## abcam

### Product datasheet

# Recombinant Human Alpha-synuclein protein aggregate (Active) ab218819

6 References 9 图像

描述

产品名称 重组人Alpha-synuclein蛋白aggregate (Active)

生物活性 Endogenous alpha-synuclein phosphorylation.

100  $\mu$ M alpha synuclein protein monomer (**ab218818**) seeded with 10  $\mu$ M alpha synuclein protein aggregate (ab218819) in 25  $\mu$ M Thioflavin T (**ab120751**) (PBS pH 7.4, 100  $\mu$ I reaction volume) generated a fluorescence intensity of 13,000 Relative Fluorescence Units 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.

Endotoxin Level: 10-20 EU/mL

纯**度** > 95 % SDS-PAGE.

ab218819 was purified by ion-exchange.

内毒素水平 <=20.000 Eu/ml 表达系统 Escherichia coli

Accession P37840

**蛋白长度** Full length protein

无动物成分 No

性质 Recombinant

**种属** Human

序列 MDVFMKGLSK AKEGVVAAAE KTKQGVAEAA

GKTKEGVLYV GSKTKEGVVH GVATVAEKTK EQVTNVGGAV VTGVTAVAQK TVEGAGSIAA ATGFVKKDQL GKNEEGAPQE GILEDMPVDP

DNEAYEMPSE EGYQDYEPEA

预**测分子量** 14 kDa **氨基酸** 1 to 140

额外的序列信息 (NP\_000336.1) (GenelD 6622)

描述 重组人Alpha-synuclein蛋白(Active)

技术指标

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Our Abpromise guarantee covers the use of ab218819 in the following tested applications.

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

应用 Western blot

**Functional Studies** 

SDS-PAGE

形式 Liquid

补充说明 For best results, sonicate immediately prior to use. Sonication of PFFs is required in order to

obtain fibrils with an average length of ~50nm, the optimal size for pathology.

制备和贮存

稳定性和存储 Shipped on Dry Ice. 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.

常规信息

功能 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.

组织特异性 Expressed principally in brain but is also expressed in low concentrations in all tissues examined

except in liver. Concentrated in presynaptic nerve terminals.

疾病相关 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

序列相似性 Belongs to the synuclein family.

结**构域** 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.

翻译后修饰 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

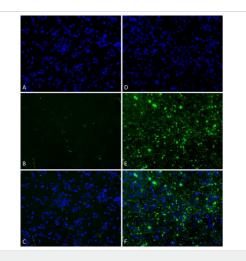
oligomers.

 $\label{thm:conjugate} \mbox{ Ubiquitinated. The predominant conjugate is the diubiquitinated form.}$ 

Acetylation at Met-1 seems to be important for proper folding and native oligomeric structure.

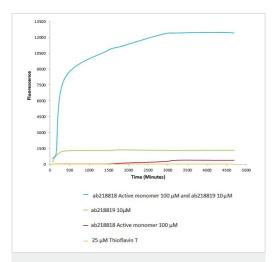
细胞定位 Cytoplasm, cytosol. Membrane. Nucleus. Cell junction, synapse. Secreted. Membrane-bound in

#### 图片



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Recombinant human Alphasynuclein protein aggregate (Active) (ab218819)

Immunohistochemical analysis of primary rat hippocampal neurons showing lewy body inclusion formation when treated with active Alpha Synuclein Protein Aggregate (ab218819) at 4 µg/ml (D-F), but not when treated with control Alpha Synuclein Protein Aggregate (ab218817) 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 hour at RT. Counterstain: Hoechst (blue) nuclear stain at 1/4000 for 1 hour at RT. Localization: Lewy body incluscions. Magnification: 20x.



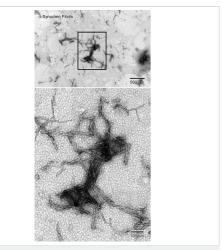
Functional Studies - Recombinant Human Alphasynuclein protein aggregate (Active) (ab218819)

ab218819 seeds the formation of new alpha synuclein fibrils from the pool of 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.

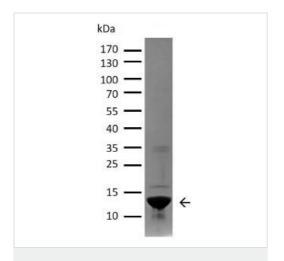
Thioflavin T emission curves show increased fluorescence (correlated to alpha synuclein protein aggregation) over time when 10  $\mu$ M of ab218819 is combined with 100  $\mu$ M of alpha synuclein monomer, as compared to ab218819 alone and alpha synuclein monomer alone.

Thioflavin T ex = 450 nm, em = 485 nm.



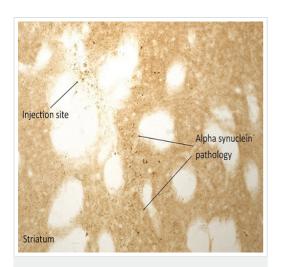
Electron Microscopy - Recombinant human Alphasynuclein protein aggregate (Active) (ab218819)

TEM of active human alpha synuclein preformed fibrils (ab218819). Fibrils were sonicated and treated with uranyl acetate.



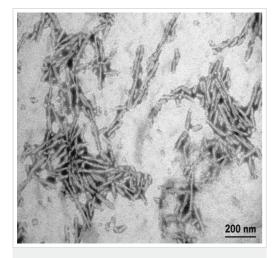
SDS-PAGE - Recombinant human Alpha-synuclein protein aggregate (Active) (ab218819)

SDS-PAGE analysis of ab218819.



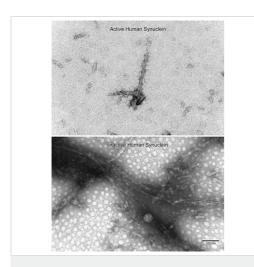
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Recombinant human Alphasynuclein protein aggregate (Active) (ab218819)

Immunohistochemistry analysis of rat brain injected with active human alpha synuclein PFFs (ab218819). Species: Female Sprague-Dawley Rat. Rat was injected with 2µL active human alpha synuclein PFFs (ab218819) 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. Alpha synuclein pathology is seen in the striatum close to an injection site.



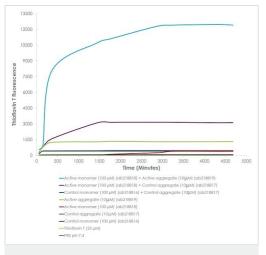
Electron Microscopy - Recombinant human Alphasynuclein protein aggregate (Active) (ab218819)

TEM of ab218819.



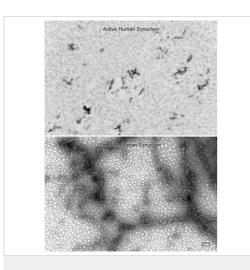
Electron Microscopy - Recombinant human Alphasynuclein protein aggregate (Active) (ab218819)

TEM of active human alpha synuclein preformed fibrils (ab218819) (top) and control (inactive) human alpha synuclein preformed fibrils (ab218817) (bottom). Fibrils were sonicated and treated with uranyl acetate. The active fibrils are shorter than the inactive fibrils.



Functional Studies - Recombinant Human Alphasynuclein protein aggregate (Active) (ab218819)

ThT emission curves show increased fluorescence (correlated to alpha-synuclein aggregation) over time when 10  $\mu$ M of active alpha-synuclein aggregate (ab218819) is combined with 100  $\mu$ M of active alpha-synuclein monomer (ab218818) (light blue), as compared to when 100  $\mu$ M of active alpha-synuclein monomer is combined with 10  $\mu$ M of control alpha-synuclein aggregate (purple line), or 100  $\mu$ M of control alpha-synuclein monomer (ab218816) is combined with 10  $\mu$ M of control alpha-synuclein aggregate (ab218817) (dark blue). ThT ex = 450 nm, em = 485 nm. View protocol.



Electron Microscopy - Recombinant human Alphasynuclein protein aggregate (Active) (ab218819)

TEM of active human alpha synuclein preformed fibrils (ab218819) (top) and control (inactive) human alpha synuclein preformed fibrils (ab218817) (bottom). Fibrils were sonicated and treated with uranyl acetate. The active fibrils are shorter than the inactive fibrils.

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