

APA045Ra01 200µg

Active Colony Stimulating Factor 2, Granulocyte Macrophage (GMCSF)

Organism Species: *Rattus norvegicus* (Rat)

Instruction manual

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

1th Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Ala18~Lys144

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

Purity: >95%

Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 1mM EDTA, 0.01% sarcosyl, 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: 6.3

Predicted Molecular Mass: 44.5kDa

Accurate Molecular Mass: 45kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

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.

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]

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APT RSPNPVTRPW KHVDAIKEAL SLLNDMPRALE  
NEKNEDVDII SNEFSIQRPT CVQTRLKLYK QGLRGNLTKL NGALTMIAASH  
YQTNCPPPTPE TDCEIEVTTF EDFIKNLKGF LFDIPFDCWK PVQK
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[ACTIVITY]

Granulocyte-macrophage colony-stimulating factor (GM-CSF), also known as colony stimulating factor 2 (CSF2), is a monomeric glycoprotein secreted by macrophages, T cells, mast cells, natural killer cells, endothelial cells and fibroblasts that functions as a cytokine. GM-CSF stimulates stem cells to produce granulocytes (neutrophils, eosinophils, and basophils) and monocytes. It also has some effects on mature cells of the immune system. These include, for example, inhibiting neutrophil migration and causing an alteration of the receptors expressed on the cells surface. GM-CSF signals via signal transducer and activator of transcription, STAT5. In macrophages, it has also been shown to signal via STAT3. Besides, Colony Stimulating Factor 2 Receptor Alpha (CSF2Ra) has been identified as an interactor of GM-CSF, thus a binding ELISA assay was conducted to detect the interaction of recombinant rat GM-CSF and recombinant rat CSF2Ra. Briefly, GM-CSF were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100uL were then transferred to CSF2Ra-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-GM-CSF pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were aspirated and

washed 3 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37°C. Finally, add 50µL stop solution to the wells and read at 450nm immediately. The binding activity of GM-CSF and CSF2Ra was shown in Figure 1, and this effect was in a dose dependent manner.

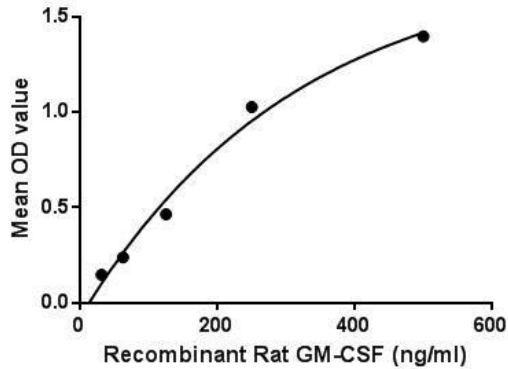


Figure 1. The binding activity of GM-CSF with CSF2Ra.

[IDENTIFICATION]

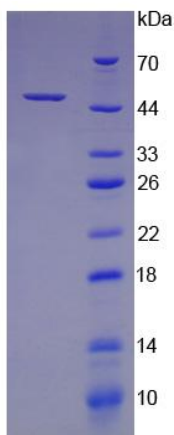


Figure 2. SDS-PAGE

Sample: Active recombinant GMCSF, Rat

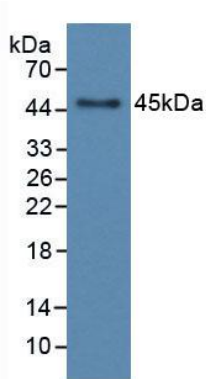


Figure 3. Western Blot

Sample: Recombinant GMCSF, Rat;

Antibody: Rabbit Anti-Rat GMCSF Ab (PAA045Ra01)

[IMPORTANT NOTE]

The kit is designed for in vitro and research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.