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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

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Alpha Synuclein E114C Mutant Pre-formed Fibrils: ATTO 488

StressMarq Biosciences INC.

Discovery through Partnership | Excellence through Quality

Human Recombinant Alpha Synuclein E114C Mutant Pre-formed Fibrils: ATTO 488
Catalog No. SPR-518-A488

Product Name

Alpha Synuclein E114C Mutant Pre-formed Fibrils: ATTO 488

Description

Human Recombinant Alpha Synuclein E114C Mutant Pre-formed Fibrils: ATTO 488

Applications

WB, Native PAGE, In vitro Assay, In vivo Assay

Concentration

Lot/batch specific. See included datasheet.

Conjugates

ATTO 488

Nature

Recombinant

Species

Human

Expression System

E. coli

Amino Acid Sequence

10% of mixture (mutant conjugated form): MDVFMKGLSKAKEGVVAAAEKTKQGVAEAAGKTKEGVLYVGSKTKEGVVHGVATVAEKTKEQVTNVGGAVVTGVTAVAQKTVEGAGSIAAATGFVKKDQLGKNEEGAPQEGILCDMPVDPDNEAYEMPSEEGYQDYEPEA
90% of mixture (wildtype): MDVFMKGLSKAKEGVVAAAEKTKQGVAEAAGKTKEGVLYVGSKTKEGVVHGVATVAEKTKEQVTNVGGAVVTG[redacted]VAQKTVEGAGSIAAATGFVKKDQLGKNEEGAPQEGILEDMPVDPDNEAYEMPSEEGYQDYEPEA

Purity

>95%

Protein Length

140 AA

Protein Size

14.434 kDa

Field Of Use

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

Properties

Storage Buffer

1X PBS pH 7.4

Storage Temperature

-80ºC

Shipping Temperature

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

Purification

Ion-exchange & SEC purified

Cite This Product

Human Recombinant Alpha Synuclein E114C Mutant Pre-formed Fibrils: ATTO 488 (StressMarq Biosciences Inc., Victoria BC CANADA, Catalog # SPR-518)

Certificate Of Analysis

Protein certified >95% pure on SDS-page and nanodrop analysis, endotoxin below 5 EU/mL at 2 mg/mL on starting monomer material.

Other Relevant Information

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

Biological Description

Alternative Names

Alpha synuclein pre-formed fibril, Alpha-synuclein PFF, Alpha synuclein protein fibrils, 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

Research Areas

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

Swiss Prot

P37840

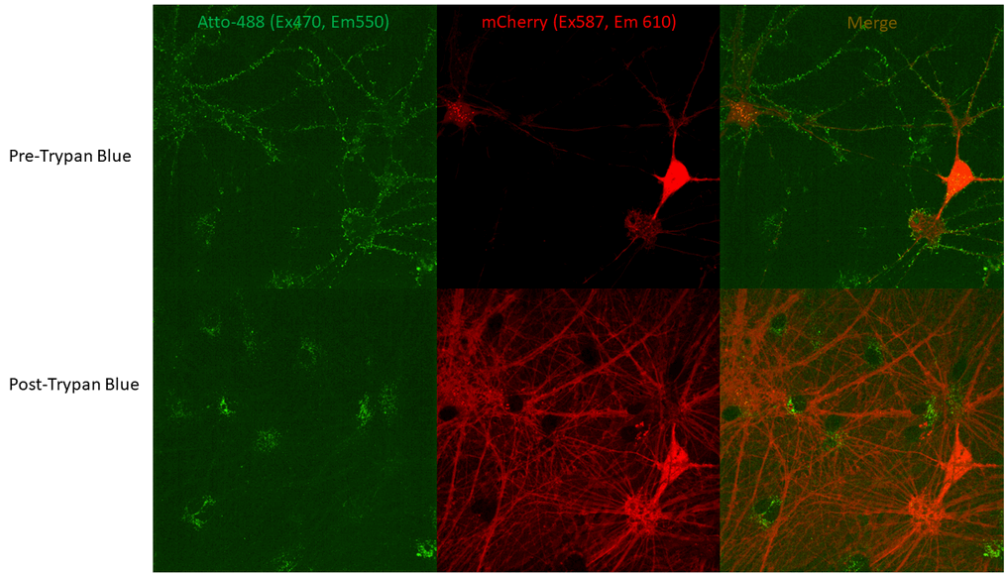
Scientific Background

The alpha-synuclein (aSyn) E114C mutation facilitates a single site-specific conjugation with ATTO-488 maleimide that avoids any hindrance on fibrilization or cell entry that may be conferred by non-specific lysine targeting conjugations. This conjugation is ideal due to internal position relative to C-terminal truncation sites, proximity to the NAC, and lack of interference with recruitment in vitro or in primary neurons (1, 2). Pre-formed fibrils (PFFs) generated with 5-25% fluorescently tagged E114C mutants have demonstrated a relative potency >80% compared to wild-type aSyn for inducing misfolding of endogenous aSyn, indicating no significant perturbation of seeding in living cells (1). Atto-488 is a useful tool for identifying cell entry, as the addition of Trypan Blue to cultures prior to imaging will quench fluorescence of extracellular Atto-488 conjugated aSyn (3). Our aSyn E114C-Atto-488 PFFs, which are formed from 10% fluorescently tagged E114C mutants and 90% wild-type monomers, are an excellent tool for studying cell entry and localization, with demonstrated entry into neurons after trypan blue quenching.

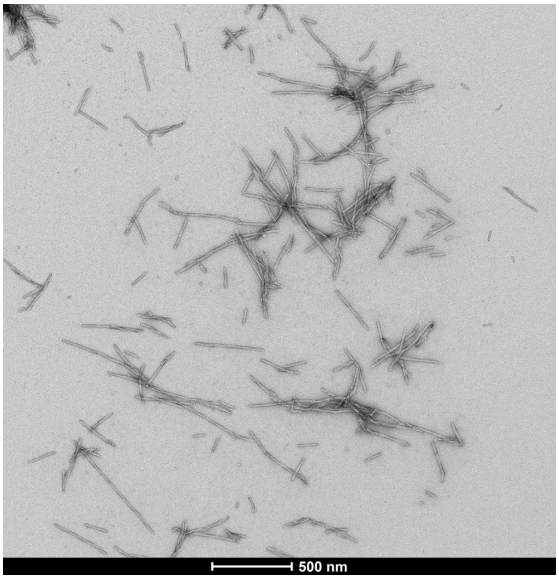
References

1.,Haney et al. 2016. Comparison of strategies for non-perturbing labeling of α-synuclein to study amyloidogenesis. Organic & Biomolecular Chemistry. DOI: 10.1039/c5ob02329g
2.,Karpowicz et al. 2017. Selective imaging of internalized proteopathic a-synuclein seeds in primary neurons reveals mechanistic insight into transmission of synucleinopathies. JBC. DOI: 10.1074/jbc.M117.780296
3.,Pieri et al. 2016. Structural and functional properties of prefibrillar α-synuclein oligomers. Scientific Reports. DOI: 10.1038/srep24526

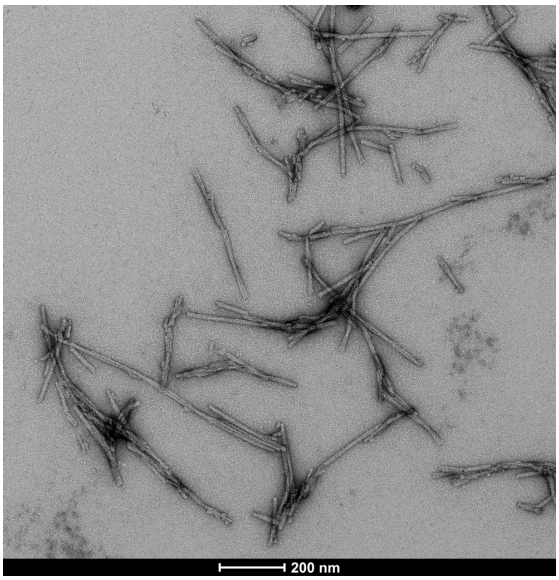
Product Images



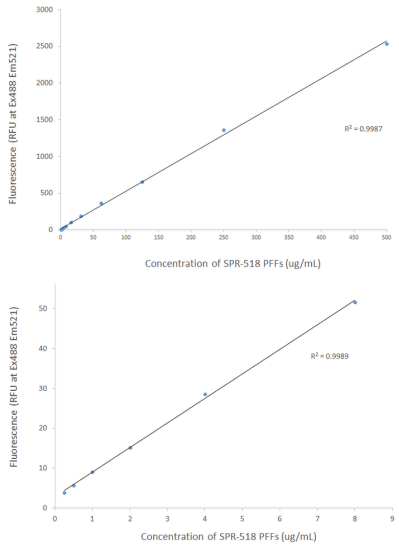
Neuronal uptake of ATTO-488 conjugated Alpha Synuclein E114C Pre-Formed Fibril (PFFs) (SPR-518-A88) visible by fluorescence after Trypan Blue quenching. Neurons expressing mCherry via AAVs (division 19) were treated with SPR-518 and then Trypan Blue to quench extracellular PFF fluorescence. ATTO-488 fluorescence present after (bottom row) Trypan Blue addition is from internalized PFFs. Note: greater mCherry signal post-Trypan Blue addition attributed to overlap of excitation/emission spectra (mCherry maxima ex 587/em610, Trypan Blue ex maxima 560/em630).



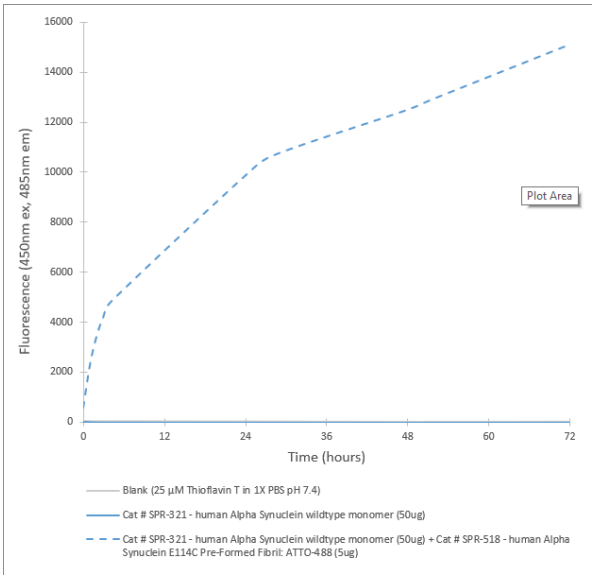
Representative TEM image of ATTO-488 conjugated Alpha Synuclein E114C Pre-Formed Fibrils (SPR-518-A88), 500nm scale. Negative stain transmission electron microscopy images of SPR-518 acquired at 80 Kv on carbon coated 400 mesh copper grids using phosphotungstic acid and uranyl acetate stain.



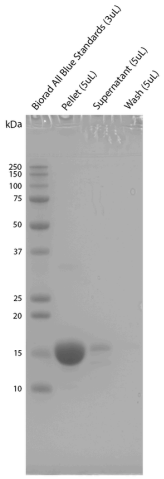
Representative TEM image of ATTO-488 conjugated Alpha Synuclein E114C Pre-Formed Fibrils (SPR-518-A88), 200nm scale. Negative stain transmission electron microscopy images of SPR-518 acquired at 80 Kv on carbon coated 400 mesh copper grids using phosphotungstic acid and uranyl acetate stain.



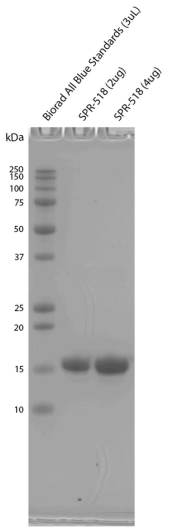
Fluorescent signal of alpha-synuclein E114C-ATTO-488 PFFs (SPR-518-A88). SPR-518 concentration and fluorescence (excitation 488nm, emission 521 nm) exhibit a linear relationship at all concentrations tested (250 ng/mL – 500 µg/mL).



In vitro seeding activity of human Alpha Synuclein wildtype monomer in ThT assay (SPR-518-A88). SPR-518 (ATTO-488 conjugated Alpha Synuclein E114C fibrils) seed fibril formation of SPR-321 (Type 1 Alpha Synuclein wildtype monomer) over 72 hours. Reactions (100uL) shaken at 600 rpm in Greiner-Bio 96 Well Non-Binding Cell Culture Microplates, Black (Greiner-Bio Catalog #655900) at 37oC and read with an XPS Microplate Reader set at 450nmex/485nmem.



Sedimentation assay on ATTO-488 conjugated Alpha Synuclein E114C Pre-Formed Fibrils (SPR-518-A88). Samples were pelleted at 15,000 x g, washed, and then re-centrifuged. Fibril samples are prepared in denaturing conditions prior to running on the gel. SDS-PAGE analysis on a 12% Bis-Tris gel shows that the majority of fibril is insoluble.



SDS-PAGE analysis of ATTO-488 conjugated Alpha Synuclein E114C Pre-Formed Fibrils (SPR-518-A88) on a 12% Tris-Glycine gel. Fibril samples are prepared in denaturing conditions prior to running on the gel. Note: PFFs are not SDS-stable, consistent with wild-type PFFs.

Product Citations

Reviews

There are no reviews yet.