



POSTER PRESENTATION

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Recombinant soluble CCR5 AND CXCR4 chemokine receptors as anti-HIV drug targets

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Background

The aim of current work was to produce recombinant soluble CCR5 and CXCR4 chemokine receptors that could be used in screening of potential HIV-1 inhibitors.

Methods

Recombinant DNA constructs were produced by using polymerase chain reaction (PCR) technique and cloning. Proteins were expressed in bacteria and purified using immobilized metal ion affinity chromatography. To validate functionality of the recombinant proteins immunoprecipitation, immunoblot and ELISA assays were performed.

Results

We designed recombinant soluble CCR5 and CXCR4 proteins where functionally important regions of native receptors were connected with artificial linkers. These proteins were expressed in *E.coli* Origami 2(DE3) cells, refolded and purified. We have shown that recombinant proteins are functionally similar to native receptors since they bind to specific anti-CCR5 and anti-CXCR4 antibodies in immunoblot, immunoprecipitation and ELISA. Using the recombinant proteins for immunization we have obtained specific rabbit polyclonal serum. Finally, we established a competitive ELISA assay to search for the inhibitors of antigen and antibody binding. By applying this assay we performed screening of combinative drug library and found potential inhibitors.

Discussion

It is well known that CCR5 and CXCR4 chemokine receptors play a central role in the mechanism by which HIV binds to and enters white blood cells, and therefore represent key targets in the search for effective novel

treatments for HIV infection and AIDS. The current results indicate that recombinant soluble chemokine receptors are functionally active and can be used in screening of potential HIV-1 inhibitors.

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