

APA652Hu61 100µg

Active Cluster Of Differentiation 28 (CD28)

Organism Species: Homo sapiens (Human)

Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Eukaryotic expression.

Host: 293F cell

Residues: Met1~Pro152

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

Purity: >90%

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: 8.7

Predicted Molecular Mass: 29.8kDa

Accurate Molecular Mass: 44-60kDa 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]

MLRLLALNL FPSIQVTGNK ILVKQSPMLV AYDNAVNLSC KYSYNLFSRE FRASLHKGLD SAVEVCVVYG NYSQQLQVYS KTGFNCDGKL GNESVTFYLQ NLYVNQTDIY FCKIEVMYPP PYLDNEKSNG TIIHVKGKHL CPSPLFPGPS KP

[ACTIVITY]

The cluster of differentiation (CD) antigen CD28 is a member of the immunoglobulin subfamily. CD28 is a central co-stimulatory molecule for TCR-mediated activation such as cytokine production and T-cell proliferation upon ligand binding and TCR stimulation. CD80 and CD86 are expressed on antigen presenting cells and CD86 is primary ligand for CD28. Thus a functional binding ELISA assay was conducted to detect the interaction of recombinant human CD28 and recombinant human CD86. Briefly, CD28 was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100 $\,\mu$ I were then transferred to CD86-coated microtiter wells and incubated for 1h at 37 $^{\circ}\!\!$ C. Wells were washed with PBST and incubated for 1h with anti-CD28 pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody for 1h at 37 $^{\circ}\!\!$ C, wells were aspirated and washed 5 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37 $^{\circ}\!\!$ C. Finally, add 50 μ l stop solution to the wells

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and read at 450/630 nm immediately. The binding activity of recombinant human CD28 and recombinant human CD86 was shown in Figure 1, the EC50 for this effect is 2.5 ug/mL.

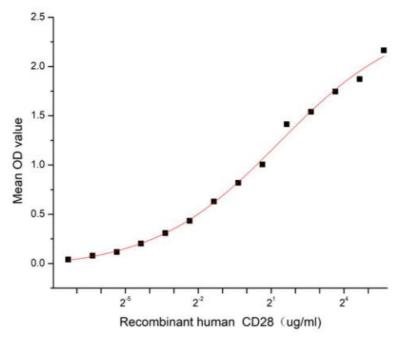


Figure 1. The binding activity of recombinant human CD28 and recombinant human CD86

[IDENTIFICATION]

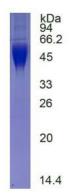


Figure 2. SDS-PAGE

Sample: Active recombinant CD28, 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.