



12th International Meeting on Microbial  
Epidemiological Markers (IMMEM XII)  
Dubrovnik, Croatia  
18–21 September 2019

## 28 PCR-based typing methods for detecting *Streptococcus pneumoniae* serotypes in Bulgarian children

Mariya Malcheva<sup>1</sup>, Ivan Simeonovski<sup>1</sup>, Viktoria Levterova<sup>1</sup>, Todor Kantardjiev<sup>1</sup>

<sup>1</sup>National Center of Infectious and Parasitic Diseases, Microbiology, Sofia, Bulgaria

**Background:** PCR-based serotyping methods utilize the heterogeneity within the capsular operon and could accurately determine more than 70 serotypes. Subsequent multiplex serotyping assays have been expanded to cover more serotypes and to determine the quantity of DNA and therefore density of colonization. Generally, PCR-based typing has been shown to be reliable and accurate, and can often be used directly on clinical specimen. Bulgarian children are vaccinated with PCV10 from 2010.

**Materials/methods:** We undertook a *S. pneumoniae* serotype carriage study to know the locally prevalent serotypes in vaccinated children n=764. Swabs from the nasopharynx were transferred to the microbiology laboratory in transport medium. Specimens were cultivated and from every swab DNA was directly isolated. Culture negative samples were screened with Real Time PCR for pneumococcal DNA and if positive they were typed. From culture positives DNA was isolated and analyzed with conventional PCR and the Operon S. Pneumo Strip commercial kit based again on the capsule operon of pneumococci.

**Results:** Overall carriage was calculated to be 52% with culture positive 21% of the samples. From the directly isolated DNA 3,5% of all samples were with more than one serotype - with two serotypes or with three serotypes carried at the same time. Below 10% of all positive samples were nontypeable for the primers and methods used. Prevalent carriage of two vaccine types was found - 4 and 23F, and 6C prevalence in non vaccine serotypes.

**Conclusions:** In conclusion we determined high overall carriage characterized mainly with non vaccine serotypes. With the methods used we were able to define more than 90% of the serotypes/serogroups carried in the analyzed children. Although reduced, vaccine type colonization in children remains persistent eight years after the introduction of PCV10.