

**APA073Po61 100µg**

**Active Interleukin 2 (IL2)**

**Organism Species: *Sus scrofa*; *Porcine (Pig)***

***Instruction manual***

FOR RESEARCH USE ONLY

NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

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12th Edition (Revised in Aug, 2016)

## **[ PROPERTIES ]**

**Source:** Eukaryotic expression.

**Host:** 293F cell

**Residues:** Ala21~Thr154

**Tags:** N-terminal His-tag

**Purity:** >90%

**Traits:** Freeze-dried powder

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

**Buffer Formulation:** PBS, pH7.4, containing 5% trehalose.

**Original Concentration:** 200µg/mL

**Applications:** Cell culture; Activity Assays.

(May be suitable for use in other assays to be determined by the end user.)

**Predicted isoelectric point:** 5.2

**Predicted Molecular Mass:** 16.8kDa

**Accurate Molecular Mass:** 17&20kDa 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 (pH7.6) 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 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.

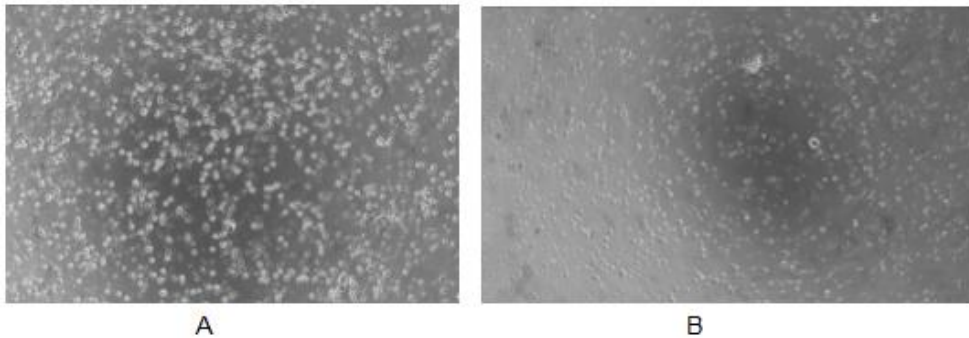
## **[ SEQUENCE ]**

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          APTSSSTKNT KKQLEPLLLD LQLLLKEVKN  
YENADLSRML TFKFYMPKQA TELKHLQCLV EELKALEGVL NLGQSKNSDS  
ANIKESMNNI NVTVLELKGS ETSFKCEYDD ETVTAVEFLN KWITFCQSIY  
STLT
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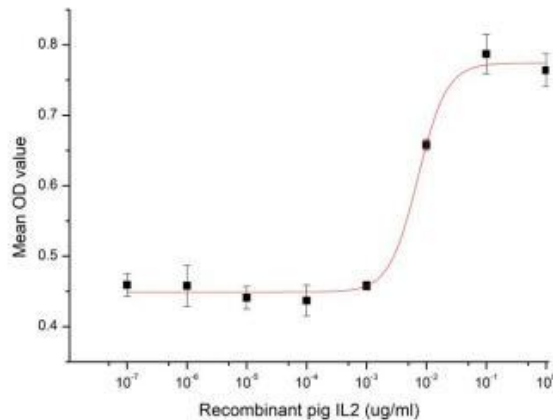
## **[ ACTIVITY ]**

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 pig 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 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 pig IL-2. After incubated for 96h, cells were observed by inverted microscope and cell proliferation was measured by Cell Counting Kit-8 (CCK-8). 10 µl 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 96h observed by inverted microscope was shown in Figure 1. The dose-effect curve of recombinant pig IL-2 was shown in Figure 2. It was obvious that recombinant pig IL-2 significantly promoted cell proliferation of Splenic lymphocytes cells. The EC50 for this effect is typically 0.0072-0.0079 ug/ml.



**Figure 1. Cell proliferation of splenic lymphocytes cells after stimulated with IL-2.**  
(A) Splenic lymphocytes cells cultured in 1640, stimulated with 1 ug/ml IL-2 for 96h;  
(B) Unstimulated Splenic lymphocytes cells cultured in 1640 for 96h.



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

## [ IDENTIFICATION ]

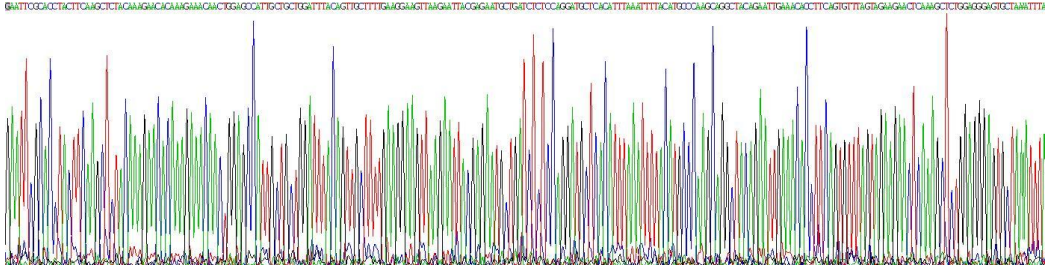


Figure 3. Gene Sequencing (extract)

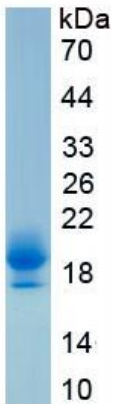


Figure 4. SDS-PAGE

Sample: Active recombinant IL2, Pig

## [ 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.