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APB041Ra01100µg Active Vitronectin (VTN) 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: Gln21~Arg400 Tags: N-terminal His Tag Purity: >90% Endotoxin Level: <1.0EU per 1µg (determined by the LAL method). Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 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: 6.3 Predicted Molecular Mass: 47.1kDa Accurate Molecular Mass: 55kDa as determined by SDS-PAGE reducing conditions.

[<u>USAGE</u>]

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.

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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]

QESCKGRCTQ GFMASKKCQC DELCTYYQSC CVDYMEQCKP QVTRGDVFTM PEDEYWSYDY PEETKNSTNA GVQSENTSLH FNLKPRAEET IKPTTPDPQE QSNTQEPEVG QQGVAPRPDT TDEGTSEFPE EELCSGKPFD AFTDLKNGSL FAFRGEYCYE LDETAVRPGY PKLIQDVWGI EGPIDAAFTR INCQGKTYLF KGSQYWRFED GVLDPDYPRN ISEGFSGIPD NVDAALALPA HSYSGRERVY FFKGKQYWEY EFQQQPSQEE CEGSSLSAVF EHFALLQRDS WENIFELLFW GRSSDGAKGP QFISRDWHGV PGKVDAAMAG RIYITGSTFR SVQAKKQKSG RRSRKRYRSR RGRGHSRSRS RSMSSRRPSR

[ACTIVITY]

Vitronectin (VTN) is a glycoprotein of the hemopexin family which is abundantly found in serum, the extracellular matrix and bone. It is a secreted protein and exists in either a single chain form or a clipped, two chain form held together by a disulfide bond. VTN can binds to integrin alpha-V beta-3 and thus promotes cell adhesion and spreading. It also binds to plasminogen activator inhibitor-1 (PAI-1) and serves to regulate proteolysis initiated by plasminogen activation. Besides, Insulin Like Growth Factor Binding Protein 5 (IGFBP5) has been identified as an interactor of VTN, thus a binding ELISA assay was conducted to detect the interaction of recombinant Rat VTN and recombinant Rat IGFBP5. Briefly, VTN were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100ul were then transferred to IGFBP5-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-VTN pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were aspirated and washed 3 times. With the addition of substrate solution, 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 VTN and IGFBP5 was shown in Figure 1, and this effect was in a dose dependent Cloud-Clone Corp.

manner.



Figure 1. The binding activity of VTN with IGFBP5

[IDENTIFICATION]



Figure 2. SDS-PAGE Sample: Active recombinant VTN, Rat

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Figure 3. Western Blot Sample: Recombinant VTN, Rat; Antibody: Rabbit Anti- Rat VTN (PAB041Ra01)

[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.