth. The prevalence of tuberculosis was 14.4 million in 2006 and an estimated 0.5 million cases of multidrug resistant TB (MDR-TB).[1] In Pakistan, the prevalence of tuberculosis reported in 2006 was 263 cases per million population and the incidence of 181 per million population per year. According to latest WHO report, the incidence of MDR-TB in Pakistan in new and previously treated cases is 3.4% and 36% respectively.[1] MDR-TB, defined as resistance of Mycobacterium tuberculosis (MTB) to at least Isoniazid and Rifampicin,[2] is a major threat to the tuberculosis control measures. Multi-drug resistant strains of MTB are particularly problematic in developing countries because alternative agents, such as cycloserine, amikacin, and the quinolones are expensive, less effective, and less well tolerated than the standard counterparts.[3-5] Acquired drug resistance of MTB is defined as the acquisition of resistance to anti-tuberculosis drugs by the multiplication of the resistant mutant strains of bacteria as a result of inadequate chemotherapy. Persons at risk for MDR-TB include: previously treated for tuberculosis, contacts of patients previously treated and/or known to have MDR-TB, dwellers from developing countries, and patients with AFB positive sputum after 3 months of therapy.[6] Epidemiologic studies have denoted that multi-drug resistance is a man-created phenomenon. Exposure to in-appropriate antituberculous therapy selectively allows for multiplication of resistant bacilli, thus becoming dominant and leading to therapeutic failure. Resistant strains may be transmitted from person to person.[7] MDR-TB strains usually acquire resistance through a series of single locus mutations.[8] W-Beijing family, the most common single MDR-TB genotype that originated in Asia has become the dominant strain.[9] The W-Beijing family has spread throughout the world and epidemics have been widely reported, including the United States. The Beijing-strain related to strain W from New York City has been increasing in prevalence and is associated with high levels of treatment failure and acquired drug resistance.[10] WHO has estimated that on average a person with MDR-TB infects up to 20 people in lifetime.[11] In this study, we aimed to determine the resistance pattern of mycobacterium tuberculosis isolates among category I and II patients of

pulmonary tuberculosis (PTB).

Subjects and Methods

This prospective study was conducted at the Department of Medicine, Liaquat University of Medical and Health Sciences Jamshoro, from November 2008 to September 2009. Total 890 suspected PTB cases were selected for the study who fulfilled all of the following criteria: i) clinical history suggestive of tuberculosis. ii) Sputa positive for AFB. iii) CXR findings consistent with pulmonary tuberculosis. iv) Patients with past and present history of antituberculous drugs who had discontinued the drugs because of unknown reason. Smear negative patients were not included. Clinical suspicion of tuberculosis was based upon history; such as low grade fever with evening rise, night sweats, anaemia, malaise, weight loss, elevated ESR and organ specific findings. Category I was defined as i). New smear positive PTB ii). New smear negative PTB with extensive parenchymal involvement. And category II defined as previously treated smear positive PTB i.e.; relapse, treatment interruption, and treatment failure.[12] One spot sputum specimen (about 5 ml) and another morning specimen were collected under the guidance of a doctor. Both the specimens were sent to the laboratory as soon as possible. Two direct smears were prepared from each of the two specimens for Ziehl-Nielsen (Z-N) staining. Each specimen was processed for culture by digestion, decontamination and concentration following modified Petroff's method and was inoculated on Lowenstein-Jensen (L-J) media for six weeks.[13,14] The DST was performed for INH, RIF, EMB and SM after isolation by using the resistance proportion method on the 7H10 Middlebrook medium. The estimation of growth or no growth of mycobacterium tuberculosis strains were based on the presence of single critical concentration of antituberculous drug. The critical concentration of an antituberculous drug represented the lowest concentration of the drug in the medium that inhibited the susceptible strains. The resistance was defined if over 1% of the mycobacteria strains were able to grow.[15,16] The study protocol was approved by ethics committee of the institute. Informed written consent was taken from all the patients. MDR- TB was defined as simultaneous resistance of an isolate to isoniazid and rifampicin. The quantitative and qualitative variables were calculated by student's t-test and Chi square test respectively. The data was analyzed on SPSS version 10.0 for windows. A p-value of <0.05 was taken as significant.

Results

Out of 890 pulmonary tuberculosis (PTB) cases, the growth on culture media was obtained in 285 cases. Remaining specimens were rejected due to contamination or some other different reasons. Out of 285 cases, 176 (61.75%) were male and 109 (38.24%) female. Mean age was 37 ± 19.90 years. The details of demographic data and other

Table-1: Demographic data in the study group (n=285).

Variable	Category I (n=85)	Category II (n=200)	Total (n=285)	p-value
Male	78(91.26%)	98 (49%)	176 (61.75%)	0.59
Female	7(8.23%)	102(51%)	109 (38.24%)	0.61
Age (years)	28±19.3	38±17.9	37±19.9	0.70
Family history (TB)	24(28.23%)	67(33.5%)	91(31.92%)	0.001
Previous ATT *	10(11.7%)	177(88.5%)	187(65.6%)	0.001
Diabetes mellitus	15(17.6%)	39(19.5%)	54(18.94%)	0.04
Chronic liver disease	34(40%)	56(28%)	90(31.57%)	0.61
Haemoptysis	12(14.1%)	34(12%)	46(16.14%)	0.03
Pleural effusion	19(22.35%)	43(21.5%)	62(21.75%)	0.06
Cavitations	17(20%)	87(43.5%)	104(36.49%)	0.05
Pneumothorax	0(0%)	13(6.5%)	13(4.56%)	0.07

* anti-tuberculosis therapy.

Table-2: Drug resistant patterns in the study group (n=285).

Variable	Category I (n=85)	Category II (n=200)	Total (n=285)	p-value
Drug sensitive	70(82.3%)	10(5%)	80(28.05%)	0.001
Drug resistance	15(17.64%)	190(95%)	205(71.92%)	0.01
◆ Isoniazid	06(7.05%)	140(70%)	146(51.22%)	0.03
◆ Rifampicin	04(4.07 %)	85(42.5%)	89(15.4%)	0.04
◆ Ethambutol	03(3.52 %)	35(17.57%)	38(13.33%)	0.02
◆ Pyrazinamide	03(3.52 %)	23(11.5%)	26(9%12)	0.01
◆ Streptomycin	0(0%)	11(5.5%)	11(3.85%)	0.03
MDR-TB *	5(5.88%)	115(57.50%)	120(42.10%)	0.0001

* Multidrug resistant tuberculosis.

variables are shown in Table-1. The DST showed drug sensitive and drug resistant isolates in 80 (28.05%) and 205 (71.92%) cases respectively (p=0.001). The DR-TB rates for individual drugs; INH, RIF, EMB, PZA and SM were 51.22%, 15.4%, 13.33%, 9%12, and 3.85%respectively (p=0.03). The MDR-TB isolates were detected in 120 (42.10%) cases, including 5 (5.88%) in category I and 115 (57.50%) in category II patients (p=0.0001). The details of DR-TB and MDR-TB are shown in Table-2. The association of DR-TB and MDR-TB was found highly significant with family history of tuberculosis, previous antituberculosis therapy and concomitant diabetes mellitus (p=0.01) (Table-1).

Discussion

Globally MDR-TB has increased all over the world over the past few decades. World Health Organization (WHO) estimates show that over 50 million people in the world are presently infected with multi-drug resistant tuberculosis.[11] Pakistan presently ranks eighth among the list of high tuberculosis burden countries, with an annual TB related death rate of 43/100,000.[1] Development of multi-drug resistance is attributed to inappropriate treatment regimen, sub-standard antimicrobials and/or poor patient compliance.[17] The drug resistance (DR-TB) rates for individual drugs; INH, RIF, EMB, PZA & SM were 51,22%, 15.4%, 13.33%, 9%12, and 3.85% respectively (p=0.03). The highest drug resistance rate was observed for INH. These results are comparable with previous studies.[18-21] The MDR-TB isolates were detected in 120 (42.10%) cases, including 5 (5.88%) in category I and 115 (57.50%) in category II patients (p=0.0001). Our results of MDR-TB are comparable to the study of Irfan S et al[18] who reported primary resistance, secondary resistance and MDR-TB of 39%, 79% and 79% respectively, while Ikram A et al[11] had reported 47.6%. One study from Bangladesh has reported similar results of MDR-TB.[21] Another local study of Khoharo HK et al[22] had reported MDR-TB rate of 51.66%, matching closely to our results. Our high levels of drug resistance are mainly because most of our subjects belonged to chronic cases/drug defaulter, which are harbouring more resistant isolates (secondary drug resistance). This poses a growing threat to the public health further burdening the resources especially when the MDR-TB burden is already high in the country. Recent hospital-based studies from Mumbai (India) and Aga Khan University Karachi (Pakistan) reported 51% and 47% MDR-TB rates respectively[17] comparable to our study. The optimal approach would be primary prevention through vaccination, continued surveillance and appropriate therapeutic decisions with monitoring of the patients.[11] Akhtar S et al had reported DR-TB to at least one antituberculous drug of 65.5%. In this study primary, initial and acquired drug resistance to one or the other agent was reported as 7%, 21.1% and 32.3% respectively comparable to our results.[23] Primary drug resistance for INH, RIF, EMB, PZA and SM was reported as 16%, 8%, 16,% 2%, and 26% respectively in a study from Karachi.[24] These results of primary drug resistance are higher when compared with most of local studies. In Pakistan tuberculosis is mostly diagnosed on clinical suspicion and on therapeutic response to anti-tuberculosis drugs, rather than on the basis of culture isolation. This results in inappropriate use of anti-tuberculosis drugs. Furthermore, compliance with treatment remains poor. Despite TB having been declared a national emergency in 2001, implementation of the national TB control programme has been hampered by under-developed health facilities, lack of resources and poor management. The DOTS strategy adopted in 1995 has shown some progress with DOTS coverage at 24% in 2001 but with high drop-out rates of 17%.[11] All these factors appear to be responsible for the high levels of MDR-TB in our population.

Conclusions

Drug resistant and multidrug resistant tuberculosis was observed mainly in category II patients. However, primary MDR was also observed in category I patients and reflects dissemination of MDR cases within the community. In the absence of an action plan, an extensively drug resistant TB (XDR-TB) is likely to emerge and treatment options may then be non-available

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