APA080Si01 100µg

Active Interleukin 8 (IL8)

Organism Species: *Rhesus monkey (Simian)*

Instruction manual

FOR RESEARCH USE ONLY

NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Ala23~Pro101

Tags: N-terminal His-tag

Purity: >92%

Endotoxin Level: <1.0EU per 1µg (determined by the LAL method).

Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 5% Trehalose.

Applications: Cell culture; Activity Assays.

(May be suitable for use in other assays to be determined by the end user.)

Predicted isoelectric point: 9.2

Predicted Molecular Mass: 12.5kDa

Accurate Molecular Mass: 14kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

Reconstitute in 10mM PBS (pH7.4) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

[STORAGE AND STABILITY]

Storage: Avoid repeated freeze/thaw cycles.

Store at 2-8°C for one month.

Aliquot and store at -80°C for 12 months.

Stability Test: The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37°C for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration date under appropriate storage condition.

[SEQUENCE]

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                    AVLPRSAK ELRCECIKTY SKPFHPKFIK
ELRVIESGPH CANTEIIVKL SDGRELCLDP KEPWVQRVVE KFVKRAENQN
P
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[ACTIVITY]

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.Thus, 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 (100ul cell suspension,10^6 cells/ml in RPMI 1640 with FBS free) and recombinant Simian IL8 (1ng/ml, 10ng/ml ,100ng/ml and 1000ng/ml diluted separately in serum free RPMI 1640) was added in lower chamber with a polycarbonate filter(8um pore size) used to separate the two compartments. After incubation at 37℃ with 5% CO_2 for 90min, the filter was removed,then cells in low chamber were observed by inverted microscope at low magnification (×100) and the number of migrated cells were counted at high magnification(×400) randomly (five fields for each filter). Result shows IL8 is able to induce migration of Jurkat cells. The migrated Jurkat cells in low chamber at low magnification(×100) were shown in Figure 1. Five fields of each chamber were randomly chosen, and the migrated cells were counted at high magnification(×400). Statistical results were shown in Figure 2. The optimum chemotaxis of IL8 occurs at 10-100ng/ml.
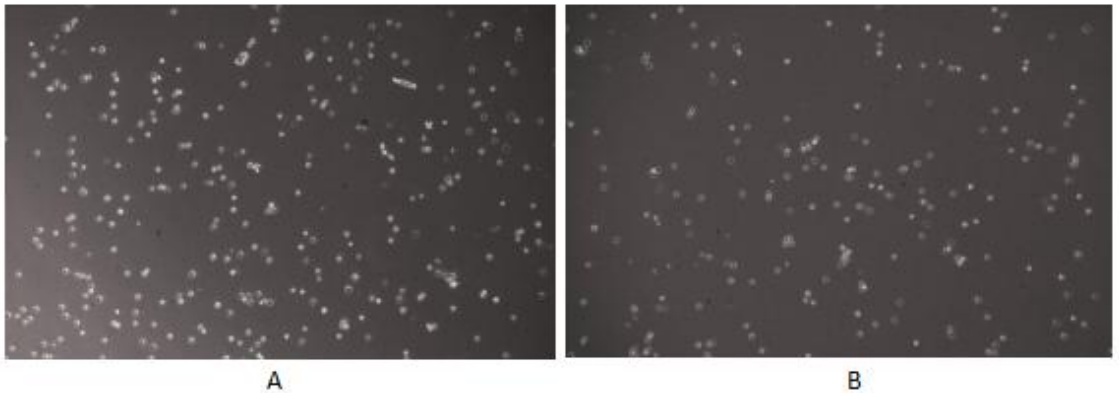
Figure 1.The chemotactic effect of IL8 on Jurkat cells

(A)Jurkat cells were seeded into the upper chambers and serum free RPMI 1640 with 10ng/ml recombinant Simian IL8 was added in lower chamber, then cells in lower chamber were observed at low magnification (×100) after incubation for 90min;

(B)Jurkat cells were seeded into the upper chambers and serum free RPMI 1640 without recombinant Simian IL8 was added in lower chamber, then cells in lower chamber were observed at low magnification (×100) after incubation for 90min.
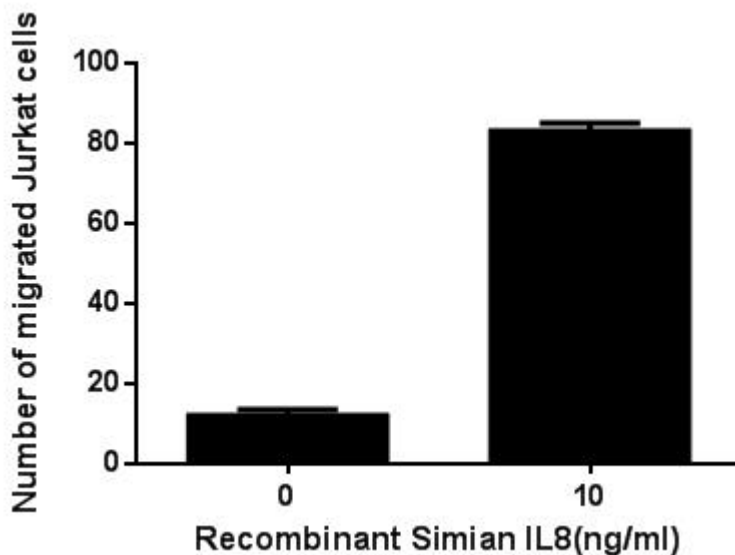

Figure 2.The chemotactic effect of recombinant Simian IL8 on Jurkat cells
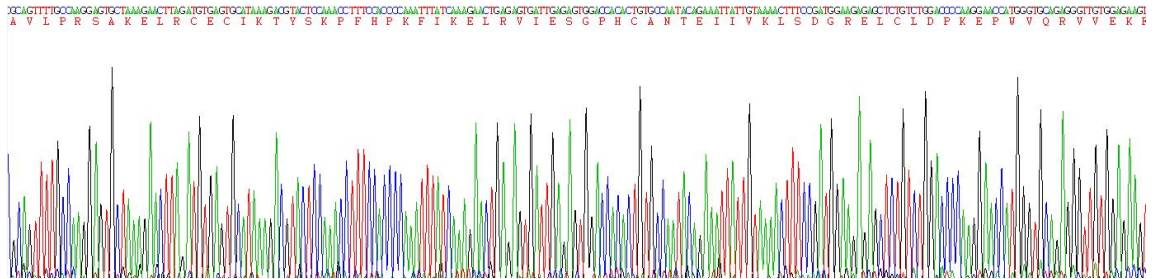
[IDENTIFICATION]

GCCAGTTTTGCCAGGAGTGCTAAAGAACTTAGATGTGAGTGCATAAAGACGTACTCCAAAACTTTCCACCCCAAATTTATCAAAGAACTGAGAGTGATTGAGAGTGGACCACACTGTGCAATACAGAAATTATTGTAAAACTTTCCGATGGAAGAGAGCTCTGTCTGGACCCCAAGGAACCATGGGTGCAGAGGGTTGTGGAGAAGT

A V L P R S A K E L R C E C I K T Y S K P F H P K F I K E L R V I E S G P H C A N T E I I V K L S D G R E L C L D P K E P V V Q R V V E K F

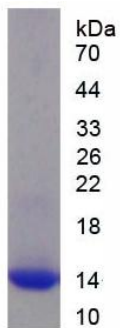**Figure 3. Gene Sequencing (extract)**

Figure 4. SDS-PAGE

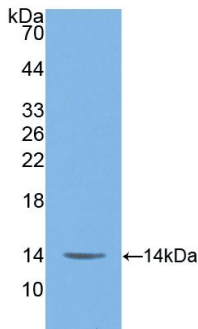**Sample: Active recombinant IL8, Simian**

Figure 5. Western Blot

Sample: Recombinant IL8, Simian;

Antibody: Rabbit Anti- Simian IL8 Ab (PAA080Si01)

[IMPORTANT NOTE]

The kit is designed for research use only, we will not be responsible for any issue
if the kit was used in clinical diagnostic or any other procedures.