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APB305Hu02 100µg Active Complement C4-B (C4B) 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: Gln19~Leu333 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% SKL, 5%Trehalose . Original Concentration: 200µg/mL Applications: Cell culture; Activity Assays. (May be suitable for use in other assays to be determined by the end user.) Predicted isoelectric point: 9.4 Predicted Molecular Mass: 39.1kDa Accurate Molecular Mass: 39kDa as determined by SDS-PAGE reducing conditions.

[<u>USAGE</u>]

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 2-8°C for one month.

Aliquot and store at -80°C for 12 months.

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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.

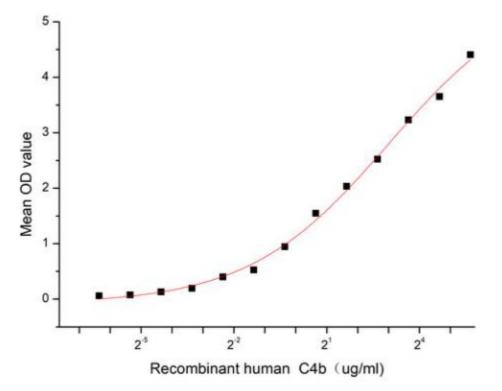
[SEQUENCE]

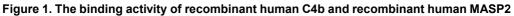
QK PRLLLFSPSV VHLGVPLSVG VQLQDVPRGQ VVKGSVFLRN PSRNNVPCSP KVDFTLSSER DFALLSLQVP LKDAKSCGLH QLLRGPEVQL VAHSPWLKDS LSRTTNIQGI NLLFSSRRGH LFLQTDQPIY NPGQRVRYRV FALDQKMRPS TDTITVMVEN SHGLRVRKKE VYMPSSIFQD DFVIPDISEP GTWKISARFS DGLESNSSTQ FEVKKYVLPN FEVKITPGKP YILTVPGHLD EMQLDIQARY IYGKPVQGVA YVRFGLLDED GKKTFFRGLE SQTKLVNGQS HISLSKAEFQ DALEKLNMGI TDL

[ACTIVITY]

Complement Component 4b (C4b) is a component of the complement system, which is activated by the recognition of foreign pathogens, such as bacteria and viruses, or altered self-cells. In the classical activation pathway of complement, C4b is produced by the proteolytic cleavage of the precursor protein C4 by the activated enzyme C1s, and it can bind to C2a to form C3 invertase (C4b2a) which is responsible for the cleavage of C3. Besides, the binding of MASP2 to C4b is an important step in the lectin pathway of the complement system, thus a functional binding ELISA assay was conducted to detect the interaction of recombinant human C4b and recombinant human MASP2. Briefly, C4b was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100 µ I were then transferred to MASP2-coated microtiter wells and incubated for 1h at 37 °C . Wells were washed with PBST and incubated for 1h with anti-C4b pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody for 1h at $37 \,^{\circ}$ C, wells were aspirated and washed 5 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37 $^\circ$ C. Finally, add 50 μ L stop solution to the wells and read at 450/630 nm immediately. The binding activity of recombinant human C4b and recombinant human MASP2 was shown in Figure 1, Cloud-Clone Corp.

the EC50 for this effect is 1.39 ug/mL.





[IDENTIFICATION]

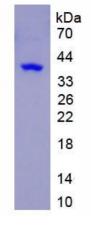


Figure 2. SDS-PAGE

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Sample: Active recombinant C4B, Human

[<u>IMPORTANT NOTE</u>]

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.