

APC284Mu01 100µg

Active Afamin (AFM)

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: Thr210~Glu403

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 .

Original Concentration: 200µg/mL

Applications: Cell culture; Activity Assays.

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

Predicted isoelectric point: 6.2

Predicted Molecular Mass: 23.5kDa

Accurate Molecular Mass: 23kDa 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]

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T QYLKASSSYQ RNVCGALIKF GPKVLNSINV AVFSKKFKPKI
GFKDLTTLLE DVSSMYEGCC EGDVVHCIRS QSQVNVHICS KQDSISSKIK
VCCEKKTLE EACIINANKD DRPEGLSLRE AKFTESENVQ QERDSDPKF
FAEFIYEYSR RHPDLSTPEL LRITKVYMDF LEDCCSRENP AGCYRHVEDK
FNE
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[ACTIVITY]

Afamin (AFM, also known as Alpha -Albumin), a bioactive protein, is expressed in the liver and secreted into the bloodstream. Human afamin is a 87 kDa protein which is encoded by the fourth member of the human albumin gene family. Mouse AFM shares 66% aa sequence identity with Full-length human AFM. This protein can bind and transport vitamin E across the blood-brain barrier in body fluids under conditions where the lipoprotein system is not sufficient, it also can act as a carrier for hydrophobic molecules in body fluids. Essential for the solubility and activity of lipidated Wnt family members. Thus a functional binding ELISA assay was conducted to detect the interaction of recombinant mouse AFM and recombinant human WNT3A. Briefly, AFM was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100 μ l were then transferred to WNT3A-coated microtiter wells and incubated for 1h at 37°C. Wells were washed with PBST and incubated for 1h with anti-AFM 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 μ L stop solution to the wells and read at 450/630

nm immediately. The binding activity of recombinant mouse AFM and recombinant human WNT3A was shown in Figure 1, the EC50 for this effect is 0.01 ug/mL.

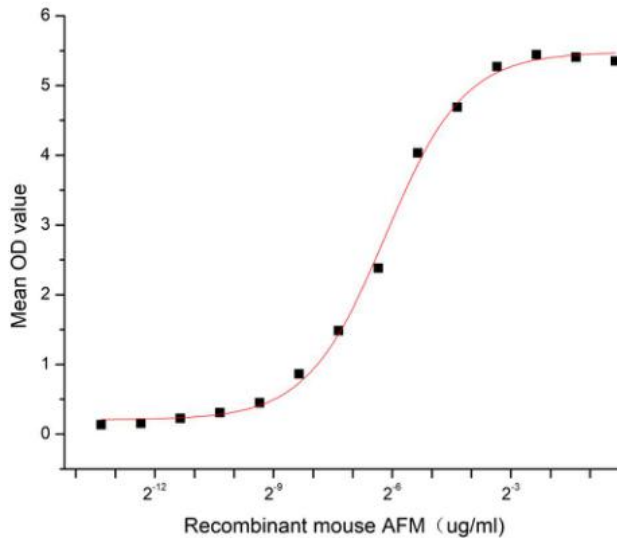


Figure 1. The binding activity of recombinant mouse AFM and recombinant human WNT3A

[IDENTIFICATION]

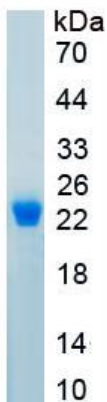


Figure 2. SDS-PAGE

Sample: Active recombinant AFM, 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.