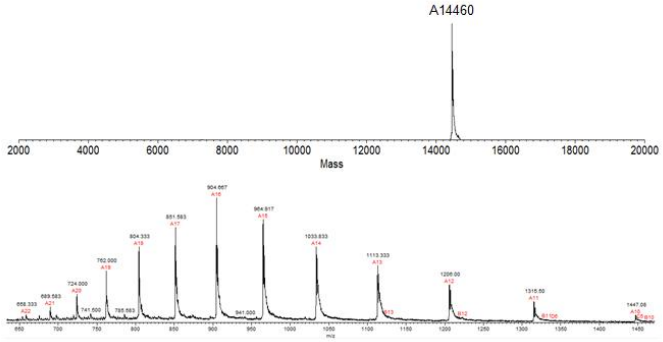
Product Datasheet

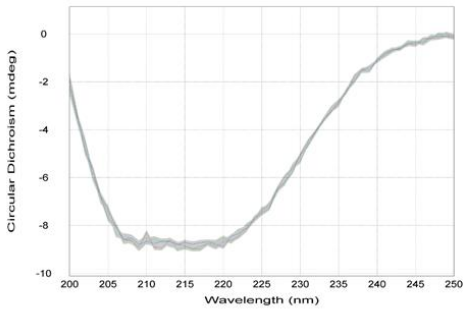
Unmodified Human α-Synuclein Oligomers

Sequence	MDVFMKGLSKAKEGVVAAAEKTKQGVAEAAGKTKEGVL YVGSKTKEGVVHGVATVAEKTKEQVTNVGGAVVTGVTA VAQKTVEGAGSIAAATGFVKKDQLGKNEEGAPQEGILED MPVDPDNEAYEMPSEEGYQDYEPEA
Swiss Prot	P37840
Gene ID	6622
Accession #	NP_000336.1
Species	Human
Amino acids	1-140, full length protein
Conjugates/Tags	No Tag
Molecular weight	14 kDa (14,460 Da)
Nature	Recombinant, expressed in Escherichia coli.
Certificate of analysis	Certified > 95 % purity by SDS-PAGE. Full characterization provided in Figure 1.
Field of Use	Not for use in humans. For research purposes only.
Applications	In vitro assays, cellular assays, animal studies or as standards in WB, SDS-PAGE, ELISA, and other immunoassays.
Form	Shipped lyophilized on dry ice.
Preparation	Protein was lyophilized in PBS. For 25 µg and 50 µg aliquots, 225 µl or 450 µl was lyophilized, respectively. We recommend resuspending in water at initial lyophilized volume to maintain salt balance.
Storage	Store at -80°C upon receipt. Following resuspension, aliquot and store at -80°C.
Handling	Protein is stable for up to 3 freeze/thaw cycles. We recommend avoiding repeated thawing cycles.
Product Citation	In case of publication or scientific presentations using this product, please cite as “Unmodified Human α-Synuclein Oligomers (ND Biosciences SA, Switzerland, Catalogue #ND002, Lot #09/20-002.004)”. Characterization data (Figure 1) remains property of ND Biosciences and is not to be used in any publications without written permission from ND Biosciences.
Safety measures	This product is an active protein and may elicit a biological response in vivo, handle with caution.
References	Kumar ST, Donzelli S, Chiki A, Syed MMK, Lashuel HA. A simple, versatile and robust centrifugation-based filtration protocol for the isolation and quantification of α-synuclein monomers, oligomers and fibrils: Towards improving experimental reproducibility in α-synuclein research. J Neurochem. 2020;153(1):103-119. doi:10.1111/jnc.14955.

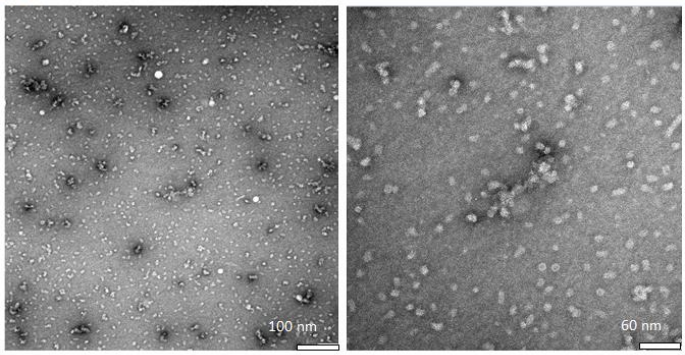
(A)


A14460
Mass

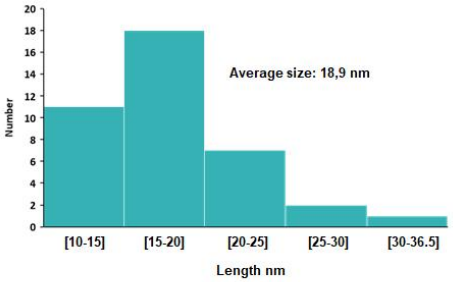
(B)


Circular Dichroism (mdeg)
Wavelength (nm)

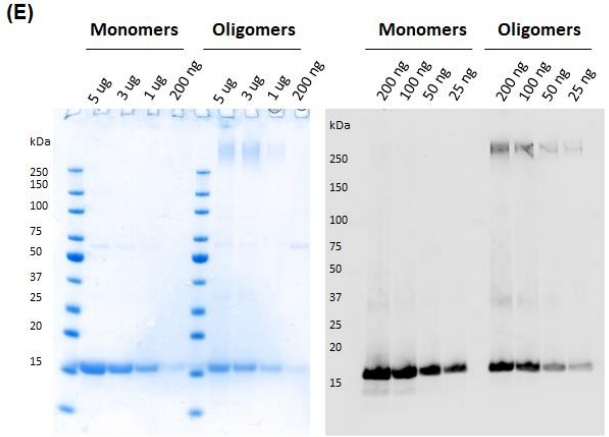
(C)


100 nm
60 nm

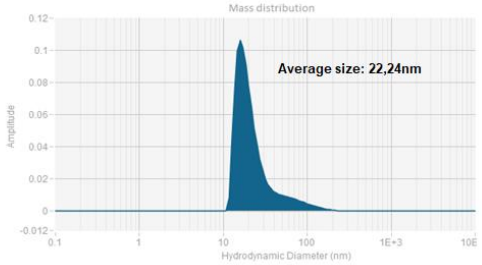
(D)


Average size: 18,9 nm
Number
[10-15]
[15-20]
[20-25]
[25-30]
[30-36.5]
Length nm

(E)


Monomers
Oligomers
Monomers
Oligomers
5 ug
3 ug
1 ug
200 ng
200 ng
100 ng
50 ng
25 ng
kDa

(F)


Mass distribution
Average size: 22,24nm
Amplitude
Hydrodynamic Diameter (nm)

(G)

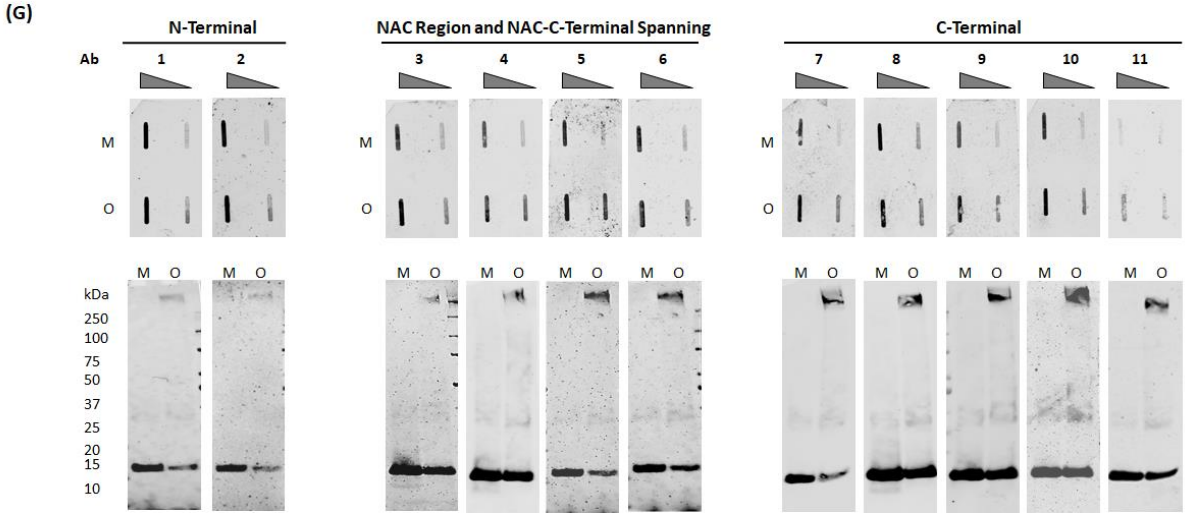

N-Terminal
NAC Region and NAC-C-Terminal Spanning
C-Terminal
Ab
M
O
kDa

Figure 1. Characterization of α-Syn oligomers. (A) Mass spectrometry analysis confirms the integrity of α-Syn protein used to generate oligomers, and shows a mass of ~14460 Da. **(B)** Circular dichroism (CD) analysis shows a spectrum with a minimum at 217 nm, establishing β-sheet conformation of α-Syn oligomers. **(C)** Transmission electron microscopy (TEM) of uranyl acetate stained α-Syn oligomers shows a population of annular pores and spherical/tubular-like particles. **(D)** The size distribution of oligomers visualized by TEM is between 10 and 36.5 nm, with an average size of 18.9 nm. **(E)** Coomassie staining (left panel), and western blot analysis using the 610787 antibody (Syn1, right panel) at different loaded amounts shows that α-Syn oligomers migrate as SDS-resistant high molecular weight (HMW) species (>250kDa), with some oligomers breaking down to monomers at ~14 kDa. Monomeric α-Syn was loaded at similar amounts as control. **(F)** Dynamic light scattering analysis of α-Syn oligomers shows a homogenous population in terms of size distribution, with average size of 22.24 nm. **(G)** Screening of reactivity of α-Syn oligomers with a panel of antibodies targeting the N -terminus, NAC region and C terminus of α-Syn. α-Syn monomers were included as controls. Upper panel: Slotblots of α-Syn monomers (M) and oligomers (O) at two concentrations (200 and 25 ng). Lower panel: Western Blots of 100 ng of α-Syn monomers (M) and oligomers (O). Antibodies: Ab1 (Abexa Abx013202, Epitope: 15-64), Ab2 (ABnostics AB-0904, Epitope: 44-57), Ab3 (Cosmobio SNP-08, Epitope 75-91), Ab4 (Biolegend 848302, Epitope: 80-96), Ab5 (Invitrogen PA5-13397, Epitope: 75-104), Ab6 (Saint John's laboratory STJ115317, Epitope: 61-140), Ab7 (Biolegend 838201, Epitope: 87-110), Ab8 (BD Biosciences 610787, Epitope: 91-99), Ab9 (clone 2F12, Epitope: 125-135), Ab10 (clone SOY-1, Epitope: 91-100), Ab11 (clone 1H9, Epitope: 131-140).

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