APH757Ra01 100μg
Active Klotho (KL)
Organism Species: *Rattus norvegicus (Rat)*
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: Leu517~Leu956
Tags: N-terminal His-tag
Purity: >94%
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: 6.4
Predicted Molecular Mass: 54.8kDa
Accurate Molecular Mass: 55kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

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 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]

LEGT FPCDFAWGVV DNYIQVDPTL SQFTDPNVYL
WDVHHSKRLI KVDGVVAKKR KPYCVDFSAI RPQITLLREM RVTHFRFSLD
WALILPLGNQ TQVNRTVLHF YRCMVSELVH ANITPVVALW QPATPHQGLP
HALAKHGAWE NPHTALAFAD YANLCFEELG HWVKFWITIN EPNSRNMTYR
AGHHLLKAHA LAWHLYDDKF RAAQKGKISI ALQVDWIEPA CPFSQKDKEV
AERVLEFDVG WLAEPIFGSG DYPHVMREWL NQKNNFLLPY FTEDEKKLIR
GSFDFLALSH YTTILVDWEK EDPIKYNDYL EVQEMTDITW LNSPNQVAVV
PWGLRKALNW LRFKYGDLPM FVTANGIDDD PHAEQDSLRM YYIKNYVNEA
LKAYVLDGIN LCGYFAYSLS DRSVPKSGFY RYAANQFEPK PSIKHYRKII
DNNGFL

[ACTIVITY]

Klotho (KL) is a transmembrane protein that, in addition to other effects, provides some control over the sensitivity of the organism to insulin and appears to be involved in aging. The Klotho protein is a novel β-glucuronidase capable of hydrolyzing steroid β-glucuronides. Genetic variants in KLOTHO have been associated with human aging, and Klotho protein has been shown to be a circulating factor detectable in serum that declines with age. The binding of certain fibroblast growth factors (FGF's) viz., FGF19, FGF20, and FGF23, to their fibroblast growth factor receptors, is promoted via their interactions as co-receptors with β-Klotho. Besides, Fibroblast Growth Factor 23 (FGF23) has been identified as an interactor of KL, thus a binding ELISA assay was conducted to detect the interaction of recombinant rat KL and recombinant rat FGF23. Briefly, KL were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of

100μL were then transferred to FGF23-coated microtiter wells and incubated for 2h at 37℃. Wells were washed with PBST and incubated for 1h with anti-KL 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℃. Finally, add 50μL stop solution to the wells and read at 450nm immediately. The binding activity of KL and FGF23 was shown in Figure 1, and this effect was in a dose dependent manner.

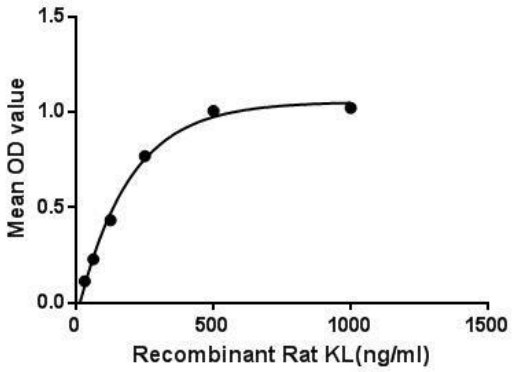

Figure 1. The binding activity of KL with FGF23.

[IDENTIFICATION]

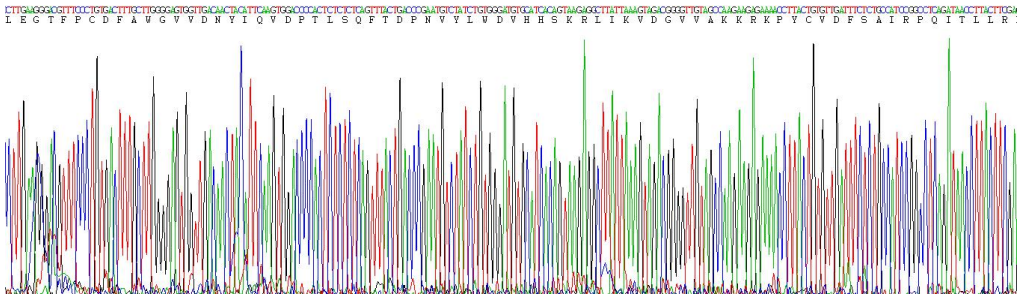

Figure 2. Gene Sequencing (extract)

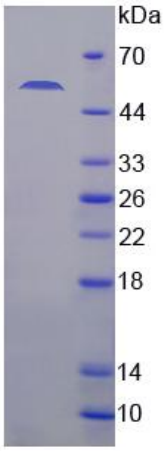

Figure 3. SDS-PAGE

Sample: Active recombinant Klotho, Rat

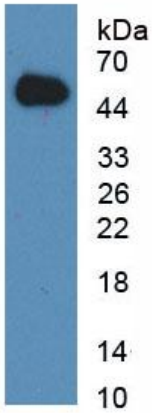

Figure 4. Western Blot

Sample: Recombinant Klotho, Rat;

Antibody: Rabbit Anti-Rat Klotho Ab (PAH757Ra01)

[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.