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Genotyping of Nucleotide Sequences eccB₅ Gene of Mycobacterium tuberculosis complex

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Tuberculosis (TB) is an infectious disease caused by the *Mycobacterium* tuberculosis complex (MTBC) and major of the health world's problems. Indonesia is the big five countries of incidence TB case in 2014 and 2015. MTBC has many virulence factors such as $EccB_5$ encoded by $eccB_5$ gene. $EccB_5$ is a transmembran protein conserved membrane protein and could play a role of induced damage host cell, macrofage infection, and maybe corelate with the active disease. The aims of this research were to analyze of DNA sequences of $eccB_5$ gene of MTBC.

Methods: The isolat of *Mycobacterium* spp were collected from clinical microbiology of RSUD Dr. Soetomo hospital Surabaya Indonesia. DNA extraction were using TE boil extraction method and to be continued of nucleic acid amplification using PCR techniques. Spesific primer were designed by *software Clone Manager* 9 versi 9.2 (*Scietific & Educational software*) (forward 5'GCAGTGGCAAATACCCAGGTCAATGTC 3' and reversed 5'GGCAATATTCTCGGGCTTGATGACC 3'). The positivity of DNA spesific showed revealed of amplicon in 1592 bp and PCR product were sequenced by 1st Base. The sequence analysis using Genetyx-Win versi 10.0 (Genetyx Corporation, Tokyo, Japan).

Result: The total isolates of *Mycobacteria* spp were 28 and showed the positive of MTBC were 24 and 4 NTM using ICT test. The number of homology from MTBC using blast NCBI were 99%-100%. In this reseach proposed of one SNPs from the isolate with the SNPs position in 1277 (A/T) and change of amino acid (N/I) in 426 of codon position. **Conclusion**: The frequency distribution of patients with pulmonary TB for male and female were 46.43% and 39.29% respectively from 24 isolates. The sequence of $eccB_5$ gene of MTBC showed high significantly homology and proposed of SNPs.

Keywords: Tuberculosis, *Mycobacteria* spp, *Mycobacterium tuberculosis* complex, eccB₅ gene, SNPs