

Pengklonaan dan ekspresi fragmen gen Apobec3G pada sistem prokariotik = The clone and expression gene fragments Apobec3G in prokaryotic system

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Abstrak

Latar Belakang : Sistem in vitro pendeteksi interaksi protein Vif HIV-1 dengan Apobec3G akan mempermudah identifikasi obat anti HIV-1 yang dapat meghambat replikasi HIV-1 melalui fungsi protein intrinsik Apobec3G. Protein Apobec3G rekombinan yang diperoleh melalui ekspresi pada sistem prokariota dapat digunakan bersama-sama dengan protein vif rekombinan untuk pengembangan sistem identifikasi substansi penghambat interaksi Apobec3G dan Vif HIV-1 dalam rangka eksplorasi protein bahan alam sebagai penghambat infeksi HIV.

Metode : Gen Apobec3G diklona ke dalam vektor plasmid pGEX6P-1 dalam E.coli TOP10, kemudian dilanjutkan dengan ekspresi pada sel E.coli BL21 untuk memperoleh protein rekombinan. Proses induksi dilakukan pada suhu 37°C dengan konsentrasi IPTG 0,5 mM, waktu induksi 2 dan 4 jam.

Hasil : Gen apobec3G telah berhasil diklona ke dalam vektor ekspresi prokariot, tetapi protein belum berhasil diekspresikan.

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Background: A system for detection of in vitro interaction of HIV-1 Vif protein with apobec3G protein will facilitate the identification of novel anti HIV-1 drug that inhibit the virus replication through functional intrinsic Apobec3G protein. Recombinant Apobec3G protein that is obtained through expression in prokaryotic expression system can be utilized in combination with recombinant HIV-1 Vif protein for the development of a sistem for identification of an inhibitory substance for interaction of Apobec3G and HIV-1 Vif, in order to explore the potential of natural substances as inhibitors of HIV infection.

Methods: The gene encoding the Apobec3G protein was cloned into the plasmid vector pGEX6P-1 in Top10 E. coli, followed by expression in BL21 E. coli to obtain the recombinant protein. Induction of expression was performed at 37oC with IPTG concentration of 0.5 mM for 2 and 4 hours.

Results: The gene encoding the Apobec3G has been successfully cloned in the prokariotic expression vector, however the expression of the corresponding recombinant protein has not been successful.