

Human alpha Synuclein protein (active, Pre-Formed Fibrils)

Cat. No. GTX17669-pro

Applications	Functional Assay	Package
Species	Human	200 µg, 100 µg

PRODUCT

Summary Active Human Recombinant Alpha Synuclein Pre-formed Fibrils (Type 1)

Applications

Application Note

Endogenous alpha-synuclein phosphorylation. 100 µM alpha synuclein protein monomer (GTX17668-pro) seeded with 10 nM alpha synuclein protein PFF (GTX17669-pro) in 25 µM Thioflavin T (PBS pH 7.4, 100 µl reaction volume) generated a fluorescence intensity of 13,000 Relative Fluorescence Units after incubation at 37°C with shaking at 600 rpm. Fluorescence was measured by excitation at 450 nm and emission at 485 nm on a Molecular Devices Gemini XPS microplate reader.

*For best results, sonicate immediately prior to use.

Properties

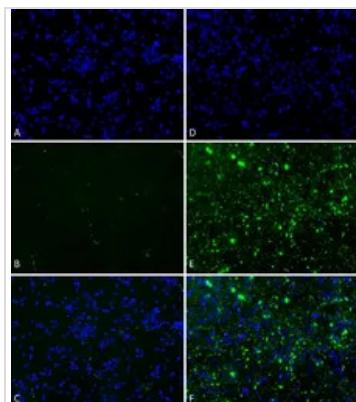
Form	Liquid
Buffer	PBS
Preservative	No preservatives
Storage	Store as concentrated solution. Aliquot and store at -80°C. Avoid freeze-thaw cycles.
Concentration	Batch dependent (Please refer to the vial label for the specific concentration.)
Region/Sequence	Full-length without tagged; MDVFMKGLSK AKEGVAAAEE KTKQGVAAEA GKTKEGVLYV GSKTKEGVVH GVATVAEKT EQVTNVGGAV VTGVTAVAQK TVEGAGSIAA ATGFVKKDQL GKNEEGAPQE GILEDMPVDP DNEAYEMPSE EGYQDYEPEA
Expression System	E. coli
Purification	Purified by ion-exchange chromatography
Purity	> 95% by SDS-PAGE
Endotoxin	<5 EU/mL
Conjugation	Unconjugated
Note	For laboratory research use only. Not for any clinical, therapeutic, or diagnostic use in humans or animals. Not for animal or human consumption.
	Purchasers shall not, and agree not to enable third parties to, analyze, copy, reverse engineer or otherwise attempt to determine the structure or sequence of the product.



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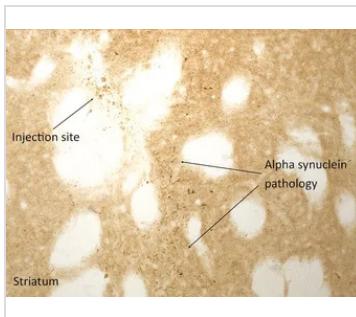
DATA IMAGES



GTX17669-pro Functional Assay Image

Primary rat hippocampal neurons show Lewy body inclusion formation when treated with active Alpha Synuclein Protein Preformed Fibrils (GTX17669-pro) at 4 µg/ml (D-F), but not when treated with control Alpha Synuclein Protein Preformed Fibrils (GTX17669-pro) at 4 µg/ml (A-C).

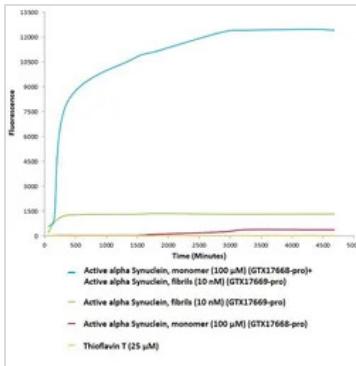
Tissue: Primary hippocampal neurons. Species: Sprague-Dawley rat. Fixation: 4% formaldehyde from PFA. Primary Antibody: Mouse anti-pSer129 Antibody at 1:1000 24 hours at 4°C. Secondary Antibody: FITC Goat Anti-Mouse (green) at 1:700 for 1 hours at RT. Counterstain: Hoechst (blue) nuclear stain at 1:4000 for 1 hour at RT. Localization: Lewy body inclusions. Magnification: 20x.



GTX17669-pro Functional Assay Image

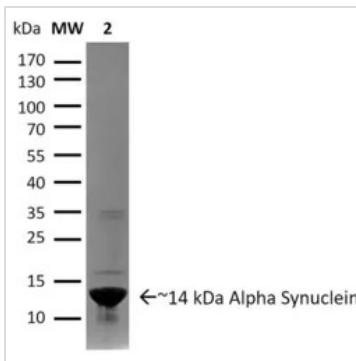
Immunohistochemistry analysis of rat brain injected with active human alpha synuclein PFFs (GTX17669-pro). Species: Female Sprague-Dawley Rat. Rat was injected with 2 µL active human alpha synuclein PFFs (GTX17669-pro) in each of 2 injection sites: AP+1.6, ML+2.4, DV-4.2 from skull; and AP-1.4, ML+0.2, DV-2.8 from skull. 30-days post-injection. Fixation: Saline perfusion followed by 4% PFA fixation for 48 hrs.

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 striatum close to an injection site.



GTX17669-pro Image

Active alpha synuclein preformed fibrils (GTX17669-pro) seed the formation of new alpha synuclein fibrils from the pool of alpha synuclein monomers (GTX17668-pro). 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 active alpha synuclein preformed fibrils (GTX17669-pro) is combined with 100 µM of alpha synuclein monomer (GTX17668-pro), as compared to active alpha synuclein preformed fibrils (GTX17669-pro) alone and alpha synuclein monomer (GTX17668-pro) alone. Thioflavin T ex = 450 nm, em = 485 nm.



GTX17669-pro Image

SDS-PAGE of ~14 kDa active Human Recombinant Alpha Synuclein Protein Preformed Fibrils (GTX17669-pro). Lane 1: Molecular Weight Ladder (MW). Lane 2: active Alpha Synuclein Protein Preformed Fibrils (GTX17669-pro).



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