ABSTRAK

OPTIMASI AMPLIFIKASI DAN KLONING GEN *Chaperonin 60.1* PADA *Mycobacterium tuberculosis*

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Bakteri *Mycobacterium tuberculosis* merupakan salah satu bakteri paling patogen di dunia yang menyebabkan penyakit tuberkulosis. Sepertiga penduduk dunia telah diestimasi telah terinfeksi oleh basil tuberkulosis. Kegagalan vaksin BCG dalam mengontrol penyakit tuberkulosis di dunia, mencetuskan penemuan vaksin baru yang lebih efektif. Protein Chaperonin 60.1 *Mycobacterium tuberculosis* dipercaya dapat digunakan sebagai kandidat vaksin baru yang efektif.

Melalui penggunaan metode PCR, gen *Chaperonin 60.1* diamplifikasi. PCR dilakukan sebanyak 30 siklus, dengan denaturasi 94°C selama 60 detik, annealing 52°C selama 30 detik, dan extension 72°C selama 120 detik. Produk PCR dianalisis dengan elektroforesis dan didapatkan pita yang mendekati ukuran 1620 pasangan basa (pb). Kemudian hasil PCR ini diligasikan ke dalam plasmid pGEM-T *Easy Cloning Vector*, hasil ligasi ditransformasikan ke dalam *Escherichia coli* DH5α. Dari hasil kloning didapatkan 7 koloni putih dan 24 koloni biru. Untuk melihat koloni putih yang mengandung DNA sisipan maka digunakan metode lisis cepat. Dari hasil lisis cepat didapatkan 2 plasmid yang berasal dari koloni nomor 4 dan 6 yang diduga mengandung DNA target.

Kata kunci: *Mycobacterium tuberculosis*, gen Chaperonin 60.1, PCR, Kloning, Lisis Cepat
ABSTRACT

OPTIMIZATION OF AMPLIFICATION TECHNIQUE AND GENE CLONING OF Chaperonin 60.1 ON Mycobacterium tuberculosis

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Mycobacterium tuberculosis is one of the most pathogens in the world that cause tuberculosis disease. Estimates are that roughly one third of the world’s population is infected with the bacillus. The failure of BCG vaccination to control the global tuberculosis epidemic underline the need for better vaccine. Chaperonin 60.1, a protein base from Mycobacterium tuberculosis was believed to be a new effective vaccine for tuberculosis disease.

The Chaperonin 60.1 gene was amplified by PCR method. The PCR technique was performed in 30 cycles, denaturation at 94°C for 60 second; annealing at 52°C for 30 second; and extension at 72°C for 120 second. Analysis by gel agarose electrophoresis shown PCR fragment in the expected size 1620bp. Subsequently, resulted PCR product was ligated into pGEM-T Easy Cloning Vector and transformed into Escherichia coli DH5α. The result of cloning were 7 white colonies and 24 blue colonies. In order to examined the recombinant plasmid in white colonies contain target insert, quick lysis method was performed. It was found that two plasmids from the colony number 4 and 6 were predicted to be insert with DNA target.

Keyword: Mycobacterium tuberculosis, Chaperonin 60.1 gene, PCR, Cloning, Quick lysis
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