

# Produktinformation



Forschungsprodukte & Biochemikalien
Zellkultur & Verbrauchsmaterial
Diagnostik & molekulare Diagnostik
Laborgeräte & Service

Weitere Information auf den folgenden Seiten! See the following pages for more information!



Lieferung & Zahlungsart siehe unsere Liefer- und Versandbedingungen

# Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

# SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien T. +43(0)1 489 3961-0 F. +43(0)1 489 3961-7 <u>mail@szabo-scandic.com</u> www.szabo-scandic.com Alpha Synuclein Protein

94 /100 5 Citations



Discovery through Partnership | Excellence through Quality

Human Recombinant Alpha Synuclein Preformed Fibrils (Type 1) Catalog No. SPR-322

Powered by Bioz

#### **Product Name**

Alpha Synuclein Protein

#### Description

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

#### Applications

WB, SDS-PAGE, In vivo assay, In vitro assay

#### Concentration

Lot/batch specific. See included datasheet.

#### Conjugates

No tag

#### Nature

Recombinant

#### Species

Human

#### **Expression System**

E. coli

#### **Amino Acid Sequence**

MDVFMKGLSK AKEGVVAAAE KTKQGVAEAA GKTKEGVLYV GSKTKEGVVH GVATVAEKTK EQVTNVGGAV VTGVTAV AQK TVEGAGSIAA ATGFVKKDQL GKNEEGAPQE GILEDMPVDP DNEAYEMPSE EGYQDYEPEA

## Purity

>95%

#### **Protein Length**

**Full Length** 

#### **Biological Activity**

Endogenous alpha-synuclein phosphorylation. 100  $\mu$ M alpha synuclein protein monomer (SPR-321) seeded with 10 uM alpha synuclein protein PFF (SPR-322) in 25  $\mu$ M Thioflavin T (PBS pH 7.4, 100  $\mu$ l 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.

#### **Field Of Use**

Not for use in humans. Not for use in diagnostics or therapeutics. For in vitro research use only.

## **Properties**

#### **Storage Buffer**

PBS

#### **Storage Temperature**

-80°C

#### **Shipping Temperature**

Dry Ice. Shipping note: Product will be shipped separately from other products purchased in the same order.

#### Purification

Ion-exchange Purified

#### **Cite This Product**

Human Recombinant Alpha Synuclein Protein (StressMarq Biosciences, Canada, Cat # SPR-322)

#### Certificate Of Analysis

Certified >95% pure using SDS-PAGE analysis.

#### **Other Relevant Information**

For best results, sonicate immediately prior to use. Refer to the Neurodegenerative Protein Handling Instructions on our website, or on the product datasheet for further information.

# **Biological Description**

## **Alternative Names**

Alpha synuclein PFFs, Alpha synuclein aggregates, Alpha synuclein PFF, Alpha synuclein protein aggregates, Alpha synuclein aggregates, Alpha-synuclein protein, Non-A beta component of AD amyloid protein, Non-A4 component of amyloid precursor protein, NACP protein, SNCA protein, NACP protein, PARK1 protein, SYN protein, Parkinson's disease familial 1 Protein, Alpha synuclein protein seed

#### **Research Areas**

Alzheimer's Disease, Neurodegeneration, Neuroscience, Parkinson's Disease, Synuclein, Tangles & Tau, Multiple System Atrophy

#### **Cellular Localization**

Cytoplasm, Membrane, Nucleus

Accession Number	
NP_000336.1	
Gene ID	
6622	
Swiss Prot	
P37840	

## Scientific Background

Alpha-Synuclein (SNCA) is expressed predominantly in the brain, where it is concentrated in presynaptic nerve terminals (1). Alpha-synuclein is highly expressed in the mitochondria of the olfactory bulb, hippocampus, striatum and thalamus (2). Functionally, it has been shown to significantly interact with tubulin (3), and may serve as a potential microtubule-associated protein. It has also been found to be essential for normal development of the cognitive functions; inactivation may lead to impaired spatial learning and working memory (4). SNCA fibrillar aggregates represent the major non A-beta component of Alzheimers disease amyloid plaque, and a major component of Lewy body inclusions, and Parkinson's disease. Parkinson's disease (PD) is a common neurodegenerative disorder characterized by the progressive accumulation in selected neurons of protein inclusions containing alpha-synuclein and ubiquitin (5, 6).

#### References

- 1. "Genetics Home Reference: SNCA". US National Library of Medicine. (2013).
- 2. Zhang L., et al. (2008) Brain Res. 1244: 40-52.
- 3. Alim M.A., et al. (2002) J Biol Chem. 277(3): 2112-2117.
- 4. Kokhan V.S., Afanasyeva M.A., Van'kin G. (2012) Behav. Brain. Res. 231(1): 226-230.
- 5. Spillantini M.G., et al. (1997) Nature. 388(6645): 839-840.
- 6. Mezey E., et al. (1998) Nat Med. 4(7): 755-757.

# Product Images



Primary rat hippocampal neurons show lewy body inclusion formation when treated with Type 1 Alpha Synuclein Protein Pre-formed Fibrils (SPR-322) at 4  $\mu$ g/ml (D-F), but not when treated with Type 2 Alpha Synuclein Protein Pre-formed Fibrils (SPR-317) at 4  $\mu$ g/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.



Immunohistochemistry analysis of rat brain injected with Type 1 human alpha synuclein PFFs (SPR-322). Species: Female Sprague-Dawley Rat. Rat was injected with 16µg Type 1 human alpha synuclein PFFs (SPR-322) 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. 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 striatum close to an injection site.



Type 1 alpha synuclein Pre-formed Fibrils (SPR-322) seed the formation of new alpha synuclein fibrils from the pool of alpha synuclein monomers (SPR-321). 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  $\mu$ M of Type 1 alpha synuclein Pre-formed Fibrils (SPR-322) is combined with 100  $\mu$ M of alpha synuclein monomer (SPR-321), as compared to Type 1 alpha synuclein Pre-formed Fibrils (SPR-322) alone and alpha synuclein monomer (SPR-321) alone. Thioflavin T ex = 450 nm, em = 485 nm.



SDS-PAGE of ~14 kDa Type 1 Human Recombinant Alpha Synuclein Protein Pre-formed Fibrils (SPR-322). Lane 1: Molecular Weight Ladder (MW). Lane 2: Type 1 Alpha Synuclein Protein Pre-formed Fibrils (SPR-322).



TEM of Type 1 Alpha Synuclein Pre-formed Fibrils (PFFs) (SPR-322)



TEM of Type 1 Alpha Synuclein Pre-formed Fibrils (PFFs) (SPR-322)



Toxicity results comparing Active Human Recombinant Alpha Synuclein Pre-formed Fibrils (Type 2) (Catalog No. SPR-317) and Active Human Recombinant Alpha Synuclein Pre-formed Fibrils (Type 1) (Catalog No. SPR-322). Data was graphed after live cell imaging results were obtained using the following procedure: After 8 days in vitro, primary rat mixed cortical neuron cells were washed with 1X PBS and treated with 500 µg/ml of Type 1 and Type 2 Alpha Synuclein Proteins for 20 hours at 37°C. Following treatements, cells were washed with 2X PBS and incubated with a staining solution (2.0 µM Cell Event + 2.5 µM Ethidium homodimer + 2.5 µg/ml Hoechst 33342 in sterile HBSS) for 30 minutes at 37°C. The addition of the Type 2 Alpha Synuclein Proteins resulted in a significant increase in cell death.



ATTO633 fluorescently-labelled alpha synuclein PFFs (SPR-322) were taken up, transported into the soma, and induced alpha synuclein aggregation in mouse neurocortical primary cells. (A) Neurites filled with fluorescently-labelled alpha synuclein seeds in a microfluidic co-culture system after 24 hours. (B) Alpha synuclein seeds within the soma and neurites of mouse neurocortical primary cells after 24 hours. Experiment and imaging courtesy of Cellectricon.



Confocal imaging shows NeuN+ (mature) primary cortical neurons filled with ATTO633 fluorescentlylabelled alpha synuclein PFFs (SPR-322). ATTO-633 alpha synuclein PFFs seed endogenous alpha synculein phosphorylation after 7 days. Experiment and imaging courtesy of Cellectricon.



Immunocytochemistry/Immunofluorescence analysis of human iPSC-derived neurons treated with alpha synuclein pre-formed fibrils (SPR-322). Primary Antibody: Mouse Anti-Alpha Synuclein (pSer129) Monoclonal Antibody (SMC-600) at 1:1000 for O/N at 4°C. Secondary Antibody: Anti-Rabbit: A488 at 1:1000 for 1 hour at RT. Magnification: 40X. Nuclear stain: Hoechst- 20 min, RT (blue). Actin stain: Phalloidin-647- 20 min, RT (magenta). 4K cells per well. iPSC neurons: Applied StemCell Catalog # ASE-9321K. A) negative control; no fibrils added to well. B) 7 days after addition of active recombinant human pre-formed fibrils (Type 1). Fibrils were sonicated before use and applied 2.5 ug per well



TEM of Type 1 Alpha Synuclein Pre-formed Fibrils (PFFs) (SPR-322)



Alpha Synuclein Oligomers Have Distinct Secondary Structure Differences Compared to Fibrils. UV-CD data suggests that StressMarq's Alpha Synuclein Oligomers have distinct secondary structure differences compared to our monomers and fibrils. More specifically, StressMarq's Kinetically Stable Alpha Synuclein Oligomers (SPR-484) show a significantly higher alpha helix content and lower beta sheet/turn content than our Alpha Synuclein Pre-formed Fibrils (Type 1) (SPR-322). StressMarq's Alpha Synuclein Monomers (SPR-316) show a strong negative signal at 200 nm indicative of a disordered protein state (low secondary structure content).



Far-UV CD spectra of human alpha-synuclein

UV-CD data suggests StressMarq's Alpha Synuclein Pre-formed Fibrils (PFF), Type 1 (cat#SPR-322) and Type 2 (cat#SPR-317) both have a high beta sheet/turn content, yet do have small secondary structure differences. StressMarq's Alpha Synuclein Monomers (cat#SPR-316) show a strong negative signal at 200 nm indicative of a disordered protein state (low secondary structure content). For this experiment, pre-formed fibrils (PFF) were subjected to 10 cycles of sonication prior to UV-CD to ensure solubility prior to measurement.



StressMarq's Alpha Synuclein PFFs (red) cat#SPR-322, were shown to be taken up by SH-SY5Y cells and transmitted to neuronal iPSCs within 14 days. Blue: Hoechst/DNA; Green: SHSY5Y-GFP; Red: alpha-synuclein PFFs-555 (cat#SPR-322); Purple: Tubulin.

# **Product Citations**



"PrePrint: Unless otherwise stated, recombinant protein generated in-house was applied at 10 nM concentration concentration supplemented with 1% bovine serum albumin, conditioned media was applied neat for 20 min an

**Granulovacuolar degeneration bodies are independently induced by tau and α-synuclein pathology** Marta Jorge-Oliva, Jasper F. M. Smits, ..., Wiep Scheper

Alzheimer's Research & Therapy | 2022 Dec 14 | PubMed ID: 36517915 | Read Article 🖉

"scher UP200St ultrasonic homogeniser (two cycles of 1 min in pulses of 10 s and 5 s rest). The resulting PFFs further diluted in PBS into the required concentration and re-sonicated (1 min in pulses of 10 s and 5 s rest). PH

Rational Generation of Monoclonal Antibodies Selective for Pathogenic Forms of Alpha-SynucleinEbrima Gibbs, Beibei Zhao, ..., Nikolaos N. LourosBiomedicines | 2022 Sep 02 | PubMed ID: 36140270 | Read Article Image: Article Snippet

".. SEC ( ).The preparation was confirmed to contain primarily tetramers by SEC ( ).. Full length human α StressMarq (Victoria, BC, Canada, SPR-322) and were sonicated to generate small soluble seeding fibrils a

# **Reviews**

Based on validation through cited publications.



**StressMarq Biosciences** January 25, 2021: