Anti-CaMKII alpha Antibody [JM11-07]

ET1703-54

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IP, FC, IHC-P, IF-Tissue, IF-Cell, IHC-Fr
Molecular Wt:	Predicted band size: 54 kDa
Clone number:	JM11-07

Description: The Ca2+/calmodulin-dependent protein kinases (CaM kinases) comprise a structurally related subfamily of serine/threonine kinases which include CaMKI, CaMKII and CaMKIV. CaMKII is a ubiquitously expressed serine/threonine protein kinase that is activated by Ca2+and calmodulin (CaM) and has been implicated in regulation of the cell cycle and transcription. There are four CaMKII isozymes designated α, β, γ and δ, which may or may not be co-expressed in the same tissue type. CaMKIV is stimulated by Ca2+ and CaM but also requires phosphorylation by a CaMK for full activation. Stimulation of the T cell receptor CD3 signaling complex with an anti-CD3 monoclonal antibody leads to a 10-40 fold increase in CaMKIV activity. An additional kinase, CaMKK, functions to activate CaMKI through the specific phosphorylation of the regulatory Threonine residue at position 177.

Immunogen: Synthetic peptide within Human CaMKII alpha aa 37-71 / 478.

Positive control: Mouse brain tissue, rat brain tissue lysates, rat testis tissue, mouse cerebellum tissue, rat brain tissue, Neuro-2a.

Subcellular location: Cell junction, Cell projection, Synapse.

Database links: SwissProt: Q9UQM7 Human | P11798 Mouse | P11275 Rat

Recommended Dilutions:

WB	1:1,000-1:2,000
IHC-P	1:50-1:200
FC	1:50-1:100
IP	1:10-1:50
IF-Tissue	1:50
IF-Cell	1:100
IHC-Fr	1:200

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

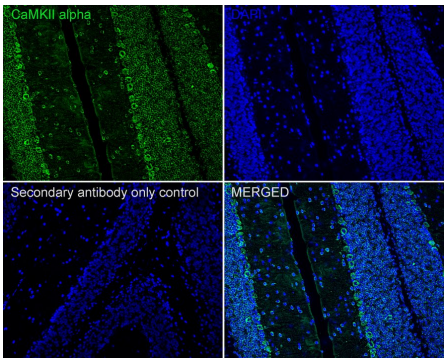

Fig1: Immunofluorescence analysis of frozen mouse cerebellum tissue with Rabbit anti-CaMKII alpha antibody (ET1703-54) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (ET1703-54, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

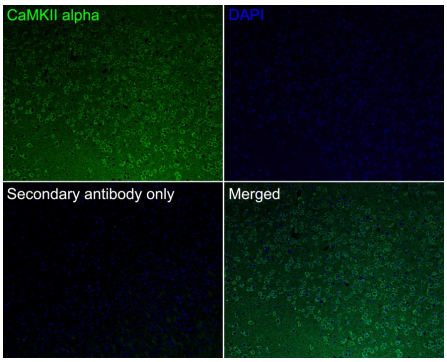

Fig2: Immunofluorescence analysis of paraffin-embedded mouse brain tissue labeling CaMKII alpha with Rabbit anti-CaMKII alpha antibody (ET1703-54) at 1/50 dilution.

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 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (ET1703-54, green) at 1/50 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

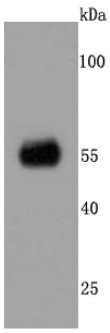

Fig3: Western blot analysis of CaMKII alpha on rat brain tissue lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (ET1703-54, 1/500) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:200,000 dilution was used for 1 hour at room temperature.

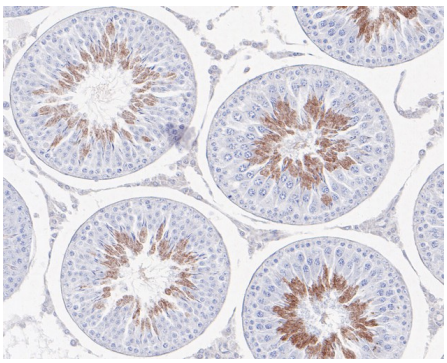

Fig4: Immunohistochemical analysis of paraffin-embedded rat testis tissue using anti-CaMKII alpha antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1703-54, 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.

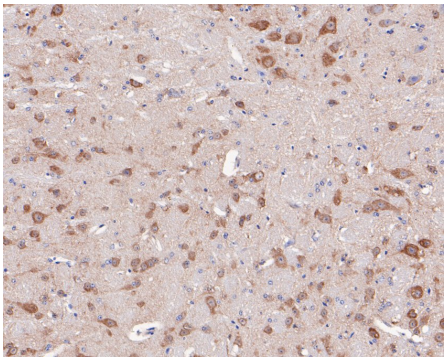

Fig5: Immunohistochemical analysis of paraffin-embedded mouse brain tissue with Rabbit anti-CaMKII alpha antibody (ET1703-54) at 1/200 dilution.

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 (ET1703-54) 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.

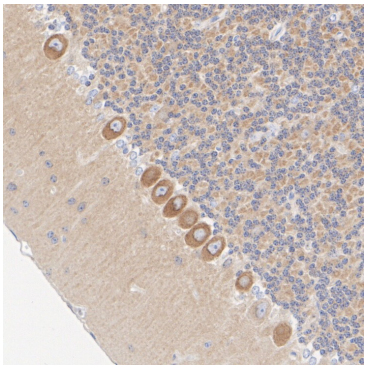

Fig6: Immunohistochemical analysis of paraffin-embedded mouse cerebellum tissue with Rabbit anti-CaMKII alpha antibody (ET1703-54) at 1/200 dilution.

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 (ET1703-54) 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.

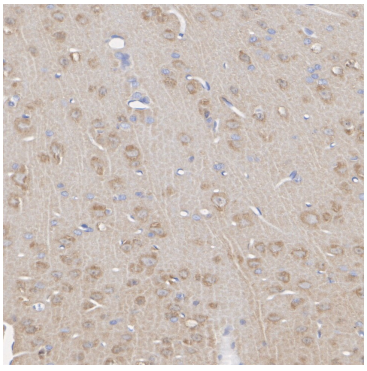

Fig7: Immunohistochemical analysis of paraffin-embedded rat brain tissue with Rabbit anti-CaMKII alpha antibody (ET1703-54) at 1/200 dilution.

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 (ET1703-54) 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.

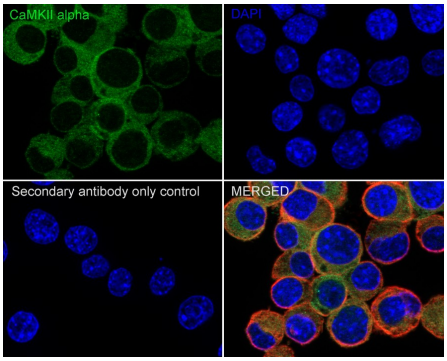

Fig8: Immunocytochemistry analysis of Neuro-2a cells labeling CaMKII alpha with Rabbit anti-CaMKII alpha antibody (ET1703-54) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 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 alpha antibody (ET1703-54) 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.

Background References

1. Thein S et al. CaMKII mediates recruitment and activation of the deubiquitinase CYLD at the postsynaptic density. PLoS One 9:e91312 (2014).
2. Dumoulin MC et al. Extracellular Signal-Regulated Kinase (ERK) Activity During Sleep Consolidates Cortical Plasticity In Vivo. Cereb Cortex N/A:N/A (2013).