APA077Ra61 50µg

Active Interleukin 4 (IL4)

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: Cys25~Ser147
Tags: N-terminal His-tag

**Purity: >95%** 

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: 600µg/mL

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

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

Predicted isoelectric point: 9.1

Predicted Molecular Mass: 15.4kDa

**Accurate Molecular Mass:** 18kDa as determined by SDS-PAGE reducing conditions.

## [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]

CNDSPL REIINTLNQV TEKGTPCTEM
FVPDVLTATR NTTENELICR ASRVLRKFYF PRDVPPCLKN KSGVLGELRK
LCRGVSGLNS LRSCTVNEST LTTLKDFLES LKSILRGKYL QSCTSMS

#### [ACTIVITY]

The interleukin 4 (IL4,) is a cytokine that induces differentiation of naive helper T cells (Th0 cells) to Th2 cells. Upon activation by IL4, Th2 cells subsequently produce additional IL4 in a positive feedback loop. IL4 has many biological roles, including the stimulation of activated B-cell and T-cell proliferation, and the differentiation of B cells into plasma cells. It is a key regulator in humoral and adaptive immunity. IL4 induces B-cell class switching to IgE, and up-regulates MHC class II production. IL4 decreases the production of Th1 cells, macrophages, IFN-gamma, and dendritic cell IL12. The activity of IL4 is usually measured by a cell proliferation assay using TF-1 cells. TF-1 cells were seeded into triplicate wells of 96-well plates at a density of 8,000 cells/well with 2% serum standard 1640 which contains various concentrations of recombinant rat IL4. After incubated for 3 days, cells were observed by inverted microscope and cell proliferation was measured by Cell Counting Kit-8 (CCK-8). Briefly, 10 µl of CCK-8 solution was added to each well of the plate, then the absorbance at 450 nm was measured using a microplate reader after incubating the plate for 2-4 hours at 37  $\,^\circ\mathrm{C}$  . Proliferation of TF-1 cells after incubation with IL4 for 3 days observed by inverted microscope was shown in Figure 1. Cell viability was assessed by CCK-8 (Cell Counting Kit-8) assay after incubation with recombinant rat IL4 for 3 days.

The result was shown in Figure 2. It was obvious that IL4 significantly increased cell viability of TF-1 cells. The ED50 is 13.76 ng/ml.

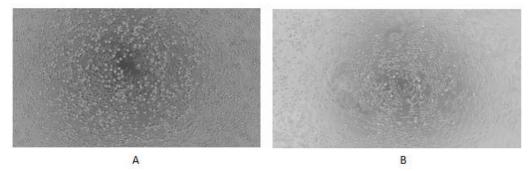


Figure 1. Cell proliferation of TF-1 cells after stimulated with IL4.

- (A) TF-1 cells cultured in 1640, stimulated with 100 ng/ml IL4 for 3 days;
- (B) Unstimulated TF-1 cells cultured in 1640 for 3 days.

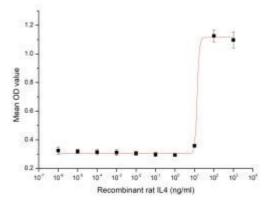


Figure 2. Cell proliferation of TF-1 cells after stimulated with IL4.

# [ IDENTIFICATION ]

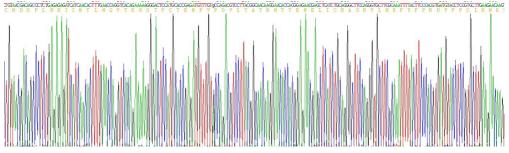


Figure 3. Gene Sequencing (extract)

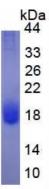


Figure 4. SDS-PAGE

Sample: Active recombinant IL4, Rat

## [ 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.