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APA056Ra01 100µg Active Interleukin 10 (IL10) Organism Species: *Rattus norvegicus* (Rat) *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: Ser19~Asn178

Tags: N-terminal His-tag

Purity: >95%

Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 0.05% sarcosyl and 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: 9.1

Predicted Molecular Mass: 22.3kDa

Accurate Molecular Mass: 22kDa as determined by SDS-PAGE reducing conditions.

[<u>USAGE</u>]

Reconstitute in 20mM Tris, 150mM NaCl (pH8.0) 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]

SK GHSIRGDNNC THFPVSQTHM LRELRAAFSQ VKTFFQKKDQ LDNILLTDSL LQDFKGYLGC QALSEMIKFY LVEVMPQAEN HGPEIKEHLN SLGEKLKTLW IQLRRCHRFL PCENKSKAVE QVKNDFNKLQ DKGVYKAMNE FDIFINCIEA YVTLKMKN

[ACTIVITY]

Interleukin 10 (IL10), also known as human cytokine synthesis inhibitory factor (CSIF), is an anti-inflammatory cytokine. IL10 is a cytokine with multiple, pleiotropic, effects in immunoregulation and inflammation. It downregulates the expression of Th1 cytokines, MHC class II antigens, and co-stimulatory molecules on macrophages. It also enhances B cell survival, proliferation, and antibody production. IL10 can block NF-κB activity, and is involved in the regulation of the JAK-STAT signaling pathway. Investigation also has shown that IL-10 predominantly inhibits lipopolysaccharide (LPS) and bacterial product mediated induction of the pro-inflammatory cytokines TNF α , IL-1 β , IL-12 and IFN γ secretion from Toll-Like Receptor (TLR) triggered myeloid lineage cells. Besides, Interleukin 10 Receptor Alpha (IL10Ra) has been identified as an interactor of IL10, thus a binding ELISA assay was conducted to detect the interaction of recombinant rat IL10 and recombinant rat IL10Ra. Briefly, IL10 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100uL were then transferred to IL10Ra-coated microtiter wells and incubated for 2h at 37 °C. Wells were washed with PBST and incubated for 1h with anti-IL10 pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were aspirated

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and washed 3 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37° C. Finally, add 50μ L stop solution to the wells and read at 450nm immediately. The binding activity of IL10 and IL10Ra was shown in Figure 1, and this effect was in a dose dependent manner.







Figure 2. Gene Sequencing (extract)

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Figure 3. SDS-PAGE

Sample: Active recombinant IL10, Rat



Figure 4. Western Blot Sample: Recombinant IL10, Rat; Antibody: Rabbit Anti-Rat IL10 Ab (PAA056Ra01)

[<u>IMPORTANT NOTE</u>]

The kit is designed for in vitro and research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.