

APA079Ca61 100μg Active Interleukin 6 (IL6)

Organism Species: Canis familiaris; Canine (Dog)

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: Ala18~Met207 Tags: N-terminal His-tag

**Purity: >90%** 

**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: 6.0

Predicted Molecular Mass: 22.8kDa

Accurate Molecular Mass: 33kDa 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]

ATAFPTPGPLAGDSKDDATSNSLPLTSANKVEELIKYILGKISALRKEMCDKFNKCEDSKEALAENNLH LPKLEGKDGCFQSGFNQETCLTRITTGLVEFQLHLNILQNNYEGDKENVKSVHMSTKILVQMLKSKVKN QDEVTTPDPTTDASLQAILQSQDECVKHTTIHLILRSLEDFLQFSLRAVRIM

## [ACTIVITY]

Interleukin 6 (IL-6) is an interleukin that acts as both a pro-inflammatory cytokine and an anti-inflammatory myokine. Interleukin 6 is secreted by T cells and macrophages to stimulate immune response and also plays a role in fighting infection. It supports the growth of B cells and is antagonistic to regulatory T cells. To test the effect of IL6 on cell proliferation, 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 dog IL6. 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  $\mu$ l 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 1-4 hours at 37  $^{\circ}\mathrm{C}$ .

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Proliferation of TF-1 cells after incubation with IL6 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 dog IL6 for 3 days. The result was shown in Figure 2. It was obvious that recombinant dog IL6 significantly increased cell viability of TF-1 cells, the EC50 was 8.34 ng/ml.

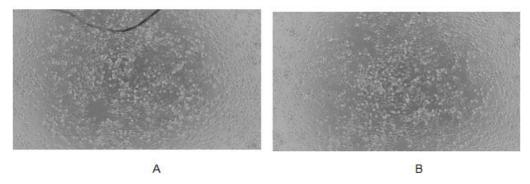


Figure 1. Cell proliferation of TF-1 cells after stimulated with recombinant dog IL6.

- (A) TF-1 cells cultured in 1640, stimulated with 10 ng/ml IL6 for 3 days;
- (B) Unstimulated TF-1 cells cultured in 1640 for 3 days.

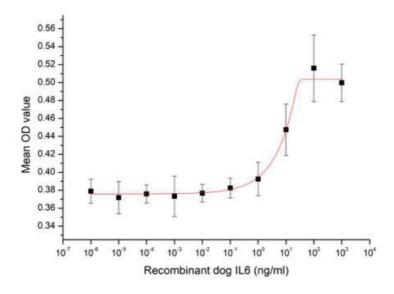


Figure 2. Cell proliferation of TF-1 cells after stimulated with recombinant dog IL6.

## [ IDENTIFICATION ]

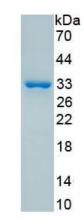


Figure 3. SDS-PAGE

Sample: Active recombinant IL6, Dog

## [ 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.