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APA821Ca01 100µg Active C Reactive Protein (CRP) Organism Species: *Canis familiaris; Canine (Dog)* 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: Gln20~Ser223 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: 5.8 Predicted Molecular Mass: 26.7kDa Accurate Molecular Mass: 29kDa 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]

Q IDLDEKAFVF PRESENSYVI LFPQLQKPMK AFTVCLQVYT DLTRPHSLFS YATKSQSNEI LLFKERPGLF SVSVGGSDAF INFPQKFYAP QHFCVTWESV TGLTELWVDG KPMVRASLRR GYTVGSGASI VLGQEQDSFG GGFDKNQSLV GDIEDVNMWD FVLSPSQILT LYTTRALSPN VLNWRNLRYE TRGEVFLKKE LWS

[ACTIVITY]

C reactive protein (CRP) is an annular (ring-shaped), pentameric protein found in blood plasma, whose levels rise in response to inflammation. It is an acute-phase protein of hepatic origin that increases following interleukin-6 secretion by macrophages and T cells. lts physiological role bind is to to lysophosphatidylcholine expressed on the surface of dead or dying cells (and some types of bacteria) in order to activate the complement system via C1q. Besides, Coagulation Factor II (F2) has been identified as an interactor of CRP, thus a functional binding ELISA assay was conducted to detect the interaction of recombinant dog CRP and recombinant rat F2. Briefly, CRP was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100 µI were then transferred to F2-coated microtiter wells and incubated for 1h at 37 °C. Wells were washed with PBST and incubated for 1h with anti-CRP 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 °C. Finally, add 50 µL stop solution to the wells and read at 450/630 nm immediately. The binding activity of recombinant dog CRP and

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recombinant rat F2 was shown in Figure 1, the EC50 for this effect is 0.82 ug/mL.

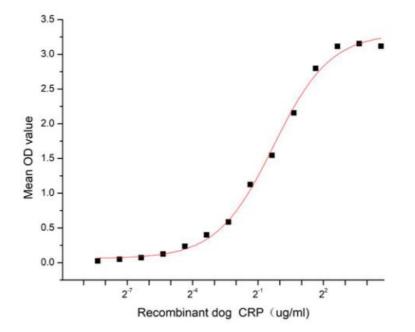


Figure 1. The binding activity of recombinant dog CRP and recombinant rat F2

[IDENTIFICATION]

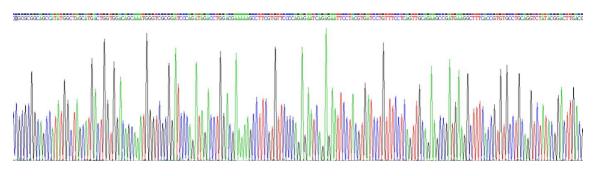


Figure 2. Gene Sequencing (extract)

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kDa 70
44
33
26
22
18
14
10

Figure 3. SDS-PAGE

Sample: Active recombinant CRP, Dog

[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.