Anti-CaMKII Antibody [SU03-57]

ET1608-47

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat, Cynomolgus monkey, Pig
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IHC-Fr, FC
Molecular Wt:	Predicted band size: 54 kDa
Clone number:	SU03-57

Description: Ca2+/calmodulin-dependent protein kinase II (CaM kinase II or CaMKII) is a serine/threonine-specific protein kinase that is regulated by the Ca2+/calmodulin complex. CaMKII is involved in many signaling cascades and is thought to be an important mediator of learning and memory. CaMKII is also necessary for Ca2+ homeostasis and reuptake in cardiomyocytes, chloride transport in epithelia, positive T-cell selection, and CD8 T-cell activation. Misregulation of CaMKII is linked to Alzheimer's disease, Angelman syndrome, and heart arrhythmia.

Immunogen: Synthetic peptide within Human CaMKⅡ aa 201-250 / 478.

Positive control: SHG-44 cell lysate, PC-12 cell lysate, Neuro-2a cell lysate, PC-12, rat brain tissue, rat cerebellum tissue, mouse brain tissue, mouse cerebellum tissue.

Subcellular location: Cytoplasm, Sarcoplasmic reticulum membrane, Cell membrane, Cell junction.

Database links: SwissProt: Q13554 Human | Q13555 Human | Q13557 Human | Q9UQM7 Human | P11798 Mouse | P28652 Mouse | Q6PHZ2 Mouse | Q923T9 Mouse | P08413 Rat | P11275 Rat | P11730 Rat | P15791 Rat

Recommended Dilutions:

WB	1:500-1:2,000
IF-Cell	1:100
IF-Tissue	1:50-1:200
IHC-P	1:50-1:200
IHC-Fr	1:100
FC	1:1,000

Storage Buffer: 1*PBS (pH7.4), 0.1% BSA, 40% Glycerol, 0.2% Proclean 950.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). Store at -20℃ long term.

Purity: Protein A affinity purified.

Images

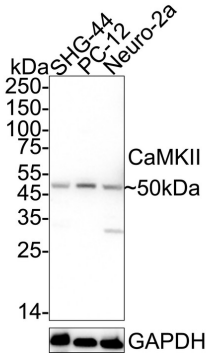

Fig1: Western blot analysis of CaMKII on different lysates with Rabbit anti-CaMKII antibody (ET1608-47) at 1/1,000 dilution.

Lane 1: SHG-44 cell lysate
Lane 2: PC-12 cell lysate
Lane 3: Neuro-2a cell lysate

Lysates/proteins at 30 µg/Lane.

Predicted band size: 54 kDa
Observed band size: 50 kDa

Exposure time: 14 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1608-47) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

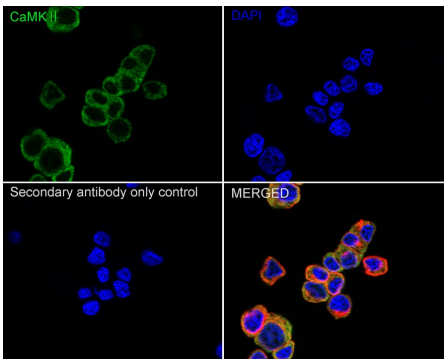

Fig2: Immunocytochemistry analysis of PC-12 cells labeling CaMKII with Rabbit anti-CaMKII antibody (ET1608-47) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-CaMKII antibody (ET1608-47) at 1/100 dilution in 1% BSA in PBST 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.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

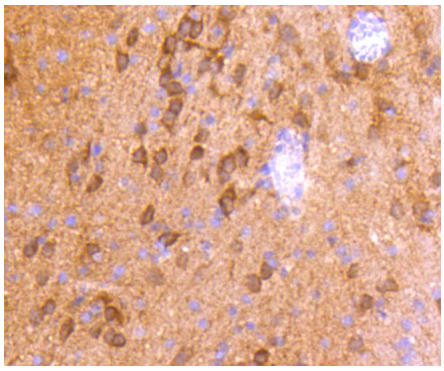

Fig3: Immunohistochemical analysis of paraffin-embedded rat brain tissue using anti-CaMKII antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1608-47, 1/50) for 30 minutes 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.

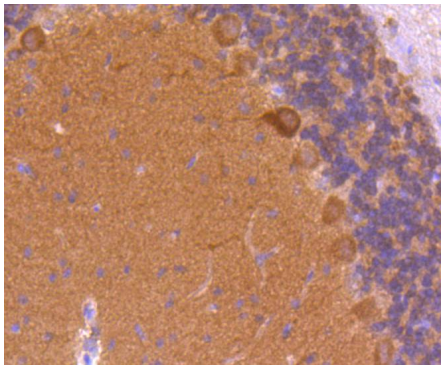

Fig4: Immunohistochemical analysis of paraffin-embedded rat cerebellum tissue using anti-CaMKII antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1608-47, 1/50) for 30 minutes 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.

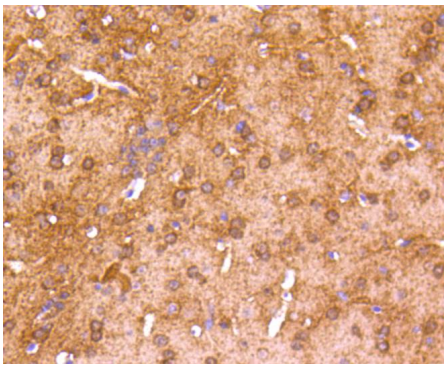

Fig5: Immunohistochemical analysis of paraffin-embedded mouse brain tissue using anti-CaMKII antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1608-47, 1/50) for 30 minutes 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.

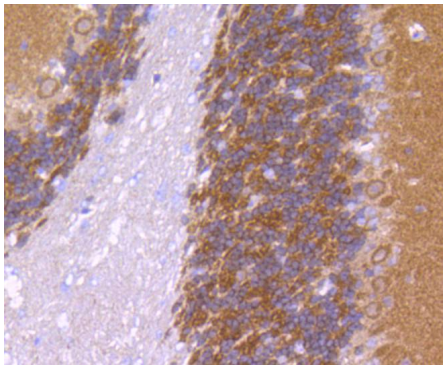

Fig6: Immunohistochemical analysis of paraffin-embedded mouse cerebellum tissue using anti-CaMKII antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1608-47, 1/50) for 30 minutes 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.

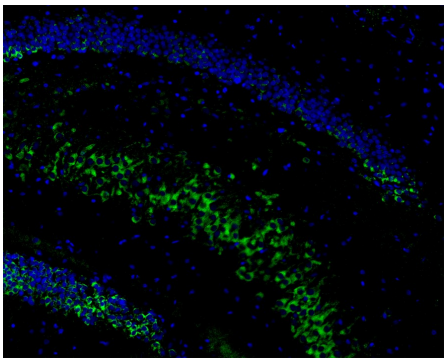

Fig7: Immunofluorescence analysis of frozen mouse hippocampus tissue labeling CaMKⅡ with Rabbit anti-CaMKⅡ antibody (ET1608-47).

The tissues were blocked in 3% BSA for 30 minutes at room temperature, washed with PBS, and then probed with the primary antibody ((ET1608-47, green) at 1/100 dilution overnight at 4℃, washed with PBS. Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) was used as the secondary antibody at 1/200 dilution. Nuclei were counterstained with DAPI (blue). Image acquisition was performed with KFBIO KF-FL-400 Scanner.

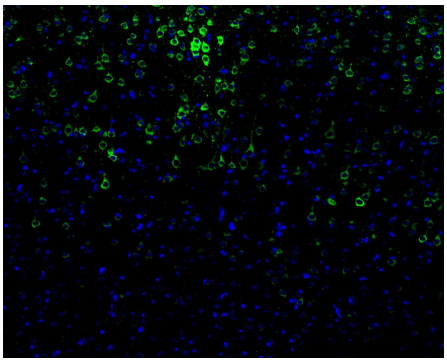

Fig8: Immunofluorescence analysis of frozen mouse cerebral cortex tissue labeling CaMKⅡ with Rabbit anti-CaMKⅡ antibody (ET1608-47).

The tissues were blocked in 3% BSA for 30 minutes at room temperature, washed with PBS, and then probed with the primary antibody ((ET1608-47, green) at 1/100 dilution overnight at 4℃, washed with PBS. Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) was used as the secondary antibody at 1/200 dilution. Nuclei were counterstained with DAPI (blue). Image acquisition was performed with KFBIO KF-FL-400 Scanner.

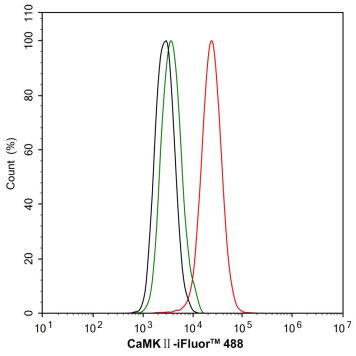

Fig9: Flow cytometric analysis of PC-12 cells labeling CaMKII.

Cells were fixed and permeabilized. Then stained with the primary antibody (ET1608-47, 1/1,000) (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).

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Wang Y et al. EPHB4 Protein Expression in Vascular Smooth Muscle Cells Regulates Their Contractility, and EPHB4 Deletion Leads to Hypotension in Mice. J Biol Chem 290:14235-44 (2015).
2. Cook-Snyder DR et al. A retrograde adeno-associated virus for collecting ribosome-bound mRNA from anatomically defined projection neurons. Front Mol Neurosci 8:56 (2015).