abcam

Product datasheet

Recombinant mouse Alpha-synuclein protein aggregate Type 1 (Active) ab246002

4 References 6 Images

Description

Product name	Recombinant mouse Alpha-synuclein protein aggregate Type 1 (Active)
Biological activity	Endogenous alpha-synuclein phosphorylation. 100 µM alpha synuclein protein monomer seeded with 10 nM alpha synuclein protein PFF (ab246002) in 25 µM Thioflavin T (PBS pH 7.4, 100 µl reaction volume) generated an increased fluorescence intensity after incubation at 37°C with shaking at 600 rpm for 24 hours. Fluorescence was measured by excitation at 450 nm and emission at 485 nm on a microplate reader.
Purity	> 95 % SDS-PAGE. Ion-exchange purified.
Endotoxin level	< 5.000 Eu/ml
Expression system	Escherichia coli
Accession	O55042
Protein length	Full length protein
Animal free	No
Nature	Recombinant
Species	Mouse
Sequence	MDVFMKGLSKAKEGVVAAAEKTKQGVAEAAGKTKEGVL YVGSKTKEGVVH GVTTVAEKTKEQVTNVGGAVVTGVTAVAQKTVEGAGNIA AATGFVKKDQM GKGEEGYPQEGILEDMPVDPGSEAYEMPSEEGYQDYEP EA
Predicted molecular weight	15 kDa
Amino acids	1 to 140
Additional sequence information	NP_001035916.1

Specifications

Our **Abpromise guarantee** covers the use of **ab246002** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Applications	Immunocytochemistry

	Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections)
	Functional Studies
	Electron Microscopy
	SDS-PAGE
Form	Liquid
Additional notes	Active Mouse recombinant Alpha Synuclein Pre-Formed Fibrils (Type 1).
	For best results, sonicate immediately prior to use.

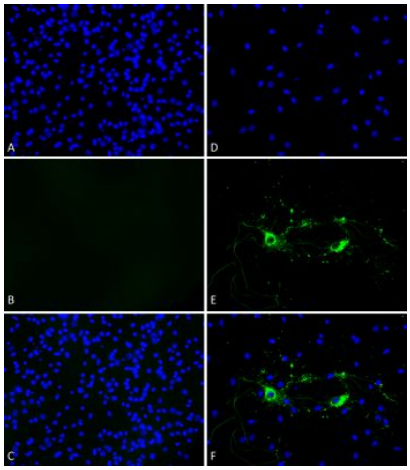
Preparation and Storage

Stability and Storage	Shipped on Dry Ice. Upon delivery aliquot. Store at -80°C. Avoid freeze / thaw cycle.
	Constituent: 95% PBS
	This product is an active protein and may elicit a biological response in vivo, handle with caution.

General Info

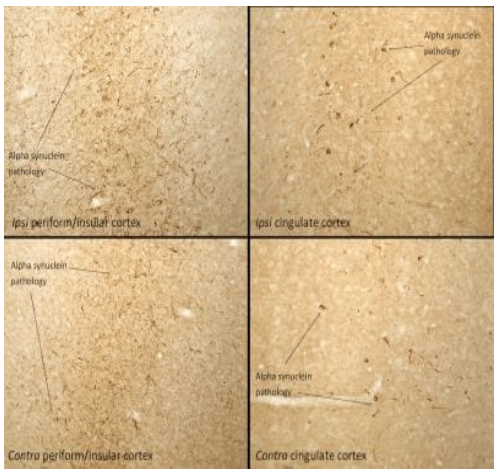
Function	May be involved in the regulation of dopamine release and transport. Induces fibrillization of microtubule-associated protein tau. Reduces neuronal responsiveness to various apoptotic stimuli, leading to a decreased caspase-3 activation.
Tissue specificity	Expressed principally in brain but is also expressed in low concentrations in all tissues examined except in liver. Concentrated in presynaptic nerve terminals.
Involvement in disease	Genetic alterations of SNCA resulting in aberrant polymerization into fibrils, are associated with several neurodegenerative diseases (synucleinopathies). SNCA fibrillar aggregates represent the major non A-beta component of Alzheimer disease amyloid plaque, and a major component of Lewy body inclusions. They are also found within Lewy body (LB)-like intraneuronal inclusions, glial inclusions and axonal spheroids in neurodegeneration with brain iron accumulation type 1. Parkinson disease 1 Parkinson disease 4 Dementia Lewy body
Sequence similarities	Belongs to the synuclein family.
Domain	The 'non A-beta component of Alzheimer disease amyloid plaque' domain (NAC domain) is involved in fibrils formation. The middle hydrophobic region forms the core of the filaments. The C-terminus may regulate aggregation and determine the diameter of the filaments.
Post-translational modifications	Phosphorylated, predominantly on serine residues. Phosphorylation by CK1 appears to occur on residues distinct from the residue phosphorylated by other kinases. Phosphorylation of Ser-129 is selective and extensive in synucleinopathy lesions. In vitro, phosphorylation at Ser-129 promoted insoluble fibril formation. Phosphorylated on Tyr-125 by a PTK2B-dependent pathway upon osmotic stress. Hallmark lesions of neurodegenerative synucleinopathies contain alpha-synuclein that is modified by nitration of tyrosine residues and possibly by dityrosine cross-linking to generated stable oligomers. Ubiquitinated. The predominant conjugate is the diubiquitinated form. Acetylation at Met-1 seems to be important for proper folding and native oligomeric structure.
Cellular localization	Cytoplasm, cytosol. Membrane. Nucleus. Cell junction, synapse. Secreted. Membrane-bound in dopaminergic neurons.

Images



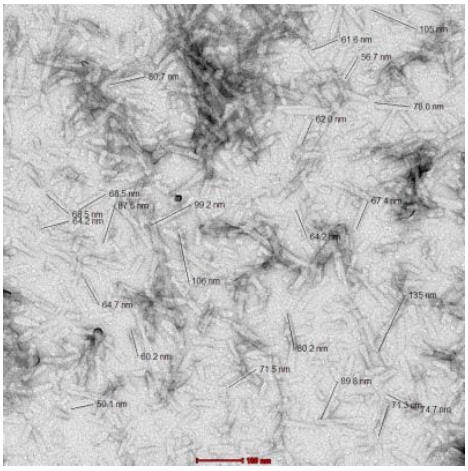
Immunocytochemistry - Recombinant mouse Alpha-synuclein protein aggregate (Active) (ab246002)

Primary rat hippocampal neurons (DIV16) show lewy body inclusion formation and loss of cells when treated with ab246002 at 4 µg/ml (D-F) on DVI2, but not when treated with a control (A-C). Tissue: Primary hippocampal neurons. Species: Sprague-Dawley rat. Fixation: 3% formaldehyde from PFA for 20 min. Blocker: 1:1 PBS:proprietary block and 30 mL/mL of 0.1% triton-X 100 for 30 min. Primary Antibody: Mouse anti-pSer129 Antibody (1/1000) and Rabbit anti-pSer129 (1/800) for 24 hours at 4°C. Secondary Antibody: ATTO 546 Donkey Anti-Mouse (1/700) and ATTO 488 Donkey Anti-Rabbit (1/700) for 1 hour at room temperature (composite green). Counterstain: Hoechst (blue) nuclear stain at 1/3000 for 1 hour at room temperature. Localization: Lewy body incluscions. Magnification: 20x.



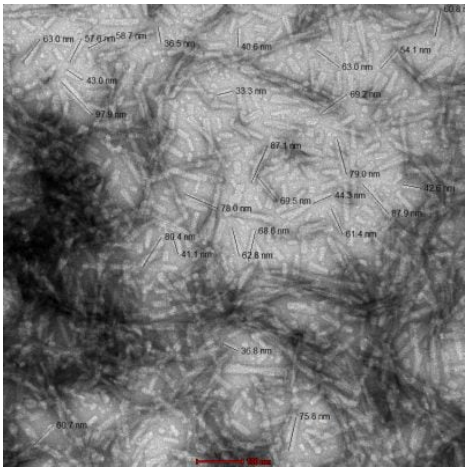
Immunohistochemistry (PFA fixed) - Recombinant mouse Alpha-synuclein protein (Active) (ab246002)

Immunohistochemistry analysis of rat brain injected with ab246002. Species: Female Sprague-Dawley Rat. Rat was injected with 2µL ab246002 in each of 2 injection sites: AP+1.6, ML+2.4, DV-4.2 from skull; and AP-1.4, ML+0.2, DV-2.8 from skull. 30 days post-injection. Fixation: Saline perfusion followed by 4% PFA fixation for 48 hours. Primary antibody: rabbit monoclonal anti-pSer129 alpha synuclein. Secondary Antibody: Biotin-SP Donkey Anti-Rabbit IgG (H+L) at 1/500 for 2 hours in cold room with shaking. ABC signal amplification, DAB staining. Magnification: 20x. Alpha synuclein pathology is seen in the periform/insular cortex and the cingulate cortex on both the same (ipsi) and opposite (contra) sides as the injection sites.



Electron Microscopy - Recombinant mouse Alpha-synuclein protein (Active) (ab246002)

TEM of ab246002. Fibrils were sonicated and image was taken at 100kx magnification.



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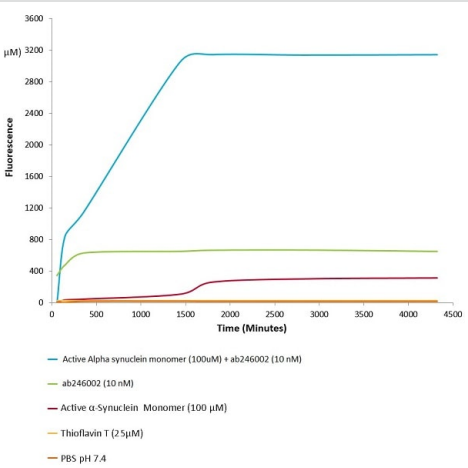
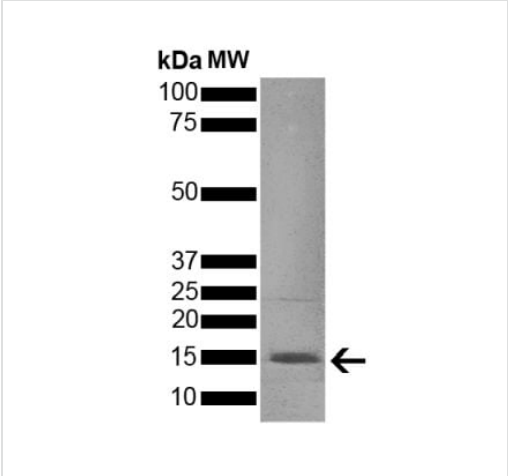


Functional Studies - Recombinant mouse Alpha-synuclein protein (Active) (ab246002)

ab246002 seeds the formation of new Alpha synuclein fibrils from the pool of active alpha synuclein monomers. Thioflavin T is a fluorescent dye that binds to beta sheet-rich structures, such as those in alpha synuclein fibrils. Upon binding, the emission spectrum of the dye experiences a red-shift, and increased fluorescence intensity. Thioflavin T emission curves show increased fluorescence (correlated to alpha synuclein protein aggregation) over time when 10 nM of ab246002 is combined with 100 µM of active Alpha synuclein monomer, as compared to ab246002 and active alpha Synuclein monomer alone. Thioflavin T ex = 450 nm, em = 485 nm.



SDS-PAGE - Recombinant mouse Alpha-synuclein protein (Active) (ab246002)

SDS-PAGE analysis of ab246002 (2 µg).

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