APA859Hu03 10μg
Active Mannose Associated Serine Protease 2 (MASP2)
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: Ile445~Ile683
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: 1000μg/mL
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: 5.7
Predicted Molecular Mass: 27.4kDa
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 2-8ºC 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 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]

IYGGQK
AKPGDFPWQV LILGGTTAAG ALLYDNWVLT AAHAVYEQKH DASALDIRMG
TLKRLSPHYT QAWSEAVFIH EGYTHDAGFD NDIALIKLNN KVVINSNITP
ICLPRKEAES FMRTDDIGTA SGWGLTQRGF LARNLMYVDI PIVDHQKCTA
AYEKPPYPRG SVTANMLCAG LESGGKDSCR GDSGGALVFL DSETERWFVG
GIVSWGSMNC GEAGQYGVYT KVINYIPWIE NII

[ACTIVITY]

MASP2 (Mannan-binding lectin serine protease 2) is a serum protease that plays an important role in the activation of the complement system via mannose-binding lectin. The preproprotein of MASP2 is proteolytically processed to generate A and B chains that heterodimerize to form the mature protease, which is able to associate with MBL2. Thus, a functional binding ELISA assay was constructed to detect the association of rhMASP2 with MBL2. Briefly, rhMASP-2 were diluted serially in 10 mM Tris-HCl, 1 M NaCl, 5mM CaCl2, and 0.05% Triton X-100(pH 7.4). Duplicate samples of 100 ul were then transferred to MBL2-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1 h with anti-MASP-2 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 for 15-25 minutes at 37°C. Finally, add 50 μL stop solution to the wells and read at 450 nm immediately. The binding activity of MASP2 with MBL2 was shown in Figure 1 and this effect was in a dose dependent manner.

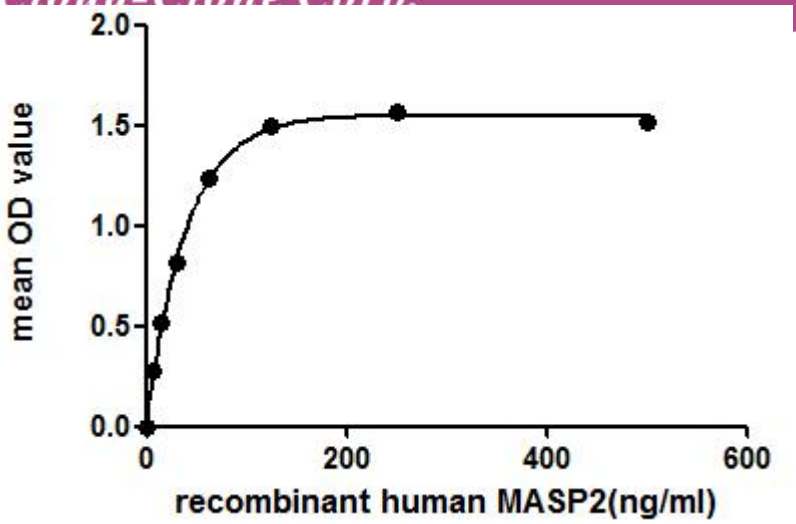

Figure 1. The binding activity of MASP2 with MBL2

The activity of MASP2 was also measured by its ability to cleaves a thioester substrate Z-Lys-SBzl•HCl. The reaction was performed in 0.05 M Tris, pH 8.5 (assay buffer), initiated by addition 50 µL of various concentrations of MASP2 (diluted by assay buffer) to 50 µL of substrate and DTNB mixture (equal volumes mixed by 0.4 mM substrate and 0.4 mM DTNB). The final well serves as a negative control with no MASP2, replaced with 50 µL assay buffer. Incubated at 25℃ for 5min, then read at a wavelength of 405 nm. The specific activity of recombinant human MASP2 is >30 pmol/min/µg.

$$\text{Specific Activity (pmol/min/ug)} = \frac{\text{Adjusted } V_{max}^{*} \text{ (OD/min) x well volume (L) x } 10^{12} \text{ pmol/mol}}{\text{ext. coeff}^{**} \text{ (M}^{-1}\text{cm}^{-1}\text{) x path corr.}^{***} \text{ (cm) x amount of enzyme (ug)}}$$

*Adjusted for Substrate Blank
**Using the extinction coefficient 13260 $M^{-1}cm^{-1}$
***Using the path correction 0.320 cm

[IDENTIFICATION]

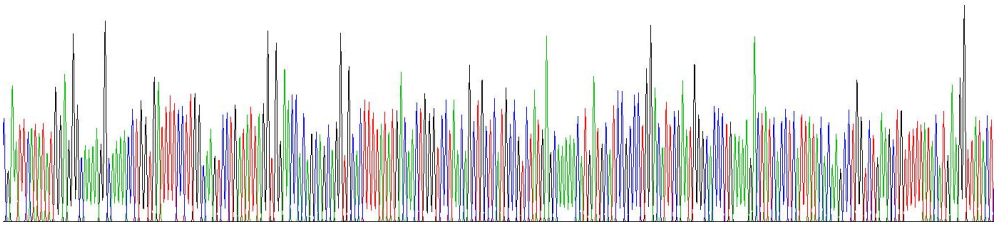

Figure 2. Gene Sequencing (extract)

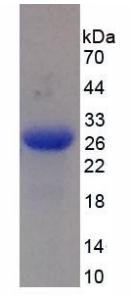

Figure3. SDS-PAGE

Sample: Active recombinant MASP2, 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.