

APA259Ra01 50µg
Active Annexin A5 (ANXA5)
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: Met1~Asp319

Tags: N-terminal His Tag

Purity: >95%

Endotoxin Level: <1.0EU per 1µg (determined by the LAL method).

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: 5.2

Predicted Molecular Mass: 37.0kDa

Accurate Molecular Mass: 35kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

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.

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]

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MALRGTVTDF SGFDGRADAE VLRKAMKGLG TDEDSILNLL TARSNAQRQQ  
IAEEFKTLFG RDLVNDMKSE LTGKFEKLIV ALMKPSRLYD AYELKHALKG  
AGTDEKVLTE IIASRTPEEL RAIKQAYEEE YGSNLEDDVV GDTSGYYQRM  
LVVLLQANRD PDTAIDDAQV ELDAQALFQA GELKWGTDEE KFITILGTRS  
VSHLRRVFDK YMTISGFQIE ETIDRETS GN LENLLLAVVK SIRSIPAYLA  
ETLYYAMKGA GTDDHTLIRV IVSRSEIDLF NIRKEFRKNF ATSLYSMIKG  
DTSGDYKKAL LLLCGGEDD
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[ACTIVITY]

Annexin A5 (ANXA5) is a cellular protein in the annexin group. In flow cytometry, ANXA5 is commonly used to detect apoptotic cells by its ability to bind to phosphatidylserine, a marker of apoptosis when it is on the outer leaflet of the plasma membrane. ANXA5 has been proposed to play a role in the inhibition of blood coagulation by competing for phosphatidylserine binding sites with prothrombin and also to inhibit the activity of phospholipase A1. Besides, Tumor Protein p53 (TP53) has been identified as an interactor of ANXA5, thus a binding ELISA assay was conducted to detect the interaction of recombinant rat ANXA5 and recombinant rat TP53. Briefly, ANXA5 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100µL were then transferred to TP53-coated microtiter wells and incubated for 2h at 37 °C. Wells were washed with PBST and incubated for 1h with anti-ANXA5 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 ANXA5 and TP53 was shown in Figure 1, and this effect was in a dose dependent manner.

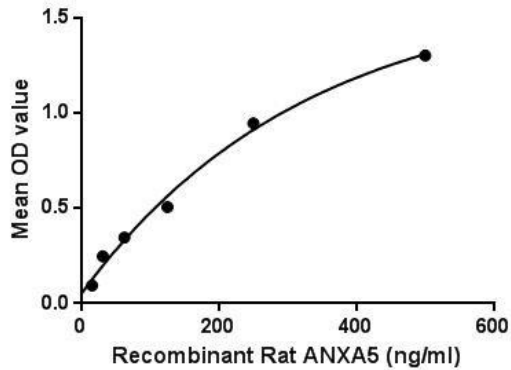


Figure 1. The binding activity of ANXA5 with TP53

[IDENTIFICATION]

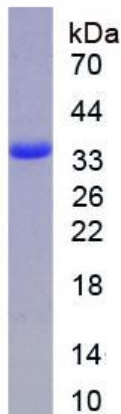


Figure 2. SDS-PAGE

Sample: Active recombinant ANXA5, Rat

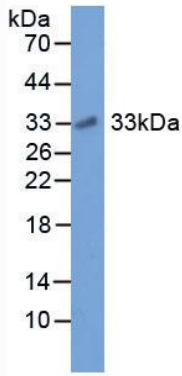


Figure 3. Western Blot

Sample: Recombinant ANXA5, Rat;

Antibody: Rabbit Anti-Rat ANXA5 Ab (PAA259Ra01)

[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.