PREVALENCE OF DRUG RESISTANT TUBERCULOSIS IN ARMED FORCES-STUDY FROM A TERTIARY REFERRAL CHEST DISEASES HOSPITAL AT PUNE

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ABSTRACT

This study was conducted to find out the prevalence and pattern of primary and acquired resistance to antimycobacterial drugs among patients of pulmonary tuberculosis, in Armed Forces. Out of 2562 clinically diagnosed patients of tuberculosis, in a span of three years, 1146 were bacteriologically positive. The study included only 1120 smear and culture positive cases, and excluded 26 cases in which no growth was obtained on culture. 192 out of 1120 cases (17.14%), showed overall resistance to one or more antituberculous drugs (ATD). Primary drug resistance (PDR) was observed in 161 (14.37%) and acquired drug resistance (ADR) in 31 isolates (2.77%). Of the resistant cases on short course chemotherapy (SCC), single drug resistance was observed in 99 (51.56%), resistance to any two drugs in 63 (32.81%), and three or more drugs in 30 (15.62%) cases. Analysis of resistance to specific drug revealed 26.56% for streptomycin (S), 15.10% for rifampicin (R), 7.29% for isoniazid (H), 2.08% for pyrazinamide (P) and 0.52% for ethambutol (E). Resistance to H and R was present in 4.16% strain and their combination with other drugs resistance was in 16.14% of the drug resistant strains, thus constituting 2.76% of the total sputum positive cases. A group of 26 cases is also discussed, where there was discrepancy in clinical status and bacteriological parameters and treatment for multi-drug resistant tuberculosis (MDR-TB) was instituted.

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KEY WORDS : Pulmonary tuberculosis, Drug resistance

Introduction

The emergence of resistance to antitubercular drugs (ATD) has been known since ancient times. Researchers had found out the existence of resistance and selection of mutants in the wild strains of *Mycobacterium tuberculosis* (MTB) under certain treatment conditions. However, the first clinically drug resistant tuberculosis case was not described until 1970[1]. There have been reports from various parts of India [1-11] and some parts of the world [12,13] regarding recent outbreaks of MDR-TB. The rapidity with which it is spreading and high case fatality are cause of concern. The initially reported outbreaks involved small number of cases among close contacts, who had prolonged exposure to the source of infection. However, later studies showed large number of patients in the outbreaks [14,15]. Initially, well-documented reports of MDR-TB were from USA but soon after reports from other countries flooded the literature and reported developing countries-the worst sufferer [16].

In global surveillance project, the surveys conducted from 1985-94 under WHO revealed, the highest rate of incidence of MDR-TB in Nepal (48%), Gujarat (33.8%) and in some segments of population of New York (30.1%) followed by relatively lower incidence from other parts of the world [12,17]. The ADR usually results from erratic or inadequate therapy in patients of drug susceptible tuberculosis but re-infection with drug resistant TB is possible [13,18]. Poor compliance to drug therapy is prevalent widespread in our country [3]. Nosocomial transmission amongst HIV infected patients [19] and health workers have further flared up the issue [15].

We report here the incidence of primary and acquired drug resistance and the pattern of resistance to antimycobacterial drugs in patients comprising soldiers and their families, representing various geographical areas of our country.

Material and Methods

The study was conducted in a tertiary care chest hospital at Pune for a period of 3 years form July 95 to July 98. Early morning on spot sputum specimen from 2562 clinically suspected tuberculosis patients were collected on three consecutive days and processed by modified Petroff's method using 4% NaOH. The smears were subjected to acid fast stain by Ziehl Neelsen method and examined by light microscopy. The smears were graded 1+,2+ and 3+ as per CDC criteria, if 1-9 organisms were seen in 100, 10,1 high power field examination respectively and 4+, if more than 9 organisms were seen in all fields. Out of 1146, only 1120 smear positive patients in which *Mycobacterium tuberculosis* was isolated on culture were included in the study. The clinician at the centre thoroughly interrogated all such patients, regarding history

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of contact with patients of pulmonary TB and previous history of chemotherapy with ATD.

The sputum specimens were cultured on Lowenstein Jensen (LJ) slants in duplicate and incubated for isolation of mycobacterial strains. Identification and subsequent biochemical tests were performed in accordance with the manual of laboratory methods (1987), Tuberculosis Research Centre (ICMR), Chennai [20]. Indirect susceptibility tests were carried out by incorporating required concentration of the drugs in the media which was inoculated by taking standardized inoculum [20]. Control strain H37 RV was set up with each batch of drug sensitivity testing. The strains were defined resistant if a growth of >20 colonies was observed at the following concentration. Isoniazid: MIC>1 μg/ml, Streptomycin: Resistance ratio ≥ 8, Rifampicin: MIC >64 μg/ml, Ethambutol: MIC >8 μg/ml, Pyrazinamide: MIC >100 μg/ml and Ethionamide: MIC> 114 μg/ml.

Failure during the chemotherapy was defined as a smear positive case, who remained or became smear positive again at 16-18 weeks after commencing the treatment. Relapse was said to have occurred if a person declared cured/quiescent at the end of SCC became bacteriologically positive after at least 3 months of bacteriological quiescence [21]. Drug resistance was classified as single, double or multiple drug resistance and MDR was defined as resistance to H and R with or without resistance to other drugs.

Results

Of a total of 1120 strains of MTB tested, 192 (17.14%) were resistant to one or more ATD. 143 strains of these 192 resistant strain, were from the indoor cases and rest 49 were from the outdoor cases. The indoor patients included 130 serving soldiers and 13 were wives of serving soldiers. 49 cases who received treatment as outdoor patients were females, compliance of their treatment and subsequent follow-up could not be controlled adequately. It is interesting to note that drug resistance was observed in 18 (37%) of these 49 patients.

The study included 130 males and 62 female patients. Majority of the patients were in the age group between 20 - 53 years. The youngest patient was 18 and oldest 55 years old (mean age 36). No association could be established between the age or sex of the patients and the pattern of drug resistance.

Out of the 192 resistant strains, ADR was observed in 31 (16.15%) and PDR in 161 (83.85%) cases constituting 2.77% and 14.37% of the total MTB isolates respectively (Table-1). Past history of pulmonary TB was available in only 29 (2.58%) cases that were treated in-door with SCC for a period of 6 to 9 months at our center. They were labelled as relapse cases. In 31 (2.76%) patients, history of irregular treatment with ATD from general practitioners was present, for a period of 1-2 years before reporting to this hospital. 10(0.9%) patients gave history of having been in close contact with their relatives who were suffering from pulmonary TB. The remaining 122 drug resistant cases denied any such history in the past (Table-2).

In addition to pulmonary TB, 7 patients had diabetes mellitus, as associated illness. Two patients had HbsAg positive viral hepatitis during their period of hospitalization and blood for HIV was positive in 5 patients.

In 169 out of 192 (88%) patients, sputum was positive on smear and culture examination, whereas in 23 (12%) though the smear was negative for acid fast bacilli but growth of MTB was obtained on culture (Table-3). In addition to 192 drug resistance cases, there were 26 patients who could not be studied further in the laboratory due to negative culture report. Majority of the sputum smears were graded 3+, as per CDC criteria for bacillary count, accounting for 31.25% (Table-4)

Antibiotic susceptibility in 192 cases of drug resistance revealed single drug resistance in 99 patients (8.84%), two drugs in 63 (5.62%) and 30 patients (2.68%) showed resistance to three or more drugs (Table-5). The commonest single drug resistance was to S in 4.55% followed by R in 2.59% and H in 1.25% cases. In 28 patients (2.5%) S and R were the commonest double drug resistance. Next common drug resistance was to S and H in 1.43% of the patients. MDR (H and R) was present in 8 patients (0.71%) and in 30 patients (2.68%), the resistance to H and R was associated with resistance to other drugs (Table-6).

TABLE 1
Incidence of primary and acquired drug resistance TB

Category of drug resistance	Total no. of MTB strains	No. of drug resistant strains	% to 1120 MTB strains
Primary	1120	161 (83.85%)*	14.37
Aquired		31 (16.15%)*	2.77
Total		192	17.14

*Figures in brackets indicates % of strains to the number of drug resistant strains.

TABLE 2
Past history of drug resistant cases (n=192)

Past h/o the patients		No. of drug resistant strains	% to total MTB strains
(a)	Relapse (after complete treatment in the past)	29	2.58
(b)	History of irregular treatment with ATD	31	2.76
(c)	History of close contract with a known case of TB	10	0.9
(d)	No h/o exposure to ATD/ pulmonary TB patients	122	10.9

TABLE 3
Sputum status of 192 drug resistant patients

Sputum / culture Status of patients	No. of cases	% to drug resistant strains
Sputum and culture +ve	169	88
Sputum -ve and culture +ve	23	12

TABLE 4
Bacillary count of drug resistant strains in sputum (n=192)

Bacillary count	No of patients	% to drug resistant strains
4+	21	10.94
3+	60	31.25
2+	34	17.71
1+	54	28.12
—	23	11.98

TABLE 5
Antibiotics sensitivity pattern of drug resistant strains

Total isolates of MTB	Strains resistant to 1 or more drugs				Total resistant strains
	1 Drug	2 Drugs	3 Drugs	>3 Drugs	
	99	63	17	13	192
1120	(51.56%)	(32.81%)	(8.86%)	(6.77%)	
	(8.84%)*	(5.62%)*	(1.62%)*	(1.62%)*	(17.14%)*

(*Percentage of drug resistance to the total number of MTB isolates)

TABLE 6
Pattern of drug resistance in cases (n=192)

Strains resistant to 1 Drug			Strains resistant to 2 Drugs	Strains resistant to 3 and more drugs	
S-	51	(4.55%)	SH - 16 (1.43%)	HRE - 02 (1.18%)	
H -	14	(1.25%)	SR - 28 (2.50%)	SHR - 11 (0.98%)	
R -	29	(2.59%)	RH - 08 (0.71%)	SRE - 04 (0.35%)	
Z -	04	(0.36%)	SE - 04 (0.36%)	SHRE - 05 (0.45%)	
E -	01	(0.09%)	SZ - 01 (0.09%)	SHRZ - 03 (0.27%)	
Eth-	0		RZ - 02 (0.18%)	5 drugs - 05 (0.45%)	
			HE - 03 (0.27%)	(all including R)	
			RE - 01 (0.09%)		
Total - 99		(8.84%)	63 (5.62%)	30 (2.68%)	=17.14%

(Figures in brackets indicate % of drug resistant strains to total MTB isolates)

157 of these cases required treatment with second line drugs while rest 35 patient responded well to first line drugs. Resistance to only H was obtained in 2 out of the 5 HIV positive drug resistant cases, 2 strains showed resistance to H and R and one to HRE. One of the MDR-TB patients had loculated pyopneumothorax and cavitary lesion in left upper lobe, required surgery. Out of 157 patients 133 were cured and rest 24 patients subsequently developed sequel of advanced disease like bronchiectasis and diffused fibrosis. 21 patients developed chronic TB, 7 lost to follow-up and 8 had fatal out-come.

Discussion

Tuberculosis continues to plague the world and remains the major global health problem. It is the single largest killer amongst infectious diseases and accounts for 20% of avoidable adult deaths [16]. The present study assessed the prevalence and the pattern of drug resistance amongst soldiers and families in Armed Forces. These cases were referred from various hospitals located in different geographical regions, comprising patients hailing from different states of the country thus making this study unique from surveys carried out by other workers mostly covering a particular geographical region.

The study revealed the overall drug resistance to ATD in 17.14% cases. It was interesting to note that out of 49 drug resistant females who received outdoor treatment, 18 (37%) had acquired drug resistance. The actual drug resistance among patients treated indoors with directly observed treatment (DOT) (n=143) was therefore 12.77%. The overall rate of initial drug resistance among patients with or without history of previous chemotherapy in a nationwide study conducted by ICMR [22] revealed to be as high as 32% in nine urban centers in India. In a survey conducted during 1980 at Raichur and North Arcot district, Karnataka [11] showed similar high level of drug resistance (34%). However Jena et al [10] in their study conducted in 1992-93 reported lower incidence of drug resistance (12.7%) in their patients, explaining that chances of indiscriminate exposure to ATD, prior to reporting to the hospital were very low. The higher drug resistance, in surveys conducted by other workers has been mostly attributed to factors such as deterioration in public health care, erratic and inadequate treatment due to non-availability of drugs and lack of consistency in follow up [4,6,11]. Such lapses promoted and heightened the potential for transmission of infection in the community.

In Armed Forces the patients are invariably healthier than the civil counterpart and they have to undergo a thorough periodic medical checkup in a well-established medical setup. A patient once suspected after the preliminary investigations is referred to a chest centre where complete work up is done and anti-tuberculous therapy (ATT) is started only on firmly establishing the diagnosis. Hospitalization for serving soldiers, in our setup, for the treatment of TB is mandatory and DOT is the rule. There is no lapse in the follow-up of the cases and no problem is encountered in the availability of ATD. Hospitalized patients are given highly balanced and nutritious dietary regimen. The maintenance of proper medical record and regular periodic follow-up are another likely causes of lower incidence of ADR in Armed Forces. Unfortunately it is gradually and steadily showing the rising trend reflecting it as primary infection for our soldier too. In an unpublished data of our centre in 1987, the incidence of drug resistant TB was 0.2%. It increased to 1.4% in 1989, and in the present study it has risen to 17.14% among indoor patients.

The present study revealed high incidence of PDR in 14.37% of cases. Most of the patients though denied previous history of ATT but there is remote possibility of consuming these drugs without their knowledge. The incidence of PDR in various studies has been reported around 20% [3]. Most of the patient possibly acquired infections from patients excreting drug resistant organisms. This is in contrast to other studies conducted by other workers [4-11,22].

A higher figure of 37% drug resistance among our

female patients treated in out-door without direct supervision was due to inadequate drug regimen received from private practitioners due to difficulty in coming from remote areas. In most of these cases the husband was posted in far-off stations, thus making their treatment more difficult.

We observed the highest percentage of drug resistance to S in 128 (11.43%) out of 1120 isolates. Out of these, 77 (40.10%) isolates showed resistance to S in combination with other drugs. In various studies conducted in other parts of the country, resistance to H was the commonest [5,6,11]. In an earlier study conducted at this center, Jena et al [10] reported similar higher resistance to S and attributed it to the free availability of S in Armed Forces for use in tubercular and non-tubercular disease.

The resistance to the H was observed in 67 isoloates (5.98%). In 53 isolates (4.73%), it was in combination with other drugs. The dangerous resistance pattern of H and R was found in 8 (0.71%) and in combination with other drugs in 34 patients (3.03%). Some workers [4-6,11,22] have reported higher level of drug resistance to H. The prevalence of MDR in India is lower as compared to other parts of the world. It is estimated to be about 3% in primary and 6% in acquired drug resistance except in a study conducted from Gujarat where a higher level of drug resistance was observed in 11.4% to 18.5% of the cases [4].

Resistance to R was observed in 98 strains (8.75%), out of which 69 (35.93%) showed additional resistance to various drugs. Resistance to R was observed only in 29 cases (2.59%). The increasing pattern of drug resistance to R in post 1990 era is attributed to unsupervised SCC.

The probable cause of 26 sputum positive and culture negative report in a subset of patients was due the intake of ATD, knowing or unknowingly in the past, that made them culture negative but dead bacilli continued to be expectorated resulting in sputum positivity. Smear negativity in a paucibacillary sputum sample is a well-known phenomenon as 10^5 bacilli per ml of sputum are required for organisms to be seen on light microscopy but culture shows the growth. In such situation, the drug resistance was also suspected on clinical, bacteriological (fall and rise phenomenon), and radiological grounds and the patients were treated as drug resistant cases.

Though the nationwide surveys of drug resistance reveal a rising trend of MDR cases but in India the correct evaluation and the reliable data is not available due to lack of culture facilities and proper reporting system [3,23,25,26]. Uniformity in the laboratory methodology is required in identification of MTB, preparation of culture media, concentration of drugs used in the susceptibility test and standardization of bacterial inoculum etc. Variations of results within the country by various workers may be also due to the difference in the selection of patient groups studied and quality of inquiry made regarding previous history of drug intake [27,28]. The misdiagnosis of drug resistant TB in susceptibility testing, possibly due to laboratory diagnostic errors, have been stressed to the extent that some workers feel that susceptibility tests alone are not enough to dictate the treatment. Clinical correlation is also necessary in establishing the diagnosis of drug resistance [27-29]. Another factor which contributes to the delay in the diagnosis of drugs resitant TB is the primitive methods of isolation of MTB in vogue [23]. Conventionally, a laboratory takes almost 10 weeks from the day of submitting the first sample for the isolation and culture of acid fast bacilli, to its species identification and time taken for performing antibiotic susceptibility tests.

The present study shows that the incidence of ADR in Armed Forces is still lower as compared to the studies from various civil set-ups. However, it is rising very significantly when we compare this data with our own previous data.

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