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Emergence of Extensively Drug Resistant (XDR) Strains of *Mycobacterium Tuberculosis* in TB Patients in Kerala, India

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Abstract

We report the detection of extensively drug resistant strains of *Mycobacterium tuberculosis* in Kerala, India. Earlier we had reported that 22 of 206 local isolates were multidrug resistant. Now, we tested the same isolates for their resistance to ofloxacin and amikacin. Six among the MDR isolates showed resistance to both drugs and therefore fall under XDR category.

Keywords: India, Kerala, Tuberculosis, XDR, MDR, *Mycobacterium tuberculosis*

Introduction

India harbored the largest number of tuberculosis (TB) cases in 2008[1]. TB control programs have been badly hit with the emergence of drug resistant strains of *M. tuberculosis*. Multidrug

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resistant (MDR) -TB occurs when the bacteria are resistant to at least isoniazid and rifampicin, the two most potent first-line anti-TB drugs. MDR-TB is treated using second-line drugs which include two major classes, fluoroquinolones and aminoglycosides. TB caused by *M. tuberculosis* strains resistant to any fluoroquinolone, and at least one of the three injectable second-line drugs (capreomycin, kanamycin, and amikacin), in addition to being MDR, is called extensively drug resistant tuberculosis (XDR-TB) [2]. Ten percent of MDR-TB tested worldwide was found to be XDR-TB [3]. The combination of HIV/AIDS and XDR-TB could become a great survival challenge for humanity.

To date there are only few reports on XDR-TB from India. The first report on the prevalence of XDR-TB in India was in 2007 from a retrospective study in which they showed occurrence of 7.4% XDR-TB cases among MDR-TB patients in Lucknow [4], and 8% from a tertiary care center in Mumbai [5]. Recently a low prevalence (2.4%) of XDR cases was reported in another hospital-based study from New Delhi [6]. While Gujarat had 3.2% cases [7], the scenario is alarming in the eastern states of India with 22.2% XDR cases [8].

Materials and methods

In an earlier study, we had analyzed the resistance profile of *M. tuberculosis* isolated from TB patients in Kerala. In this retrospective analysis, we studied 206 isolates of *M. tuberculosis* cultured from the sputum samples of tuberculosis patients attending TB clinics from different parts of Kerala. Of them, 92 were from new cases and 104 cases were from re-treatment cases. Treatment status of 10 patients was unknown. We observed a prevalence of MDR at 10.7% (5.4% in new cases and 16.4% in re-treatment cases) [9]. Subsequently, we analyzed the same isolates for their resistance to second-line drugs also. The susceptibility of these isolates to two second-line drugs, ofloxacin and amikacin was tested at 0.5, 1.0 and 2.0 µg/ml by resazurin microtitre assay (REMA) as described by Martin et al. [10]⁵ This study was approved by the Institutional Biosafety and Ethics Committees. As this was a retrospective study, informed consent from the patients was not required.

Results

Among the 206 isolates analysed for their DST against second line drugs, 19 were resistant to ofloxacin and 13 were resistant to amikacin (Fig 1). All the amikacin resistant ones were resistant to ofloxacin also. Of these 13 amikacin and ofloxacin resistant isolates, six were MDR and hence met the definition of being XDR. Representative strains from these clinical isolates belonging to each resistance category i.e. pansusceptible, MDR and XDR along with *M. tuberculosis* H37Rv were analyzed by automated BACTEC MGIT 960 TB system (Becton Dickinson) for validation of the drug susceptibility testing protocol and we observed 100% correlation between the results of REMA and MGIT.

Discussion

The clinical history of these six XDR patients revealed that all were re-treatment cases. Thus in this study, XDR TB was found only in re-treatment cases (5.76%, 6/104). XDR among MDR cases is 27% (6/22). As per the details provided by the physicians, two of these patients were already under treatment with second-line drugs at the time of sputum collection while the others were not. Age or sex of the patients did not show any obvious propensity to XDR TB. Among the isolates, 162 were from males and 44 from females and the age varied widely between 12-85 years. However XDR-TB was found to be in the age group of 20-58 years (5 males: 1 female). Since the drug sensitivity analysis was done on samples collected from public sector hospitals and TB clinics, blood samples were not collected from the patients to determine their HIV status. Moreover, because of the social stigma and discrimination that may ensue, the physicians do not recommend it either. Therefore, the correlation between TB infection and HIV status of the patients could not be included in this study.

World Health Organization (WHO) considers XDR-TB a serious emerging threat to global public health, especially in countries with high prevalence of HIV. Even though HIV prevalence in Kerala is low [11], tuberculosis alone can be a health hazard because of the high population density. The observation calls for immediate measures to be taken to control and contain the spread of drug

resistant strains of *M. tuberculosis*. Fortunately DOTS plus treatment has been implemented recently in Kerala [11] and we hope it will help contain the spread of the disease in this state effectively.

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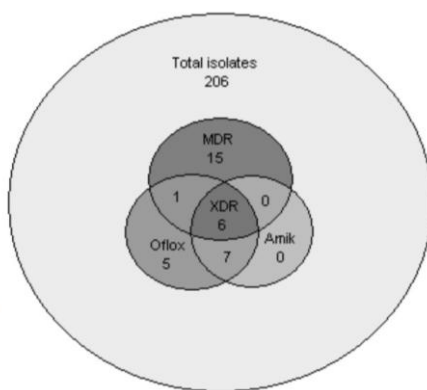


Fig 1. Extensive Drug resistance (XDR) in *M. tuberculosis* isolated from TB patients in Kerala. MDR - Multidrug resistant isolates, Oflox - isolates resistant to Ofloxacin, Amik- isolates resistant to Amikacin.