SUSCEPTIBILITY PATTERN OF 2ND LINE ANTI TUBERCULOSIS DRUGS AT RAHIM YAR KHAN

Hafiz Muhammad Rizwan,[1] Masood Ul Haq,[1] Muhammad Ahmad,[1] Imran Bashir,[1] Abdul Salam,[1] Rana Nasir Ali,[1] Arsalan Ahmad Khan Durrani[1]

ABSTRACT

Background: Pakistan has a high burden of drug resistant TB. Effective management of these cases needs the inclusion of 2nd line anti-tuberculosis drugs. A comprehensive knowledge of susceptibility pattern to these drugs is mandatory to formulate the best possible regimen. **Objective:** To determine the susceptibility pattern of 2nd line anti tuberculosis drugs. **Methodology:** This cross sectional study was carried out at Department of Pulmonology, Sheikh Zayed Medical College / Hospital, Rahim Yar Khan on smear positive cases of PTB. This study was conducted from 1st March 2010 to 30th April 2011. A total of 64 smear positive tuberculosis patients of any age and sex regardless of previous treatment with 1st line ATT & no history of prior exposure to 2nd line ATT were included. Sociodemographic data like age, sex, marital status and income were recorded. Early morning sputum samples were cultured on LJ medium at a reference lab. Drug susceptibility testing (DST) was done for ethionamide, amikacin, kanamycin, capreomycin and ofloxacin to determine the presence of resistance. The data was analyzed on SPSS version 15. **Results:** In this study, out of total 64 cases, 36 (56.25%) were males and 28 (43.75%) females with age range of 9 to 76 years. Thirteen cases (20.31%) had previous exposure to 1st line ATT. Twelve (18.8%) were resistant to one or more drugs. Resistance was highest for ofloxacin (14.1%) followed by ethionamide (6.3%), capreomycin (3.1%), amikacin (1.6%) and kanamycin (1.6%). Sociodemographic characteristics also did not show any statistically significant association with drug resistance. **Conclusion:** There is high frequency of resistance to ofloxacin and ethionamide. To avoid addition of further resistance, DST should be available as early as possible by conventional methods or by rapid genotypic methods at the start of treatment.

Keywords: 2nd line ATT, Drug resistance, DST.

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INTRODUCTION

Tuberculosis is 2nd leading cause of death due to infections worldwide. Global Tuberculosis report 2015 by World Health Organization reveals that 9.6 million people got Tuberculosis and 1.5 million people died of the disease during 2014.[1] According to World Health Organization estimates for burden of tuberculosis, Pakistan has a prevalence rate of 341 per 100,000 population, an incidence rate of 270 per 100,000 population and mortality rate of 28 per 100,000 population.[1]

Effective treatment with combination of anti-tuberculosis drugs remained the mainstay for control of disease. Recommended treatment for newly diagnosed TB cases for 6 months can cure the disease.[2] In 2013, treatment success rate was reported to be 86% globally. However, emergence of drug resistance has major detrimental effect on TB control situation. Treatment success rates have gone down in parallel with extent of resistance to anti-tuberculosis drugs. Resistance to one drug may not pose a big problem but poly resistance especially Multi Drug Resistance (MDR) is a major threat. Treatment has to be continued for upto two years with less effective & poorly tolerated drugs at a cost which is 100 times higher than the drug susceptible TB. Therefore, treatment success rate goes down to 50%. Extensively drug resistant TB (XDR-TB) has even the worst prognosis. XDR-TB has been reported from 115 countries by 2015 while MDR – TB has been reported from all over the world. According to World Health Organization estimates, there were 480,000 new MDR-TB cases and 190,000 deaths during 2014.[1]

In presence of drug resistance, one have to choose combination which contain 2nd line drugs like, ethionamide, cycloseine, PAS, amikacin, kanamycin, capeomycin & quinolones.[3] Many of

1.Department of Pulomonology, Sheikh Zayed Medical College/Hospital, Rahim Yar Khan, University of Health Sciences Lahore, Pakistan.

Correspondence:
Dr. Masood ul Haq, Associate Professor, Department of Pulmonology, Sheikh Zayed Medical College/Hospital, Rahim Yar Khan, Pakistan.

E-mail: masoodulhaq_pk@yahoo.com **Mobile:**+92-3347324576 **Received:** 01-07-2016 **Accepted:** 08-07-2016

these drugs have been used previously for treatment of tuberculosis but later on, their routine use was abandoned as these were less effective, more toxic and have to be taken for longer period. However, in cases of drug resistant TB, depending upon the number of 1st line TB drugs which are not available due to presence of resistance, one or more of these 2nd line drugs may be needed to complete the regimen. But resistance to 2nd line ATT is also fast emerging due to multiple reasons. These include poor prescribing practices of treating doctors, noncompliance of patients, poor quality, non-availability and adverse effects of drugs etc.[4-7] Various previous studies on resistance of 2nd line anti-Tuberculosis drugs were conducted either on proven MDR-TB cases or those with strong suspicion of drug resistance; hence incidence of resistance was high.[8-11] There was no study conducted in general population to detect resistance to these drugs. Knowledge of sensitivity pattern of these drugs in a community may help to decide which drug / drugs can be included in a regimen to treat drug resistant TB cases. There are only few laboratories capable of doing Drug Susceptibility Testing (DST) for 2nd line anti-tuberculosis drugs. According to World Health Organization, there are only seven such laboratories in Pakistan.[12] Therefore, it is impossible to get DST for all suspected drug resistant cases and treatment has to be started empirically. In this situation, periodic surveillance with regional and national surveys on representative samples become important to guide the treatment in drug resistant TB cases. Therefore, we decided to conduct this study to know the sensitivity pattern of 2nd line antituberculosis drugs in our patients.

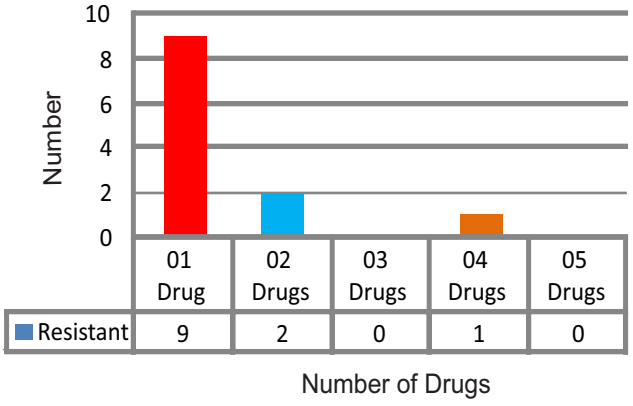
METHODOLOGY

This cross sectional study was carried out at Department of Pulmonology, Sheikh Zayed Medical College / Hospital, Rahim Yar Khan, Pakistan after approval of Institutional Review Broad. This study was conducted from 1st March 2010 to 30th April 2011. A total of 64 smear positive tuberculosis patients of any age and either sex regardless of previous treatment with 1st line ATT & no history of exposure to 2nd line ATT, who presented at our outpatient department from catchment area of DOTS diagnostic and treatment centre of Sheikh Zayed Hospital, Rahim Yar Khan were included in this study. An informed consent was taken from the patients for inclusion into this study. Sociodemographic data like age, sex, marital status and income were recorded on a performa. Patients were instructed to bring a morning sample of sputum in a sterile container. Samples were sent for a culture on LJ medium at a reference laboratory. DST was done for ethionamide, amikacin, kanamycin, capreomycin and ofloxacin. The concentrations of drugs used per ml of medium were Ethionamide 25 mcg/ml, Amikacin 06 mcg/ml, Kanamycin 06 mcg/ml, Capreomycin 10 mcg/ml and Ofloxacin 01 mcg/ml. Data was analyzed using SPSS version 15 and chi square test was applied to see any significant association among certain risk factors and resistance. P value of 0.05 was taken as significant.

RESULTS

In this study, a total of 64 isolates were tested for five 2nd line anti-tuberculosis drugs, ofloxacin, ethionamide, capreomycin, amikacin and kanamycin. Out of 64 there were 36 (56.25%) males and 28 (43.75%) females with age range of 09 to 76 years. Twelve out of 64 isolates (18.8%) were found resistant to one or more drugs while 52 (81.2%) were sensitive to all drugs tested. (Figure I).

Figure I: Overall Sensitivity (Susceptibility) To One or More 2nd Line Anti Tuberculosis Drugs



Mono resistance was present in 09 cases, in which 07 were resistant to ofloxacin alone, and one each to capreomycin and ethionamide. Poly resistance was found in three cases, two had resistance to two drugs, ofloxacin and ethionamide and one had resistance to 04 drugs i.e. capreomycin, kanamycin, amikacin and ethionamide. No isolate in this study had simultaneous resistance to ofloxacin (quinolone) and any of the injectable (amikacin, capreomycin and

kanamycin), so no possibility of X-DR-TB in this study. However, 11 (17.2%) cases had resistance either to one of injectables or a quinolone, hence may be considered pre XDR-TB. Thirteen out of 64 (20.3%) have previous history of treatment 1st line anti-tuberculosis drugs.

Table I: Presence of drug resistance to one or more drugs with history of anti-tuberculosis treatment

Resistance	H/O ATT n=13	NO H/O ATT n=51	Total n=64
Resistant to One or More Drugs	**04(30.8%)**	**08 (15.7%)**	**12(18.8%)**
Resistant to one drug 09	02	07	
Resistant to 02 drugs 02	01	01	
Resistant to 03 drugs 00	00	00	
Resistant to 04 drugs 01	01	00	
Resistant to 04 drugs 00	00	00	
Fully sensitive to all 05 2nd Line Drugs	**09 (69.2%)**	**43 (84.3%)**	**52(81.2%)**

Table I shows the presence of drug resistance to one or more drugs with history of anti-tuberculosis treatment.

Table II: Resistance to individual drugs versus history of anti-tuberculosis treatment

Drugs	Secondary Resistance	Primary Resistance	Total n=64	P Value
	H/O ATT n=13	No H/O ATT n=51		
Ofloxacin	03 (23.1%)	06 (11.8%)	09 (14.1%)	0.26
Ethionamide	02 (15.4%)	04 (3.4%)	04 (6.3%)	0.18
Capreomycin	01 (7.7%)	01 (2.0%)	02 (3.1%)	0.36
Amikacin	01 (7.7%)	00 (00.0%)	01 (1.6%)	0.20
Kanamycin	01 (7.7%)	00 (00.0%)	01 (1.6%)	0.20

Table II shows resistance to individual drugs both overall and in relation with history of anti-tuberculosis treatment. There was no statistically significant difference of resistance between two groups with or without treatment. Table III shows sociodemographic characteristics of participants of this study as well as comparison of the data between groups having resistant and sensitive drugs.

Table III: Sociodemographic characteristics of participants of the study

Variables	Total Patients	Resistant	Sensitive	P Value
	64	12	52	
Age (Years)				
Range	9-76	15-60	9-76	---
Median	30.00	24.00	30.00	---
Mean	33.70	28.75	34.85	--
Sex				
Male	36	06	30	**0.43**
Female	28	06	22	
Residence				
Urban	49	09	40	**0.57**
Rural	15	03	12	
Education				
Uneducated	33	08	25	**0.20**
Educated	31	04	27	
Marital Status				
Never Married	23	04	19	**0.55**
Married	41	08	33	
Monthly Family Income (Rs)				
Less Than 15000	55	11	44	**0.46**
More Than 15000	09	01	08	

DISCUSSION

According to WHO guidelines for Programmatic Management of Drug Resistant TB (PMDT), an effective combination should include one of the quinolones (levofloxacin, moxifloxacin, gatifloxacin and ofloxacin), one from the injectable group (kanamycin, amikacin and capreomycin) and two or more of the drugs from the group 04 (ethionamide, prothionamide, cycloserine and PAS) of WHO ATT classification of anti-tuberculosis drugs.[3]

There is significant cross resistance between various quinolones as well as various members of injectable especially kanamycin and amikacin. Among the group 04 of anti-tuberculosis drugs, ethionamide is considered most important. In this study, DST was done for ofloxacin (quinolones) and all the injectable agents and ethionamide. All of these drugs are very important component of drug resistant treatment regimen.

If resistance to any one of the quinolones and any of these injectable is present in an MDR-TB patient (who had resistance to Rifampicin & Isoniazid) it is labeled as extensively drug resistant TB (XDR-TB), which is the most difficult form to treat.[13]

On the other hand if resistance to either quinolone or an injectable is detected in a MDR-TB case, it is considered as Pre XDR-TB. Fortunately, none of isolate in this study was simultaneously resistant to ofloxacin and injectables but 11 out of 64 (17.2%) cases have resistance either to one of injectables or a quinolone.

So, there is no possibility of XDR-TB in this study even if resistance to rifampicin and isoniazid is detected in these patients but 17.2% (i.e. one in every six patients) will be Pre XDR-TB. This very significant pool of quinolone resistance among the TB patients is a bombshell. If these patients are not correctly managed and acquire additional resistance to other potent drugs, they may end up in either MDR or XDR TB.

In this study, resistance to ofloxacin (14.1%) is most common. It means that every one in seven patients is resistant to ofloxacin which is quite alarming. Similar results have been shown in other local studies. One study from Faisalabad, Pakistan revealed resistance to ofloxacin in 5.08% of cases,[14] while in another study from Rawalpindi Pakistan, resistance to levofloxacin was observed in 24%.[15] Results of various international studies reported in literature about the presence of resistance to quinolones differ considerably due to variable inclusion criteria. Many of these studies were done on patients having proven MDR or suspected XDR-TB cases.[16-19]

The quinolones are one of the most widely prescribed drugs for infections other than tuberculosis. In countries where TB is common, other infections are also common and antibiotics are freely available without prescription from pharmacies.[20,21] Higher resistance to quinolones in such communities can be explained on this basis. Injudicious use of quinolones adversely affects the communities in two ways, one by masking the symptoms of TB and delaying the diagnosis and secondly by producing drug resistance.[22]

There were only two cases that had resistance to injectable therapy; one having resistance of capreomycin alone and the other to all the three injectables. This strengthens the previous recommendations of considering amikacin and kanamycin as a single drugs. This was relatively low as compared to quinolones.

Amikacin and kanamycin are being prescribed for infections other than TB. Because of their availability as injectable preparation only, they are used less frequently and for shorter period of time. This lesser use might explain comparatively low resistance to these injectable drugs.

Capreomycin resistance was higher than that of amikacin and kanamycin, it is rather surprising as it is very costly and has only become recently available in Pakistan in the programmatic management of drug resistant TB (PMDT); poor TB patients are unlikely to have this previously and cross resistance with other injectable is also less likely. This 3.1% resistance to capreomycin is hard to explain.

Ethionamide is another bactericidal drug in group four. It is better than other drugs in this group like PAS and cycloserine which are bacterostatic, less effective and more toxic. In this study, resistance to ethionamide is 6.3% which is second highest after ofloxacin. This might again be due to the fact that ethionamide along with cycloserine are the two drugs which were available in Pakistan market for some times and used by many physician for drug resistant TB before the start of PMDT in Pakistan. Ethionamide along with cycloserine were very costly, their availability and affordability remained a problem, which might be a cause of high resistance. Secondly, cross resistance due to structural similarity of ethionamide with isoniazid in 15 – 20% cases might be another cause.[23]

Overall resistance in this study is quite high. Twelve out of 64 isolates (18.8%) were resistant to one or more of the five 2nd line drugs tested, which means resistance in one out of every five patients. In this study, thirteen out of 64 (20.3%) have previous history of treatment. But as all of them were treated with 1st line ATT and were not exposed to any of the drugs tested, the resistance cannot be classified as primary or secondary.

Possibly for the same reason there is no statistically significant difference for resistance to any drug in relation with history of ATT. As these drugs are also frequently used in indications other than tuberculosis, which can explain higher resistance in these patients. Failure to accumulate data about the previous use of all these drugs as antibiotics in period preceding the diagnosis of tuberculosis may be considered one limitation of this study. There are few other limitations as well like small number of participants and failure to perform DST for other group 4 drugs like cycloserine and PAS.

Therefore, it may be recommended to conduct larger studies with DST available for other 2nd line drugs and a detailed history of all antibiotics used prior to the diagnosis of TB.

CONCLUSION

In conclusion, there is a high frequency of resistance to ofloxacin and ethionamide in our community. To avoid addition of further resistance, drug susceptibility testing should be available as early as possible by conventional method or by rapid genotypic methods at the start of treatment.

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