Original Article

Resistance to Antituberculosis Drugs in Pulmonary Tuberculosis

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Abstract

Background : Incidence of drug resistance and pattern of susceptibility to antitubercular drugs in pulmonary tuberculosis amongst soldiers and their families was studied for four years at a military hospital in northwest India.
Methods : Identification and susceptibility tests were carried out as per procedures laid out in laboratory manual of Tuberculosis Research Centre (TRC), Chennai.
Results : Of the 172 strains of Mycobacterium tuberculosis (MTB) isolated from sputum samples, 150 (87.21%) were sensitive and 22 (12.79%) showed resistance to one or more antitubercular drugs. Acquired drug resistance was observed in 7 (31.82%) and primary drug resistance in 15 (68.18%) cases. Among 22 drug resistant cases, who were on short course chemotherapy (SCC), resistance to single drug was observed in 12 (54.54%), two drugs in 7 (31.82%) and to three or more drugs in 3 (13.64%) isolates. Fourteen (18.14%) strains were resistant to Streptomycin, 8 (4.65%) to Rifampicin, 11 (6.40%) to Isoniazid, 1 (0.58%) to Pyrazinamide and 2 (1.16%) to Ethambutol. Multidrug resistance was observed in 5 (2.91%) cases, of which resistance to Isoniazid and Rifampicin was present in 2 (1.16%) and their combination with other drugs in other 3 (1.74%) isolates.
Conclusions : Drug susceptibility pattern to antitubercular drugs is discussed and compared with studies from other centres.

MJAFI 2006; 62 : 325-327

Key Words : Drug resistance, Antitubercular drugs, Mycobacterium tuberculosis.

Introduction

Tuberculosis affects one third population of the world, of which 95% live in developing countries [1]. It has been estimated that 30% of the tuberculosis patients reside in India [2]. There are 13 million infected and diseased, 3.5 million are sputum positive and 2.2 million new cases added every year [3]. The situation has become alarming due to rising drug resistance in immuno-compromised including HIV patients [4-6]. It is worrying whether it is the causative organism which is acquiring resistance to antitubercular drugs (ATD) or failure to institute the direct observed therapy (DOT) in patients [7].

Materials and Method

The study included 1501 clinically suspected pulmonary tuberculosis (TB) cases, who reported to a Military Hospital from Jan 2000 to Dec 2003 . Early morning samples of sputum were collected on the spot, on three consecutive days and processed by modified Petroff's method using 4% sodium hydroxide (NaOH). The smears were subjected to acid fast stain by Ziehl- Neelson's method and examined under light microscope.

Sputum smears from 662 patients were positive for acid fast bacilli (AFB). The sputum specimens of these were cultured on Lowenstein Jensen's (LJ) slant media, in duplicate, for isolation of Mycobacterium tuberculosis (MTB). Identification and subsequent biochemical tests were conducted in accordance with the manual of laboratory methods, TRC (ICMR), Chennai [8]. Drug sensitivity tests were performed by incorporating required drug concentration of antimycobacterial drugs in the medium on which standardised inoculums of processed sputum were inoculated. Control stain, $H_{37}RV$, was set up with each batch for drug sensitivity testing and the strains were defined resistant, if more than 20 colonies were obtained on drug slant media. Resistance ratio (RR) method of drug sensitivity was carried out for Streptomycin (S) and minimum inhibitory concentration (MIC) for Isoniazid (H), Rifampicin (R), Ethambutol (E) and Pyrazinamide (Z).

The cut off value by minimum inhibitory concentration (MIC) method was >1mg/ml for H, >64mg/ml for R, >8mg/ml for E, >100mg/ml for Z and RR of ≥ 8mg/ml for S for defining resistant strains. The clinician interviewed patients regarding previous antitubercular treatment (ATT), self modification of doses or change of prescription, checked prescription for adequacy of doses, duration and appropriate combination of drugs etc. Accordingly, the drug resistance was classified as acquired or primary and the results were specified as resistant to single, double, or multi drug resistant (MDR).

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Received : 24.08.2004; Accepted :19.05.2005

Results

One hundred and seventy five culture positive patients were included in the study, of which 147 (84%) were male and 28 (16%) females, in the age group of 18-78 years. Growth of MTB was obtained in sputum samples of 175 patients, including 10 in whom the sputum was reported as AFB negative on smear examination. These were perhaps the paucibacillary sputum samples. Besides this, three isolates were identified as atypical mycobacteria, including one as rapid grower and therefore excluded from the study. In 487 sputum positive AFB samples, no growth was obtained on culture. Of the total 172 strains of MTB, 150 (88.21%) were sensitive to all drugs tested, whereas 22 (12.79%) strains were resistant to one or more drugs. Of the resistant strains, acquired drug resistance (ADR) was present in 7 (31.82%) and primary drug resistance (PDR) in 15 (68.18%) isolates (Table 1). Among noncompliance group of patients, there was one case of relapse due to mono drug therapy who had developed resistance to H and R. Two patients confessed to have self modified the treatment due to loss of appetite and drug reaction. Four cases defaulted during treatment due to vague reasons.

Antibiotic susceptibility pattern of 22 isolates showed resistance to single, double, and three or more drugs in 12 (6.98%), 7 (4.07%) and 3 (1.74%) patients respectively. The commonest single drug resistance was to S in 6 (3.49%), followed by H in 3 (1.74%) cases (Table 2). The commonest double drug resistance was to S and H in 3 (1.74%) followed by S and R / H in 2 (1.16%) cases each. MDR (H & R) was present in only 2 (1.16%) cases, out of which one patient was HIV positive. S revealed the highest resistance pattern in combination with other drugs in 14 (8.14%), followed by H in 11 (6.40%) and R in 8 (4.65 %) (Table 3).

Table 1

Pattern of drug resistance

Category of drug resistance	Total no of mycobacterial strains	No of drug resistant strains (%)	% of 172 mycobacterial strain
Primary	172	15 (68.18)	8.72
Acquired		7 (31.82)	4.07
Total		22	12.79

Table 2

Pattern of drug resistance

	Strains resistant to		
	1 drug (%)	2 drug (%)	3 or more drugs (%)
S	6 (3.49)	SH 3 (1.74)	SHR 1 (0.58)
H	3 (1.74)	HR 2 (1.16)	SHRE 2 (1.16)
R	1 (0.58)	SR 2 (1.16)	
E	1 (0.58)		
Z	1 (0.58)		
Total	12 (6.98%)	7 (4.07%)	3 (1.74%) = 12.79

Table 3

Drug resistance pattern in combination with other drugs

Drug	No of strains resistant	% of 22 resistant strains	% of 172 total isolates
S	14	63.63	8.14
H	11	50.00	6.40
R	8	36.36	4.65
E	2	9.10	1.16

Discussion

The overall incidence of drug resistance in our study, where the patients were referred from hospitals, from different states was 12.79%. There have been reports of drug resistant tuberculosis from various parts of India [9-15] and the world [16,17] but only a few reports from the armed forces [18-20]. It is interesting that almost similar figure of drug resistance (12.70%) to MTB was reported in a study conducted in 1992-93 among soldiers and their families from a tertiary chest disease centre [18]. From the same centre, higher incidence of drug resistance in 17.14% cases was reported in another study carried in 1995-98 [19]. Over all drug resistance of 13.8% in non HIV patients has recently been reported from another armed forces hospital [20].

A comprehensive review of Indian drug resistant TB was done in 1997 and later in 1999 by TRC (ICMR), Chennai [5,4]. Reinfection with MTB, and its transmission in the hospital environment has further complicated the issue in the HIV patients and even the hospital staff [21]. Medical services in armed forces are relatively well organised and ATT is not advocated until the diagnosis is established [18,19]. There is need to understand the geographical susceptibility of the host in view of the great variation in the drug susceptibility as shown in various surveys [4,5,8].

Growth of MTB obtained on culture in cases of smear negative for AFB is a known phenomenon as 10^5 bacilli per ml of sputum are required for organism to be seen on light microscope but culture may show growth. Fall and rise phenomenon in the sputum is another reason for this disparity.

Our results vary from other studies in the pattern of primary and acquired drug resistance, which is 8.72% and 4.07% respectively. It is not in agreement with many surveys conducted on civil population by other workers, where acquired resistance was invariably reported higher than those of PDR [5,11,13,14].

The resistance to S was highest in our series in 14 (8.14%) isolates, of which 8 (4.65%) showed resistance to combination of drugs. This figure is higher when compared with most studies but is in agreement with studies conducted in armed forces [18,19]. We had 11(6.40%) isolates which were resistant to H. In 8 (4.65%) isolates, there was resistance to other ATD.

The deadly double drug combination of H with R was present in 2 (1.16%) and in combination with other drugs in other 3 (1.74%) cases. Relatively higher level of drug resistance to H has been reported by some workers [12,13]. MDR strains in India are still not so high as compared to other parts of the world [4,5].

Resistance to R was observed in 8 (4.65%) isolates, 7 (4.01%) of which were in combination with other drugs. Resistance to R in combination with H is on the rise in most of the studies perhaps due to unsupervised OPD treatment [10-14]. Our figure of resistance to R is lower as compared to many studies done across the country since hospitalisation is mandatory for all cases of tuberculosis and DOT is followed meticulously. Drug resistance to R was present in combination with other drugs except in one case. Variation of results may be due to difference of selection of patient groups studied, quality of inquiry made, history taking, variations in standardisation of laboratory techniques and reliability of concentration of drugs in the media [9].

Conflicts of Interest

None identified

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