

**APB827Mu01 100µg  
Active Netrin 1 (Ntn1)**

**Organism Species: *Mus musculus (Mouse)*  
Instruction manual**

FOR RESEARCH USE ONLY  
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

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13th Edition (Revised in Aug, 2023)

## **[ PROPERTIES ]**

**Source:** Prokaryotic expression.

**Host:** *E. coli*

**Residues:** Pro313~Pro565

**Tags:** N-terminal His and GST 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:** 9.0

**Predicted Molecular Mass:** 58.5kDa

**Accurate Molecular Mass:** 58kDa 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.



**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 ]

```
PECDRCKP FHYDRPWQRA TAREANECVA CNCNLHARRC  
RFNMELYKLS GRKSGGVCLN CRHNTAGRHC HYCKEGFYRD MGKPITHRKA  
CKACDCHPVG AAGKTCNQTT GQCPCKDGVV GITCNCAKG YQQRSPIAP  
CIKIPVAPPT TAASSVEEPE DCDSYCKASK GKLKMNMKKY CRKDYAVQIH  
ILKADKAGDW WKFTVNIISV YKQGTSRIRR GDQSLWIRSR DIACKCPKIK  
PLKKYLLGN AEDSP
```

## [ ACTIVITY ]

Netrin 1 (Ntn1) is included in a family of laminin-related secreted proteins. Mouse Ntn1 is a 66-76 kDa glycoprotein that is well known for its involvement in axonal guidance during embryonic development and as an enhancer of cancer cell metastasis. Ntn1 is also involved in outgrowth and migration orientation in the developing CNS and plays a significant role in the morphogenesis of endothelial cells and vascular smooth-muscle cells. It is also involved in the processes of cytoskeleton reorganization, angiogenesis, epithelial cell adhesion, and cell migration in the lungs, mammary gland, and pancreas. Slit Homolog 1 (Slit1) can act as a potent promoter of both Netrin-1 attractive and repulsive activities on distinct neuronal cell types, thereby opening novel perspectives on the role of combinations of cues in brain wiring. Thus a functional binding ELISA assay was conducted to detect the interaction of recombinant mouse Ntn1 and recombinant human Slit1. Briefly, Ntn1 was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100  $\mu$ l were then transferred to Slit1-coated microtiter wells

and incubated for 1h at 37 °C. Wells were washed with PBST and incubated for 1h with anti-Ntn1 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 450/630 nm immediately. The binding activity of recombinant mouse Ntn1 and recombinant human Slit1 was shown in Figure 1, the EC50 for this effect is 0.03 ug/mL.

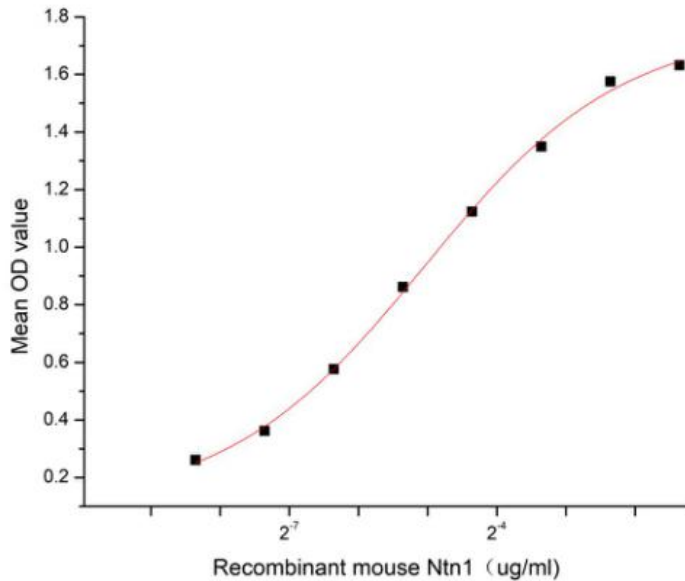


Figure 1. The binding activity of recombinant mouse Ntn1 and recombinant human Slit1

**[ IDENTIFICATION ]**

6 AATTTCCTG AATGGACCGTTTGAGGCCCTTGCATACGACCGCCCTGGCAGCGCCCGCGCCGCGGAGGCGCMCGAGTGCCTGCTGCACTGCAAGCTCCATGCTCGCCGCTGCAGATTCAACATGGAGCTCTATAAGCTATCA GGGCGCAGAGCGGGGAGTCTGTCT

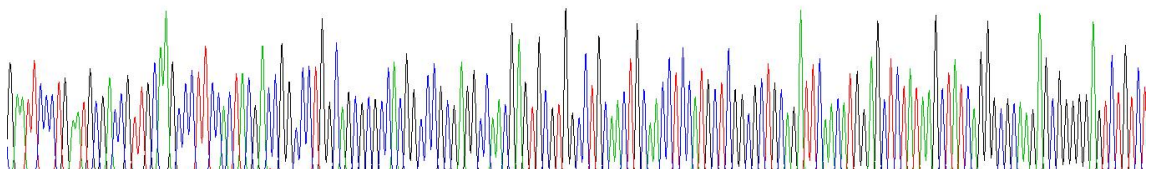
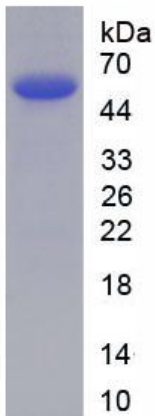


Figure 2. Gene Sequencing (extract)



**Figure 3. SDS-PAGE**

**Sample: Active recombinant Ntn1, 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.