

APL534Ra01 100µg

Active Growth Arrest And DNA Damage Inducible Protein Alpha (GADD45a)

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~Arg165

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: 4.5

Predicted Molecular Mass: 22.2kDa

Accurate Molecular Mass: 22kDa 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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MTLEEFSAAE QKIERMDTVG DALEEVLSKA RSQRTITVGV YEAAKLLNVD
PDNVVLCCLA ADEDDDRDVA LQIHFTLIRA FCCENDINIL RVSNPGR LAE
LLLLLENDKSP AESGGLA QTP DLHCVLVTNP HSSQWKDPAL SQLICFCRES
RYMDQWVPVI NLPER
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[ACTIVITY]

Growth arrest and DNA-damage-inducible protein GADD45 alpha (GADD45a) is a member of the GADD45 family. It may affect PCNA interaction with some CDK (cell division protein kinase) complexes; stimulates DNA excision repair in vitro and inhibits entry of cells into S phase. In T-cells, functions as a regulator of p38 MAPKs by inhibiting p88 phosphorylation and activity. Besides, Proliferating Cell Nuclear Antigen (PCNA) has been identified as an interactor of GADD45a, thus a binding ELISA assay was conducted to detect the interaction of recombinant rat GADD45a and recombinant rat PCNA. Briefly, GADD45a were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100µL were then transferred to PCNA-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-GADD45a 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 GADD45a and PCNA was shown in Figure 1, and this effect was in a dose dependent manner.

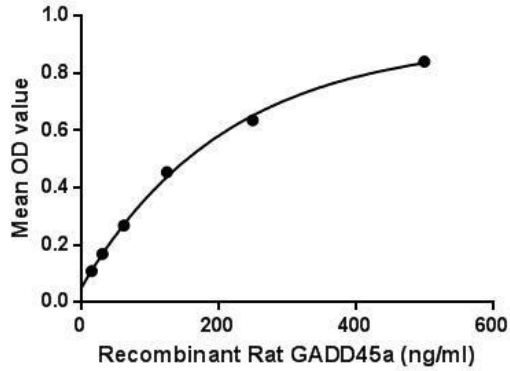


Figure 1. The binding activity of GADD45a with PCNA.

[IDENTIFICATION]

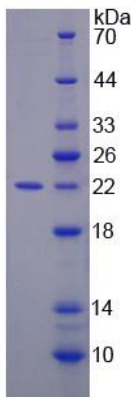


Figure 2. SDS-PAGE

Sample: Active recombinant GADD45a, Rat

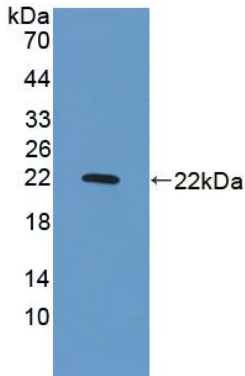


Figure 3. Western Blot

Sample: Recombinant GADD45a, Rat;

Antibody: Rabbit Anti-Rat GADD45a Ab (PAL534Ra01)

[IMPORTANT NOTE]

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.