Anti-alpha+beta Synuclein Antibody [ST05-21] ET1609-66

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:	ST05-21

Description: The synucleins, including α-synuclein (also designated NACP for nonamyloid component precursor), β-synuclein (also designated PNP 14 for phospho-neuroprotein 14) and γ-synuclein (also designated persyn or BCSG1 for breast cancer-specific gene 1) are presynaptic proteins abundant in neurons. Synucleins are predominantly expressed in the brain and are speculated to be involved in synaptic regulation and neuronal plasticity. α-Synuclein, identified as a component of Alzheimer's disease amyloid plaques, is localized to neuronal cell bodies and synapses. Coordinate expression of α-synuclein and β-synuclein may be important during hematopoetic cell differentiation. A mutant form of α-synuclein is found in patients with early onset Parkinson's disease. γ-Synuclein is associated with axonal pathology in Parkinson's disease.

Immunogen: Synthetic peptide within Human alpha+beta Synuclein aa 2-50 / 140.

Positive control: Mouse brain tissue lysate, rat brain tissue lysate, Hela, N2A, SH-SY5Y, human brain tissue, mouse brain tissue, rat brain tissue.

Subcellular location: Cytoplasm, Nucleus, Membrane, Secreted, Cell junction.

Database links: SwissProt: P37840 Human | Q16143 Human | O55042 Mouse | Q91ZZ3 Mouse | P37377 Rat | Q63754 Rat

Recommended Dilutions:

WB	1:1,000-1:2,000
IF-Cell	1:50-1:200
IF-Tissue	1:50-1:200
IHC-P	1:5,000
FC	1:50-1:100

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

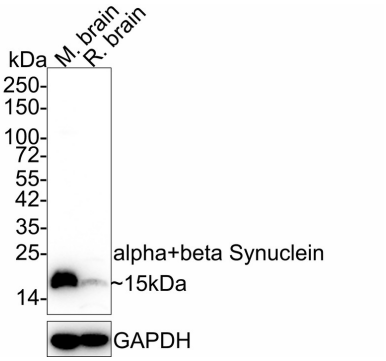

Fig1: Western blot analysis of alpha+beta Synuclein on different lysates with Rabbit anti-alpha+beta Synuclein antibody (ET1609-66) at 1/1,000 dilution.

Lane 1: Mouse brain tissue lysate
Lane 2: Rat brain tissue lysate

Lysates/proteins at 40 µg/Lane.

Predicted band size: 14 kDa
Observed band size: 15 kDa

Exposure time: 43 seconds;

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 (ET1609-66) 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.

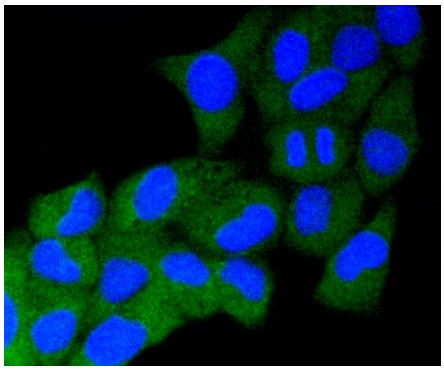

Fig2: ICC staining of alpha+beta Synuclein in Hela cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1609-66, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

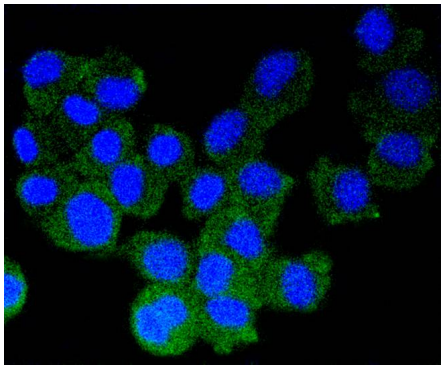

Fig3: ICC staining of alpha+beta Synuclein in N2A cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1609-66, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

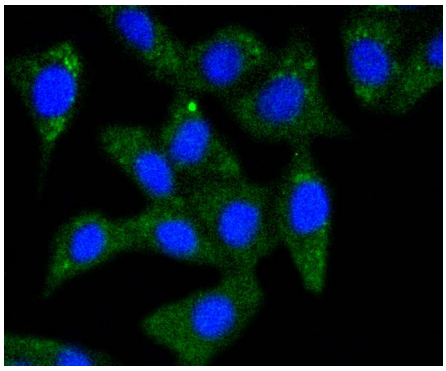

Fig4: ICC staining of alpha+beta Synuclein in SH-SY5Y cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1609-66, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

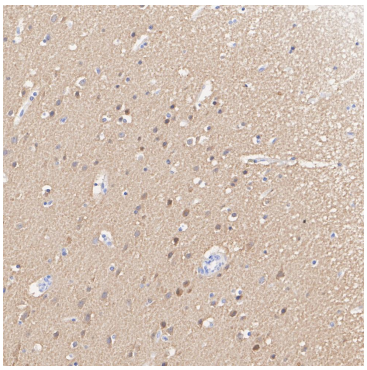

Fig5: Immunohistochemical analysis of paraffin-embedded human brain tissue with Rabbit anti-alpha+beta Synuclein antibody (ET1609-66) at 1/5,000 dilution.

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 (ET1609-66) at 1/5,000 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.

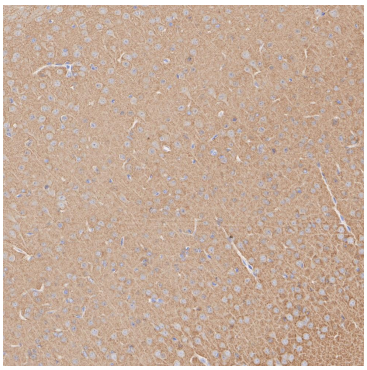

Fig6: Immunohistochemical analysis of paraffin-embedded mouse brain tissue with Rabbit anti-alpha+beta Synuclein antibody (ET1609-66) at 1/5,000 dilution.

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 (ET1609-66) at 1/5,000 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.

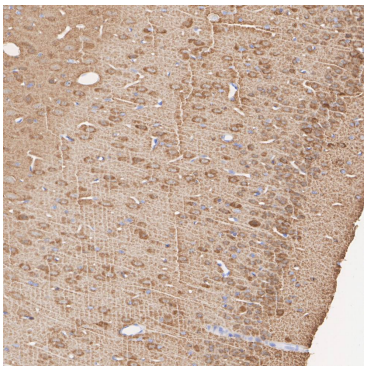

Fig7: Immunohistochemical analysis of paraffin-embedded rat brain tissue with Rabbit anti-alpha+beta Synuclein antibody (ET1609-66) at 1/5,000 dilution.

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 (ET1609-66) at 1/5,000 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.

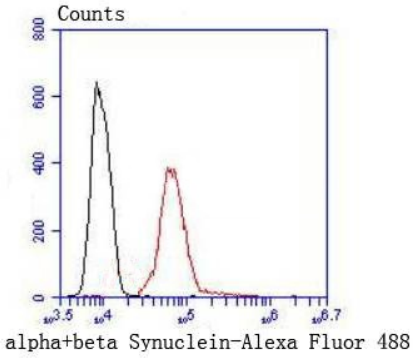

Fig8: Flow cytometric analysis of alpha+beta Synuclein was done on Hela cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1609-66, 1/50) (red). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes.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. Hall H et al. Characterization of cognitive deficits in rats overexpressing human alpha-synuclein in the ventral tegmental area and medial septum using recombinant adeno-associated viral vectors. PLoS One 8:e64844 (2013).
2. Ejlerskov P et al. Tubulin polymerization-promoting protein (TPPP/p25a) promotes unconventional secretion of a-synuclein through exophagy by impairing autophagosome-lysosome fusion. J Biol Chem 288:17313-35 (2013).