



APA181Hu01 200µg

Active Neutrophil Elastase (NE)

Organism Species: Homo sapiens (Human)

Instruction manual

FOR RESEARCH USE ONLY

NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Ile30~Gln247

Tags: Two N-terminal Tags, His-tag and GST-tag

Purity: >80%

Endotoxin Level: <1.0EU per 1µg (determined by the LAL method).

Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 5% Trehalose.

Original Concentration: 50µg/mL

Applications: Activity Assays.

(May be suitable for use in other assays to be determined by the end user.)

Predicted isoelectric point: 8.7

Predicted Molecular Mass: 53.3kDa

Accurate Molecular Mass: 53kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

Reconstitute in ddH₂O to a concentration of 0.1-0.5mg/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]

I VGGRRARPHA WPFMVSLQLR
GGHFCGATLI APNFVMSAAH CVANVNVRAV RVVLGAHNLS RREPTRQVFA
VQRIFENGVD PVNLLNDIVI LQLNGSATIN ANVQVAQLPA QGRRLLGNGVQ
CLAMGWLLG RNRGIASVLQ ELNVTVTSL CRRSNVCTLV RGRQAGVCFG
DSGSPLVCNG LIHGIASFVR GGCASGLYPD AFAPVAQFVN WIDSIIQ

[ACTIVITY]

Neutrophil Elastase (NE), elastase 2 is a serine proteinase in the same family as chymotrypsin and has broad substrate specificity. Secreted by neutrophils and macrophages during inflammation, it destroys bacteria and host tissue. The neutrophil form of elastase is 218 amino acids long, with two asparagine-linked carbohydrate chains (see glycosylation). Neutrophil elastase may play a role in degenerative and inflammatory diseases by its proteolysis of collagen-IV and elastin of the extracellular matrix. This protein degrades the outer membrane protein A (OmpA) of *E. coli* as well as the virulence factors of such bacteria as *Shigella*, *Salmonella* and *Yersinia*. Besides, Collagen Type XVII (COL17) has been identified as an interactor of NE, thus a binding ELISA assay was conducted to detect the interaction of recombinant human COL17 and recombinant human NE. Briefly, NE were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100ul were then transferred to COL17-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-NE 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 NE and COL17 was shown in Figure 1, and this effect was in a dose dependent manner.

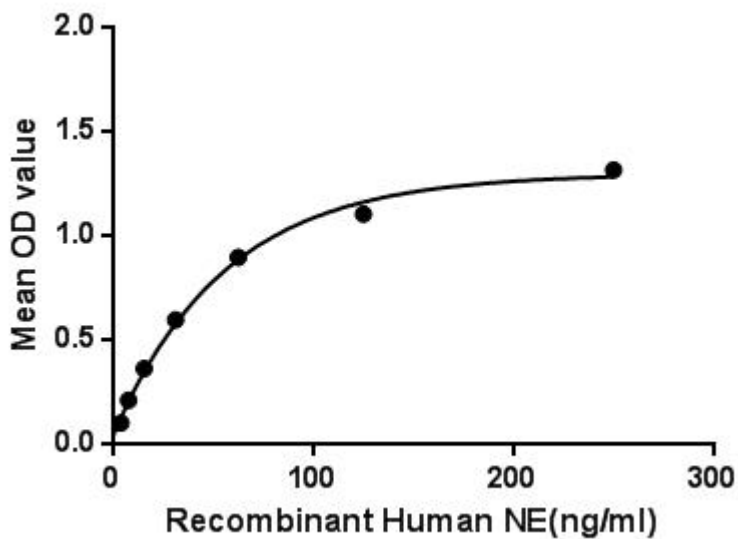


Figure 1. The binding activity of NE with COL17

[IDENTIFICATION]

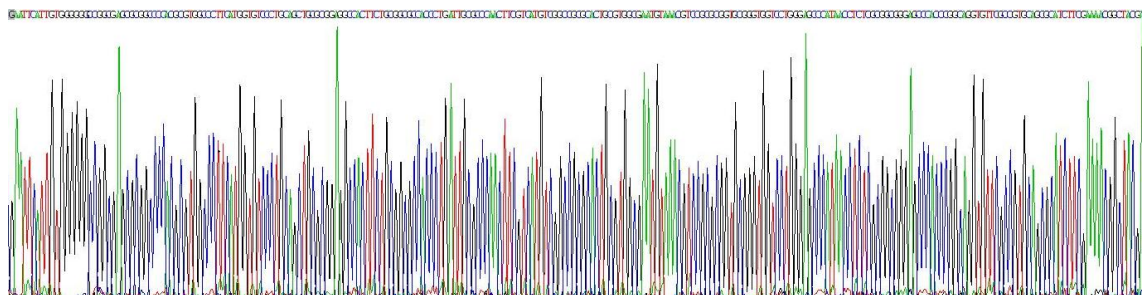


Figure 2. Gene Sequencing (extract)

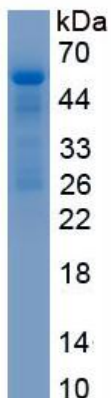


Figure 3. SDS-PAGE

Sample: Active recombinant NE, 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.