

APB697Mu01 100µg

Active Alpha-1-Antitrypsin (a1AT)

Organism Species: *Mus musculus* (Mouse)

Instruction manual

FOR RESEARCH USE ONLY

NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Glu25~Lys413

Tags: N-terminal His-tag

Purity: >80%

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

Buffer Formulation: PBS, pH7.4, containing 0.01% Sarcosyl, 5%Trehalose .

Original Concentration: 200µg/mL

Applications: Activity Assays.

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

Predicted isoelectric point: 5.7

Predicted Molecular Mass: 45.0kDa

Accurate Molecular Mass: 45&12kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

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.

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]

```
EDVQET DTSQKDQSPA SHEIATNLGD
FAISLYRELV HQSNTSNIFF SPVSIATAFA MLSLGSKGDT HTQILEGLQF
NLTQTSEADI HKSFQHLLQT LNRPDSELQL STGNGLFVNN DLKLVEKFLE
EAKNHYYAEV FSVNFAESEE AKKVINDFVE KGTQGKIAEA VKKLDQDTVF
ALANYILFKG KWKKPDPEN TEEAEFHVDE STTVKVPMMT LSGMLHVHHC
STLSSWVLLM DYAGNATAVF LLPDDGKMQH LEQTLSEKELI SKFLLNRRRR
LAQIHFPRLS ISGEYNLCTL MSPLGITRIF NNGADLSGIT EENAPLKLSQ
AVHKAULTID ETGTEAAAVT VLQMPMSMP PILRFDHPFL FIIFEHTQS
PIFLGKVVDP THK
```

[ACTIVITY]

Alpha-1-Antitrypsin (a1AT), a major serine protease inhibitor synthesized primarily in the liver, is crucial for protecting tissues from excessive proteolytic damage. Circulating in human plasma, it targets and inhibits a range of proteases — including neutrophil elastase, trypsin, and chymotrypsin — which, if left unregulated, can degrade essential structural proteins. Beyond its protective function, A1AT is involved in modulating inflammatory responses and maintaining pulmonary homeostasis; deficiencies in A1AT are associated with conditions such as emphysema and liver disorders. Notably, A1AT can directly bind to and inhibit Renin (REN), a central enzyme in the renin-angiotensin system, thereby modulating its proteolytic activity. To detect the activity of recombinant a1AT, a functional ELISA assay was performed to evaluate the interaction between recombinant mouse a1AT and recombinant dog REN. Briefly, a1AT was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100µl were then transferred to REN-coated microtiter wells and incubated for 1h at 37 °C. Wells

were washed with PBST and incubated for 1h with anti-a1AT pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody for 1h at 37 °C, wells were aspirated and washed 5 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37 °C. Finally, add 50 µL stop solution to the wells and read at 450/630nm immediately. The binding activity of recombinant mouse a1AT and recombinant dog REN was shown in Figure 1, the EC50 for this effect is 0.054 µg/mL.

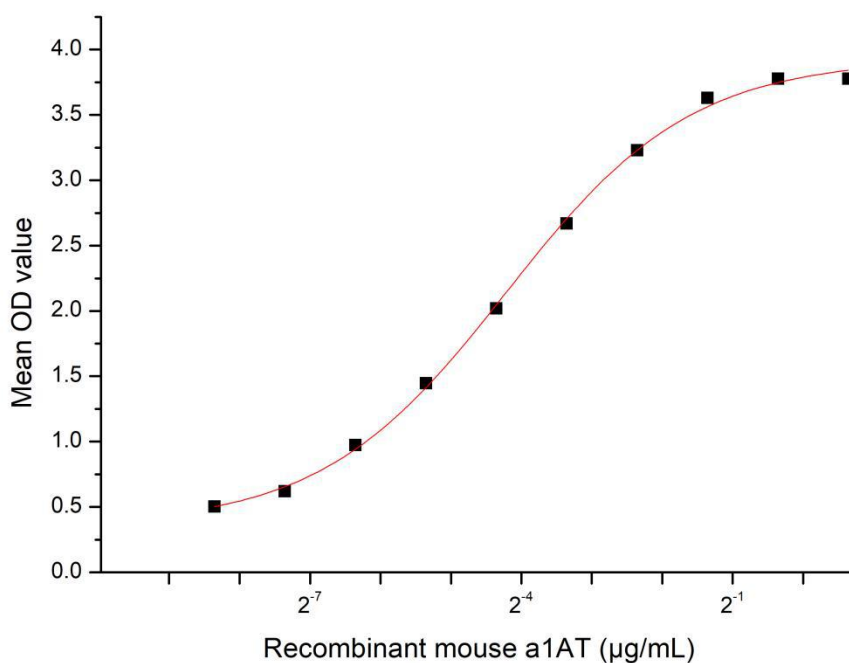


Figure 1. The binding activity of recombinant mouse a1AT and recombinant dog REN

Seq1 TGGCGAGTTCGAGGAGGACCTCCGAGCGTGATGTCCCGCCTCCGTGAAGTCTCAAACTCCGGGACTTTCCGATCGGCATDCCGGGACCTGGTCATGTCACCTTCACAACTCTCTCTCCCGAGGAGATTGCCAGCCCTTCTTAGCTCTCCCTAGGGACWAGGGGACCTCAAGCCGAGATCTAGAGCCCTCCGATTCCTCCGAAAC

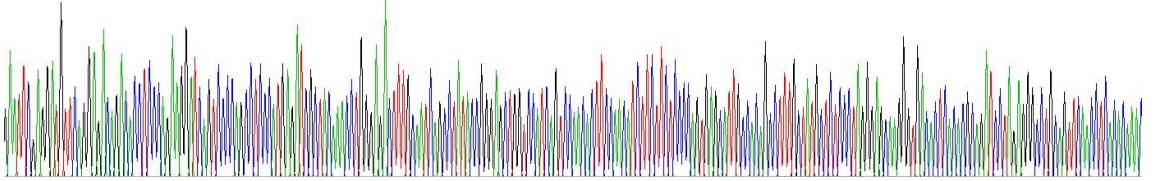


Figure 2. Gene Sequencing (extract)

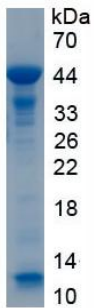


Figure 3. SDS-PAGE

Sample: Active recombinant a1AT, Mouse

[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.