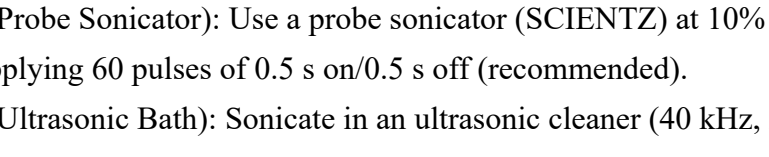
Synonym

SNCA, NACP, PARK1, alpha-Synuclein

Source

Human Alpha-Synuclein Pre-formed Fibrils, Tag Free(ALN-H5115) is expressed from E. coli cells. It contains AA Met 1 - Ala 140 (Accession # P37840-1). Predicted N-terminus: Met 1

Molecular Characterization



This protein carries no "tag".

The protein has a calculated MW of 14.5 kDa. The protein migrates as 15-16 kDa when calibrated against Star Ribbon Pre-stained Protein Marker under reducing (R) condition (SDS-PAGE).

Application

1. Sonication Conditions, Dissolution, Aliquoting, Storage, and Notes for PFF
Thawing: Thaw PFFs rapidly in a 37 °C water bath, or allow to thaw at room temperature.
Aliquoting: Since PFFs are supplied as a suspension, pipette up and down thoroughly before aliquoting to ensure homogeneity.
Storage: Store at -80 °C at all times; avoid storage at 4 °C or -20 °C, which can induce fibril depolymerization. * α-Syn fibrils cold-denatured to monomers at 0-20 °C and heat-denatured at 60-110 °C.
Sonication: The sonication protocols below are based on cell-based assay conditions.
-Option 1 (Probe Sonicator): Use a probe sonicator (SCIENTZ) at 10% power (-95 W), applying 60 pulses of 0.5 s on/0.5 s off (recommended).
-Option 2 (Ultrasonic Bath): Sonicate in an ultrasonic cleaner (40 kHz, 200-400 W) at 37 °C for 1 hour; avoid performing the treatment at 4 °C or 20 °C.
Note: The above sonication protocols are based on cell-based assays; other applications (e.g., in vivo injections) may require optimization-users should determine their optimal settings for their specific use case.
2. Recommendations for PFF Use in Animal Models
Minimize Freeze-Thaw Cycles: For animal studies, it is recommend to use the PFF at once or aliquot before use to avoid repeated freeze-thaw cycles.
PFF Maintenance During Injections: During stereotaxic injections, keep the sonicated PFFs in a 37 °C water bath to prevent re-aggregation or sedimentation.

Endotoxin

Less than 1.0 EU per μg by the LAL method / rFC method.

Purity

>90% as determined by SDS-PAGE.

Formulation

Supplied as 0.2 μm filtered solution in PBS, pH7.4 with trehalose as protectant.

Contact us for customized product form or formulation.

Shipping

This product is supplied and shipped with dry ice, please inquire the shipping cost.

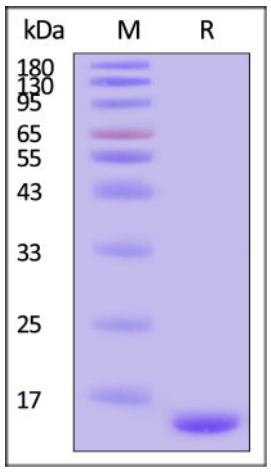
Storage

Please avoid repeated freeze-thaw cycles.

This product is stable after storage at:

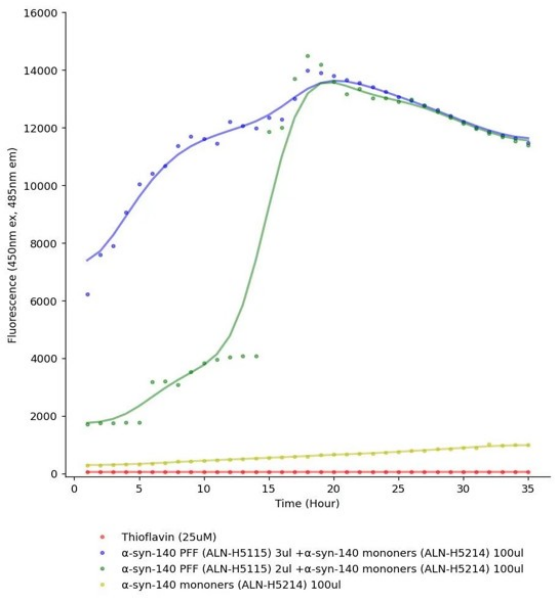
- The product should be stored at -70°C or room temperature for short storage. Do not store fibrils on ice or at 4°C;
- The unsonicated fibril is validated to be stable after storage at -70°C for 1 year under sterile conditions;
- The sonicated fibril should be stored at -70°C for not more than 8 weeks.

SDS-PAGE



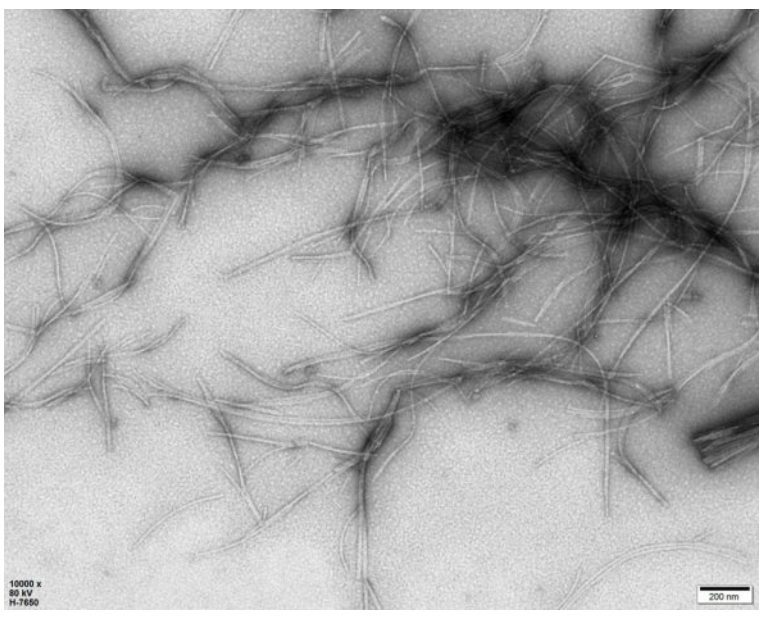
Alpha-Synuclein monomer on SDS-PAGE under reducing (R) condition. The gel was stained overnight with Coomassie Blue. The purity of the protein is greater than 90% (With Star Ribbon Pre-stained Protein Marker).

Bioactivity-ThT Assay



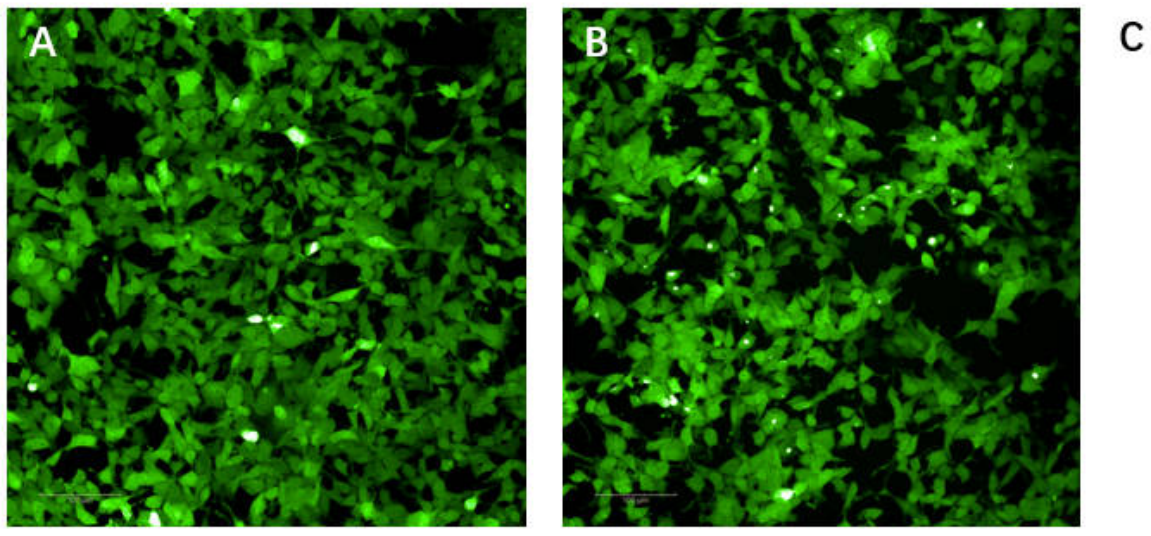
Thioflavin T emission curves show increased fluorescence (correlated to alpha-syn aggregation) over time when alpha-syn wild-type monomers (Cat. No. ALN-H5214) are combined with tau wild-type Pre-formed Fibrils (Cat. No. ALN-H5115) (Routinely tested).

Electron Microscope



Transmission electron microscopy (TEM) of Alpha-Synuclein Pre-formed Fibrils (Cat. No. ALN-H5115). Fibril structure is visible on negative stain TEM images of ALN-H5115 (Routinely tested).

Bioactivity-CELL BASE



HEK293/Human Alpha-Synuclein (GFP) Stable Cell Line (Cat. No. CHEK-ATP085) were transduced with Human Alpha-Synuclein Pre-formed Fibrils Protein, Tag Free (Cat. No. ALN-H5115). The fluorescence of GFP-Alpha-Synuclein (Green) were detected by confocal microscope. A. Lipo2000 transduction. B. Lipo2000 and Human Alpha-Synuclein Pre-formed Fibrils Protein, Tag Free (Cat. No. ALN-H5115) transduction. C. Score calculated from spot number/cell number. Scale bars, 100 μm.

Background

Alpha-synuclein is a neuronal protein that plays several roles in synaptic activity such as regulation of synaptic vesicle trafficking and subsequent neurotransmitter release. It acts also as a molecular chaperone in its multimeric membrane-bound state, assisting in the folding of synaptic fusion components called SNAREs (Soluble NSF Attachment Protein REceptors) at presynaptic plasma membrane in conjunction with cysteine string protein-alpha/DNAJC5. Abnormalities in alpha-synuclein are implicated in the pathogenesis of Parkinson's disease (PD). Alpha-synuclein is present in Lewy-bodies, the neuropathological hallmark of PD, and the protein and its aggregation have been widely linked to neurotoxic pathways that ultimately lead to neurodegeneration.