

Expression of HIV-1 Protein Recombinant p24 in Plasmid PQE80L of Escherichia Coli for development of HIV-1 Diagnostic System = Ekspresi Protein Rekombinan HIV-1 p24 menggunakan Plasmid PQe80L pada Escherichia coli untuk pengembangan sistem diagnostic HIV-1

Nuha Rahmania Zafirah, author

Deskripsi Lengkap: <https://lib.ui.ac.id/detail?id=20529066&lokasi=lokal>

Abstrak

Latar Belakang: P24 protein adalah salah satu protein milik HIV-1 yang membentuk kapsid bagian dalam dan dirilis ketika Gag protein di belah oleh protease virus. Kadar antigen p24 umumnya akan meningkat di darah pada fase akut infeksi. Oleh sebab ini, produksi protein rekombinan p24 yang efektif dan memiliki imunogenisitas serta imunoreaktivitas seperti protein p24 natural merupakan hal yang penting, untuk deteksi antibodi p24 dan membuat antibodi monoklonal p24. Dalam penelitian ini, dilakukan optimasi ekspresi protein p24 subtype HIV-1 CRF01_AE.

Metode : Penelitian ini menguji beberapa factor untuk ekspresi optimal dari protein rekombinan p24 pada plasmid pQE-80L milik Escherichia coli, termasuk konsentrasi inductor, durasi induksi, dan kultur media yang digunakan. Deteksi, quantifikasi, dan analisis protein dilakukan dengan SDS Page dan di analisis menggunakan Imajelab.

Hasil : p24 recombinant protein from HIV-1 Subtype CFR01_AE terekspresikan secara optimal menggunakan media Terrific Broth dengan di induksi 1mM Isopropyl-beta-D-thiogalactopyranoside (IPTG) selama 6 hours pada suhu 37°C.

Kesimpulan : Strategi dan Metode yang digunakan untuk mengekspresikan protein rekombinan dapat berbeda antar sistem dan antar jenis protein yang diekspresikan. Penelitian ini menunjukkan bahwa plasmid pQE80L-p24 yang sudah dikembangkan di PRVKP FKUI memiliki kemampuan untuk mengekspresikan protein rekombinan p24, dan teroptimasi pada media ekspresi Terrific Broth, konsentrasi Isopropyl-beta-D-thiogalactopyranoside (IPTG) 1mM, selama 6 jam pada suhu 37°C.

.....Background: P24 protein is one of HIV-1 protein that forms inner capsid and is released during cleave of Gag protein by viral protease. P24 antigen level will increase in the blood during the acute stage of infection. Therefore, an effective production of p24 recombinant protein that have the same immunogenicity and immunoreactivity of natural p24 protein are very important for detecting of p24 antibody and producing monoclonal p24 antibody. In this research, expression of p24 obtained from HIV-1 Subtype (HIV-1 CRF01_AE) was optimized to find the best expression system for the above protein.

Methods: In this research, several factors will be tested for optimal expression of recombinant p24 protein in plasmid pQE-80L of Escherichia coli, including the inducer concentration, duration of induction, and culture medium. Detection, quantitation, and analysis of the protein will be done using SDS Page and documented using Gel Documentation System to compare the effect of these factors.

Result: p24 recombinant protein from HIV-1 Subtype CFR01_AE is optimally expressed in Terrific Broth Medium in 1mM Isopropyl-beta-D-thiogalactopyranoside (IPTG), for 6 hours.

Conclusion: The strategies and method for protein expression may vary between a system, and a protein, to another. From this research it could be concluded that pQE80L-p24 plasmid that has been developed in

PRVKP FKUI is able to express p24 recombinant protein. Protein expression is optimized in Terrific Broth Medium in 0.5mM Isopropyl-beta-D-thiogalactopyranoside (IPTG), for 4 hours in 37°C.