Emergence of Macrolide-Resistant *Streptococcus pneumoniae* non-PCV10 serotypes among pediatric patients in Bangladesh

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Introduction

Streptococcus pneumoniae (SPN) is the leading bacterial cause of Community Acquired Pneumonia (CAP) among children in Bangladesh. The emergence and spread of macrolide-resistant SPN have limited the treatment options for severe infections, leading to its inclusion on the WHO's 2024 list of priority pathogens. Besides protecting against selected SPN serotypes, the Pneumococcal Conjugate Vaccine-10 (PCV10) might also have induced dominant pathogen-strain replacement. Therefore, regional monitoring of SPN evolution and adaptation has become a priority for effective treatment and vaccination policy. Here, we aim to identify mutations linked to phenotypic plasticity in SPN through whole genome sequencing and molecular characterization

Figure 1. Tertiary level hospital from where CAP patients were enrolled.

Patient enrollment at hospital facility

"High prevalence of macrolide-resistant non-PCV10 serotypes carriage"

Methodology:

Nasopharyngeal swabs were collected and processed for SPN from 226 CAP patients (age<5) at a tertiary level hospital in Dhaka, Bangladesh, between March 2022 and April 2023. Antimicrobial susceptibility test (AST) was performed by disk diffusion test. After AST, forty-four selected SPN isolates were subjected to whole genome sequencing using the Illumina MiSeq platform. The sequencing raw reads were then assembled and annotated using the bactopia pipeline to identify serotypes, multi-locus sequence types (MLST), resistant genes, and plasmids. A core-genome alignment was obtained with Snippy, and the phylogenetic tree was calculated using IQ-TREE

Results:

S. pneumoniae was detected in 45% (101/226) and 85% (86/101) of isolates were macrolide (azithromycin or erythromycin) resistant. Genomic data revealed that the serotypes were diverse and majority of the circulating SPN strains were non-PCV10 types. The most prevalent serotype was 19A, accounting for 27%, followed by 35B, 15B/C, 18, and 6C/D. The presence of resistance plasmid repUS43 (n=35) was very frequent, while the plasmid rep36 or repUS60 were incidental. Most isolates, with a couple of exceptions, carried the tet(M) tetracycline resistance gene along with one or more macrolide resistance genes. The AST pattern was 100% concordant with the genomic data. Phylogenomic analysis revealed the presence of a distinct and recently evolved macrolide-resistant 19A and 6A/B/C clade.

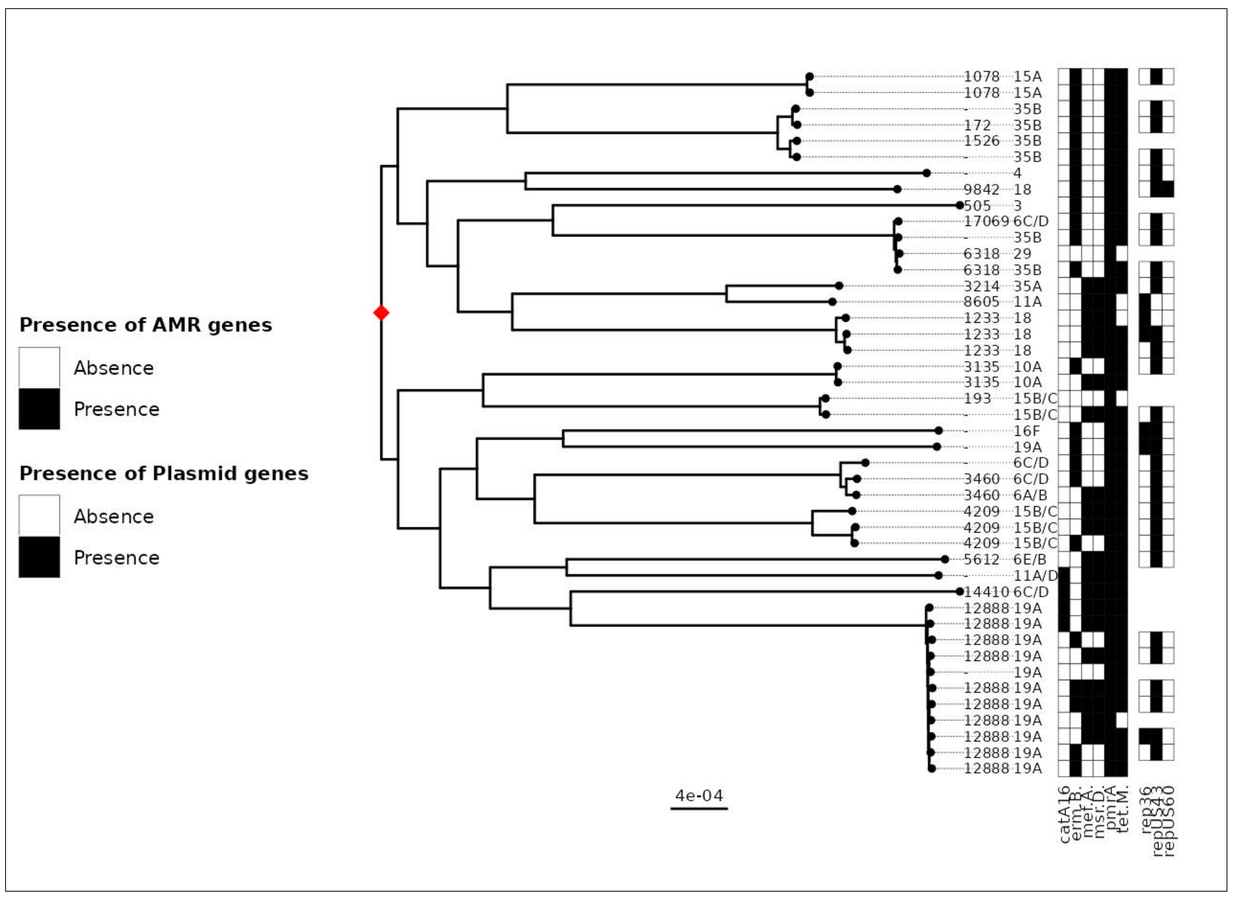

Figure 2. Phylogenetic tree representing the S. pneumoniae isolates. Midpoint-rooted phylogenetic tree of S. pneumoniae isolates with diverse serotype based on capsular polysaccharides, genoypic AMR and plasmids are indicated by squares in the outermost column.

Conclusion

Our data indicated a high prevalence of macrolide-resistant non-PCV10 serotypes circulating among hospital-based CAP patients in Bangladesh

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