APD207Hu01 50µg Active Complement Component 1, Q Subcomponent A (C1qA) Organism Species: *Homo sapiens* (Human) *Instruction manual*

FOR RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression. Host: *E. coli* Residues: Ala28~Ala245 Tags: N-terminal His-tag Purity: >95% Endotoxin Level: <1.0EU per 1µg (determined by the LAL method).

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: 9.6

Predicted Molecular Mass: 26.8kDa

Accurate Molecular Mass: 30kDa 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.

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[<u>USAGE</u>]

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°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.

[<u>SEQUENCE</u>]

APD GKKGEAGRPG RRGRPGLKGE QGEPGAPGIR TGIQGLKGDQ GEPGPSGNPG KVGYPGPSGP LGARGIPGIK GTKGSPGNIK DQPRPAFSAI RRNPPMGGNV VIFDTVITNQ EEPYQNHSGR FVCTVPGYYY FTFQVLSQWE ICLSIVSSSR GQVRRSLGFC DTTNKGLFQV VSGGMVLQLQ QGDQVWVEKD PKKGHIYQGS EADSVFSGFL IFPSA

[ACTIVITY]

The complement component 1q (C1q) is composed of 18 polypeptide chains: six A-chains, six B-chains, and six C-chains. Complement Component 1, Q Subcomponent A (C1qA) is six A-chains of C1q. Complement component 1q (C1q is a protein complex involved in the complement system, which is part of the innate immune system. C1q together with C1r and C1s form the C1 complex. It is potentially multivalent for attachment to the complement fixation sites of immunoglobulin. The sites are on the CH2 domain of IgG and, it is thought, on the CH4 domain of IgM. IgG4 cannot bind C1q, but the other three IgG types can. Besides, C Reactive Protein (CRP) has been identified as an interactor of C1qA,

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thus a binding ELISA assay was conducted to detect the interaction of recombinant human C1qA and recombinant human CRP. Briefly, C1qA were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100µL were then transferred to CRP-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-C1qA 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 C1qA and CRP was shown in Figure 1, and this effect was in a dose dependent manner.



Figure 1. The binding activity of C1qA with CRP.

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[IDENTIFICATION]





Sample: Active recombinant C1qA, Human



Figure 3. Western Blot

Sample: Recombinant C1qA, Human;

Antibody: Rabbit Anti-Human C1qA Ab (PAD207Hu01)

[IMPORTANT NOTE]

The kit is designed for in vitro and research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.