

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

Cat. No. GTX17669-pro

Applications Functional Assay**Species** Human**Package**

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 AKEGVVAAA KTKQGVAAEA GKTKEGVLYV GSKTKEGVWH GVATVAEKT
EQVTNVGGAV VTGVTAVAQK TVEGAGSIAA ATGFVKKDQL GKNEEGAPQE GILEDMPVDP DNEAYEMPSE EGYQDYEP**Expression System** E. coli**Purification** Purified by ion-exchange chromatography**Purity** > 95% by SDS-PAGE**Endotoxin** <5 EU/mL**Conjugation** Unconjugated

Note

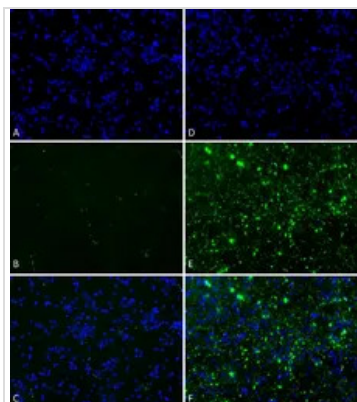
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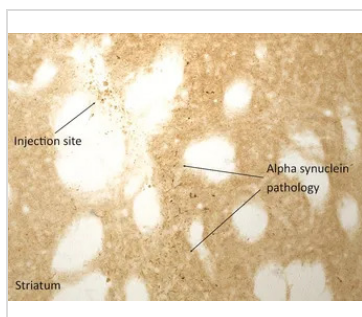


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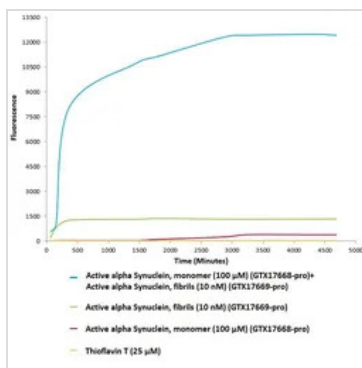
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DATA IMAGES

GTX17669-pro Functional Assay Image

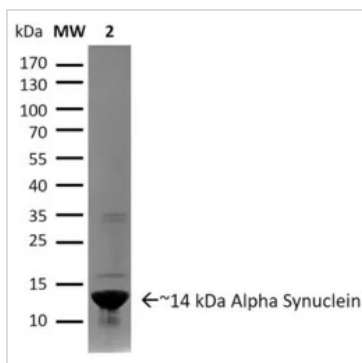
Primary rat hippocampal neurons show lewy body inclusion formation when treated with active Alpha Synuclein Protein Preformed Fibrils (GTX17669-pro) at 4 µg/ml (D-F), but not when treated with control Alpha Synuclein Protein Preformed Fibrils (GTX17667-pro) at 4 µg/ml (A-C).
Tissue: Primary hippocampal 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 2µL 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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