SynAging

Au coeur de la mémoire.

α-Synuclein Aggregate Preparations Induce Neurodegeneration and Cognitive Decline: A Novel Model for Parkinson's Disease

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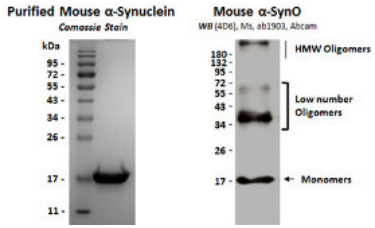
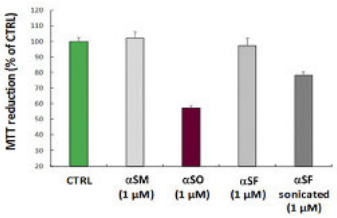
- α-synuclein pathology is clearly linked to Parkinson's disease (PD) and related dementia
- PD drug discovery needs translational in vitro and in vivo models that are recapitulating sporadic disease onset
- Here, we report novel models based on neurodegeneration induction by minute amounts of highly reproducible α-synuclein oligomer (αSO) or fibrillar (αSF) preparations
- αSO and αSF induce dose and time-dependent neurodegeneration in primary mouse neurons and iPS cell-derived human neuronal cultures
- Neurodegeneration is more pronounced for αSO than αSF and stronger on striatal neurons than hippocampal neurons
- αSO/F-induced neurodegeneration is dose dependently attenuated by brain-derived neurotrophic factor, providing a positive control for assays
- A single striatal intra-striatal injection of αSO or αSF induce cognitive decline in the NOR assay. Deficits start at day 90 for αSF, and after day 15 and up to the maximum investigated time period (3 months) for αSO, indicating neurodegeneration in the perirhinal cortex. However, up to three month, no deficit is detected in the pre-frontal cortex based Y-maze assay.
- αSF induces clear and progressing α-synuclein pathology after day 15, whereas αSO does not induce α-synuclein pathology within three month

Aggregates of α-Synuclein Proteins



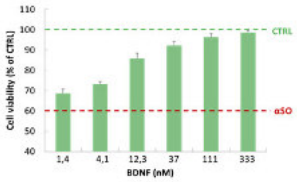
- Mouse and human α-synuclein are produced and purified at large scale to >97% purity (left figure).
- Monomers are treated to generate oligomers (αSO) and fibrils (αSF, not shown) in an highly reproducible manner. Oligomers are stable in SDS PAGE and can be detected by monomer directed antibodies (right figure).

αSO/F-Induced Neurodegeneration in Mouse Striatal Primary Neurons



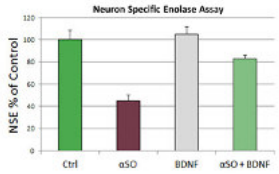
- Primary mouse striatal neurons are grown in 48-well plates
- At day 10, medium is supplemented with α-synuclein monomers (αSM), oligomers (αSO), or fibrils (αSF)
- Neurons are incubated for 72 h
- Read-out: MTT assay

BDNF Dose-Dependently Rescues αSO-Induced Neurodegeneration



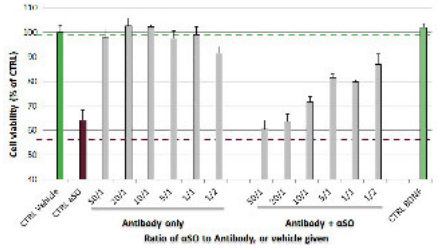
- Primary mouse striatal neurons were grown in 48-well plates, in triplicates.
- At day 10, increasing concentrations of BDNF were added to the medium
- Neurons were challenged for 72h with αSO
- Read-out: MTT assay

αSO-Induced Neurodegeneration in IPS Cell-Derived Human Neurons



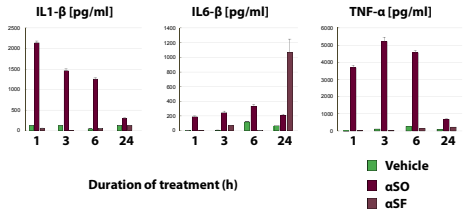
- Human neuronal cultures, containing approximately 40% neurons and astroglia, were differentiated from iPS cells for five weeks.
- The culture was treated for 72h with 0.3μM human αSO and/or BDNF
- Survival readout by ELISA for a neuron specific marker: Neuron Specific Enolase (NSA)

BDNF and αS-Antibodies Prevent αSO- Induced Neurodegeneration



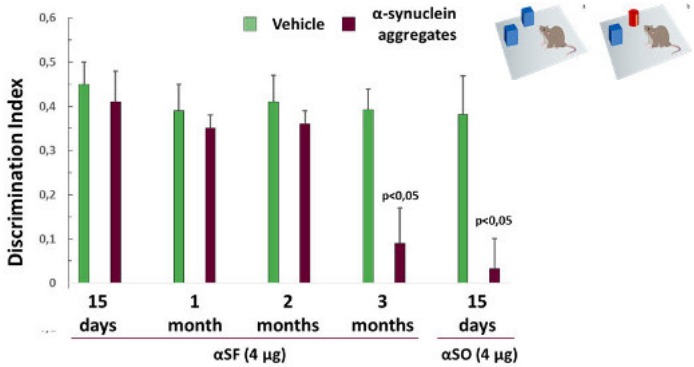
- Primary mouse striatal neurons are grown in 48-well plates
- At day 10, medium is supplemented with vehicle (left group), or αSO (right group)
- Different dilutions of α-synuclein antibodies were used to treat neurons
- A clear dose dependent rescue of the antibodies was observed

αSO and αSF Induce Different Immune Response in Mouse Astrocytes

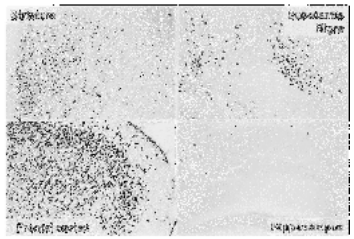


- Mouse primary cortical astrocytes are cultured in 6-well plates
- Treatment by 10 μM α-synuclein aggregates (conc. based on monomer) starts at DIV 6 and are harvested at the indicated times.
- Treatments are performed in triplicates
- Read-out: ELISA assays in conditioned medium

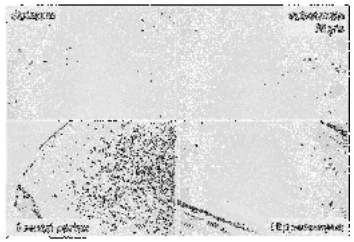
α-Synuclein-Aggregate Induced Cognitive Deficit in the Novel Object Recognition Assay



A Single αSF Injection into the Striatum induced spreading in Mice



- 15 weeks old male C57BL/6 mice were injected intra-striatal at day 0 with mouse α-synuclein fibrils
- Top figures: 'Spreading' of α-synuclein aggregates is shown for the **ipsilateral** side after 60 days for striatum, substantia nigra, frontal cortex and hippocampus
- 'Spreading' was already seen 15 days after a single αSF injection into the striatum and extended continuously until the maximal investigated period of 90 days
- αSO injection into the striatum or hippocampus did not result in any 'spreading' until the maximal investigated period of 90 days



- Bottom figures: 'Spreading' of α-synuclein aggregates is shown for the **contralateral** side after 60 days for striatum, substantia nigra, frontal cortex and hippocampus
- Whereas 'spreading' into to contralateral frontal cortex was strong, almost not transfer to the contralateral side was obtained for the striatum and the substantia nigra
- Spreading into the hippocampus was very sparse, similarly in both, the ipsilateral and contralateral side

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