PRECLINICAL NEURONAL MODELS TO SCREEN THE EFFECTIVENESS OF DRUGS DIRECTED TO SLOW DOWN ALZHEIMER'S AND PARKINSON'S DISEASE PROGRESSION

JOLIEN BEEKEN[1,2], ANAÏS LUYKX[1], TEUN VAN NUNEN[1], YANICK FANTON[3]

[1] Neurology research group, InnoSer Belgium
[2] Study Director Neurology and Cardiometabolic diseases, InnoSer Belgium
[3] Chief Scientific Officer, InnoSer Belgium
Email: jbeeken@innoserlaboratories.com

InnoSer

VIEW OUR NEUROLOGY PORTFOLIO HERE

THE NEED FOR MORE REPRESENTATIVE PRECLINICAL MODELS

Alzheimer's and Parkinson's disease drug development is held back due to lack of in vitro reproducible models that represent human complexity and can be used to screen disease-modifying therapeutics in a relatively inexpensive, efficient, and fast fashion.

Here, we present validated cellular models of neurodegenerative diseases to enable efficient screening of disease-modifying candidate compounds for Alzheimer's and Parkinson's diseases prior to, or in conjunction to screening in animal models.

CELLULAR MODELS OF ALZHEIMER'S AND PARKINSON'S DISEASE

Microglial (HMC3) and differentiated neuronal-like cells (SH-SY5Y) were treated with pre-formed amyloid beta (Aβ) or alpha-synuclein (αSyn) fibrils to model the pathophysiology of neurodegeneration in vitro (**Figure 1**). Using these cellular models, common hallmarks of neurodegeneration such as aggregation, neurotoxicity, ROS production and phagocytosis can be screened or tested for disease-modifying drugs (**Figure 1**).

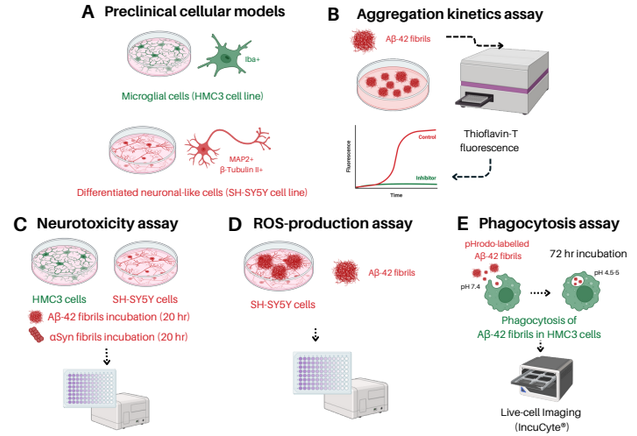

FIGURE 1. Visualisation of the validated cellular models of Alzheimer's and Parkinson's Disease. Neurotoxicity, ROS production and phagocytosis have been shown to play a crucial role in the progression of neurodegenerative diseases; this can be modelled using various in vitro assays.

VALIDATION OF CELLULAR MODELS OF ALZHEIMER'S AND PARKINSON'S DISEASE

For all experiments, microglial and/or differentiated SH-SY5Y cells were used (**Figure 2**). Prior to evaluating the effect of Aβ-42 fibrils in the cellular models, the aggregation kinetics were confirmed (**Figure 2**). Fibrillar Aβ-42 (**Figure 4**), and αSyn induce cell toxicity in both neuronal and microglial cells.

Aβ fibrils induce ROS production in neuronal cells, which can be rescued by co-treatment with Edaravone (**Figure 5**). Aβ-42 fibrils induce an increase in microglial phagocytic capacity that increases upon Aducanumab co-treatment (**Figure 6**).

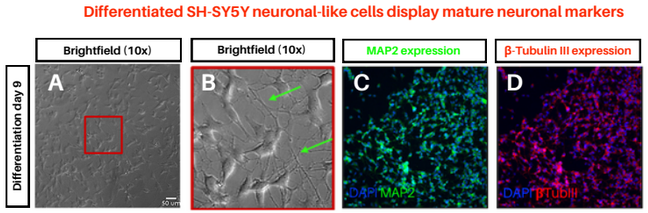

FIGURE 2. SH-SY5Y cells differentiate into mature neuronal-like cells, confirming their suitability in disease modelling. Differentiated neuronal-like SH-SY5Y cells show a mature phenotype with extensive and elongated neuritis outgrowths (green arrows), form neurite clusters (**A, B**) and express more mature neuronal markers (**C**) MAP2 (**D**) β-Tubulin III, confirming their use in all subsequent assays.

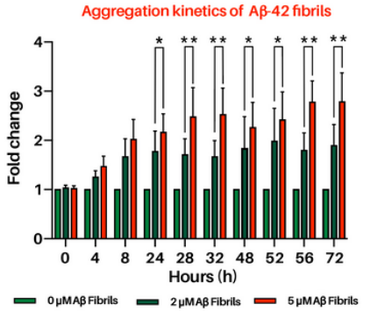

FIGURE 3. The Aggregation assay confirms Aβ-42 fibril aggregation kinetics in vitro, confirming the fibrils suitability in subsequent assays. The formation of fibril aggregates is monitored by fluorescence resulting from binding of Thioflavin T (ThT) to the aggregates. 24-hour incubation of Aβ-fibrils with the reporter ThT (20 μM) leads to significant increases in Aβ-42 fibril aggregation (M± SEM; n = 6 per condition; $*P<0.05$; $**P<0.01$).

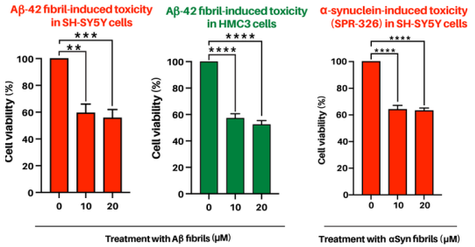

FIGURE 4. The Aβ-induced and α-synuclein-induced neurotoxicity in vitro assay can be used to test novel Alzheimer's and Parkinson's disease compounds' efficacy in rescuing cell viability. 24-hour treatment with Aβ-42 and α-synuclein fibrils induces toxicity in SH-SY5Y and HMC3 cells compared to the control condition (M± SEM; n=4 per condition; $**P=0.005$; $***P=0.001$; $****P<0.0001$).

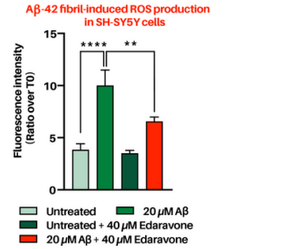

FIGURE 5. The ROS assay can be used to evaluate compounds aimed at decreasing ROS production. Significant increase in ROS production induced by Aβ-42 fibrils can be rescued by Edaravone (M±SEM; n=5 per condition; $*P<0.05$; $****P<0.0001$).

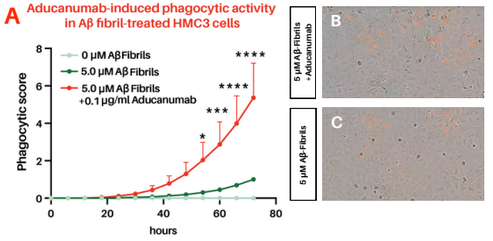

FIGURE 6. The Aβ-induced phagocytosis assay can be used to test novel Alzheimer's disease compounds' efficacy in stimulating the clearance of Aβ fibrils. (**A**) Aβ-42 fibril treatment induces significant increase in HMC3 cells' phagocytic capacity in comparison to no fibril treatment. Simultaneous treatment of 0.1 μg/ml Aducanumab and 5.0 μM fibrils (**B**) induces a significant increase in phagocytic capacity in comparison to 5.0 μM fibril treatment alone (**C**). (M± SEM; n=3 per condition; $*P<0.05$, $**P<0.005$, $***P<0.001$, $****P<0.0001$).

IMPLICATIONS FOR PRECLINICAL RESEARCH

We show that our cellular neurodegenerative disease models can serve as a highly efficient tool for testing multiple candidate test compounds before screening in vivo. As a preclinical CRO, InnoSer continuously works to expand its range of in vitro and in vivo disease offering services.

Visit us at booth #22 to discuss this poster in detail!

Contact us at: info@innoserlaboratories.com
Visit our page at: www.innoserlaboratories.com/neurology