IGF-II treatment prevents the oxidative damage derived by MPP+/MPTP administration in a cellular and animal model of Parkinson's disease

Nº4050

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Introduction

Oxidative distress and mitochondrial dysfunction are key components from the pathophysiology of different neuronal disorders such as Parkinson's disease (PD) [1]. The pleiotropic hormone insulin-like growth factor II (IGF-II), which is highly distributed throughout the brain, has proven to have a neuroprotective and antioxidant role in some neurodegenerative diseases [2]. One of the many implication of the interaction between IGF-II and its specific receptor is its involvement in the sphingosine kinase (SPHK) pathway [3].

In this work, we analyze the role of IGF-II in the mitochondrial cytoarchitecture and function, and the implication of the SPHK pathway, regarding the oxidative damage evoked by the administration of 1-methyl-4-phenylpyridinium (MPP^+) and its effects in a PD animal model.

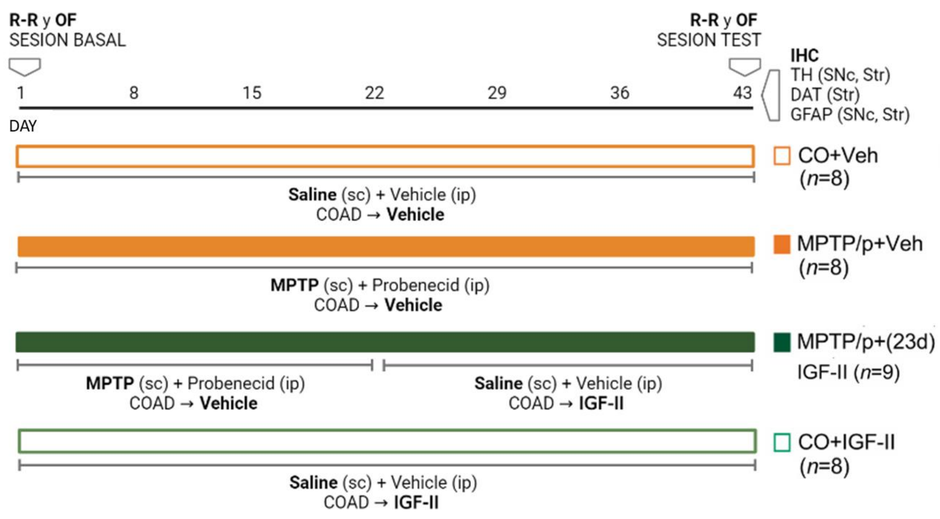
Materials and methods

CELLULAR MODEL

- Mitochondrial oxygen consumption rate. Commercial Seahorse XF cell Mito Stress test kit (Agilent Technologies, USA).
- Electron microscopy. The samples were observed with a FEI NOVA NanoSEM 450. Mitochondria were analyzed using Image J software considering six different mitochondrial indexes.
- Lactate dehydrogenase (LDH) levels. As a Cytotoxicity and cell death indicator, LDH levels were measured with a colorimetric reader (Shenzhen iCubio Biomedical Technology Co., Ltd).

ANIMAL MODEL

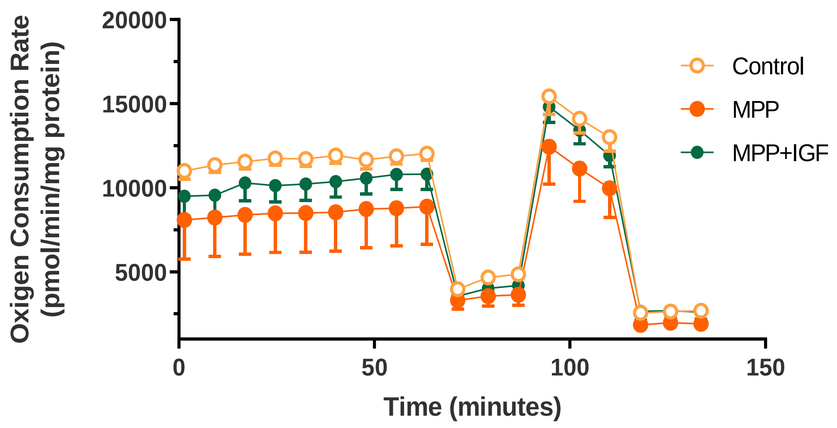
- Experimental design. 5-week administration regime.

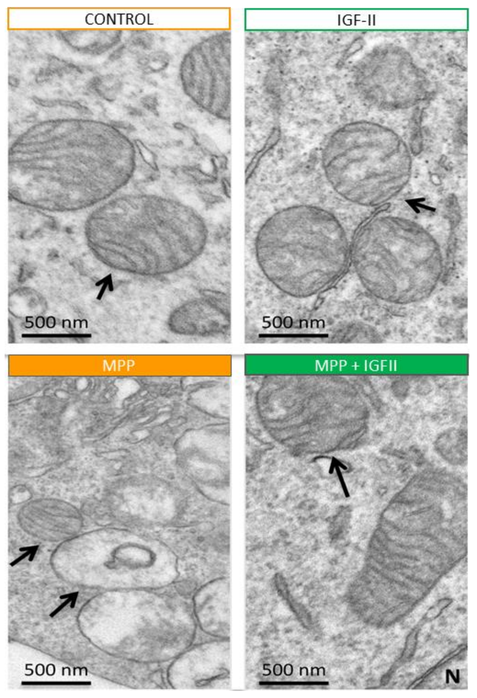


- Immunohistochemistry for astrogliosis and dopaminergic markers. TH, DAT, and glial fibrillary acidic protein (GFAP) diaminobenzidine (DAB) immunostaining was performed on striatal and SN sections.
- Behavioral tests. Motility ability was examined via self-grooming (total time) and rotarod (latency to fall).
- Data análisis. One-way ANOVA tests using GraphPad Prism 9.5.1. Statistical significance is defined by $p<0.05$. Data is expressed as Mean±SEM. Pairwise comparisons were performed using a post hoc Newman-Keuls comparison test.

1 Mitochondrial structure and function

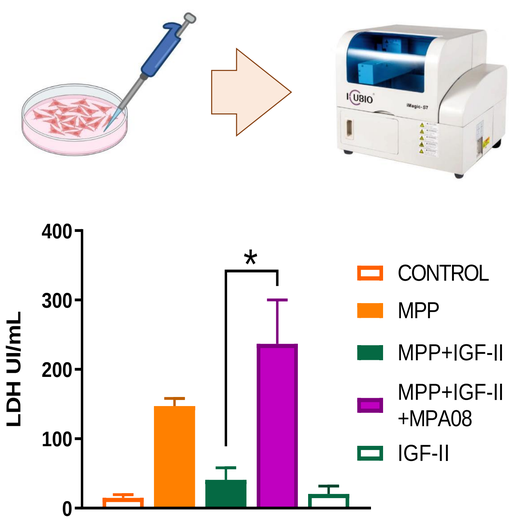
Ultrastructural analyses indicate that IGF-II treatment can attenuate the deleterious mitochondrial effects induced by MPP^+. We observed a 28% decrease in OCR in MPP^+ treated cells compared to control. Incubation of MPP^+ treated cells in presence of IGF-II reduce the OCR decrease to only 13% showing a protective effect on the damage induced by MPP^+.





2 Sphingosine kinase pathway

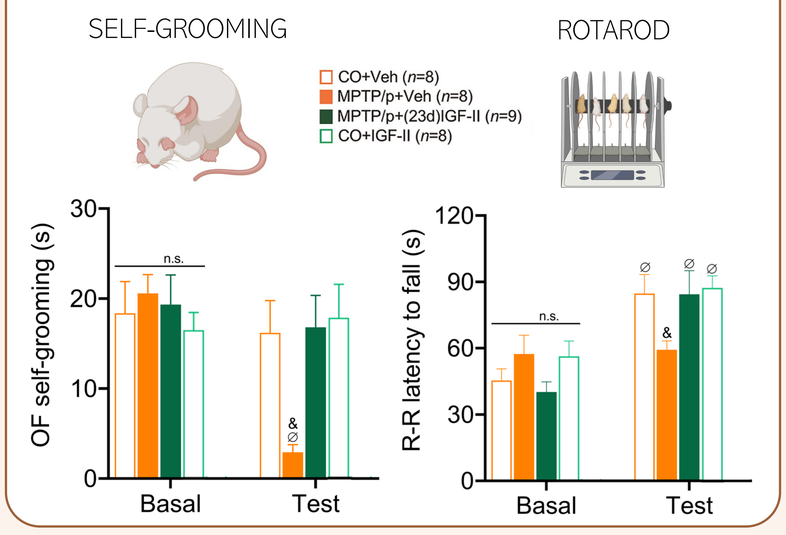
The co-incubation of MPP^+ with IGF-II reverts the damage evoked by the toxin. However, when the SPHK pathway is inhibited with a specific inhibitor (MPA08) the damage is present.



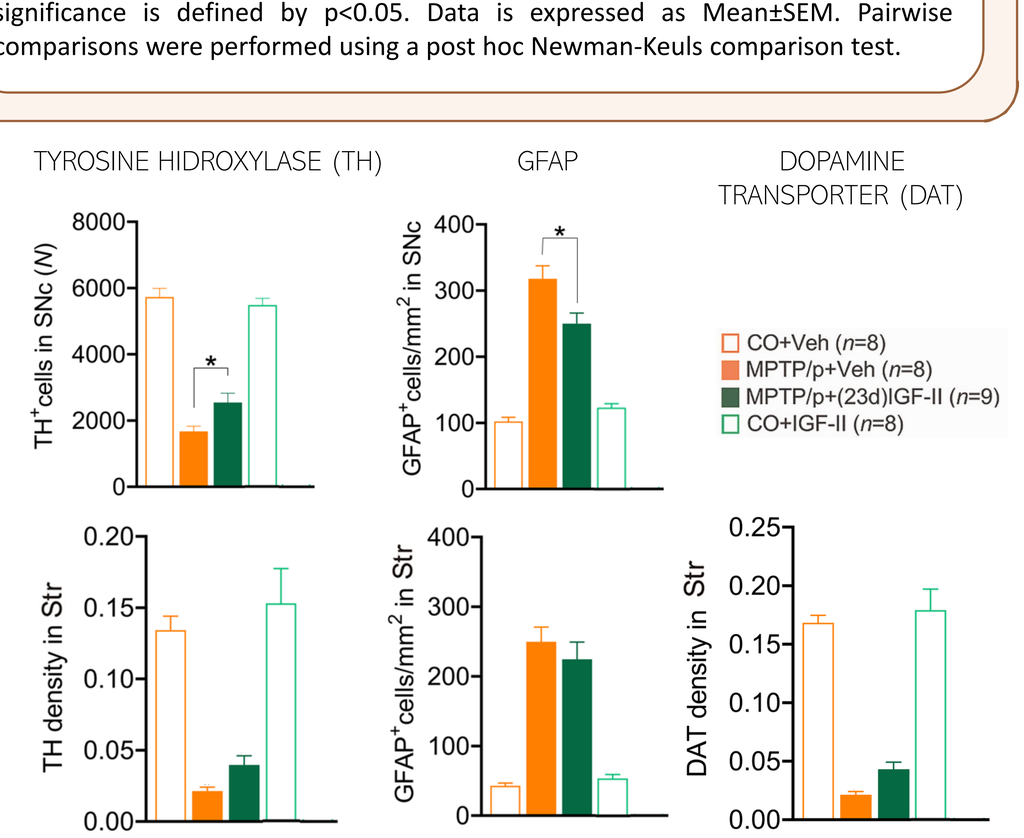
3 Behavioural tests

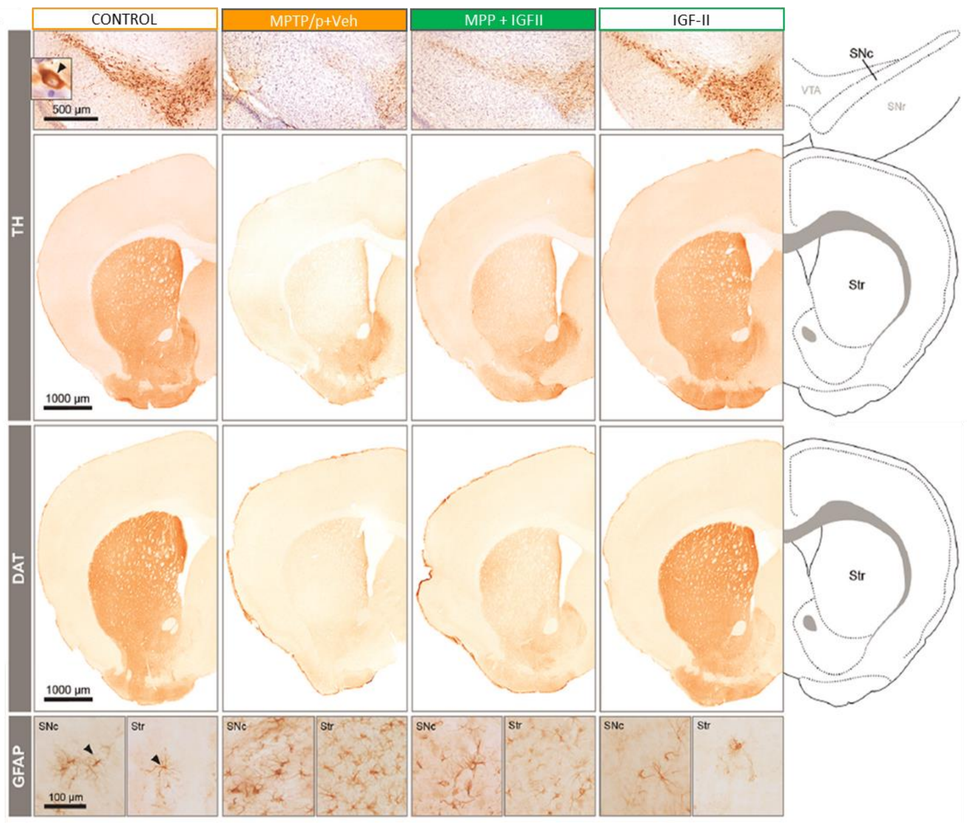
Motility impairment caused by the administration of MPTP/p is counteracted by the treatment of IGF-II, in both self-grooming (open field) and rotarod test.

There were no significant differences in the basal test performance carried out at day 1.



4 Immunohistochemistry for dopaminergic markers





We studied whether IGF-II treatment could halt the neurodegeneration of the dopaminergic nigrostriatal pathway.

To do this, we treated a cohort of mice with IGF-II, starting 3 weeks after the first dose of MPTP/p.

IGF-II mitigated the loss of dopaminergic neurons in the SNc ($p<0.05$). IGF-II coadministration also proved a tendency to recover the reduced expression of TH and DAT in the dopaminergic terminals of the striatum ($P < 0.05$). The overexpression of $GFAP^+$ astrocytes in the SNc was reduced significantly when IGF-II was also administered.

Conclusion

IGF-II counteracts the increased oxidative stress and mitochondrial dysfunction induced by the MPP^+ neurotoxin, the behavioral impairment, and the DA-NSP degeneration. Sphingosine kinase appears to be involved in this mechanism.

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[2] A. Beletskiy, E. Chesnokova, N. Bal, Insulin-like growth factor 2 as a possible neuroprotective agent and memory enhancer—its comparative expression, processing and signaling in mammalian CNS, Int. J. Mol. Sci. 22 (2021) 1849, https://doi.org/10.3390/ijms22041849.
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