# Anti-inflammatory effects of SM07883, a novel, potent, and selective oral DYRK1A inhibitor in mouse models of neuroinflammation

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**Poster #J2**

## Background

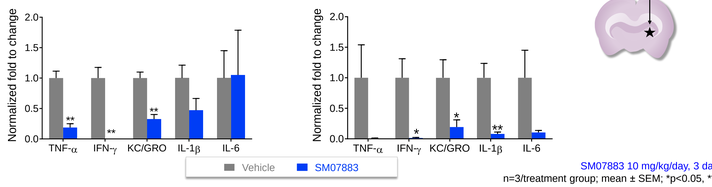
- Elevated cellular stress signals such as amyloid beta (Aβ) and tumor necrosis factor alpha (TNF-α) induce dual-specificity tyrosine phosphorylation-regulated kinase 1A (DYRK1A) activity[1-3]
- DYRK1A regulates amyloid precursor protein (APP) and tau phosphorylation (pTau), is overexpressed in Alzheimer's disease (AD) brains, and correlates with disease pathology[1-4]
- SM07883 is an oral small-molecule DYRK1A inhibitor that reduced tau and amyloid pathology and associated neuroinflammation in AD mouse models[5]
- This study assessed the effects of DYRK1A inhibition by SM07883 *in vitro* and *in vivo* on neuroinflammatory pathology in lipopolysaccharide (LPS)- and experimental autoimmune encephalomyelitis (EAE)-induced inflammation mouse models

## Conclusions

- **SM07883 ameliorated neuroinflammatory responses in preclinical models compared to vehicle**
  - **Reduced acute and chronic neuroinflammation in the absence of neurodegeneration**
  - **Not restricted to innate immunity with a potent effect on CNS-related adaptive immune responses**
- **SM07883, a small-molecule DYRK1A inhibitor, may have therapeutic potential in neurodegenerative diseases**
- **A Phase 1 human study is ongoing**
  - **ANZCTR.org.au registration #ACTRN12619000327189**

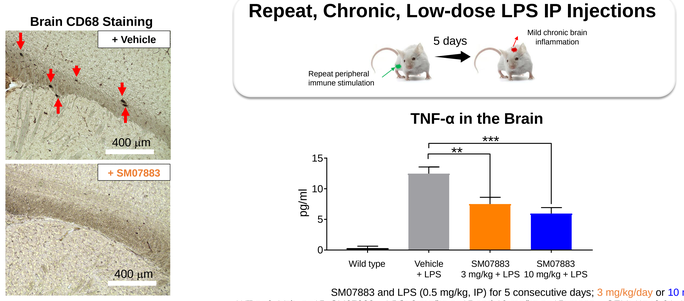
## Results

### Figure 1. SM07883 reduced acute inflammation *in vivo*



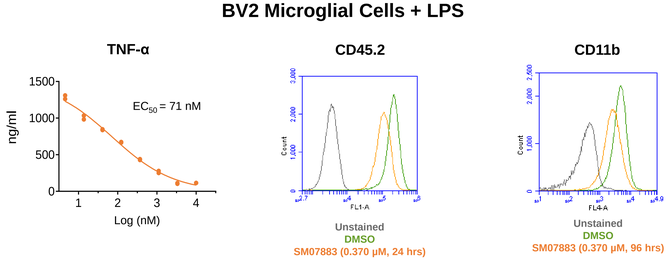

SM07883 10 mg/kg/day, 3 days prior
n=3/treatment group; mean ± SEM; *p<0.05, **p<0.01

### Figure 2. SM07883 reduced chronic neuroinflammation *in vivo*

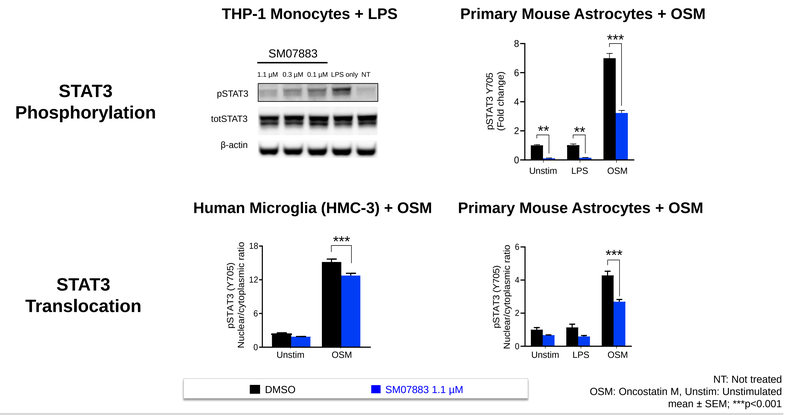



SM07883 and LPS (0.5 mg/kg, IP) for 5 consecutive days; 3 mg/kg/day or 10 mg/kg/day
WT n=2, Veh. n=15, SM07883 + LPS: 3 mg/kg n=15 and 10 mg/kg n=15; mean ± SEM; **p<0.01, ***p<0.001

### Figure 3. SM07883 reduced microglial cell activation *in vitro*

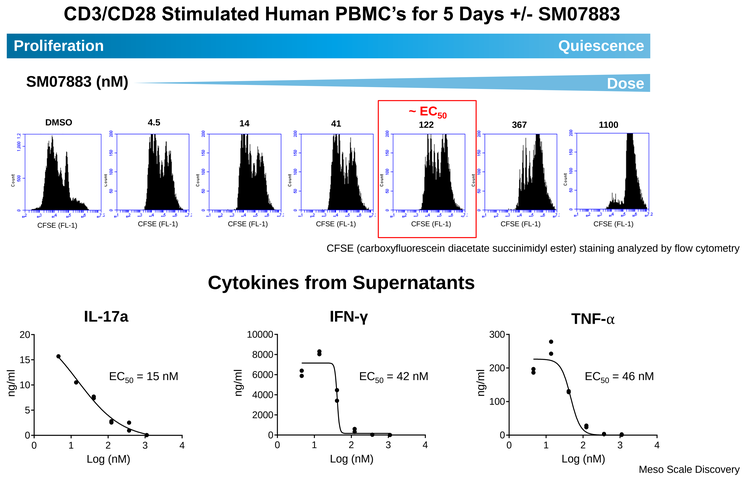



### Figure 4. SM07883 reduced STAT3 phosphorylation and translocation *in vitro*




NT: Not treated
OSM: Oncostatin M, Unstim: Unstimulated
mean ± SEM; ***p<0.001

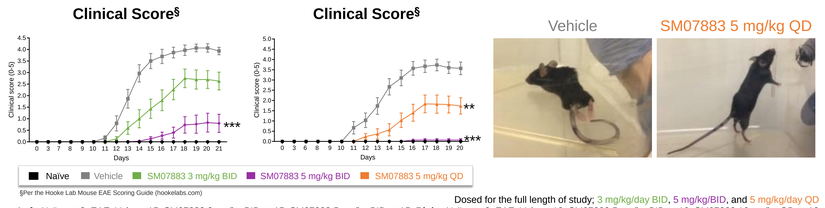
### Figure 5. SM07883 prevented T-cell proliferation and proinflammatory cytokine secretion *in vitro*




CFSE (carboxyfluorescein diacetate succinimidyl ester) staining analyzed by flow cytometry

Meso Scale Discovery

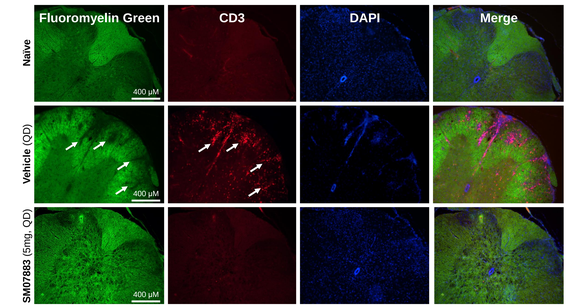
### Figure 6. SM07883 reduced MOG-induced EAE acute symptoms *in vivo*




§Per the Hooke Lab Mouse EAE Scoring Guide (hookelabs.com)

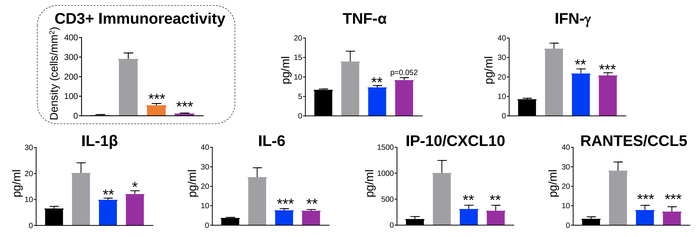
Dosed for the full length of study: 3 mg/kg/day BID, 5 mg/kg/BID, and 5 mg/kg/day QD
**Left:** Naïve n=2, EAE: Veh. n=15, SM07883 3 mg/kg BID n=15, SM07883 5 mg/kg BID n=15; **Right:** Naïve n=2, EAE: Veh. n=12, SM07883 5 mg/kg BID n=10, SM07883 10 mg/kg QD n=12
mean ± SEM; **p<0.01, ***p<0.001

### Figure 7. SM07883 reduced EAE-mediated (T-cell) damage in the spinal cord *in vivo*




Arrows indicate areas of T cell (CD3)-mediated demyelination (Fluoromyelin green)

### Figure 8. SM07883 reduced EAE-induced proinflammatory mediators in the spinal cord *in vivo*




**SM07883 also reduced IL-4, MIP-1α, MIP-1β, and GM-CSF compared to vehicle**

Wild type n=2, Veh. n=15, SM07883 5 mg/kg QD n=15, SM07883 10 mg/kg QD n=15, SM07883 5 mg/kg BID n=15; mean ± SEM; *p<0.05, **p<0.01, ***p<0.001

## Methods

- Brains from Balb/c mice administered SM07883 (PO, 10 mg/kg QD) 3 days prior to intracranial injection of 5 μL of LPS (100 ng/ml) and IFN-γ (10 U/ml) into the left striatum were collected 5 hrs and 24 hrs post injection and assayed for cytokines (Meso Scale Discovery [MSD] V-plex)[6] (**Fig. 1**)
- Brains from mice administered SM07883 (P.O., 3 or 10 mg/kg) and LPS (I.P., 0.5 mg/kg) for 5 consecutive days (n=15) were collected and one hemisphere was taken for cytokine analysis (Millipore Milliplex MYCTO-MAG-70K-PMX 25 plex) while the other hemisphere was immunohistochemically stained for monocytes with CD68 antibody[7] (**Fig. 2**)
- BV2 murine microglial cells were cultured with serial dilutions of SM07883 [1.1 μM–0.5 nM] overnight and challenged with LPS (250 ng/ml) for 5 hrs. Supernatants were tested for TNF-α (MSD). Similarly, LPS-treated BV2 cells were exposed to 0.370 μM of SM07883 for 24 hrs and 96 hrs and live cells were stained with fluorescent-labeled anti-murine CD45.2 or CD11b and analyzed by flow cytometry (**Fig. 3**)
- THP-1 monocytes were treated overnight with SM07883 before LPS (500 ng/ml) was added for 4 hrs. pSTAT3 signal was assessed by Western blot (**Fig. 4**)
- Human microglia HMC-3 and mouse primary astrocytes were treated with SM07883 overnight and LPS (100 ng/ml) or Oncostatin (OSM, 10 ng/ml) were added the next day for 15 min. pSTAT3 was assessed by AlphaLisa (Perkin-Elmer) or immunostaining (CellInsight CX5, Thermo Fisher) (**Fig. 4**)
- CFSE-labeled human PBMC's were stimulated with CD3/CD28 for 5 days and simultaneously treated with serial dilutions of SM07883 [1.1 μM-4.5 nM]. On Day 5, cells were analyzed by flow cytometry and supernatant was assayed for cytokine secretion (MSD, V-plex) (**Fig. 5**)
- C57Bl/6 mice were challenged subQ with a myelin oligodendrocyte glycoprotein $MOG_{25-55}$ emulsion with pertussis toxin IP and administered SM07883 (P.O., 5 mg/kg QD or 3 and 5 mg/kg BID) for the full length of the study. Mice were scored and weighed daily.[8] At termination, spinal cords were collected for immunohistochemistry (CD3, Fluoromyelin) and cytokine analysis (Millipore milliplex MYCTO-MAG-70K-PMX 25 plex) (**Fig.6-8**)

## References

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