

GPCR

Protein Catalog

C-X-C chemokine receptor type 4

Product specification

Acronym: CXCR4 Origin species : Human Protein reference : P61073 (UniProtKB) L01639.1 (GenBank) Family: GPCR Expression system: E.coli based CFPS Format: Proteoliposomes Protein sequence: Met1 – Ser352 Tag : Histidine tag fused to the N-terminal end of the protein Cleavage site: Factor Xa Product MW: 43 kDa

Application: Drug screening & discovery, antibody development, structural biology

Product description

Human CXCR4 (C-X-C Chemokine Receptor type 4) is an α-chemokine receptor specific for stromal-derived-factor-1 (SDF-1 also called CXCL12), a molecule endowed with potent chemotactic activity for lymphocytes. CXCR4 is essential for proper fetal development and is the major co-receptor for T-tropic strains of human immunodeficiency virus 1 (HIV-1). Additionally, SDF-1 and CXCR4 mediate cancer cell migration and metastasis. The N-terminal domain of CXCR4 is the binding site for SDF-1.

Recombinant protein sequence

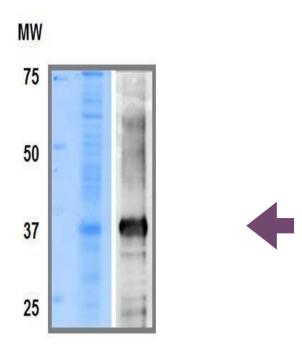
His tag – factor X cleavage site-MSGSHHHHHHSSGIEGRGRLIKHMEGISIYTSDNYTEEMGSGDYDSMKEPCFREENANFNKIFLPTIYSIIFLTGIVGNGLVILVMGYQKKLR SMTDKYRLHLSVADLLFVITLPFWAVDAVANWYFGNFLCKAVHVIYTVNLYSSVLILAFISLDRYLAIVHATNSQRPRKLLAEKVVYVGVWI PALLLTIPDFIFANVSEADDRYICDRFYPNDLWVVVFQFQHIMVGLILPGIVILSCYCIIISKLSHSKGHQKRKALKTTVILILAFFACWLPYYIG ISIDSFILLEIIKQGCEFENTVHKWISITEALAFFHCCLNPILYAFLGAKFKTSAQHALTSVSRGSSLKILSKGKRGGHSSVSTESESSSFHSS

Quality analysis

Purity: Typically > 70% as determined by SDS-Page and Coomassie Blue staining.

Purification procedure: As standard, hCXCR4 proteoliposomes are purified on a sucrose gradient. Further purification steps can be added if required.

Fig.1: hCXCR4 proteoliposome after purification (Coomassie Blue quantification and Western blot identification).



Quality controls

Cell-free expression systems provide a real alternative for membrane protein expression, enabling the study of structure and function of membrane proteins.

Methods: SPRi and Cisbio Bioassays Tag-lite® technology.

The Human CXCR4 protein was expressed in Synthelis' cell-free system in the presence of liposomes to obtain proteoliposomes in a one-step reaction. Receptor binding assay was used to characterize the interaction between the receptor and its ligands. The binding properties of CXCR4 proteoliposomes have been validated using Horiba Scientific SPRi platform and Cisbio Bioassays Tag-lite[®] technology.

Results:

SPRi technology

The binding properties of CXCR4 proteoliposomes have been validated using Horiba Scientific SPRi platform. Small molecule and peptide ligands were injected on a biochip grafted with CXCR4 biotinylated proteoliposomes. Specific interactions between CXCR4 proteoliposomes and different ligands were detected (see related <u>Application Note</u>). The signal was dose dependent. No signal was observed on the negative control spots (non-relevant proteoliposomes).

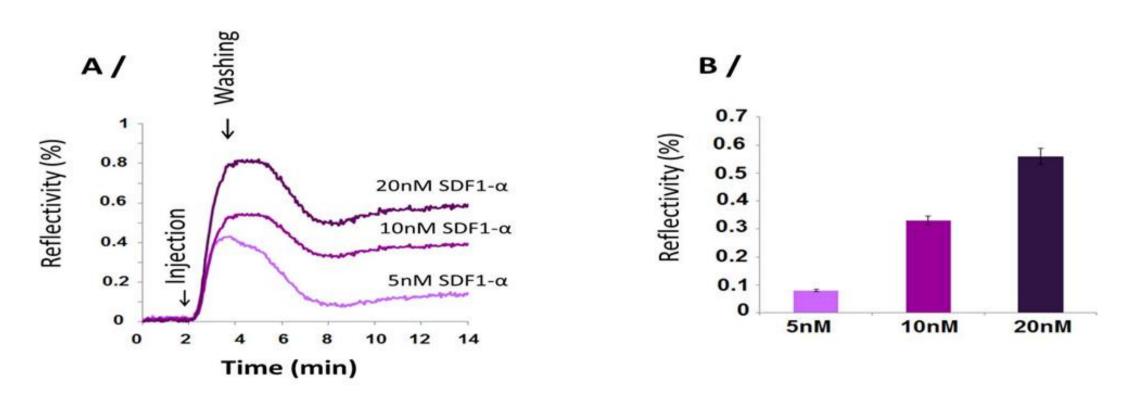


Fig.2: SPRi results. A /Sensorgrams obtained after SDF1a injections. B/ Variations of reflectivity obtained at steady state. The SDF1a peptide was injected successively at different concentrations: 5nM, 10nM then 20nM

<u>Cisbio Bioassays Tag-lite® technology</u>

CXCR4 binding properties were validated by Cisbio Bioassays Tag-lite technology. HEK293 Tag-Lite cell line overexpressing CXCR4 was used in a competitive binding assays with a constant SDF1α concentration against increasing concentrations of proteoliposomes. CXCR4 proteoliposomes are able to compete against the Tag-lite cell line for SDF1a ligand.

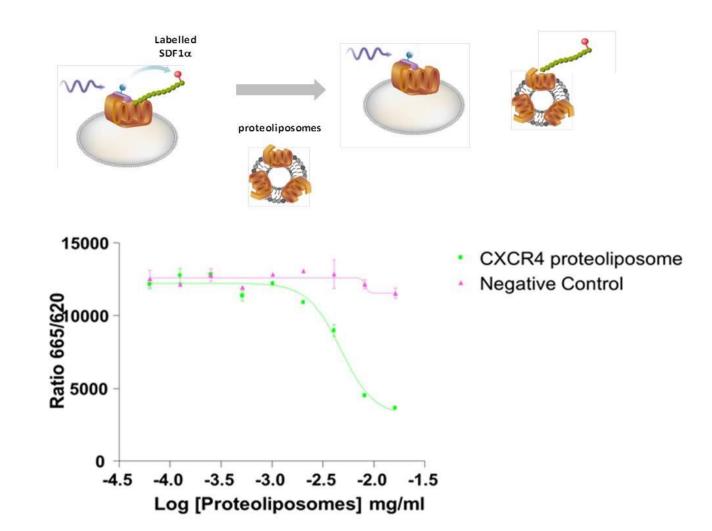


Fig.3: Competition assay between HEK293 Tag-Lite cells overexpressing CXCR4 and CXCR4 proteoliposomes

Formulation

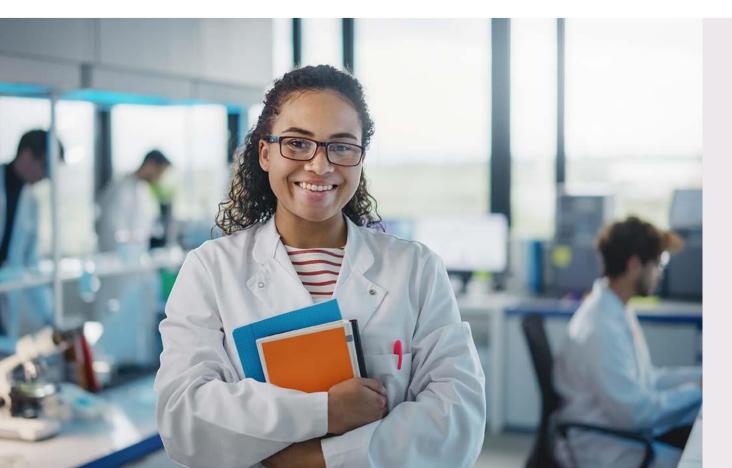
Buffer: Available in Tris 50mM, pH 7.5. Other buffers or customized formulation can be provided upon request.

Customized Hydrophobic matrix: Customized formulation with specific lipids like PEGylated or biotinylated lipids can be used upon request, as well as targeting molecules.

Storage/Stability: Store at +4°C for up to one week or several months at -80°C. Aliquot for storage. <u>Do not freeze-thaw after aliquoting.</u>

Use restrictions: For life science research use only.

Available sizes: 25µg,100 µg,500 µg, higher quantity on request.



Need a specific amount, a quote or any additional information? Contact-us



T: +33 (0)4 76 54 95 35 E: contact@synthelis.fr www.synthelis.com