Anti-Phospho-alpha Synuclein (S129) Antibody [JB22-44]

ET7107-30

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC
Molecular Wt:	Predicted band size: 14 kDa
Clone number:	JB22-44

Description: Alpha-synuclein is a protein that, in humans, is encoded by the SNCA gene. Alpha-synuclein is a neuronal protein that regulates synaptic vesicle trafficking and subsequent neurotransmitter release. It is abundant in the brain, while smaller amounts are found in the heart, muscle and other tissues. In the brain, alpha-synuclein is found mainly in the axon terminals of presynaptic neurons. Within these terminals, alpha-synuclein interacts with phospholipids and proteins. Presynaptic terminals release chemical messengers, called neurotransmitters, from compartments known as synaptic vesicles. The release of neurotransmitters relays signals between neurons and is critical for normal brain function. The human alpha-synuclein protein is made of 140 amino acids. An alpha-synuclein fragment, known as the non-Abeta component (NAC) of Alzheimer's disease amyloid, originally found in an amyloid-enriched fraction, was shown to be a fragment of its precursor protein, NACP. It was later determined that NACP was the human homologue of Torpedo synuclein. Therefore, NACP is now referred to as human alpha-synuclein.

Immunogen: Synthetic peptide within Human Phospho-alpha Synuclein (S129) aa 91-140 / 140.

Positive control: HeLa, Neuro-2a, C6, human kidney tissue, mouse brain tissue, rat brain tissue.

Subcellular location: Nucleus. Cytosol. Secreted. Membrane.

Database links: SwissProt: P37840 Human | O55042 Mouse | P37377 Rat

Recommended Dilutions:

IF-Cell	1:50-1:200
IHC-P	1:50-1:200
WB	1:500
FC	1:1,000

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

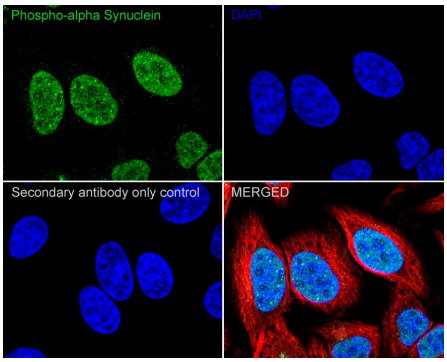

Fig1: Immunocytochemistry analysis of HeLa cells labeling Phospho-alpha Synuclein (S129) with Rabbit anti-Phospho-alpha Synuclein (S129) antibody (ET7107-30) at 1/100 dilution.

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-Phospho-alpha Synuclein (S129) antibody (ET7107-30) 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.

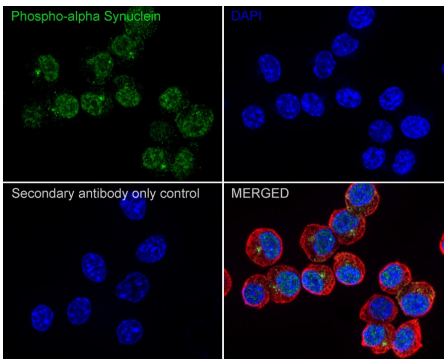

Fig2: Immunocytochemistry analysis of Neuro-2a cells labeling Phospho-alpha Synuclein (S129) with Rabbit anti-Phospho-alpha Synuclein (S129) antibody (ET7107-30) at 1/100 dilution.

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-Phospho-alpha Synuclein (S129) antibody (ET7107-30) 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.

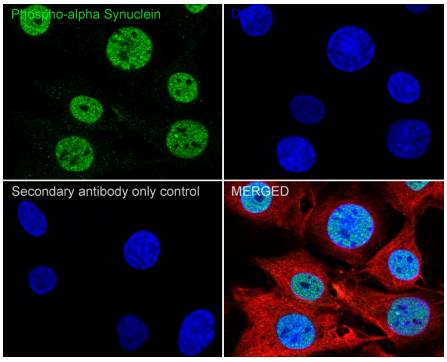

Fig3: Immunocytochemistry analysis of C6 cells labeling Phospho-alpha Synuclein (S129) with Rabbit anti-Phospho-alpha Synuclein (S129) antibody (ET7107-30) at 1/100 dilution.

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-Phospho-alpha Synuclein (S129) antibody (ET7107-30) 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.

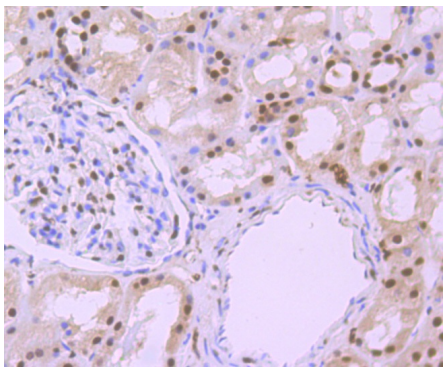

Fig4: Immunohistochemical analysis of paraffin-embedded human kidney tissue using anti-Phospho-alpha Synuclein (S129) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET7107-30, 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.

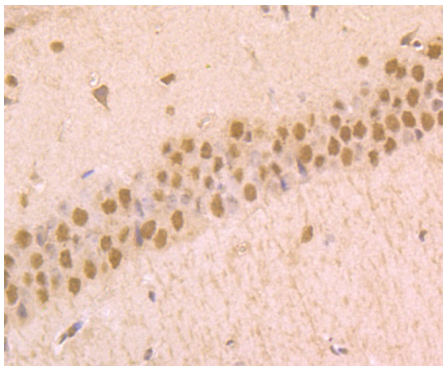

Fig5: Immunohistochemical analysis of paraffin-embedded mouse brain tissue using anti-Phospho-alpha Synuclein (S129) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET7107-30, 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.

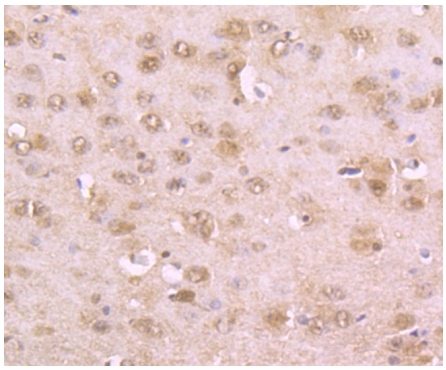

Fig6: Immunohistochemical analysis of paraffin-embedded rat brain tissue using anti-Phospho-alpha Synuclein (S129) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET7107-30, 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.

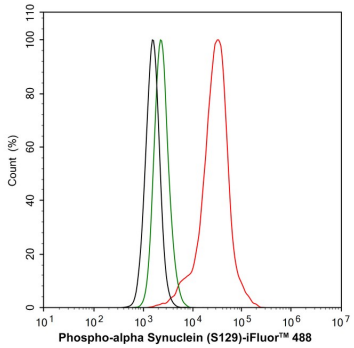

Fig7: Flow cytometric analysis of HeLa cells labeling Phospho-alpha Synuclein (S129).

Cells were fixed and permeabilized. Then stained with the primary antibody (ET7107-30, 1μg/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. Goers J et al. Nuclear localization of alpha-synuclein and its interaction with histones. Biochemistry 42:8465-8471 (2003).
2. Khalaf O et al. The H50Q mutation enhances alpha-synuclein aggregation, secretion, and toxicity. J Biol Chem 289:21856-21876 (2014).