APA223Mu01 100µg Active Interleukin 1 Receptor Antagonist (IL1RA) Organism Species: Mus musculus (Mouse) *Instruction manual*

FOR IN VITRO USE AND RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

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1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression. Host: *E. coli* Residues: Arg27~GIn178 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; In vivo assays.

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

Predicted isoelectric point: 8.5

Predicted Molecular Mass: 18.4kDa

Accurate Molecular Mass: 20kDa 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]

RPSG KRPCKMQAFR IWDTNQKTFY LRNNQLIAGY LQGPNIKLEE KIDMVPIDLH SVFLGIHGGK LCLSCAKSGD DIKLQLEEVN ITDLSKNKEE DKRFTFIRSE KGPTTSFESA ACPGWFLCTT LEADRPVSLT NTPEEPLIVT KFYFQEDQ

[ACTIVITY]

IL1RA (interleukin-1 receptor antagonist) is an agent that binds to the cell surface interleukin-1 receptor (IL-1R), which would prevent IL-1 from intracellular signal transduction. Besides, mouse IL1R1 and rat IL1R1 share similarities in amino acid sequence with the identity of 85.6%. Thus, a binding ELISA assay was constructed to detect the association of recombinant mouse IL1RA with recombinant rat IL1R1. Briefly, IL1RA were diluted serially in PBS with 0.1%BSA (pH 7.4). Duplicate samples of 100uL were then transferred to IL1R1-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-IL1RA pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were incubated 15-25 minutes at 37°C. Finally, add 50µL stop solution to the wells and read at 450nm immediately. The binding activity of IL1RA with IL1R1 was shown in Figure 1 and this effect was in a dose dependent manner.

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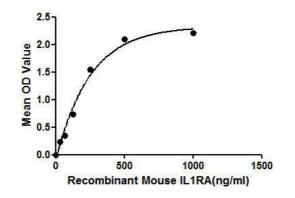


Figure 1. The binding activity of IL1RA with IL1R1.



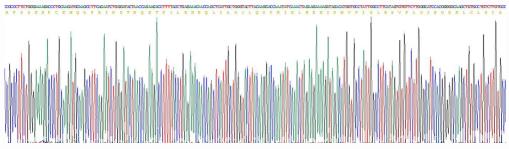


Figure 2. Gene Sequencing (extract)

kDa 70
44
33
26
22
18
14
10



Sample: Active recombinant IL1RA, Mouse

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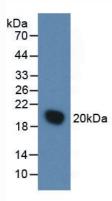


Figure 4. Western Blot

Sample: Recombinant IL1RA, Mouse;

Antibody: Rabbit Anti-Mouse IL1RA Ab (PAA223Mu01)

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