APA133Hu01 10µg Active Tumor Necrosis Factor Alpha (TNFa) Organism Species: Homo sapiens (Human) *Instruction manual*

FOR RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression. Host: *E. coli* Residues: Val77~Leu233 Tags: N-terminal His-tag Purity: >95% Buffer Formulation: 100mM NaHCO₃, 500mM NaCl, pH8.3, containing 0.01% sarcosyl, 5%Trehalose.

Applications: Cell culture; Activity Assays; In vivo assays.

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

Predicted isoelectric point: 7.0

Predicted Molecular Mass: 21.0kDa

Accurate Molecular Mass: 21kDa as determined by SDS-PAGE reducing conditions.

[<u>USAGE</u>]

Reconstitute in 100mM NaHCO₃, 500mM NaCl (pH8.3) 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.

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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]

VRSS SRTPSDKPVA HVVANPQAEG QLQWLNRRAN ALLANGVELR DNQLVVPSEG LYLIYSQVLF KGQGCPSTHV LLTHTISRIA VSYQTKVNLL SAIKSPCQRE TPEGAEAKPW YEPIYLGGVF QLEKGDRLSA EINRPDYLDF AESGQVYFGI IAL

[ACTIVITY]

TNFa (Tumor necrosis factor), is mainly secreted by macrophages and can induce cell death of certain tumor cell lines. It has been reported that TNFa can inhibit the proliferation and induce apoptosis of A549 cells, besides, the concentration of IL-1 β and IL-8 in cell supernatant will increase after stimulation. Therefore, a stimulation assay of TNFa was conducted using A549 cells. Briefly, A549 cells were incubated in DMEM with different concentrations of TNFa (1ng/mL, 10ng/mL, 100ng/mL) for 8h, after which the concentration of IL-1 β and IL-8 in the cell supernatant were detected by ELISA. IL-1 β and IL-8 levels in the cell supernatant of A549 cells increased significantly after stimulated with IL-1 β , the data was shown in Figure 1 and Figure 2 separately.

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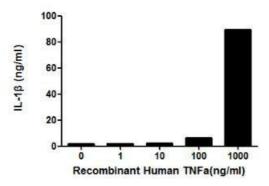
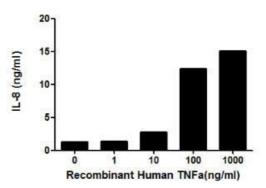
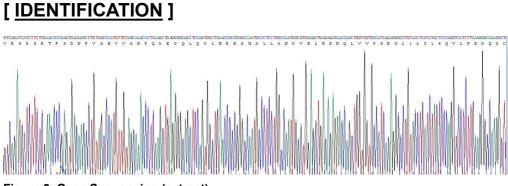
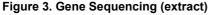


Figure 1. IL-1β level in the cell supernatant of A549 cells up-regulated by TNFa.

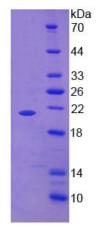








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Sample: Active recombinant TNFa, Human

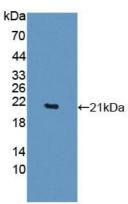


Figure 5. Western Blot Sample: Recombinant TNFa, Human; Antibody: Rabbit Anti-Human TNFa Ab (PAA133Hu01)

[<u>IMPORTANT NOTE</u>]

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.