Isolation and Phenotypic and Genotypic Study of Macrolide Resistance Among Clinical Strains of Streptococcus pneumonia in Tehran

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Background & Objectives: Like other countries worldwide, in Iran Streptococcus pneumoniae is among most common pathogens that causes pneumonia, meningitis and otitis media in children. In recent years, several reports have been published about the prevalence and antibiotic resistance of pneumococci especially for penicillin and erythromycin as the major therapeutic options in Asian countries. But due to various problems, there is no comprehensive and adequate data for the prevalence and molecular characteristics of pneumococci in Iran. The aim of the present study is to evaluate phenotypically and genotypically macrolide resistance among clinical isolates of Streptococcus pneumoniae in Tehran.

Methods: After sampling from several clinical centers in Tehran, the suspected pneumococci immediately transferred to Department of Microbiology, Faculty of Medicine, Tehran University of Medical Sciences. The isolates were identified by gram staining, optochin susceptibility and bile solubility tests. Susceptibility testing was carried out by disk diffusion Methods for penicillin, erythromycin, TMP-SMZ, vancomycin, clindamycin, tetracycline and chloramphenicol. MIC determination by E-test and phenotypic D-test using erythromycin and clindamycin discs were performed for erythromycin resistant isolates. After DNA extraction, multiplex PCR was done for detection of three erythromycin resistance genes ermA, ermB, mefA/E.

Results: From 2010 up to 2012, a total of 170 isolates as to be pneumococci were collected from 30 centers in Tehran. After biochemical and molecular confirmatory tests, 88 isolates were identified correctly as pneumococci. Most of isolates were belong to elderly (39%) and then to children (26%). Eye and blood samples had the highest and BAL and ascitis samples had the lowest frequencies. 63 percent of isolates were MDR. The highest rate of antibiotic resistance was observed for penicillin and TMP-SMZ while the lowest one was seen for vancomycin and chloramphenicol. 47% of the isolates were resistant to erythromycin and the predominant phenotype was cMLSB. As a result of multiplex PCR, the frequency of ermA, ermB, mefA/E genes was 0%, 67 % and 60%, respectively.

Conclusion: Despite clinical importance of pneumococci, difficult isolation and rapid autolysis of the colony, inappropriate optochin discs and inadequate experience of technicians have resulted in a few studies done today on this pathogen in Iran. The advantage of this study is that the strains have collected in a fairly short period of time with a few subcultures till now. The frequency of MDR especially to penicillin is very high here. Simultaneous resistance to erythromycin and tetracycline found in some isolates may indicate the presence
of composite transposons carrying both resistance genes.ermB was the most prevalent gene found corresponding to cMLSB as the commonest resistance phenotype. Respecting to pathogenic capacity and transferable macrolide resistance genes, more surveys should be done about the epidemiology of pneumococci in Iran.

**Keywords:** *Streptococcus pneumonia*; MDR; Macrolide Resistance; Multiplex PCR