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

1th 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^{o}C for one month.
Aliquot and store at -80^{o}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 37ºC 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]

LG GDEVDFTCWK SNPNIQAFMK KKGFTDFKQL
ESFYIQTLLD 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℃. 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℃. 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.

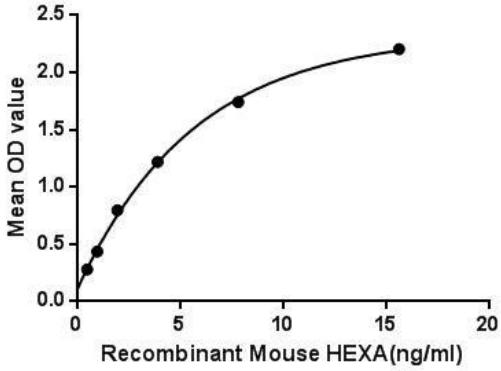

Figure 1. The binding activity of HEXA with HEXB.

[IDENTIFICATION]

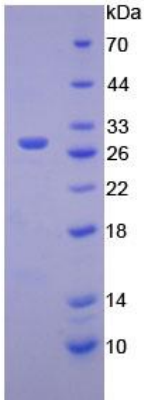

Figure 2. SDS-PAGE

Sample: Active recombinant HEXa, Mouse

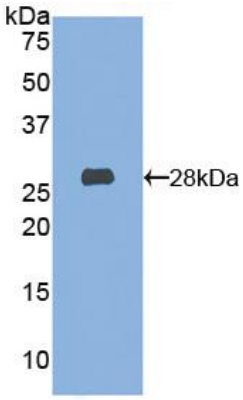

Figure 3. Western Blot

Sample: Recombinant HEXa, Mouse;

Antibody: Rabbit Anti-Mouse HEXa Ab (PAA195Mu01)