APB388Mu61 5μg
Active Neutrophil gelatinase-associated lipocalin (NGAL)
Organism Species: *Mus musculus (Mouse)*
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

12th Edition (Revised in Aug, 2016)

[PROPERTIES]

Source: Eukaryotic expression.
Host: 293F cell
Residues: Gln21~Asn200
Tags: N-terminal His-tag
Purity: >95%
Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).
Buffer Formulation: PBS, pH7.4, containing 5% Trehalose .
Original Concentration: 1000μg/mL
Applications: Cell culture; Activity Assays.
(May be suitable for use in other assays to be determined by the end user.)
Predicted isoelectric point: 9.2
Predicted Molecular Mass: 22.5kDa
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 2-8^{o}C for one month.
Aliquot and store at -80^{o}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]

QDSTQNLIPA PSLLTVPLQP DFRSDQFRGR
WYVVGLAGNA VQKKTEGSFT MYSTIYELQE NNSYNVTSIL VRDQDQGCRY
WIRTFVPSSR AGQFTLGNMH RYPQVQSYNV QVATTDYNQF AMVFFRKTSE
NKQYFKITLY GRTKELSPEL KERFTRFAKS LGLKDDNIIF SVPTDQCIDN

[ACTIVITY]

Lipocalin-2, also known as Neutrophil Gelatinase-Associated Lipocalin (NGAL), was originally identified as a component of neutrophil granules. It is a 25 kDa protein existing in monomeric and homo- and heterodimeric forms, the latter as a dimer with human neutrophil gelatinases (MMP-9). Its expression has been observed in most tissues normally exposed to microorganism, and its synthesis is induced in epithelial cells during inflammation. Lipocalin-2 has been implicated in a variety of processes including cell differentiation, tumorigenesis, and apoptosis. Studies indicate that Lipocalin-2 binds a bacterial catecholate sidropore bound to ferric ion such as enterobactin with a subnanomolar dissociation constant (Kd = 0.41 nM). The bound ferric enterobactin complex breaks down slowly in a month into dihydroxybenzoyl serine and dihydroxybenzoic acid (DHBA). It also binds to a ferric DHBA complex with much less Kd values (7.9 nM). The activity assay of recombinant mouse NGAL was measured by its ability to bind Iron(III) dihydroxybenzoic acid Fe(DHBA)3. The binding of Fe(DHBA)3 results in the quenching of Trp fluorescence in Lipocalin-2. The recombinant mouse NGAL was diluted to 100 ug/ml in assay buffer (50 mM Tris, 10 mM CaCl2, 150 mM NaCl, pH 7.5). 50 µL of the different concentrations of Fe(DHBA)3 and 50 µL of 100 ug/ml rmNGAL was loaded into the plate.

Incubated at room temperature for 30 minutes, then read at excitation and emission wavelengths of 280 nM and 340 nM in endpoint mode. The result was shown in Figure 1, the recombinant mouse Lipocalin-2 can bind >0.5 μM of Fe(DHBA)3.

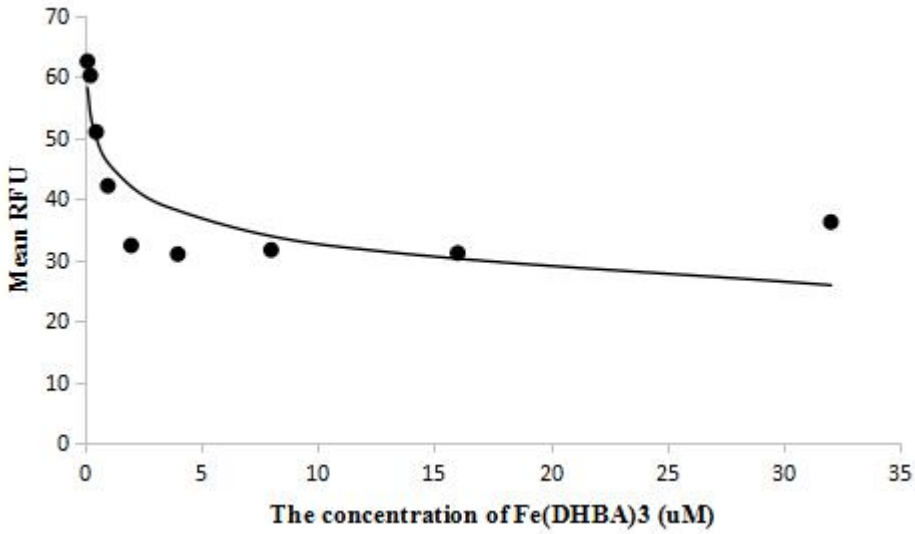

Figure 1. The binding activity of NGAL with $Fe(DHBA)_3$

[IDENTIFICATION]

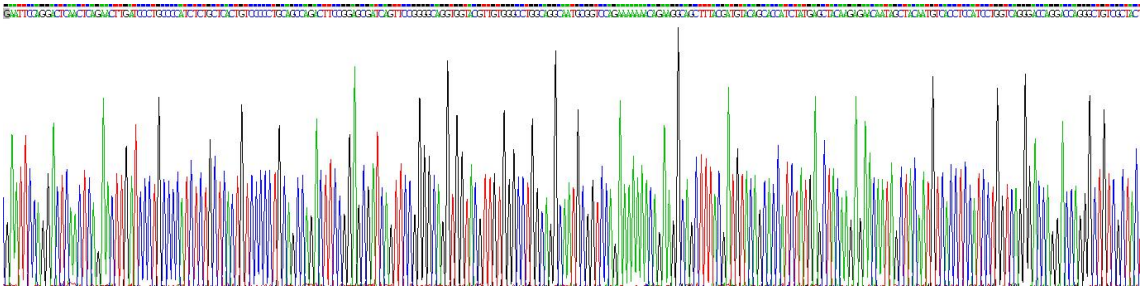

Figure 2. Gene Sequencing (extract)

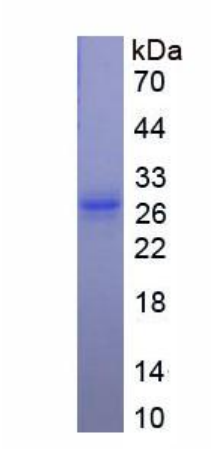

Figure 3. SDS-PAGE

Sample: Active recombinant NGAL, Mouse

[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.