

APB373Hu01 100µg
Active Defensin Beta 1 (DEFb1)
Organism Species: *Homo sapiens (Human)*
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

12th Edition (Revised in Aug, 2016)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Gly22~Lys68

Tags: N-terminal His and GST Tag

Purity: >95%

Traits: Freeze-dried powder

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: 7.6

Predicted Molecular Mass: 35.1kDa

Accurate Molecular Mass: 35kDa 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 2-8°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.

[SEQUENCE]

GNFLTGLGH RSDHYNCVSS GGQCLYSACP
IFTKIQGTCY RGKAKCKK

[ACTIVITY]

Beta Defensin-1 (DEFB1) displays potent microcidal properties, and also plays a part in other aspects of innate and adaptive immunity. DEFB1 genetic variations have been reported as contributing to hBD1 production impairment, leading to a greater susceptibility to be infected by oral pathogens, also leading to periodontitis. To counteract host immunity, cryptosporidium parvum has evolved multiple strategies to suppress host antimicrobial defense. One such strategy is to reduce the production of the antimicrobial peptide beta-defensin 1 (DEFB1) by host epithelial cells. Beta-Defensin-1, an antimicrobial peptide encoded by the DEFB1 gene, is known to play an important role in lung mucosal immunity. It is reported that DEFb2 has been identified as an interactor of DEFb1, thus a binding ELISA assay was conducted to detect the interaction of recombinant human DEFb1 and recombinant human DEFb2. Briefly, DEFb2 was diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100 µl were then transferred to DEFb1-coated microtiter wells and incubated for 1h at 37°C.

Wells were washed with PBST and incubated for 1h with anti-DEFb2 pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody for 1h at 37 °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 450/630nm immediately. The binding activity of DEFb1 and DEFb2 was shown in Figure 1. When recombinant human DEFb1 is immobilized at 2 ug/mL (100 uL/well), the concentration of rhDEFb2 that produces 50% optimal binding response is found to be approximately 0.11 ug/mL.

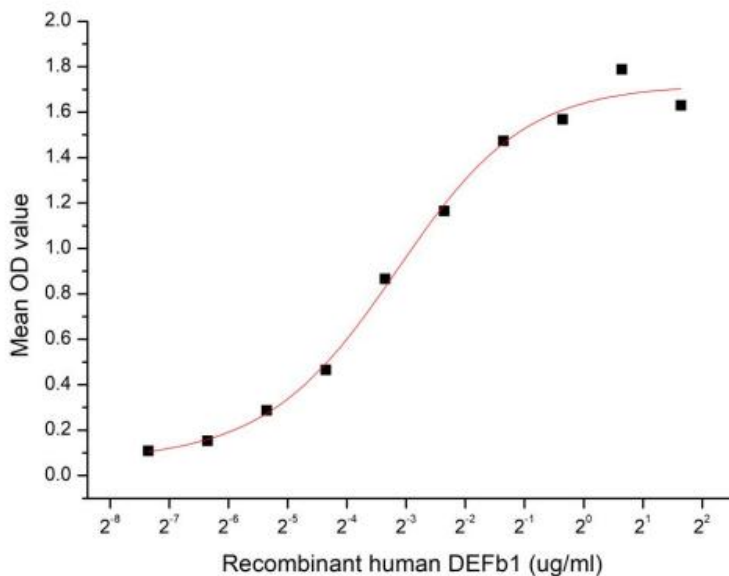


Figure 1. The binding activity of recombinant human DEFb1 with recombinant human DEFb2

[IDENTIFICATION]

GGTAACTTTCTCTCAGGCGCTTGCCAGAGATCTGATCATTAAGATTGCGTCCAGAGTGGAGGCGCAATGTCTCTATTTCTCCCTGCGCGATCTTTACGAAATTCAGGCGACTGTTCAGAGGGGAGGCCAAGTGCCTGCAAG
G N F L T G L G H R S D H Y N C V S S G G Q C L Y S A C P I F T K I Q G T C Y R G K A K C C K *

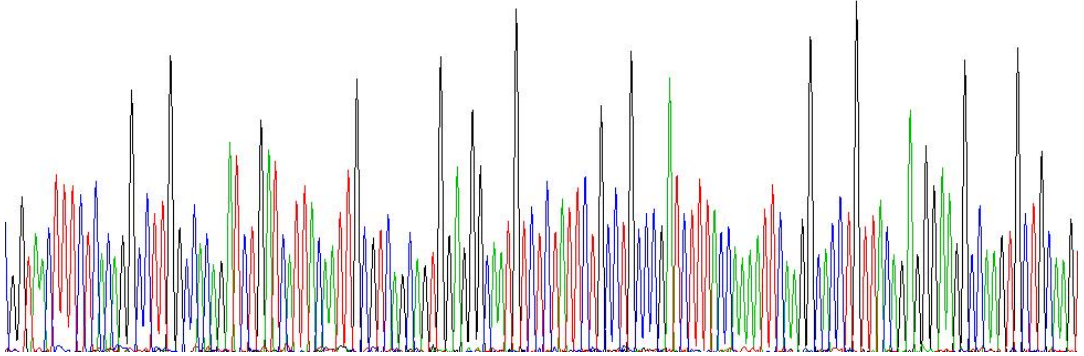


Figure 2. Gene Sequencing (extract)

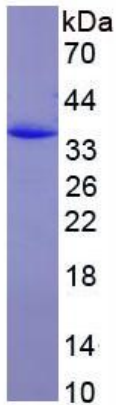


Figure 3. SDS-PAGE

Sample: Active recombinant DEFb1, 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.