APB833Ra01 100μg
Active Complement Factor D (CFD)
Organism Species: *Rattus norvegicus (Rat)*
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: Met1~Ala263
Tags: N-terminal His-tag
Purity: >95%
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: 6.6
Predicted Molecular Mass: 29.7kDa
Accurate Molecular Mass: 27kDa 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 37ºC 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]

MHSSVYLVAL VVLEAAVCVA QPRGRILGGQ EAMAHARPYM ASVQVNGTHV
CGGTLVDEQW VLSAAHCMDG VTKDEVVQVL LGAHSLSSPE PYKHLYDVQS
VVLHPGSRPD SVEDDLMLFK LSHNASLGPH VRPLPLQRED REVKPGTLCD
VAGWGVVTHA GRRPDVLQQL TVSIMDRNTC NLRTYHDGAI TKNMMCAESN
RRDTCRGDSG GPLVCGDAVE AVVTWGSRVC GNRRKPGVFT RVATYVPWIE
NVLSGNVSVN VTA

[ACTIVITY]

Complement Factor D (CFD) is a serine protease that catalyzes the initial proteolytic step in the alternative pathway of complement. Expressed in adipose tissue at high levels, factor D is also known as adipsin. It is an exceptionally specific protease and the only known protein substrate is factor B in complex with C3. Factor D protease activity is regulated by reversible conformational changes, which differs from the majority of serine proteases whose regulation involves either activation by processing of the zymogens or inactivation by binding of the inhibitors. Compared to its physiologically important proteolytic activity, factor D has much lower activity toward synthetic peptide substrates. However, thioester substrates have been routinely used for assessing factor D activity. The full-length (amino acid residues 1-263) of rat CFD was expressed which activity was measured by its ability to cleaves a thioester substrate Z-Lys-SBzl • HCl. The reaction was performed in 50 mM Tris, 1 M NaCl, pH 7.5 (Assay Buffer), initiated by addition 50 μL of various concentrations of CFD (diluted by Assay Buffer) to 50 μl substrate mixture of 0.2mM Z-Lys-SBzl • HCl and 0.2 mM DTNB. The final well serves as a negative control with no CFD, replaced with 50 μl assay buffer.

Then read in kinetic mode for 5 minutes at an absorbance of 405 nm. The specific activity of recombinant rat CFD is > 10000 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]

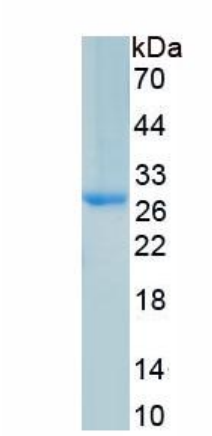

Figure 1. SDS-PAGE

Sample: Active recombinant CFD, Rat

[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.