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APA122Hu02 100µg Active Stromal Cell Derived Factor 1 (SDF1) Organism Species: Homo sapiens (Human) Instruction manual

FOR IN VITRO USE AND RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression.

Host: E. coli

Residues: Pro23~Lys89

Tags: Two N-terminal Tags, His-tag and GST-tag

Purity: >98%

Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 0.05% sarcosyl and 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: 8.5

Predicted Molecular Mass: 37.8kDa

Accurate Molecular Mass: 40kDa as determined by SDS-PAGE reducing conditions.

[<u>USAGE</u>]

Reconstitute in 20mM Tris, 150mM NaCl (pH8.0) 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.

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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]

PVSLSYRC PCRFFESHVA RANVKHLKIL NTPNCALQIV ARLKNNNRQV CIDPKLKWIQ EYLEKALNK

[ACTIVITY]

SDF1 (stromal cell-derived factor 1), also known as C-X-C motif chemokine 12, is a chemokine protein that has chemotaxis active on T-lymphocytes and monocytes. It is thought that SDF1 stimulates migration of monocytes and T-lymphocytes through its receptors, CXCR4 and ACKR3; thus, chemotaxis assay used 24-well microchemotaxis system was undertaken to detect the chemotactic effect of SDF1 on the human monocytic cell line THP-1. Briefly, THP-1 cells were seeded into the upper chambers (100uL cell suspension, 10⁶cells/mL in RPMI 1640 with 0.5% FBS) and SDF1 (10ng/mL, 30ng/mL and 60ng/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° with 5% CO_2 for 3h, 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 SDF1 is able to induce migration of THP-1 cells. The migrated THP-1 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 SDF1 occurs at 10ng/mL.

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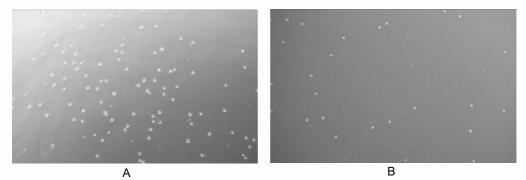


Figure 1. The chemotactic effect of SDF1 on THP1 cells.

(A) THP-1 cells were seeded into the upper chambers and serum free RPMI 1640 with 10ng/mL SDF1 was added in lower chamber, then cells in lower chamber were observed at low magnification (×100) after incubation for 3h;

(B) THP-1 cells were seeded into the upper chambers and serum free RPMI 1640 without SDF1 was added in lower chamber, then cells in lower chamber were observed at low magnification (×100) after incubation for 3h.

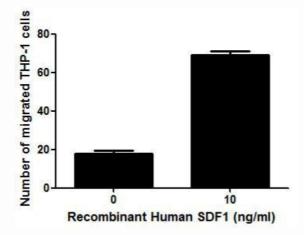
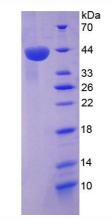


Figure 2. The chemotactic effect of SDF1 on THP-1 cells



[IDENTIFICATION]





Sample: Active recombinant SDF1, Human

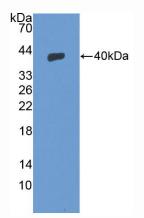


Figure 4. Western Blot

Sample: Recombinant SDF1, Human;

Antibody: Rabbit Anti-Human SDF1 Ab (PAA122Hu02)