

APA041Mu02 100µg
Active Chemokine (C-X-C Motif) Ligand 1 (CXCL1)
Organism Species: *Mus musculus* (Mouse)
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

12th Edition (Revised in Aug, 2016)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Ala25~Lys96

Tags: N-terminal His and GST Tag

Purity: >90%

Traits: Freeze-dried powder

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

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

Original Concentration: 200µg/mL

Applications: Cell culture; Activity Assays.

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

Predicted isoelectric point: 7.6

Predicted Molecular Mass: 37.9kDa

Accurate Molecular Mass: 38kDa 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]

APIANE LRCQCLQTMA GIHLKNIQSL
KVLPSGPHCT QTEVIATLKN GREACLDPEA PLVQKIVQKM LKGVPK

[ACTIVITY]

Neutrophil-activating protein 3 (NAP3) also known as chemokine (C-X-C motif) ligand 1 (CXCL1) is a small cytokine belonging to the CXC chemokine family. NAP3 is expressed by macrophages, neutrophils and epithelial cells, and has neutrophil chemoattractant activity. Thus, chemotaxis assay used 24-well microchemotaxis system was undertaken to detect the chemotactic effect of NAP3 on THP-1 the human monocytic cell line. Briefly, THP-1 cells were seeded into the upper chambers (150 ul cell suspension, 10^6 cells/ml in RPMI 1640 with FBS free) and different concentrations of recombinant mouse NAP3 diluted with serum free RPMI 1640 was added in lower chamber with a polycarbonate filter (8 um pore size) used to separate the two compartments. After incubation at 37 °C with 5% CO₂ 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 (five fields for each filter).

Result shows recombinant mouse NAP3 is able to induce migration of THP-1 cells. The migrated THP-1 cells in low chamber at low magnification ($\times 100$) were shown in Figure 1. Statistical results were shown in Figure 2. The optimum chemotaxis of NAP3 occurs at 0.001 ng/ml.

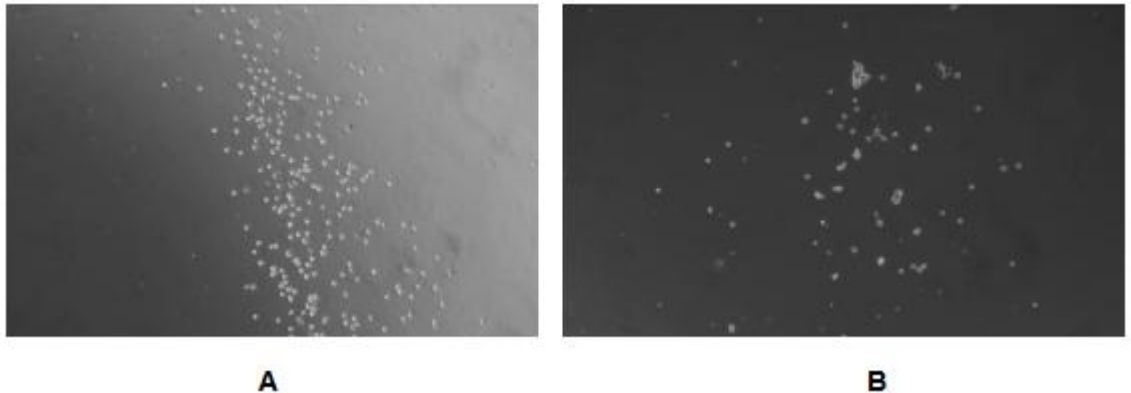


Figure 1. The chemotactic effect of NAP3 on THP-1 cells

(A) THP-1 cells were seeded into the upper chambers and serum free RPMI 1640 with 0.001 ng/ml NAP3 was added in lower chamber, then cells in lower chamber were observed at low magnification ($\times 100$) after incubation at 37 °C for 1h;

(B) THP-1 cells were seeded into the upper chambers and serum free RPMI 1640 without NAP3 was added in lower chamber, then cells in lower chamber were observed at low magnification ($\times 100$) after incubation at 37 °C for 1h.

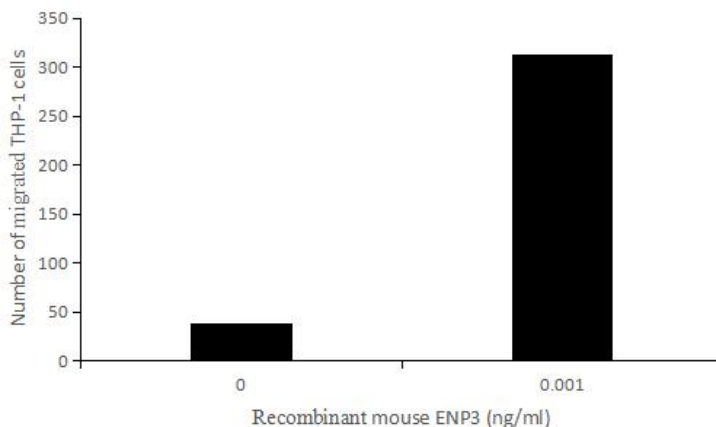


Figure 2. The chemotactic effect of NAP3 on THP-1 cells

[IDENTIFICATION]

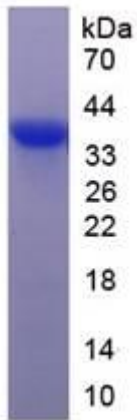


Figure 3. SDS-PAGE

Sample: Active recombinant CXCL1, Mouse

[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.