

APA080Ga01 100µg
Active Interleukin 8 (IL8)
Organism Species: *Chicken (Gallus)*
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: Ala17~Leu103

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: 9.3

Predicted Molecular Mass: 13.4kDa

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]

ALSQ GRTLVKMGNE LRCQCISTHS KFIHPKSIQD VKLTPSGPHC
KNVEIIATLK DGREVCLDPT APWQLIVKA LMAKAQLNSD APL

[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 Chicken 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 °C with 5% CO₂ for 90min, 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 (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 ($\times 100$) were shown in Figure 1. Five fields of each chamber were randomly chosen, and the migrated cells were counted at high magnification ($\times 400$). Statistical results were shown in Figure 2. The optimum chemotaxis of IL8 occurs at 10ng/ml.

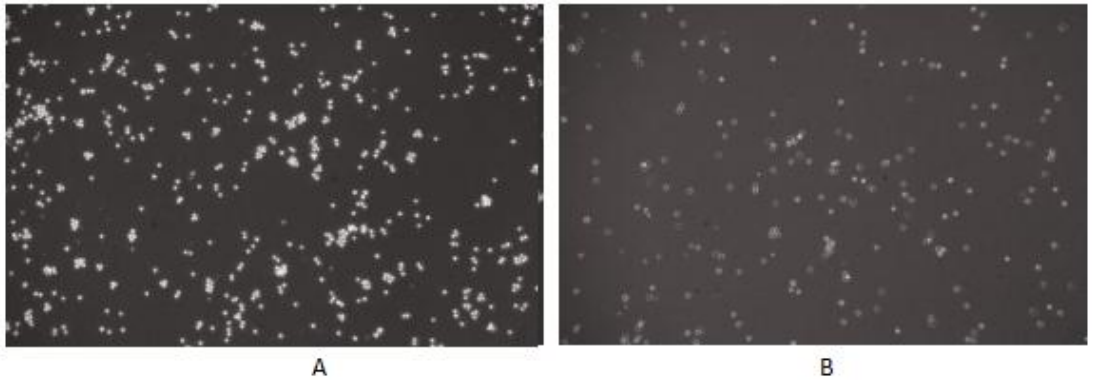


Figure 1. The chemotactic effect of IL8 on Jurkat cells

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

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

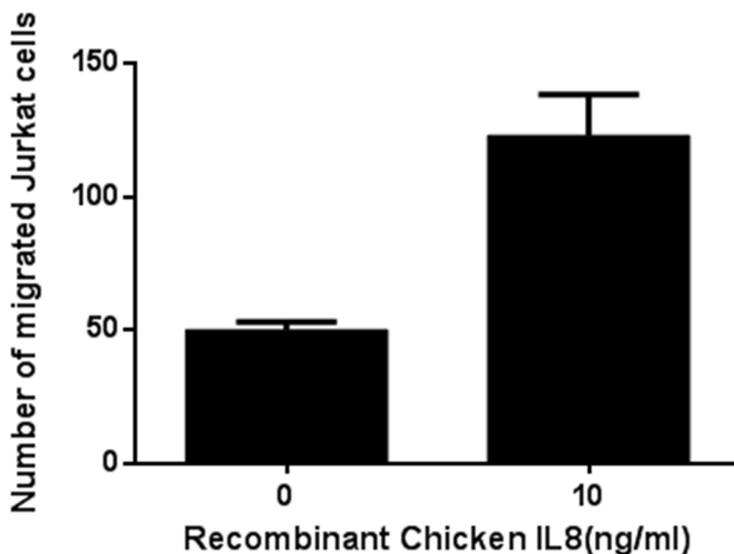


Figure 2. The chemotactic effect of recombinant Chicken IL8 on Jurkat cells.

[IDENTIFICATION]

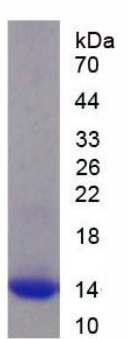


Figure 3. SDS-PAGE

Sample: Active recombinant IL8, Gallus

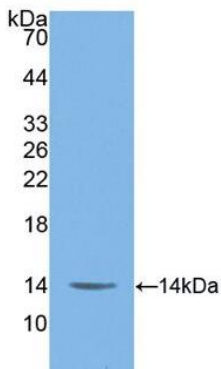


Figure 4. Western Blot

Sample: Recombinant IL8, Gallus;

Antibody: Rabbit Anti- Gallus IL8 Ab (PAA080Ga01)

[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.