

**APB929Hu01 100µg**

**Active Choline Acetyltransferase (ChAT)**

**Organism Species: *Homo sapiens* (Human)**

***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:** Asp517~Pro732

**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% 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:** 8.4

**Predicted Molecular Mass:** 27.9kDa

**Accurate Molecular Mass:** 28kDa 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.

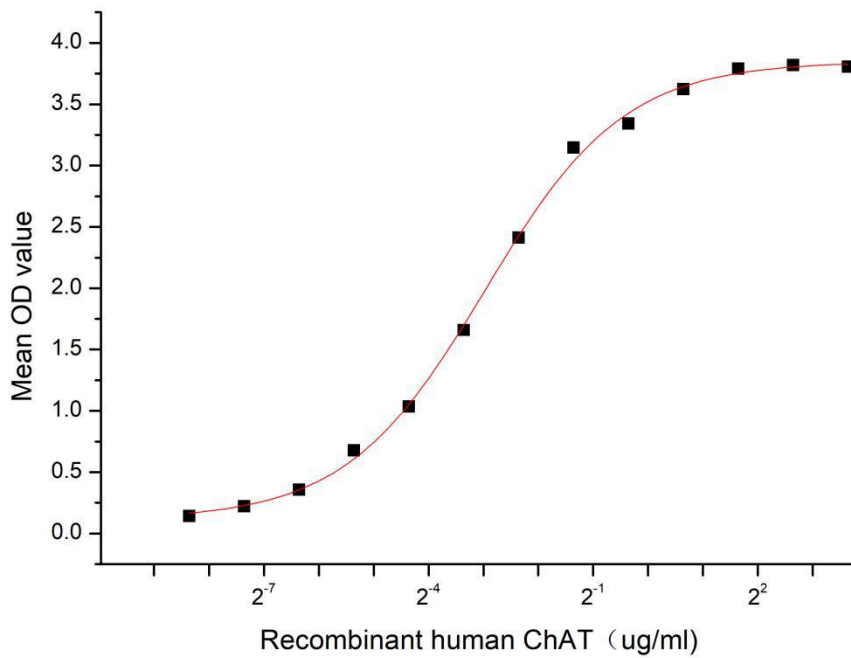
**Stability Test:** The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37°C for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration date under appropriate storage condition.

## **[ SEQUENCE ]**

```
DNYG KTFIKKQKCS PDAFIQVALQ LAFYRLHRRRL
VPTYESASIR RFQEGRDNI RSATPEALAF VRAVTDHKAA VPASEKLLLL
KDAIRAQTAY TVMAITGMAI DNHLLALREL ARAMCKELPE MFMDETYLMS
NRFVLSTSQV PTTTEMFCCY GPVVPNGYGA CYNPQPETIL FCISSFHSCK
ETSSSKFAKA VEESLIDMRD LCSLLPPTES KP
```

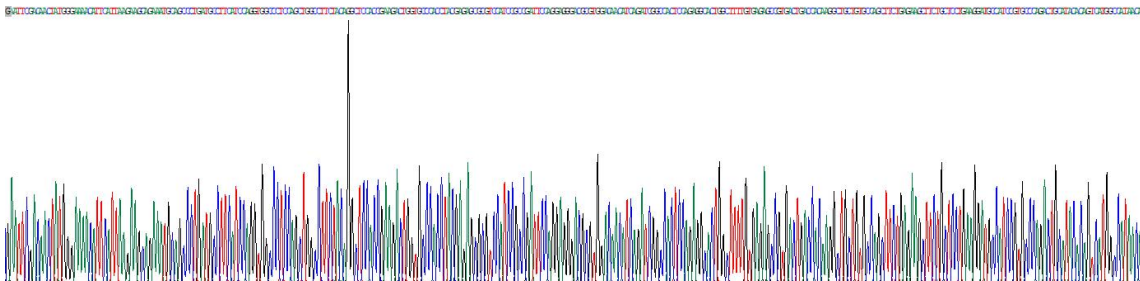
## **[ ACTIVITY ]**

Choline Acetyltransferase (ChAT) is the enzyme responsible for synthesizing acetylcholine (ACh) from acetyl-CoA and choline in presynaptic neurons. It is crucial for cholinergic neurotransmission, influencing cognitive functions, muscle activation, and autonomic processes. ChAT is primarily localized in nerve terminals and is a biomarker for cholinergic neurons. ChAT synthesizes acetylcholine, while AChE breaks it down. They work in tandem to maintain the appropriate level of acetylcholine in the synaptic cleft for proper neural signaling. Thus a functional ELISA assay was conducted to detect the interaction of recombinant human ChAT and recombinant mouse ChAT ACHE. Briefly, ChAT was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100  $\mu$ l were then transferred to ACHE-coated microtiter wells and incubated for 1h at 37 °C . Wells were washed with PBST and incubated for 1h with anti-ChAT 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  $\mu$ L stop solution to the wells and read at 450/630nm immediately. The binding activity of recombinant human ChAT and recombinant mouse ChAT ACHE was shown in Figure 1, the EC<sub>50</sub> for this effect is 0.127ug/mL.

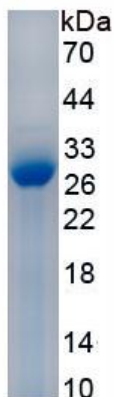


**Figure 1. The binding activity of recombinant human ChAT and recombinant mouse ChAT**

**[ IDENTIFICATION ]**



### Figure 2. Gene Sequencing (extract)



**Figure 3. SDS-PAGE**

**Sample: Active recombinant ChAT, Human**

### **[ 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.