Multiplex PCR for Detection of Capsular Polysaccharides Types of *Streptococcus pneumoniae* Clinical Isolates in Bali

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Streptococcus pneumoniae is one of respiratory infectious agents that can cause Invasive Pneumococcal Diseases (IPD), including meningitis and pneumonia. One of virulence factors involved in this bacterium's pathogenesis is capsular polysaccharides (CPS). CPS of *S. pneumoniae* is known as the pneumococcal vaccine component. Several types of *S. pneumoniae* CPS are dominant in Indonesia such as types 6, 23, 15, 33 and 12 in West Nusa Tenggara, type 7F in Jakarta, and types 6A/B dan 15B/C in Central Java. No data is reported from Bali related to *S.pneumoniae* CPS typing. Furthermore, method for detecting the CPS type simultaneously would help in determining the capsular type faster. Therefore, the aim of this study was to determine CPS types of *S. pneumoniae* isolates in Clinical Microbiology Laboratory, Sanglah General Hospital, Denpasar, Bali by using Multiplex PCR. Twenty-one isolates that were isolated from blood (11/52.4%), sputum (5/23.8%), and other clinical specimens (5/23.8%) were included in this study. Identification of *S. pneumoniae* was based on sensitivity of the isolates against optochin and presence of pneumolysin gene (*ply*) of each isolate. At first, Uniplex PCR was conducted to determine capsular type of each isolates, and then continued with Multiplex PCR 1 and 2 to determine capsular type simultaneously. Positive controls of Multiplex PCR 1 and 2 were developed in this study from DNA fragments that were positive for CPS from Uniplex PCR. All isolates were positive for the presence of *ply*, showed that all isolates were confirmed to be *S. pneumoniae*. Moreover, this study showed that type 19F was the predominant type (7 isolates (66.7%)); 2 isolates (9.5%) were positive for each type 23F and also for type 6A/B; and, there was only 1 isolate (4.8%) for each type 7F and 15B/C. Total of 8 isolates (38.1%) were found to be nontypeable isolates. By using positive controls, the Multiplex PCR successfully identified different types of CPS. Development of Multiplex PCR could help in diagnosing and identifying capsular type of *S. pneumoniae* simultaneously; therefore, could be used to figure out the predominant CPS type in the community.

Keywords: *Streptococcus pneumoniae,* Capsular Polysaccharides (CPS), Multiplex-PCR, Positive Control, Pneumococcal vaccine