Active Macrophage Inflammatory Protein 1 Alpha (MIP1a) Instruction Manual

SBPA062Hu61

Homo sapiens (Human)

Buffer Formulation	20mM Tris, 150mM NaCl, pH8.0, containing 1mM EDTA, 1mM DTT, 0.01% SKL, 5% Trehalose and Proclin300.
Traits	Freeze-dried powder
Purity	> 95%
Isoelectric Point	4.8
Applications	Cell culture; Activity Assays.

ACTIVITY TEST

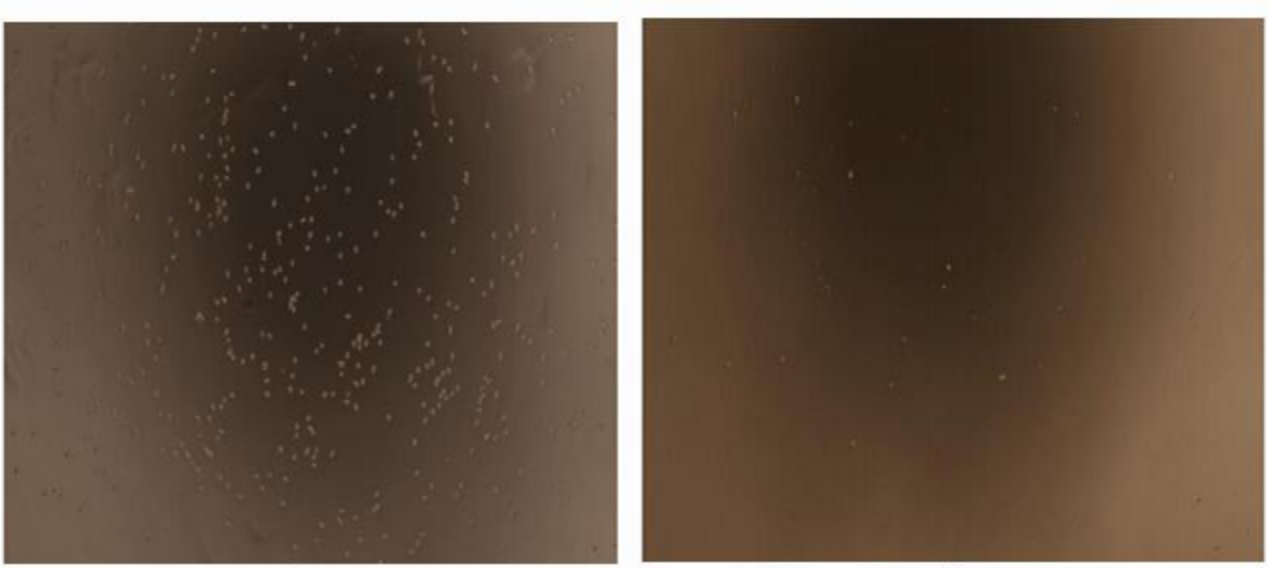


Figure 1. The chemotactic effect of MIP-1-alpha on THP1 cells

(A) THP1 cells were seeded into the upper chambers and serum free RPMI 1640 with 100ng/mL MIP-1a was added in lower chamber, then cells in lower chamber were observed at low magnification (×40) after incubation for 5h;

(B) THP1 cells were seeded into the upper chambers and serum free RPMI 1640 with no MIP-1a was added in lower chamber, then cells in lower chamber were observed at low magnification (×40) after incubation for 5h.

MIP-1a (macrophage inflammatory protein 1-alpha) also known as Chemokine (C-C motief) ligand 3 (CCL3), is a cytokine belonging to the CC chemokine family that is involved in the recruitment and activation of macrophages, monocytes and neutrophils. In this case, chemotaxis assay used 24-well microchemotaxis system was undertaken to evaluate the chemotactic effect of MIP-1a on the human monocytic cell line THP1. Briefly, THP1 cells were seeded into the upper chambers (100µl cell suspension, 106 cells/ml in RPMI 1640 with 0.5% FBS) and MIP-1a (100ng/mL, diluted 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 37oC with 5% CO2 for 5h, the filter was removed, then cells in low chamber were observed by inverted microscope at low magnification (×40) and the number of migrated cells were counted at high magnification (×400) randomly (five fields for each filter).
By counting migrated cells in low chamber at high magnification (×400) randomly, it was shown that a mean of 41.2 THP1 cells/field migrated towards serum free RPMI 1640

medium with 100ng/mL MIP-1a, while only 3.6 THP1 cells/field migrated towards serum free RPMI 1640 medium. And the migrated THP1 cells in low chamber at low magnification (×40) was shown in Figure 1.

USAGE

Reconstitute in 20mM Tris, 150mM NaCl (pH8.0) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

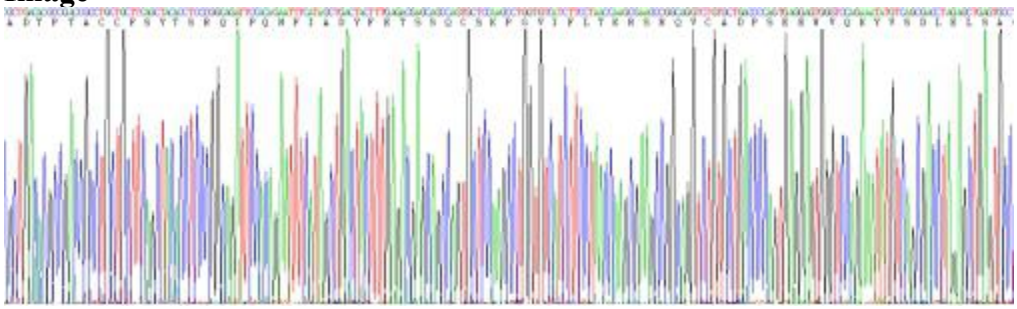
STORAGE

Avoid repeated freeze/thaw cycles. Store at 2-8°C for one month. Aliquot and store at -80°C for 12 months.

STABILITY

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.

Image



SDS-PAGE Image

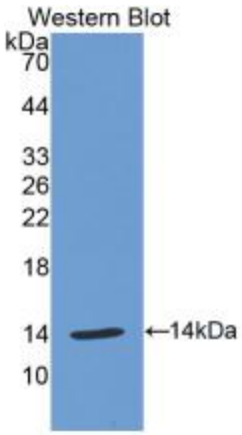


Figure. Western Blot; Sample: APA092Hu61; Antibody: PAA092Hu06.

[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.