APE913Mu01 100μg
Active Sirtuin 3 (SIRT3)
Organism Species: Mus musculus (Mouse)
Instruction manual

FOR IN VITRO USE AND RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression.
Host: *E. coli*
Residues: Gly5~Arg257
Tags: N-terminal His-tag
Purity: >92%
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: 32.2kDa
Accurate Molecular Mass: 16&29kDa as determined by SDS-PAGE reducing conditions.
Phenomenon explanation:
The possible reasons that the actual band size differs from the predicted are as follows:

1. Splice variants: Alternative splicing may create different sized proteins from the same gene.
2. Relative charge: The composition of amino acids may affects the charge of the protein.
3. Post-translational modification: Phosphorylation, glycosylation, methylation etc.
4. Post-translation cleavage: Many proteins are synthesized as pro-proteins, and then cleaved to give the active form.
5. Polymerization of the target protein: Dimerization, multimerization etc.

[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 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]

GISTPS GIPDFRSPGS GLYSNLQQYD IPYPEAIFEL GFFFHNPKPF FMLAKELYPG HYRPNVTHYF LRLLHDKELL LRLYTQNIDG LERASGIPAS KLVEAHGTFV TATCTVCRRS FPGEDIWADV MADRVPRCPV CTGVVKPDIV FFGEQLPARF LLHMADFALA DLLLILGTSL EVEPFASLSE AVQKSVPRLL INRDLVGPFV LSPRRKDVVQ LGDVVHGVER LVDLLGWTQE LLDLMQRERG KLDGQDR

[ACTIVITY]

Sirtuin 3 (SIRT3) also known as NAD-dependent deacetylase sirtuin-3 is a member of the mammalian sirtuin family of proteins. SIRT3 is a soluble protein located in the mitochondrial matrix, and contains a mitochondrial processing peptide at the N-terminus. It is a key regulator of succinate dehydrogenase (SDH), which catalyzes the oxidation of succinate to fumarate. Increased succinate concentrations and the specific G protein-coupled receptor 91 (GPR91) are involved in the activation of hepatic stellate cells (HSCs). SIRT3 agonists could help treat metabolic disorders such as obesity, metabolic syndrome and type 2

diabetes. Besides, Dihydrolipoyl Transacetylase (DLAT) has been identified as an interactor of SIRT3, thus a binding ELISA assay was conducted to detect the interaction of recombinant mouse SIRT3 and recombinant mouse DLAT. Briefly, SIRT3 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100uL were then transferred to DLAT-coated microtiter wells and incubated for 2h at 37℃. Wells were washed with PBST and incubated for 1h with anti-SITR3 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 SIRT3 and DLAT was shown in Figure 1, and this effect was in a dose dependent manner.

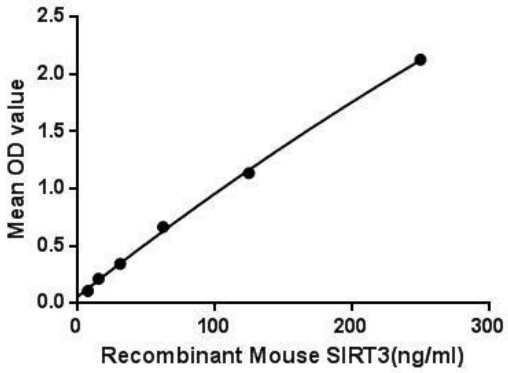

Figure 1. The binding activity of SIRT3 with DLAT.

[IDENTIFICATION]

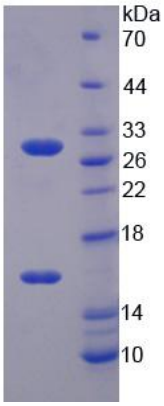

Figure 2. SDS-PAGE

Sample: Active recombinant SIRT3, Mouse

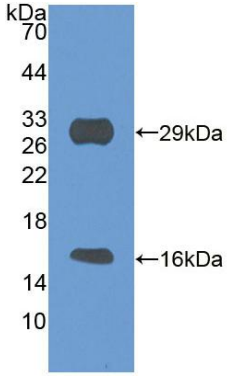

Figure 3. Western Blot

Sample: Recombinant SIRT3, Mouse;

Antibody: Rabbit Anti-Mouse SIRT3 Ab (PAE913Mu01)

[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.