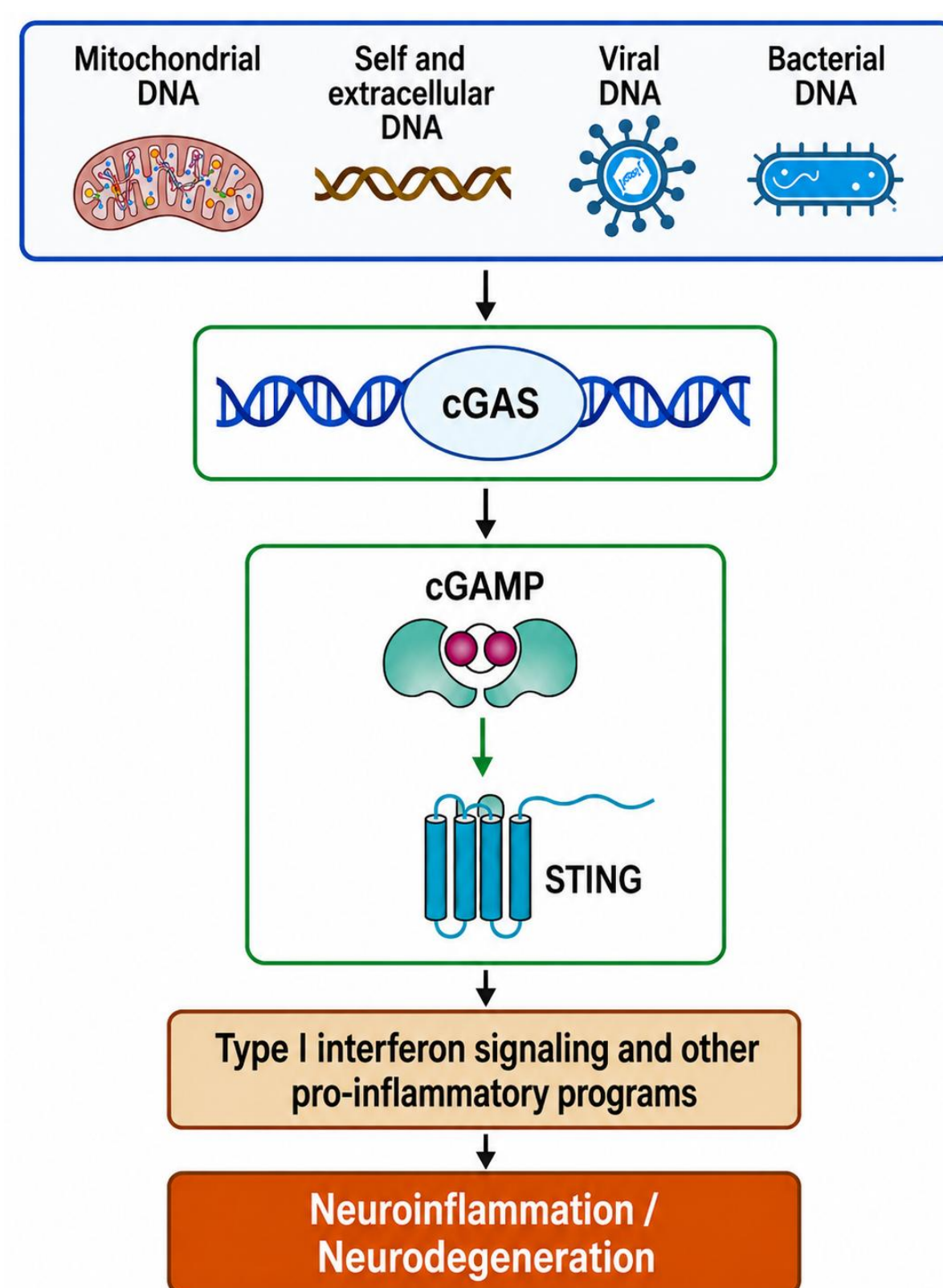


CNS penetrant cGAS inhibitors block microglia activation *in vitro* and reduce brain inflammation in TREX1-/- and 5xFAD *in vivo* models

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cGAS is a compelling target for neuroinflammatory diseases that currently lack effective treatment



Potent cGAS inhibition and biochemical/cellular translation achieved

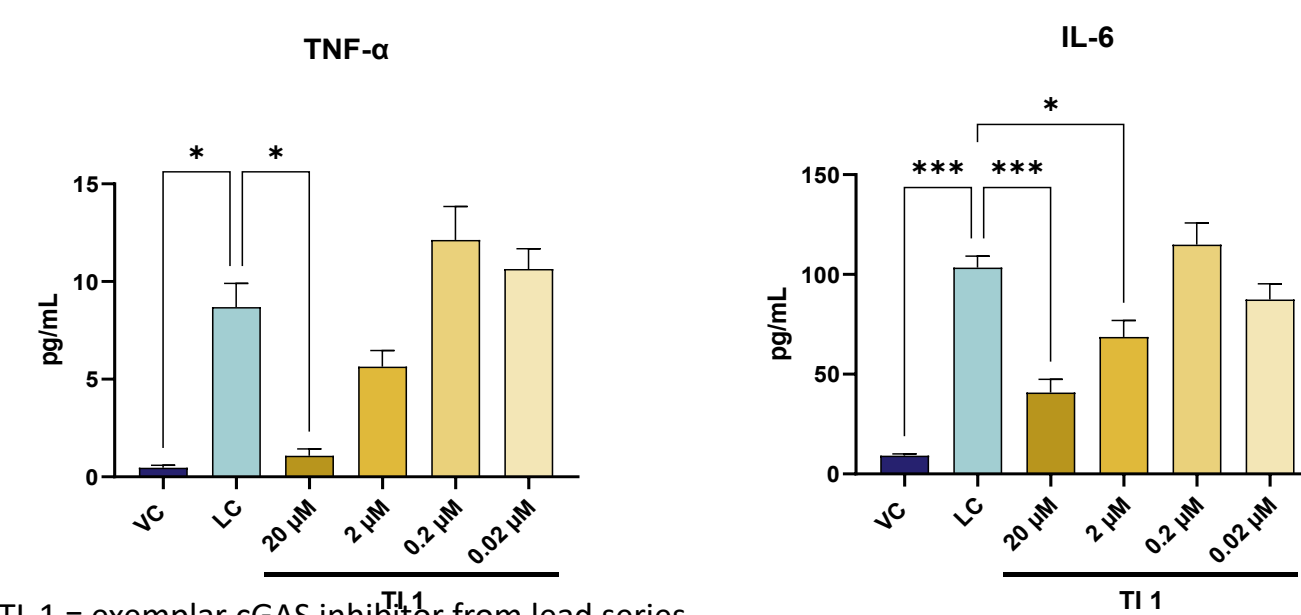
- Lead series based on highly resolved (< 2 Å) proprietary co-crystal structures
- Good cellular potency, excellent human/mouse cross reactivity and minimal serum shift
- Excellent selectivity towards related enzymes (OAS1, PDEs, kinases) and other mechanisms of pathway regulation (LPS, RNA)
- Serum shifted potencies translating to activity in human whole blood
- Excellent ADMET profile across series

Cpd ID	Human cGAS (IC ₅₀ , μM)	Human cells THP1 IRF3 reporter (IC ₅₀ , μM)		Mouse cells RAW IRF3 reporter (IC ₅₀ , μM)
	cGAMP via MS (5nM cGAS)	1% FBS	1% FBS + 25 mg/ml HSA	1% FBS
Ref Co #1	0.005	0.47	18	8.6
Ref Co #2	0.08	4.7	35	1.1
Ref Co #3	4	8.6	-	0.12
Cpd A	0.16	2.6	2.7	0.12
Cpd B	0.045	0.61	1.5	0.18
Cpd C	0.004	0.31	0.71	0.087
Cpd D	0.007	0.28	0.63	0.098
Cpd E	0.002	0.12	0.20	0.012
Cpd F	0.004	0.20	0.40	0.092

Cpd F Inhibits amyloid-β-mediated cytokine secretion in human iPSC-derived microglia

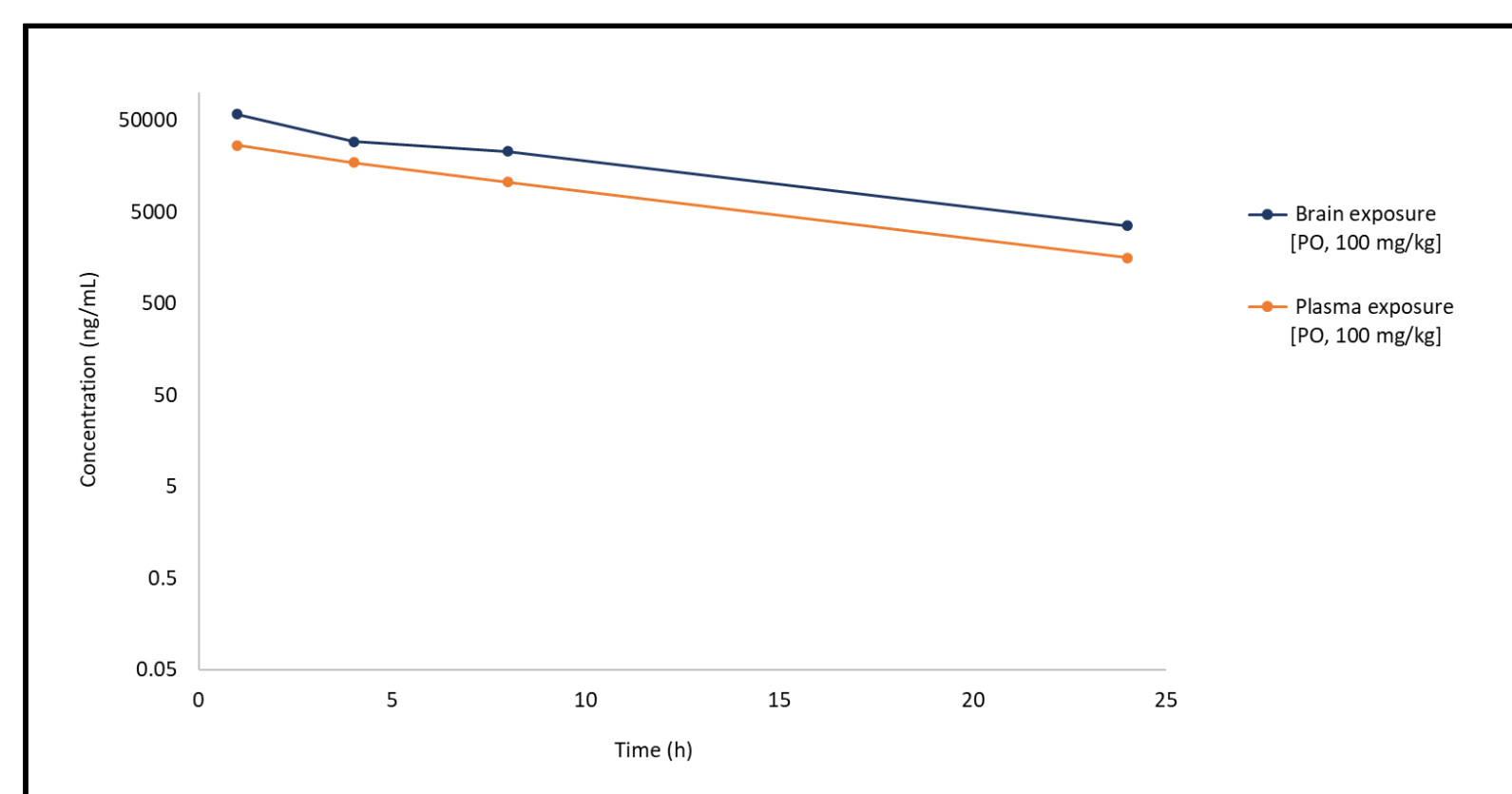
Human iPSC-derived μglia (APOE4/4 Fujifilm Cellular Dynamics), treated with brain penetrant Cpd F

- Activation with 10 μM Aβ1-42 aggregates for 24 h. Assay performed at: Scantox Neuro GmbH, Parkring 12, A-8074 Grambach, Austria



VC= vehicle
LC= lesion (Aβeta); TL 1 = exemplar cGAS inhibitor from lead series

Mouse neuro PK profile of a brain penetrant Cpd A

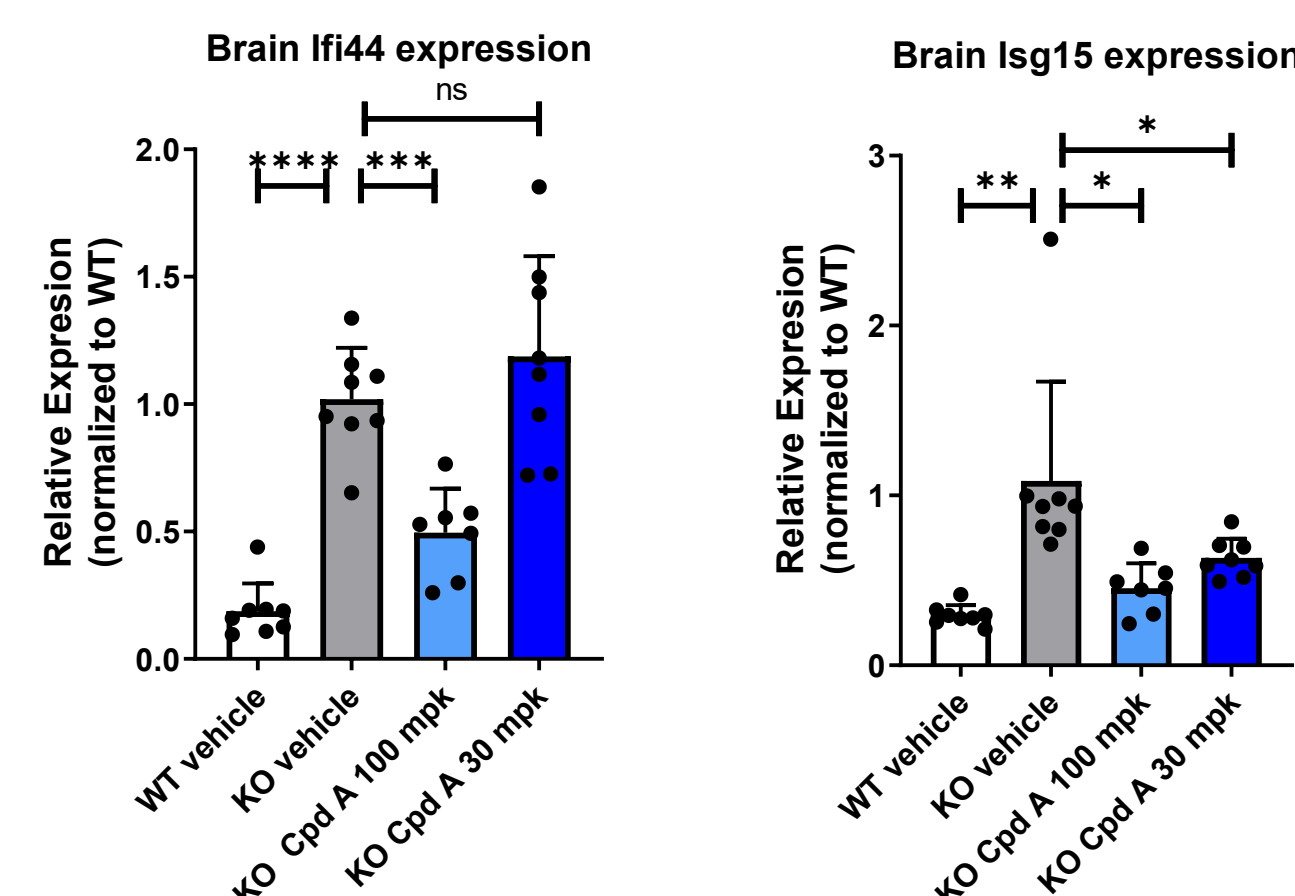


- Excellent PK
- Good oral bioavailability
- Dose dependent increase in C_{max} & AUC
- Clear brain exposure demonstrated K_{pu,u} = 1.1

CNS penetrant Cpd A robustly suppresses brain inflammation in Trex1 mouse model

6 weeks old WT or TREX1-/- mice treated 14 days BID with 100 or 30mpk Cpd A or vehicle control via oral gavage

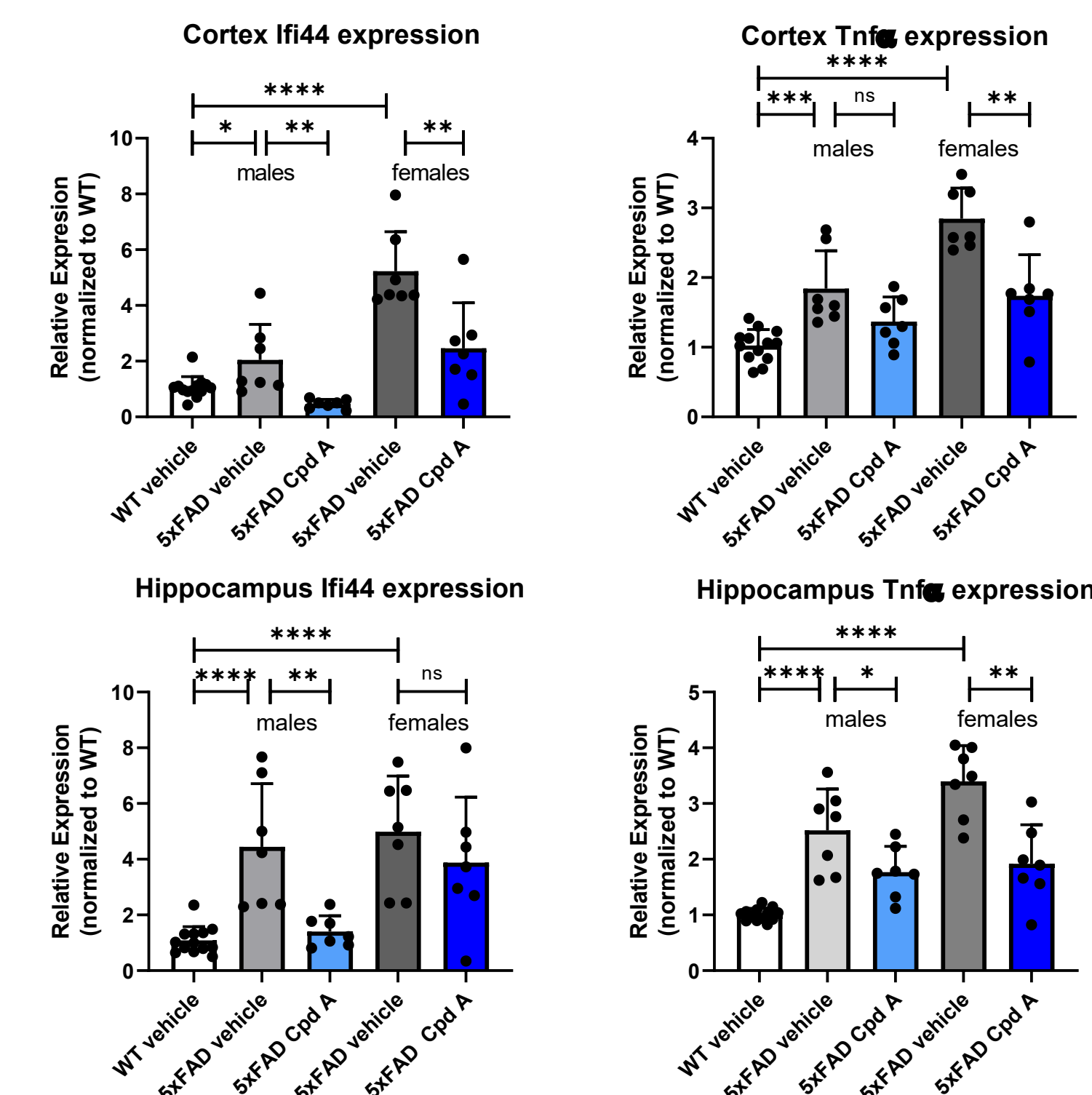
- TREX1-/- mice lack the 3'-repair exonuclease 1 gene, resulting in impaired cytosolic dsDNA degradation and chronic activation of innate immune signaling pathways
- Similar data obtained for IP10, Ufi3, Ifi44, Isg15 and Usp18 using this and other exemplar Cpd



CNS penetrant Cpd A robustly suppresses brain inflammation in 5xFAD mouse model

7.5 months old male WT or hemi 5xFAD mice treated 14 days QD with 100 mpk Cpd A or vehicle control via oral gavage

- 5xFAD mice over-express: APP KM670/671NL (Swedish), APP I716V (Florida), APP V717I (London), PSEN1 M146L, & PSEN1 L286V mutations
- Similar data obtained for IP10, Ifit3, Ifi44, Isg15 Usp18 using this and other exemplar Cpd



Summary

We have identified and developed potent and selective CNS-penetrant cGAS inhibitors capable of reducing brain inflammation in TREX1-/- and 5xFAD mouse models. These compounds are being further optimized towards identification of a pre-clinical candidate molecule.

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