APB611Hu01 100μg
Active Neuraminidase (NEU)
Organism Species: Homo sapiens (Human)
Instruction manual

FOR IN VITRO USE AND RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression.
Host: *E. coli*
Residues: Ala141~His286
Tags: N-terminal His-tag
Purity: >92%
Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 0.05% sarcosyl and 5% trehalose.
Applications: Cell culture; Activity Assays; In vivo assays.
(May be suitable for use in other assays to be determined by the end user.)
Predicted isoelectric point: 5.5
Predicted Molecular Mass: 22.6kDa
Accurate Molecular Mass: 26kDa as determined by SDS-PAGE reducing conditions.
Phenomenon explanation:
The possible reasons that the actual band size differs from the predicted are as follows:

1. Splice variants: Alternative splicing may create different sized proteins from the same gene.
2. Relative charge: The composition of amino acids may affects the charge of the protein.
3. Post-translational modification: Phosphorylation, glycosylation, methylation etc.
4. Post-translation cleavage: Many proteins are synthesized as pro-proteins, and then cleaved to give the active form.
5. Polymerization of the target protein: Dimerization, multimerization etc.

[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 2-8ºC for one month.

Aliquot and store at -80ºC 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]

AVVSDVETGV
VFLFYSLCAH KAGCQVASTM LVWSKDDGVS WSTPRNLSLD IGTEVFAPGP
GSGIQKQREP RKGRLIVCGH GTLERDGVFC LLSDDHGASW RYGSGVSGIP
YGQPKQENDF NPDECQPYEL PDGSVVINAR NQNNYH

[ACTIVITY]

NEU (Sialidase-1) is an enzyme that catalyzes the removal of sialic acid (N-acetylneuraminic acid) moities from glycoproteins and glycolipids. In the lysosome, this enzyme is part of a heterotrimeric complex together with beta-galactosidase and cathepsin A (CTSA). Thus a binding ELISA assay was conducted to detect the interaction of recombinant human NEU and recombinant human CTSA. Briefly, NEU were diluted serially in PBS, with 0.01%BSA (pH 7.4). Duplicate samples of 100uL were then transferred to CTSA-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-NEU 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 NEU and CTSA was shown in Figure 1, and this effect was in a dose dependent manner.

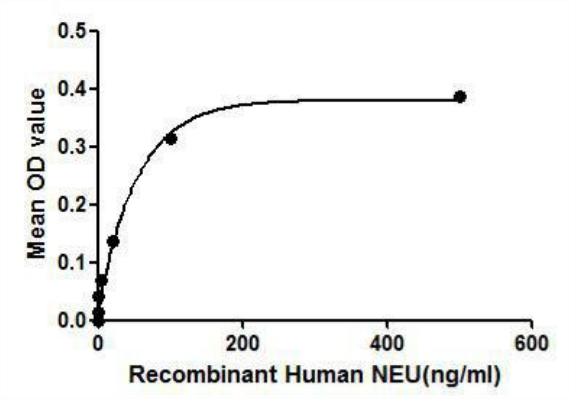

Figure 1. The binding activity of NEU with CTSA.

[IDENTIFICATION]

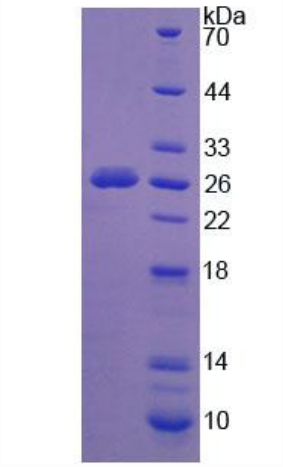

Figure 2. SDS-PAGE

Sample: Active recombinant NEU, Human

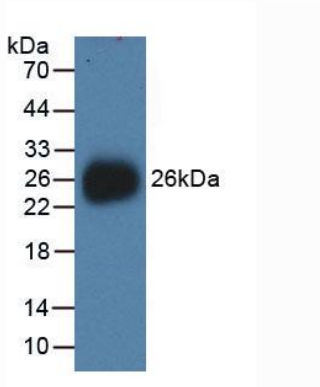

Figure 3. Western Blot

Sample: Recombinant NEU, Human;

Antibody: Rabbit Anti-Human NEU Ab (PAB611Hu01)