

APA093Hu61 100µg

Active Macrophage Inflammatory Protein 1 Beta (MIP1b)

Organism Species: *Homo sapiens (Human)*

Instruction manual

FOR RESEARCH USE ONLY

NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Eukaryotic expression.

Host: 293F cell

Residues: Ala24~Asn92

Tags: N-terminal His Tag and C-terminal Fc Region of Human IgG1

Purity: >95%

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

Buffer Formulation: PBS, pH7.4, containing 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: 4.5

Predicted Molecular Mass: 35.0kDa

Accurate Molecular Mass: 42kDa as determined by SDS-PAGE reducing conditions.

Phenomenon explanation:

The possible reasons that the actual band size differs from the predicted are as follows:

1. Splice variants: Alternative splicing may create different sized proteins from the same gene.
2. Relative charge: The composition of amino acids may affects the charge of the protein.
3. Post-translational modification: Phosphorylation, glycosylation, methylation etc.
4. Post-translation cleavage: Many proteins are synthesized as pro-proteins, and then cleaved to give the active form.
5. Polymerization of the target protein: Dimerization, multimerization etc.

[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]

APMGSDPPTACCFSYTARKLPRNFVVDYYETSS LCSQPAVVVFQTKRSKQVCADPSES~~WVQ~~EYVYDLELN

[ACTIVITY]

Macrophage Inflammatory Protein 1 Beta (MIP1b), also known as CCL4, is a cytokine belonging to the CC chemokine family that is secreted at sites of inflammation by activated leukocytes, lymphocytes, vascular endothelial cells, and pulmonary smooth muscle cells. As a chemokine, MIP1b binds to the G-protein-coupled receptors CCR5 and CCR8, acting as a chemical attractant for natural killer cells, monocytes, and various other immune cells at sites of inflected or damaged tissue. At the same time, MIP1b can also retain the abilities to induce down-modulation of surface expression of the chemokine receptor CCR5 and to inhibit the CCR5-mediated entry of HIV-1 in T-cells. Chemokine C-C-Motif Receptor 7 (CCR7) can interact with MIP1b. Thus a functional binding ELISA assay was conducted to detect the interaction of recombinant human MIP1b and recombinant mouse CCR7. Briefly, biotin-linked MIP1b were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100 ul were then transferred to CCR7-coated microtiter wells and incubated for 1h at 37 °C. Wells were washed with PBST 3 times and incubation with Streptavidin-HRP for 30min, then wells were aspirated and washed 5 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37°C. Finally, add 50 µL stop solution to the wells

and read at 450/630 nm immediately. The binding activity of recombinant human MIP1b and recombinant mouse CCR7 was shown in Figure 1, the EC50 for this effect is 1.17 ug/mL.

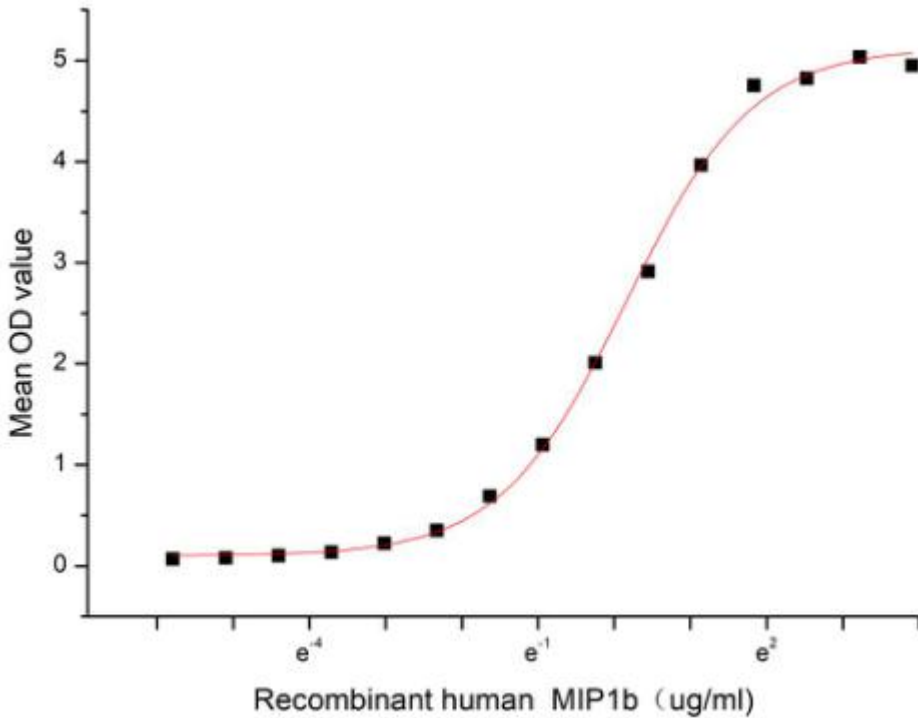


Figure 1. The binding activity of recombinant human MIP1b and recombinant mouse CCR7

[IDENTIFICATION]

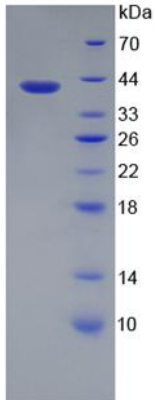


Figure 2. SDS-PAGE

Sample: Active recombinant MIP1b, Human

[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.