

Cloning and expression of pab gene of *M. tuberculosis* isolated from pulmonary TB patient in *E.coli* DH5α;

Deskripsi Lengkap: <https://lib.ui.ac.id/detail?id=20333165&lokasi=lokal>

Abstrak

Latar belakang: Antigen38 *Mycobacterium tuberculosis* merupakan agen serodiagnostik yang potensial karena mengandung dua epitop spesifik untuk sel B. Mahalnya agen diagnostik menyebabkan lambatnya realisasi diagnosis TB secara cepat di negara berkembang. Kami memproduksi antigen 38 rekombinan yang berasal dari galur lokal yang kemungkinan dapat digunakan untuk memproduksi alat serodiagnostik TB yang ekonomis.

Metode: Gen pab diisolasi dari pasien TB paru di Malang, diklon ke plasmid pGEM-Teasy menjadi pMB38. Klon *E.coli*

DH5α; yang membawa pMB38 diseleksi di medium yang ditambah dengan X-Gal. Ekspresi pab dilakukan menggunakan

pMBhis yang berasal dari pPRoExHTc dibawah kontrol promoter Trc dengan inang *E.coli* DH5α;

Hasil: Pencocokan sekuen gen pab dari klon *E.coli* DH5α; berwarna putih dengan gen pab dari *M. tuberculosis*H37Rv

memperlihatkan homologi sebesar 98%. Protein rekombinan yang sudah dihilangkan signal peptidanya ditemukan di sitoplasma.

Kesimpulan: Gen pab dari pasien TB dapat diekspresikan secara intraseluler dengan sistem heterolog

<hr>
Abstract

Background: *Mycobacterium tuberculosis* antigen38 is a potent serodiagnostic agent containing two *M. tuberculosis*specific

B-cell epitopes. The high price of imported diagnostic agents hinders realization of fast clinical TB diagnosis in

developing countries. Therefore, we produced recombinant antigen38 (recAg38M) from *M. tuberculosis* local strain, which

might be used to produce economical tuberculosis serodiagnostic kit.

Methods: Pab gene that was isolated from pulmonary TB patient in Malang was cloned into a plasmid vector (pGEMTeasy)

to construct pMB38. The *E.coli* DH5α; clone carrying pMb38 was selected on X-gal medium. The expression of pab

was mediated using pPRoExHTc under the control of Trc promoter and *E.coli* DH5α; as host.

Results: Alignment of the pab sequence from the white *E.coli* DH5α; clones with that of *M. tuberculosis* H37Rv showed

98% homology. The recombinant protein in which the signal peptide has been deleted to prevent the protein being secreted

into medium was found in the cytoplasm.

Conclusion: pab gene of *M. tuberculosis* isolated from a TB patient could be expressed in heterologous system in *E.*

coli DH5 α .