

Neurodegeneration in the systemic rotenone model of Parkinson's disease is associated with neuroinflammation, oxidative stress and gliosis

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Introduction

Parkinson's disease (PD) is the most common movement disorder, estimated to affect more than 8.5 million individuals globally. Motor symptoms of PD include bradykinesia, tremor and postural instability, and non-motor symptoms include cognitive impairment. The primary pathophysiological feature of PD is degeneration of dopaminergic neurons in the substantia nigra (SN). This neurodegenerative process is associated with phosphorylation of alpha-synuclein and accumulation of protein aggregates, neuroinflammation, oxidative stress and gliosis. This study characterised the systemic rotenone rat model of PD based on Cannon et al 2009¹, investigating the temporal progression of motor symptoms and neuropathology.

Aim:

1) To assess the effect of systemic rotenone administration for 1, 5 and 10 days on motor behaviour, neurodegeneration, gliosis, and oxidative stress in the rat

Methods

Animals and treatments: Male Lewis rats (n=8-10/group) received daily administration of rotenone by intraperitoneal injection (3.25mg/kg) for 1, 5 or 10 days.

Behavioural testing: Motor assessments (locomotor activity, postural instability and beam walk tests) were performed at baseline and on the final day of treatment prior to tissue collection.

Immunohistochemistry (IHC): Tyrosine hydroxylase (TH) positive cells were quantified in the SN by 3,3'-Diaminobenzidine (DAB) IHC. Microglia (Iba1 positive cells) were analysed by immunofluorescence.

Dopamine quantification: Dopamine (DA) concentrations in left striatal tissue samples were determined by Charles River Labs (Finland) using high performance liquid chromatography (HPLC) method with electrochemical detection.

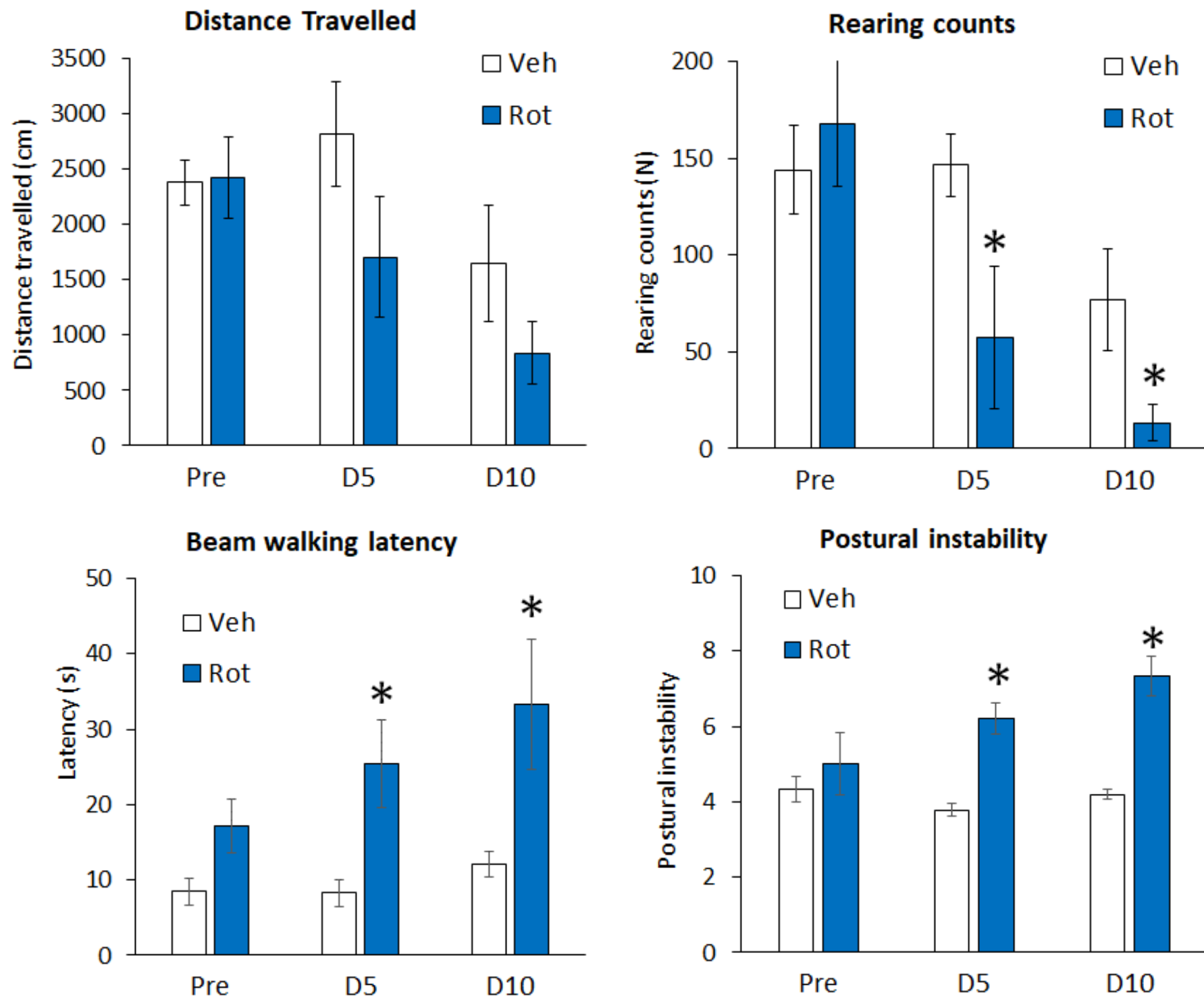
MesoScale Discovery: TNF-alpha, IL-6, IL-1beta were quantified in brain homogenate (left hemisphere). Neurofilament light chain (NfL) in fluids was analysed as a marker of neurodegeneration.

Oxidative stress: Lipid peroxidation was measured by 4-HNE quantification.

References

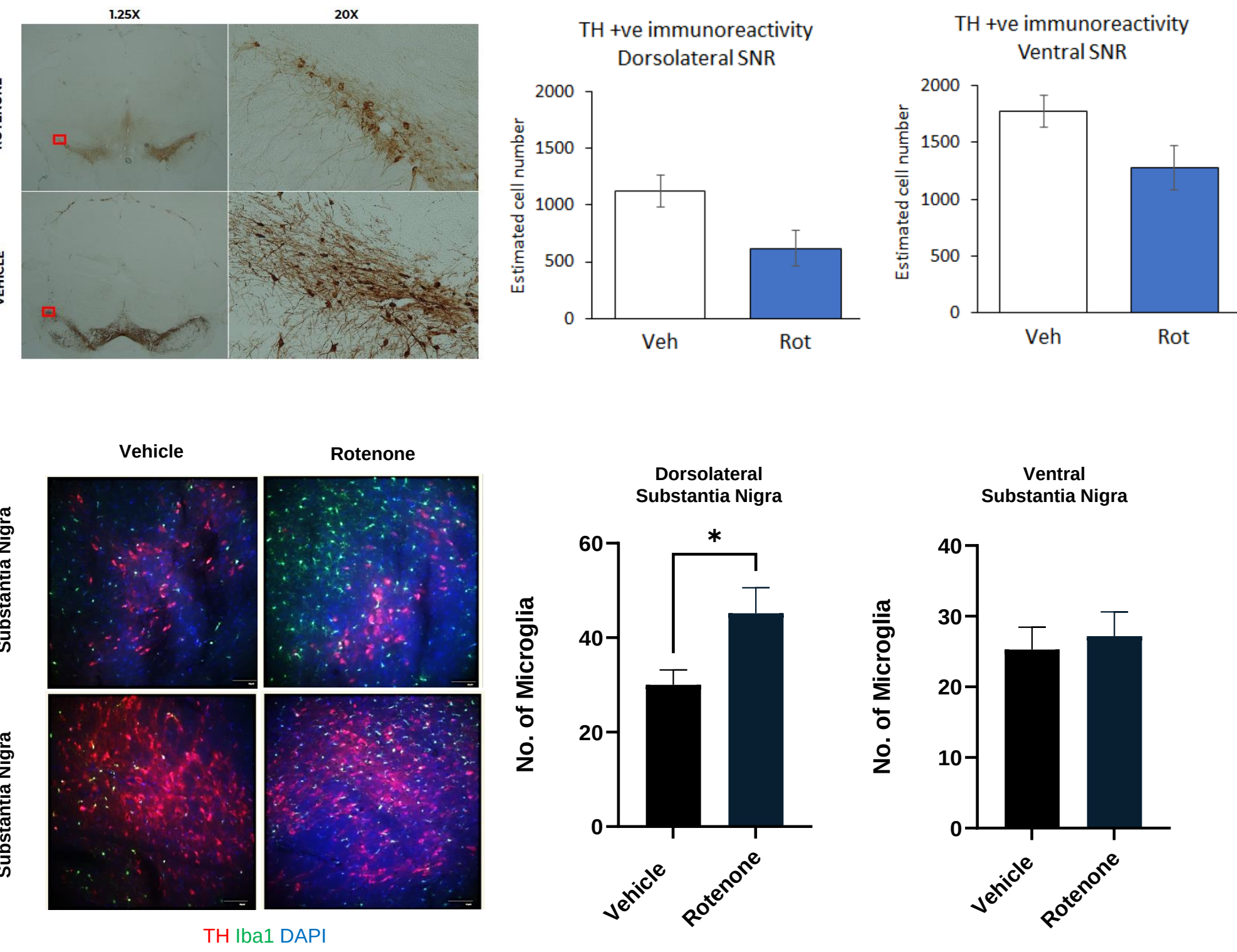
¹ Cannon JR, Tapias V, Na HM, Honick AS, Drolet RE, Greenamyre JT. A highly reproducible rotenone model of Parkinson's disease. Neurobiol Dis. 2009 May;34(2):279-90. doi: 10.1016/j.nbd.2009.01.016. PMID: 19385059; PMCID: PMC2757935.

Rotenone induces motor deficits



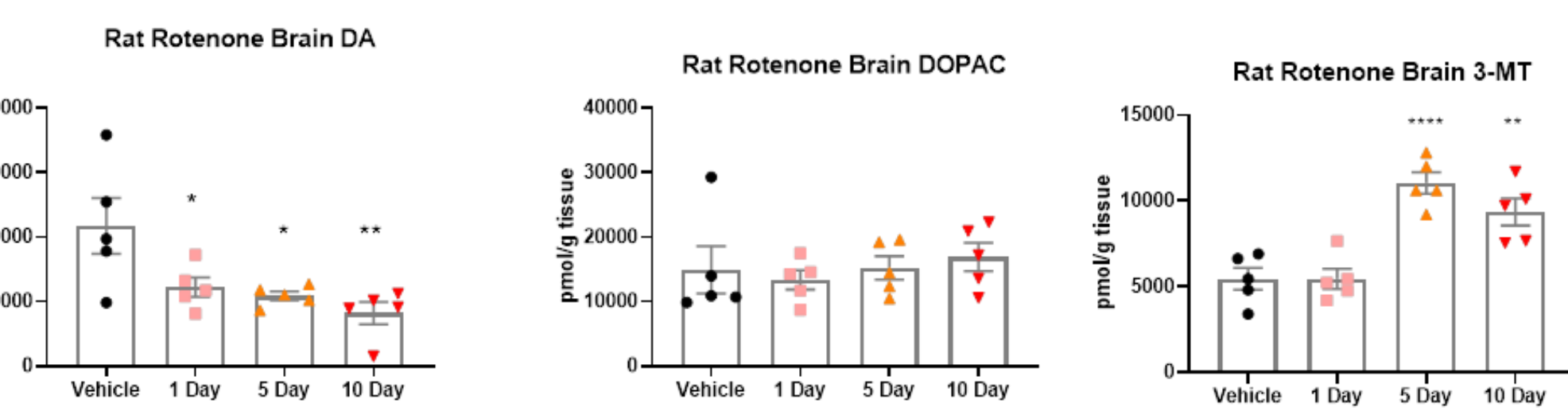
5- and 10- day treatment induced motor deficits in the open field (rearing counts), beam walking and postural instability tests. *p<0.05 vs. vehicle, Mean ± SEM, n=9-10 per group.

Rotenone induces neurodegeneration and gliosis



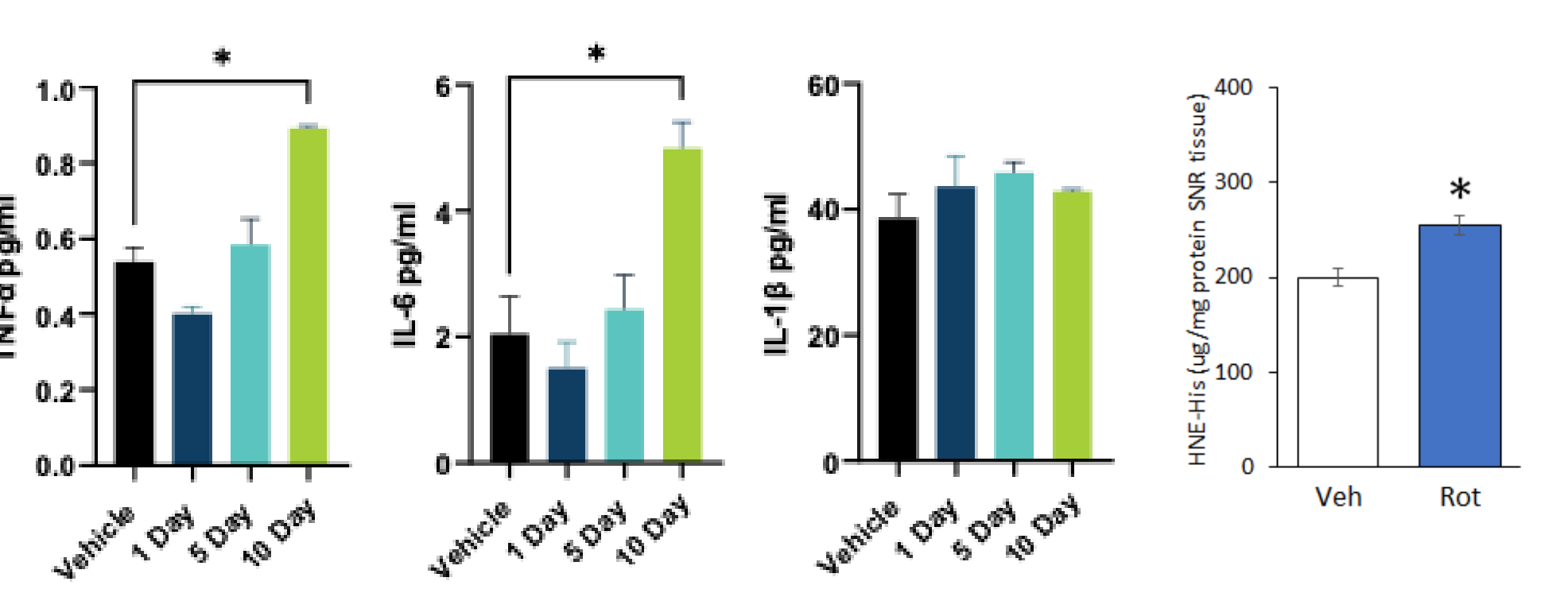
10-day treatment induced a 30-50% decrease in TH positive cells and a significant increase in Iba1 positive cells (dorsolateral SN). *p<0.05 vs. vehicle, Mean ± SEM, n=3-4 per group.

Rotenone reduces striatal dopamine concentration



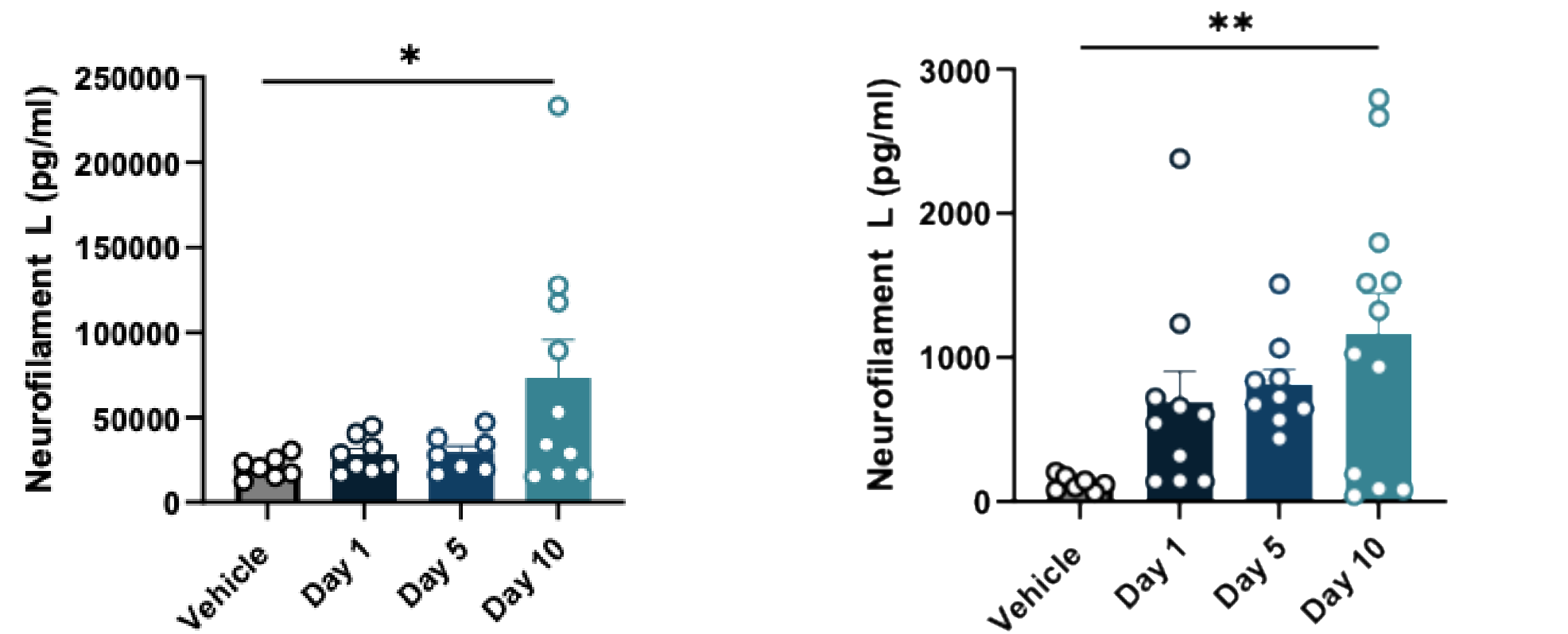
1-, 5- and 10- day treatment reduced dopamine concentration in the striatum. *p<0.05, **p<0.01, ***p<0.0001 vs. vehicle, Mean ± SEM, n=5 per group.

Rotenone increases neuroinflammatory markers and lipid peroxidation



10-day treatment increased TNF-alpha and IL-6 in brain tissue (left hemisphere). Lipid peroxidation (4-HNE) was increased at day 10. *p<0.05 vs. vehicle, Mean ± SEM, n=3-9 per group.

Rotenone increases NfL in CSF (left) and plasma (right)



10-day treatment increased NfL in CSF and plasma. *p<0.05, **p<0.01 vs. vehicle, Mean ± SEM, n=10 per group.

Conclusion

The rotenone model exhibits disease-relevant motor symptoms and dopaminergic neurodegeneration in the SN. This neurodegeneration is associated with temporal changes in markers of oxidative stress, neuroinflammation and gliosis, and a useful approach to studying treatments that may modify these processes. The present data also suggest that a single rotenone treatment regimen may have value for assessing neuroprotective drug effects.