

**APA195Mu01 100µg**

**Active Hexosaminidase A Alpha (HEXa)**

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

***Instruction manual***

FOR IN VITRO USE AND RESEARCH USE ONLY

NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

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1st Edition (Apr, 2016)

## **[ PROPERTIES ]**

**Source:** Prokaryotic expression.

**Host:** *E. coli*

**Residues:** Leu319~Thr528

**Tags:** N-terminal His-tag

**Purity:** >98%

**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:** 6.3

**Predicted Molecular Mass:** 28.2kDa

**Accurate Molecular Mass:** 28kDa 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.

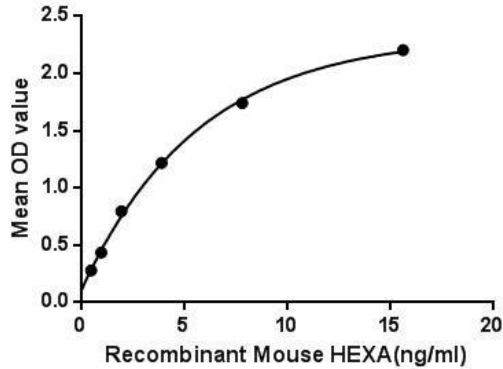
## **[ SEQUENCE ]**

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LG GDEVDFTCWK SNPNIQAFMK KKGFTDFKQL
ESFYIQTL LD IVSDYDKGYV VWQEVFDNKV KVRPDTIIQV WREEMPVEYM
LEMQDITRAG FRALLSAPWY LNRVKYGPDW KDMYKVEPLA FHGTPEQKAL
VIGGEACMWG EYVDSTNLVP RLWPRAGAVA ERLWSSNLTT NIDFAFKRLS
HFRCELVRRG IQAQPISVGY CEQEFEQT
```

## **[ ACTIVITY ]**

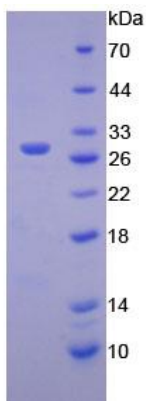
Hexosaminidase A Alpha (HEXa) is a lysosomal enzyme. There are three predominant isoenzymes: hexosaminidase A, B and S. Hexosaminidase A and the cofactor GM2 activator protein catalyze the degradation of the GM2 gangliosides and other molecules containing terminal N-acetyl hexosamines. The enzymes are composed of two alpha and/or beta subunits, which are coded by HEXA and HEXB genes, respectively. Even though the alpha and beta subunits of hexosaminidase A can both cleave GalNAc residues, only the alpha subunit which contains a key residue, Arg-424 is able to hydrolyze GM2 gangliosides. Hexosaminidase A (alpha polypeptide) plays a critical role in the brain and spinal cord (central nervous system). Besides, Hexosaminidase B Beta (HEXB) has been identified as an interactor of HEXA, thus a binding ELISA assay was conducted to detect the interaction of recombinant mouse HEXA and recombinant mouse HEXB. Briefly, HEXA were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100uL were then transferred to HEXB-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-HEXA 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 of HEXA and HEXB was shown in Figure 1, and this effect was in a dose dependent manner.



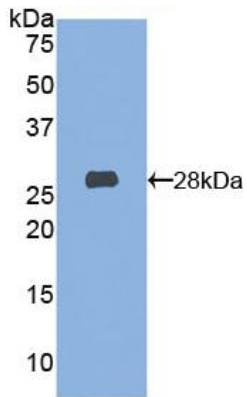
**Figure 1. The binding activity of HEXA with HEXB.**

## [ IDENTIFICATION ]



**Figure 2. SDS-PAGE**

**Sample: Active recombinant HEXa, Mouse**



**Figure 3. Western Blot**

**Sample: Recombinant HEXa, Mouse;**

**Antibody: Rabbit Anti-Mouse HEXa Ab (PAA195Mu01)**