Anti-PPP3CB Antibody [PSH0-21] - BSA and Azide free

HA750595

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, IF-Cell, FC
Molecular Wt:	Predicted band size: 59 kDa
Clone number:	PSH0-21

Description:	Calcium-dependent, calmodulin-stimulated protein phosphatase which plays an essential role in the transduction of intracellular Ca2+-mediated signals. Dephosphorylates and activates transcription factor NFATC1. Dephosphorylates and inactivates transcription factor ELK1.Dephosphorylates DARPP32. Negatively regulates MAP3K14/NIK signaling via inhibition of nuclear translocation of the transcription factors RELA and RELB. May play a role in skeletal muscle fiber type specification. Forms a complex composed of a calmodulin-dependent catalytic subunit (also known as calcineurin A) and a regulatory Ca2+-binding subunit (also known as calcineurin B). There are three catalytic subunits, each encoded by a separate gene (PPP3CA, PPP3CB, and PPP3CC) and two regulatory subunits which are also encoded by separate genes (PPP3R1 and PPP3R2). In response to an increase in Ca2+ intracellular levels, forms a complex composed of PPP3CB/calcineurin A, calcineurin B and calmodulin. Interacts (via calcineurin B binding domain) with regulatory subunit PPP3R1/calcineurin B. Interacts (via calmodulin-binding domain) with calmodulin; the interaction depends on calmodulin binding to Ca2+. Interacts with SLC12A1.
Immunogen:	Synthetic peptide within Human Serine/threonine-protein phosphatase 2B catalytic subunit beta isoform 265-318/524
Positive control:	Mouse brain tissue lysate, Jurkat cell lysate, rat brain tissue lysate, rat skeletal muscle tissue, mouse skeletal muscle tissue, MCF-7.
Subcellular location:	Cytoplasm
Database links:	SwissProt: P16298 Human | P48453 Mouse | P20651 Rat
Recommended Dilutions:	
WB	1:1,000
IHC-P	1:200-1:1,000
IF-Cell	1:200
FC	1:1000
Storage Buffer:	1*PBS (pH7.4).
Storage Instruction:	Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.
Purity:	Immunogen affinity purified.

Images

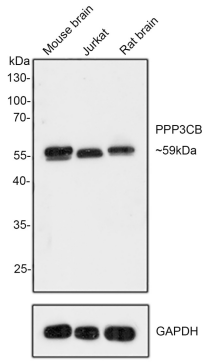

Fig1: Western blot analysis of PPP3CB on different lysates with Rabbit anti-PPP3CB antibody (HA750595) at 1/1,000 dilution.

Lane 1: Mouse brain tissue lysate
Lane 2: Jurkat cell lysate
Lane 3: Rat brain tissue lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 59 kDa
Observed band size: 59 kDa

Exposure time: 2 minutes;

10% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM for 1 hour at room temperature. The primary antibody (HA750595) at 1/1,000 dilution was used in 5% NFDM at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.

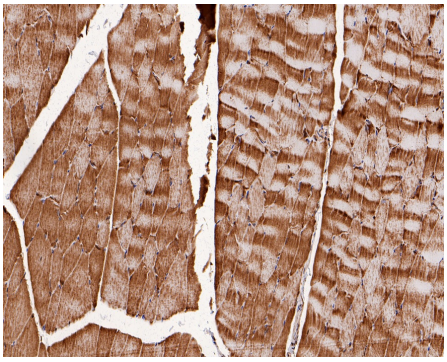

Fig2: Immunohistochemical analysis of paraffin-embedded rat skeletal muscle tissue with Rabbit anti-PPP3CB antibody (HA750595) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750595) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

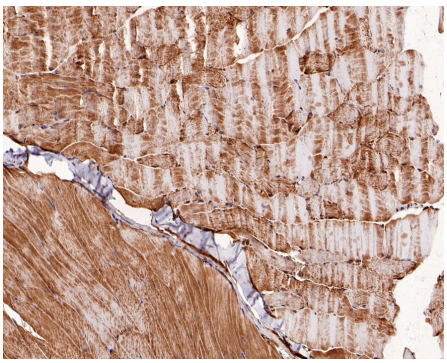

Fig3: Immunohistochemical analysis of paraffin-embedded mouse skeletal muscle tissue with Rabbit anti-PPP3CB antibody (HA750595) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750595) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

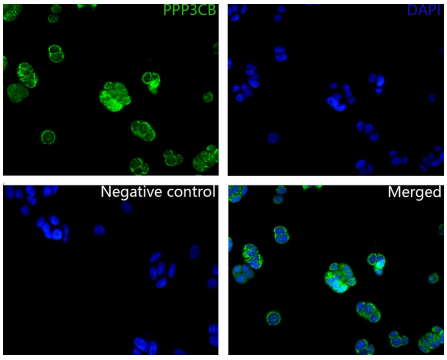

Fig4: Immunocytochemistry analysis of MCF-7 cells labeling PPP3CB with Rabbit anti-PPP3CB antibody (HA750595) at 1/200 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 ℃, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-PPP3CB antibody (HA750595) at 1/200 dilution in 2% negative goat serum overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

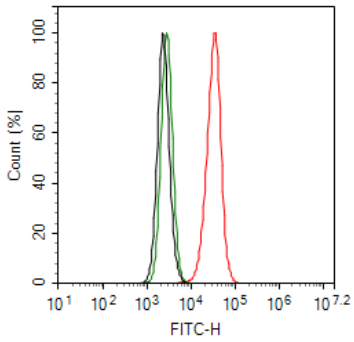

Fig5: Flow cytometric analysis of MCF-7 cells labeling PPP3CB.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA750595, 1ug/ml) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Background References

1. Susann Kilka. et al. 2009. The proline-rich N-terminal sequence of calcineurin Abeta determines substrate binding. Biochemistry. 48(9):1900-10.
2. Sheng Jie Li. et al. 2016. Cooperative autoinhibition and multi-level activation mechanisms of calcineurin. Cell Res 26(3):336-49.