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APA073Ra61 100µg Active Interleukin 2 (IL2) Organism Species: *Rattus norvegicus (Rat) 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: Ala21~Gln155 Tags: N-terminal His-tag Purity: >98% 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: 6.3 Predicted Molecular Mass: 17.1kDa Accurate Molecular Mass: 17&18kDa as determined by SDS-PAGE reducing conditions.

[<u>USAGE</u>]

Reconstitute in 10mM PBS (pH7.6) 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

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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]

APTSSPAKET QQHLEQLLLD LQVLLRGIDN YKNLKLPMML TFKFYLPKQA TELKHLQCLE NELGALQRVL DLTQSKSFHL EDAGNFISNI RVTVVKLKGS ENKFECQFDD EPATVVEFLR RWIAICQSII STMTQ

[ACTIVITY]

IL-2(Interleukin-2) is a cytokine produced by T-cells in response to antigenic or mitogenic stimulation. IL-2 is a type of signaling molecule in the immune system, that is required for both T-cell and B-cell proliferation and other activities crucial to regulation of the immune response. Therefore, in order to detect the bioactivity of recombinant rat IL-2, spleen single suspensions were prepared, activated with conA (final concentration 3 ug/ml). Cells were collected after 72h and washed with hanks. Then mouse splenic lymphocytes were were seeded into triplicate wells of 96-well plates at a density of 10,000 cells/well with or without the addition of various concentrations of recombinant rat IL-2. After incubated for 72h, cells were observed by inverted microscope and cell proliferation was measured by Cell Counting Kit-8(CCK-8). 10 μ l of CCK-8 solution was added to each well of the plate, the absorbance at 450 nm was measured using a microplate reader after incubating the plate for 1-4 hours at 37 °C . Proliferation of Splenic lymphocytes cells after incubation with IL-2 for 72h observed by inverted microscope was shown in Figure 1.

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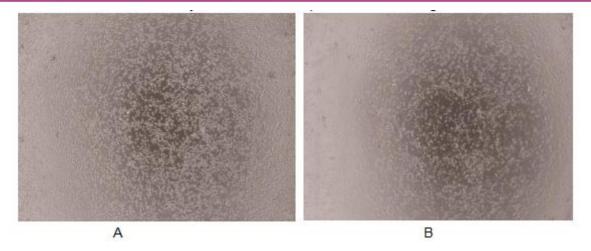


Figure 1. Cell proliferation of splenic lymphocytes cells after stimulated with IL-2.

(A) Splenic lymphocytes cells cultured in 1640, stimulated with 100ng/ml IL-2 for 72h;

(B)Unstimulated Splenic lymphocytes cells cultured in 1640 for 72h.

The dose-effect curve of recombinant rat IL-2 was shown in Figure2. It was obvious that recombinant rat IL-2 significantly promoted cell proliferation of Splenic lymphocytes cells .The ED50 for this effect is typically 1.5-1.7ng/mlng/ml.

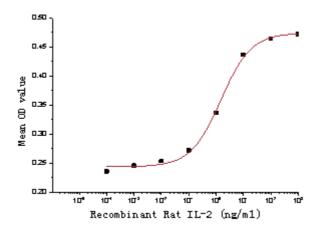


Figure 2. The dose-effect curve of IL-2 on Splenic lymphocytes cells

[IDENTIFICATION]

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kDa 70
44
33 26 22
18
14
10

Figure 3. SDS-PAGE

Sample: Active recombinant IL2, Rat

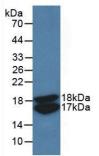


Figure 4. Western Blot

Sample: Recombinant IL2, Rat;

Antibody: Rabbit Anti-Rat IL2 Ab (PAA073Ra06)

[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.