

APB964Ra01 100µg
Active Calpain 1, Large Subunit (CAPN1)
Organism Species: Rattus norvegicus (Rat)
Instruction manual

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

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Pro75~Asp356

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: 5.3

Predicted Molecular Mass: 33.5kDa

Accurate Molecular Mass: 33kDa 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]

```
PNSSKT YGIKWKRPTE LLSNPQFIVD
GATRTDICQG ALGDCWLLAA IASLTLNETI LHRVVPYQGS FQEGYAGIFH
FQLWQFGEWV DVVVDDLLPT KDGKLVFVHS AQGNEFWSAL LEKAYAKVNG
SYEALSGGCT SEAFEDFTGG VTEWYDLQKA PSDLYQIILK ALERGSLLGC
SINISDIRDL EAITFKNLVR GHAYSVTDK QVTYQGQRVN LIRMRNPWGE
VEWKG PWSN SYEWNKVPY EREQLRVKME DGEFWMSFRD FIREFTKLEI
CNLTPD
```

[ACTIVITY]

Calpain 1, Large Subunit (CAPN1) is an intracellular protease that requires calcium for its catalytic activity. Calcium-regulated non-lysosomal thiol-protease which catalyze limited proteolysis of substrates involved in cytoskeletal remodeling and signal transduction. It has broad endopeptidase specificity. Besides, Signal Transducer And Activator Of Transcription 3 (STAT3) has been identified as an interactor of CAPN1, thus a binding ELISA assay was conducted to detect the interaction of recombinant rat CAPN1 and recombinant rat STAT3. Briefly, CAPN1 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100uL were then transferred to STAT3-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-CAPN1 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 CAPN1 and STAT3 was shown in Figure 1, and this effect was in a dose dependent manner.

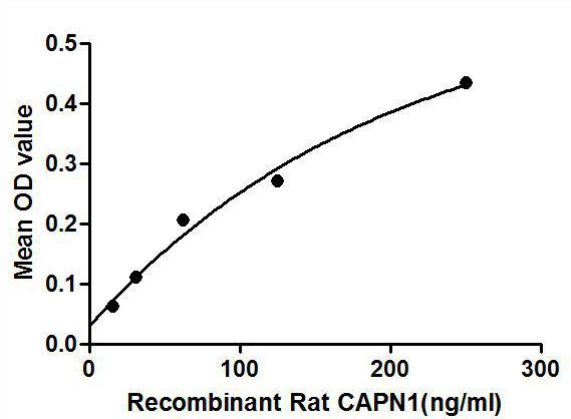


Figure 1. The binding activity of CAPN1 with STAT3.

[IDENTIFICATION]

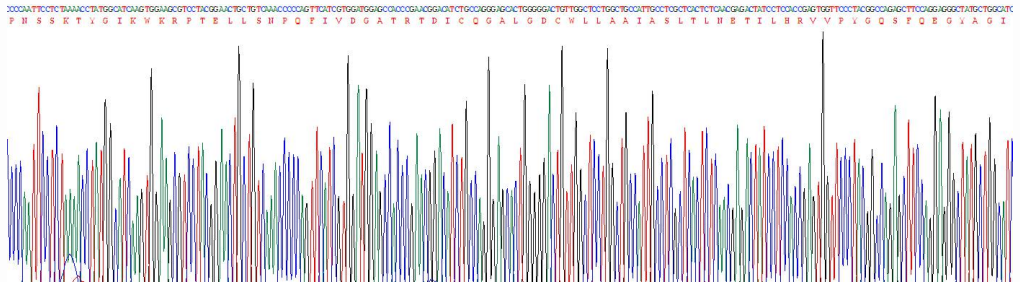


Figure 2. Gene Sequencing (Extract)

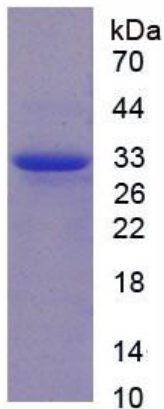


Figure 3. SDS-PAGE

Sample: Active recombinant CAPN1, Rat

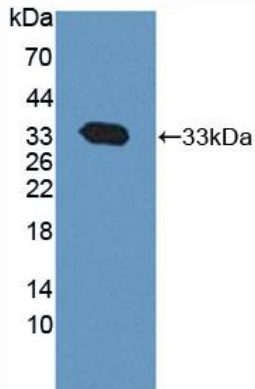


Figure 4. Western Blot

Sample: Recombinant CAPN1, Rat;

Antibody: Rabbit Anti-Rat CAPN1 Ab (PAB964Ra01)