

## Human CXCL8 / IL8 protein, His tag (active)

Cat. No. GTX00228-pro

<b>Applications</b>	Functional Assay
<b>Species</b>	Human

**Package**  
10 µg

## Applications

## Application Note

Interleukin 8 (IL8 or chemokine (C-X-C motif) ligand 8, CXCL8) is a chemokine produced by macrophages and other cell types such as epithelial cells, airway smooth muscle cells and endothelial cells. IL-8, also known as neutrophil chemotactic factor, has two primary functions. It induces chemotaxis in target cells, primarily neutrophils but also other granulocytes, causing them to migrate toward the site of infection. IL8 also induces phagocytosis once they have arrived. IL8 is also known to be a potent promoter of angiogenesis. Besides, Syndecan 1 (SDC1) has been identified as an interactor of IL8, thus a binding ELISA assay was conducted to detect the interaction of recombinant human IL8 and recombinant human SDC1. Briefly, IL8 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100 µl were then transferred to SDC1-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-IL8 pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were aspirated and washed 3 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37°C. Finally, add 50 µl stop solution to the wells and read at 450nm immediately. The binding activity of IL8 and SDC1 was in a dose dependent manner.

IL8 is a kind of neutrophil chemotactic factor, so chemotaxis assay used 24-well microchemotaxis system was undertaken to detect the chemotactic effect of IL8 on the human T-lymphocyte leukemia cell line Jurkat. Briefly, Jurkat cells were seeded into the upper chambers (100 µl cell suspension,  $1 \times 10^6$  cells/ml in RPMI-1640 with FBS free) and recombinant human IL8 (10 ng/ml, 100 ng/ml and 1000 ng/ml diluted separately in serum free RPMI-1640) was added in lower chamber with a polycarbonate filter (8µm pore size) used to separate the two compartments. After incubation at 37°C with 5% CO<sub>2</sub> for 1h, the filter was removed, then cells in low chamber were observed by inverted microscope at low magnification ( $\times 100$ ) and the number of migrated cells were counted at high magnification ( $\times 400$ ) randomly. The migrated Jurkat cells in low chamber at low magnification ( $\times 100$ ) randomly, and the migrated cells were counted at high magnification ( $\times 400$ ). IL8 is able to induce migration of Jurkat cells, and the optimum chemotaxis of IL8 occurs at 10~100 ng/ml.

**Observed MW (kDa)** 12 kDa.

## Properties

<b>Form</b>	Lyophilized powder
<b>Buffer</b>	Reconstitute with 20mM Tris and 150mM NaCl to 0.1-1.0mg/ml. Do not vortex. Lyophilized from 20mM Tris, 150mM NaCl, 1mM EDTA, 1mM DTT, 0.01% SKL, 5% Trehalose.
<b>Preservative</b>	ProClin 300
<b>Storage</b>	For short-term storage (1-2 weeks), store at 4°C. For long-term storage, store at -20°C or below. After reconstitution, keep as concentrated solution. Avoid freeze-thaw cycles.
<b>Region/Sequence</b>	N-terminal His-Tag; Ser28~Ser99 (NP_000575.1)
<b>Expression System</b>	E. coli
<b>Purity</b>	> 95%
<b>Endotoxin</b>	< 1 EU/µg
<b>Conjugation</b>	Unconjugated

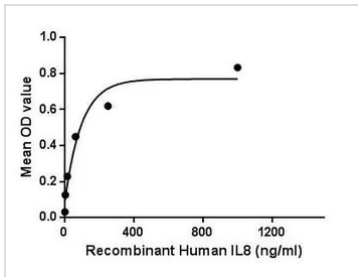


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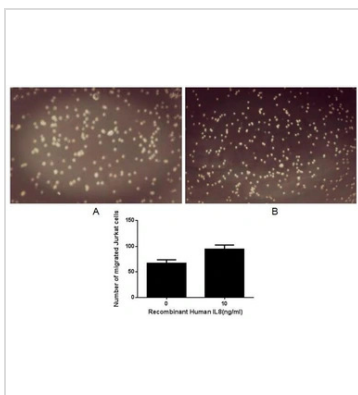
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**Note**

For laboratory use only. Not for any clinical, therapeutic, or diagnostic use in humans or animals. Not for animal or human consumption.

**DATA IMAGES**

**GTX00228-pro Functional Assay Image**

Functional ELISA analysis of GTX00228-pro Human CXCL8 / IL8 protein (active) which can bind immobilized SDC1 protein.

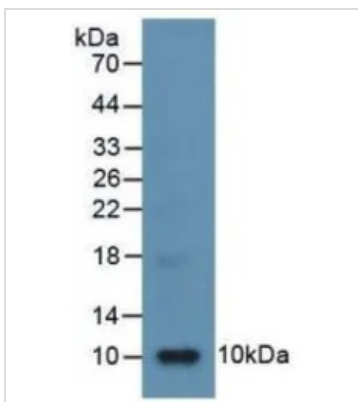

**GTX00228-pro Functional Assay Image**

The chemotactic effect of GTX00228-pro Human CXCL8 / IL8 protein (active).

Upper: (A) Jurkat cells were seeded into the upper chambers and serum free RPMI-1640 without IL8 was added in lower chamber, then cells in lower chamber were observed at low magnification ( $\times 100$ ) after incubation for 1hr.

(B) Jurkat cells were seeded into the upper chambers and serum free RPMI-1640 with 10 ng/ml IL8 was added in lower chamber, then cells in lower chamber were observed at low magnification ( $\times 100$ ) after incubation for 1hr.

Lower: The number of migrated cells were counted at high magnification ( $\times 400$ ) randomly (five fields for each filter).


**GTX00228-pro Image**

WB analysis of GTX00228-pro Human CXCL8 / IL8 protein (active).



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