A synucleinopathy rat model as a tool to investigate synaptic dysfunction and neuroinflammation in Parkinson's Disease

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Background

Parkinson's Disease (PD) is the second most common neurodegenerative disorder, is characterized by the loss of dopaminergic (DA) neurons and the presence of α-synuclein-containing aggregates in the substantia nigra [1]. Evidence continues to support that Parkinson's disease (PD) is an alpha-synucleinopathy [2]. Deposition of alpha-synuclein (αSyn) is a defining pathological feature of PD and is increasingly being understood to be involved in synaptic dysfunction [2,3]. Although the pathogenesis of Parkinson's Disease (PD) is extremely complex, several studies suggest that neuroinflammation potentially contributes to impairments in synaptic plasticity, a crucial feature in the onset and progression of motor and non-motor symptoms of PD [4,5].

The preformed fibril (PFF) synucleinopathy model in rodents recapitulates the molecular and nigrostriatal pathology associated with PD, as well as the progressive decline in dopaminergic function. However, its use as a tool to examine synaptic dysfunction and neuroinflammation in PD has not been deeply explored.

Methods

We analysed the progression of synaptic pathology by using a PFF model in female rats and evaluated endpoints at three-time points following injection of mouse alpha-synuclein-PFF (αSyn-PFF) or αSyn-monomer (control) into the striatum (Day 30, Day 60, and Day 120, all groups n=8).
Using Multiplex Infrared Western Blotting (IFWB), Li-Cor Odyssey CLX relative levels of synaptic markers (Pre-Synaptic: Synaptophysin; Post-Synaptic: PSD-95 and Spinophilin) were measured in the ventral midbrain and hippocampus. Additionally, we carried out a preliminary analysis of inflammasome activation by measuring the expression of NLRP3 in the striatum of control and αSyn-PFF animals at Day 30 and Day 120. T-tests were used to determine statistical differences between the groups.

Results

PSD-95 expression is reduced in the αSyn-PFF group

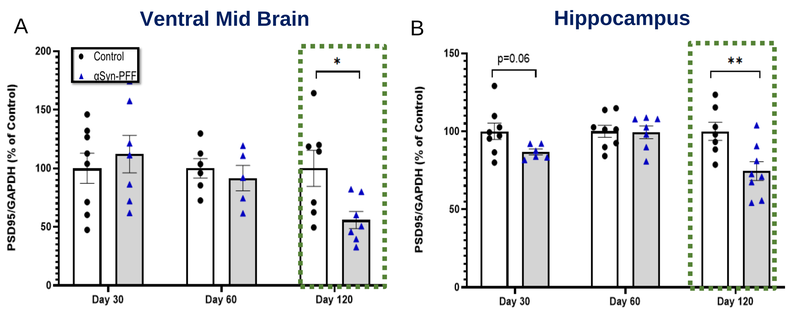

Figure 1. A) Ventral Mid Brain: PSD-95 showed a significant decrease at Day 120 in the αSyn-PFF group same group when compared against control group. B) Hippocampus: a trend toward a decrease in the αSyn-PFF group at Day 30 and a significant decrease at Day 120. Data are expressed as mean ± SEM, n= 5-8 per group. T-test *p<0.05,

Spinophilin expression is altered in the αSyn-PFF group

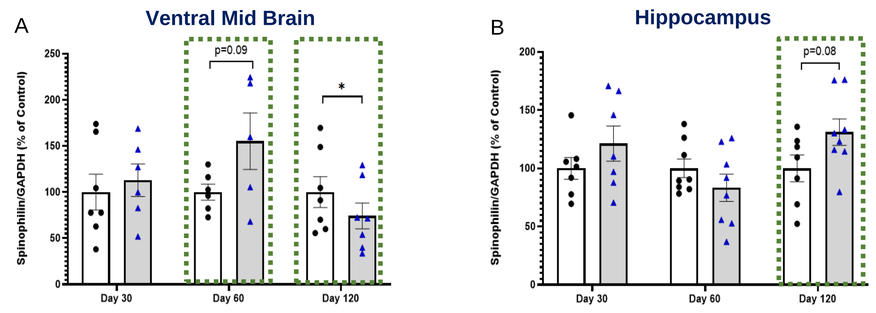

Figure 2. A) Ventral Mid Brain: There was a trend toward an increase at Day 60 in the αSyn-PFF group, while there was a significant decrease at Day 120. B) Hippocampus: there was a trend toward an increase in Spinophilin at Day 120. Data are expressed as mean ± SEM, n= 5-8 per group. T-test *p<0.05,

Synaptophysin expression is reduced in the αSyn-PFF group

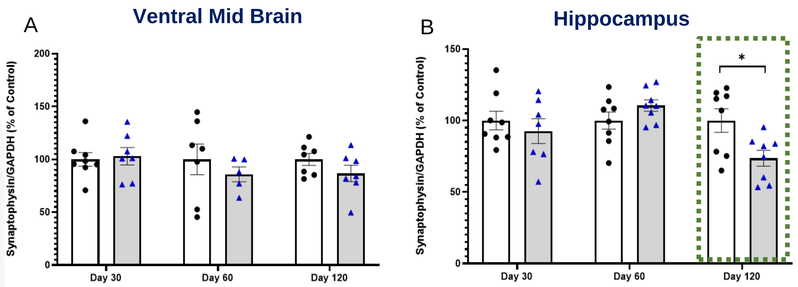

Figure 3. A) Ventral Mid Brain: No significant changes were observed in the expression of Synaptophysin between groups at any time point B) Hippocampus: There was a significant decrease in αSyn-PFF group at Day 120.Data are expressed as mean ± SEM, n= 5-8 per group. T-test *p<0.05,

Increase in striatal NLRP3 at Day 30 post PFF administration

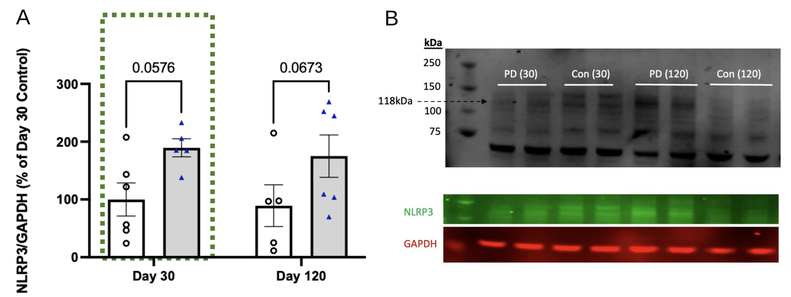

Figure 4 A) Preliminary analyses found an increase in striatal NLRP3 in αSyn-PFF group at Day 30 post PFF administration (p=0.0576) compared to control group. B) Representative WB image. Data are expressed as mean ± SEM, n= 5-6 per group. T-test *p<0.05,

Conclusion

There are significant alterations in the synaptic markers in both the ventral midbrain and hippocampus, notably at day 120 post-injection, suggesting altered synaptic plasticity in this model. In PD, it is hypothesised that synaptic dysfunction occurs prior to neurodegeneration and the possibility of identifying synaptic changes may become a promising tool in the quest for new therapeutic strategies for PD.
These results begin to form the basis of a preclinical platform using the PFF rat model to identify novel biomarkers of synaptic integrity and neuroinflammation in PD.

References : 1] Lebouvier, T., Chaumette, T., Paillusson, S., Duyckaerts, C., Bruley des Varannes, S., Neunlist, M., & Derkinderen, P. (2009). The second brain and Parkinson's disease. *The European Journal of Neuroscience*, *30*(5), 735–741. [2] Gómez-Benito, M., Granado, N., García-Sanz, P., Michel, A., Dumoulin, M., & Moratalla, R. (2020). Modeling Parkinson's Disease With the Alpha-Synuclein Protein. *Frontiers in Pharmacology*, *11*(356). [3] Picconi, B., Piccoli, G., & Calabresi, P. (2012). Synaptic dysfunction in Parkinson's disease. *Advances in Experimental Medicine and Biology*, *970*, 553–572. [4] Cardinale, A., Calabrese, V., de Iure, A., & Picconi, B. (2021). Alpha-Synuclein as a Prominent Actor in the Inflammatory Synaptopathy of Parkinson's Disease. *International Journal of Molecular Sciences*, *22*(12), 6517. [5] Chu, H.-Y. (2020). Synaptic and cellular plasticity in Parkinson's disease. *Acta Pharmacologica Sinica*, *41*(4), 447–452.