NLRP3 inflammasome inhibitor CY-09 attenuates the effects of peripheral inflammation on neuroinflammation in a transgenic Alzheimer's mouse model: A [^{18}F]DPA-714 PET study.

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Introduction

Alzheimer's disease (AD), the most common form of dementia, remains poorly understood due to its complex, multifactorial aetiology. Peripheral inflammation and neuroinflammation (NI) are recognized as major contributors in AD, but their mechanistic interplay remains unclear. The NLRP3 inflammasome represents a promising therapeutic target, though current inhibitors have relatively poor blood-brain barrier (BBB) penetration. This study aims to elucidate the impact of NLRP3 inhibition on systemically induced inflammation and AD-induced NI and AD pathology.

Material & Methods

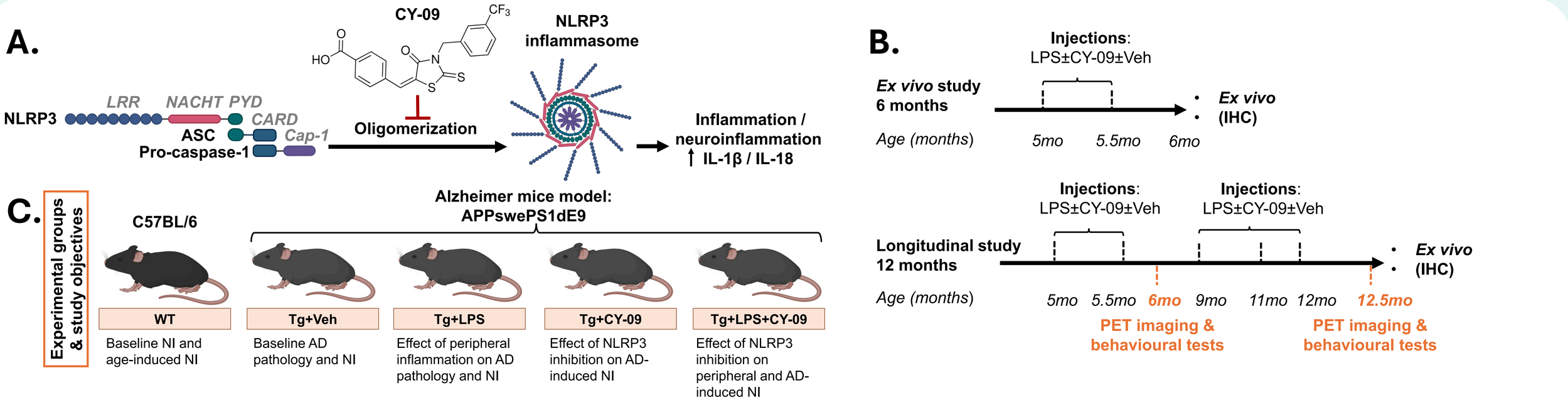


Figure 1: **Experimental design. A. Mechanism of action of the NLRP3 (NOD-like receptor family pyrin domain-containing 3) inhibitor CY-09:** CY-09 inhibits NLRP3 inflammasome activation by blocking NLRP3 oligomerization, thereby preventing caspase-1 activation and the maturation of IL-1β and IL-18, reducing inflammation and neuroinflammation. **B. Experimental timeline**: Schematic representation of the *ex vivo* and longitudinal study with age of injections. LPS (Lipopolysaccharide), PET (Positron Emission Tomography), IHC (Immunohistochemistry). **C. Experimental groups**: Wild-type (WT) and APPswePS1dE9 transgenic (Tg) mice were divided into five groups: WT, Tg mice treated with vehicle (Tg+Veh), with NLRP3 inhibitor CY-09 (Tg+CY-09), with LPS and with CY-09 (Tg+LPS+CY-09). LPS was used to induce systemic inflammation and CY-09 to inhibit NLRP3. The study assessed baseline Alzheimer's disease (AD)-related neuroinflammation (NI), the effect of peripheral inflammation, and the impact of NLRP3 inhibition.

Results

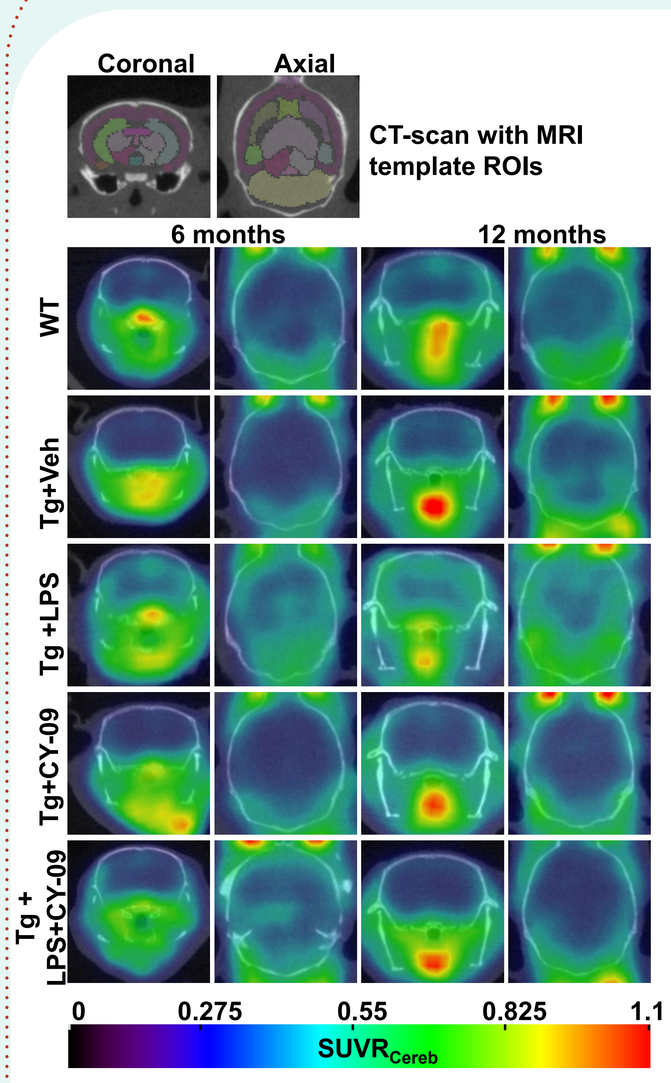
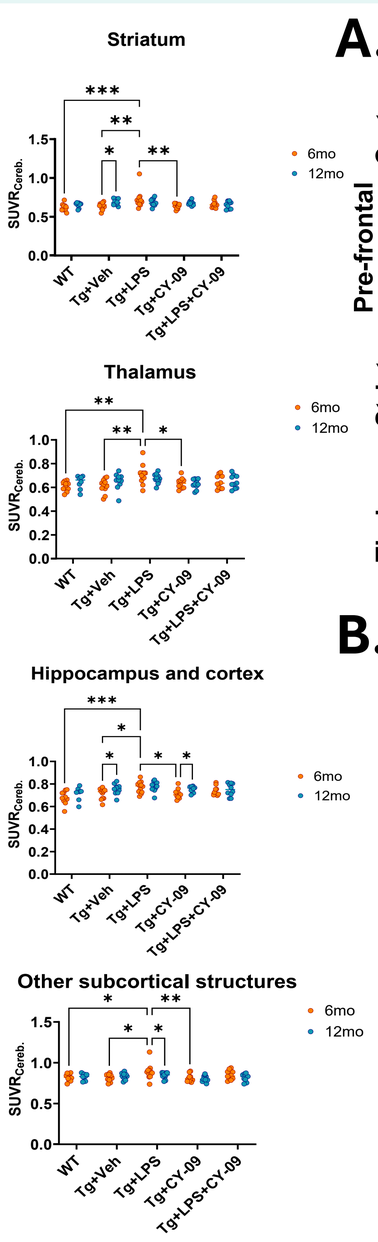


Figure 2: Longitudinal evaluation of neuroinflammation at 6 and 12 months using 18F-DPA-714 PET imaging. Data are presented as mean ± SD. Two-way ANOVA followed by Tukey's post hoc test; *p < 0.05, **p < 0.01, ***p < 0.001.



- **Significant increase in NI with age in Tg+Veh.**
- **LPS significantly accelerated NI in Tg mice at 6 months of age.**
- **CY-09 had no effect on AD-induced NI but prevented LPS-induced NI.**

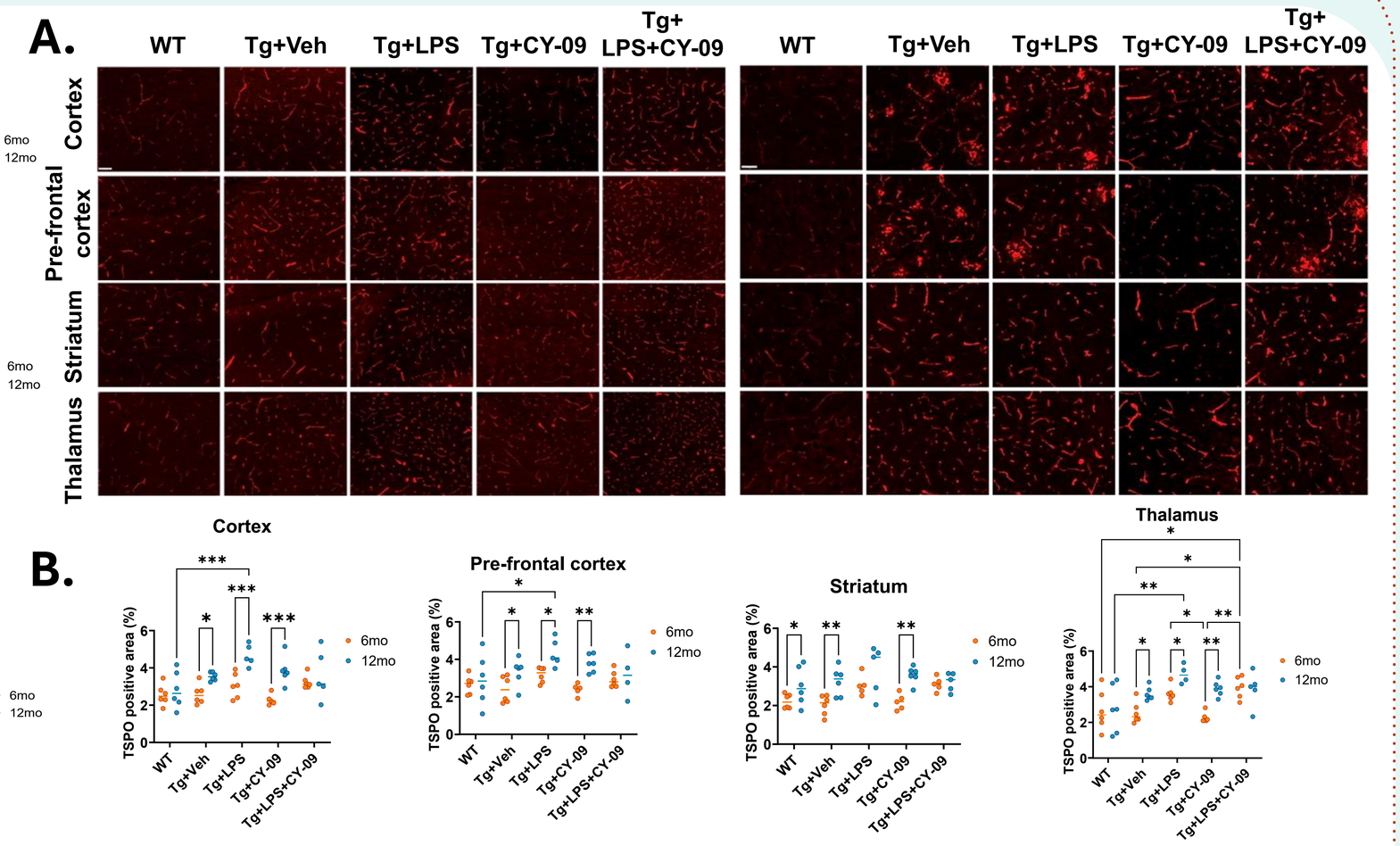


Figure 3: TSPO immunostaining at 6 and 12 months using. A. Representative brain sections showing TSPO immunostaining. **B.** Quantification of the percentage of TSPO+ cells. Scale bar: 50 µm. Data are presented as mean ± SD; each dot represents one animal. *p < 0.05; **p < 0.01; ***p < 0.001.

- **Ex vivo data confirmed TSPO PET in vivo imaging data.**

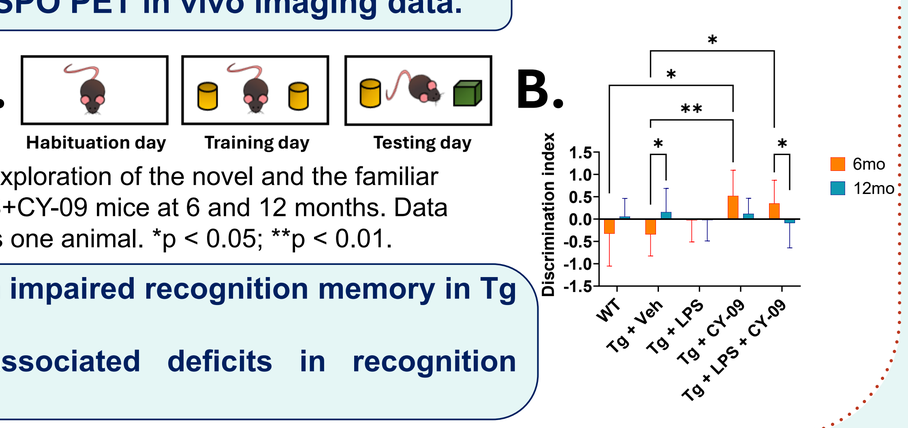


Figure 4: Novel Object Recognition Test. **A.** Schematic representation of Novel Object Recognition Test. **B.** Discrimination index (DI) of exploration of the novel and the familiar object in WT, Tg+Vehicle, Tg+CY-09, and Tg+LPS+CY-09 mice at 6 and 12 months. Data are presented as mean ± SD; each dot represents one animal. *p < 0.05; **p < 0.01.

- **LPS-induced systemic inflammation impaired recognition memory in Tg mice at 6 months.**
- **CY-09 treatment mitigated LPS-associated deficits in recognition memory at 6 months.**

Conclusion

Alzheimer's disease is a complex, multifactorial disorder in peripheral inflammation and neuroinflammation contribute to pathophysiology, although their interplay remains unclear. Targeting the NLRP3 inflammasome may help modulate this inflammatory crosstalk. This study investigates for the first time a selective NLRP3 inhibitor (CY-09) under recurrent systemic inflammatory challenges in an AD mouse model using PET imaging. We observed: ***i)*** recurrent systemic inflammation (Tg+LPS) accelerated NI in Tg mice at 6 months and that NI plateaued thereafter, whereas it increased with age in Tg+Veh; ***ii)*** CY-09 prevented the LPS-induced NI but had modest to no effect on AD-induced NI likely due to limited brain delivery; ***iii)*** CY-09 improved recognition memory and prevented LPS-induced cognitive deficits at 6 months demonstrating that ***iv)*** **NLRP3 is a good therapeutic target in AD that requires inhibitors with improved BBB/brain delivery**.

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