Journal of Alzheimer's Disease 20 (2010) 1–4

IOS Press

Supplementary Material

A Novel Transgenic Rat Model with a Full Alzheimer's-Like Amyloid Pathology Displays Pre-Plaque Intracellular Amyloid-β-Associated Cognitive Impairment

Wanda Carolina Leon[a,1], Fabio Canneva[a,1], Vanessa Partridge[a], Simon Allard[a], Maria Teresa Ferretti[a], Arald DeWilde[a], Freya Vercauteren[a,2], Ramtin Atifeh[a], Adriana Ducatenzeiler[a], William Klein[b], Moshe Szyf[a], Leena Alhonen[c] and A. Claudio Cuello[a,d,e,*]

[a]*Department of Pharmacology and Therapeutics, McGill University, Montreal, QC, Canada*

[b]*Cognitive Neurology and Alzheimer's Disease Center, Northwestern University Institute for Neuroscience, Chicago, IL, USA*

[c]*Department of Biotechnology and Molecular Medicine, A.I. Virtanen Institute for Molecular Sciences, University of Kuopio, Kuopio, Finland*

[d]*Department of Anatomy and Cell Biology, McGill University, Montreal, QC, Canada*

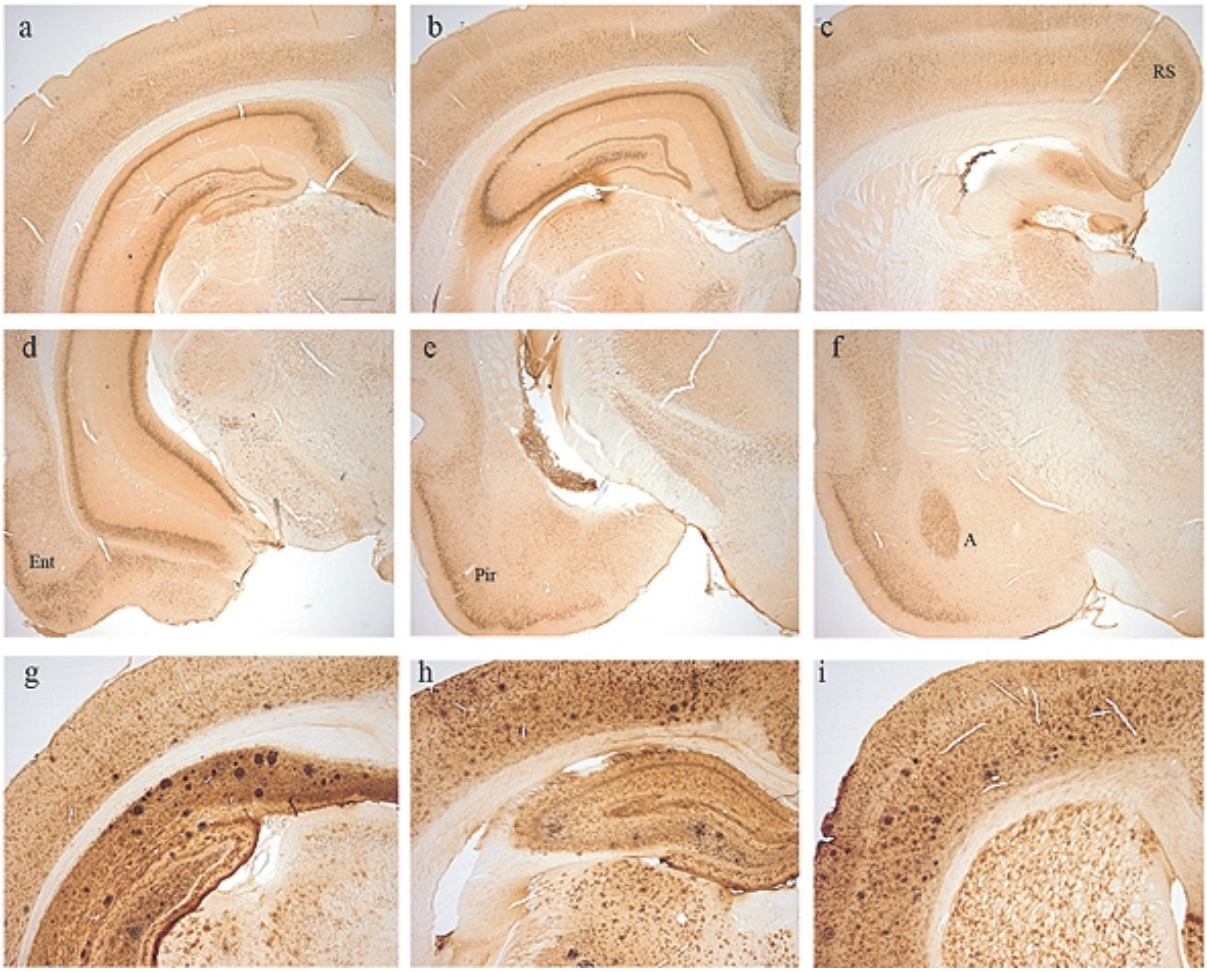
[e]*Department of Neurology and Neurosurgery, McGill University, Montreal, QC, Canada*

[1]WL and FC equally contributed to the experimental work.

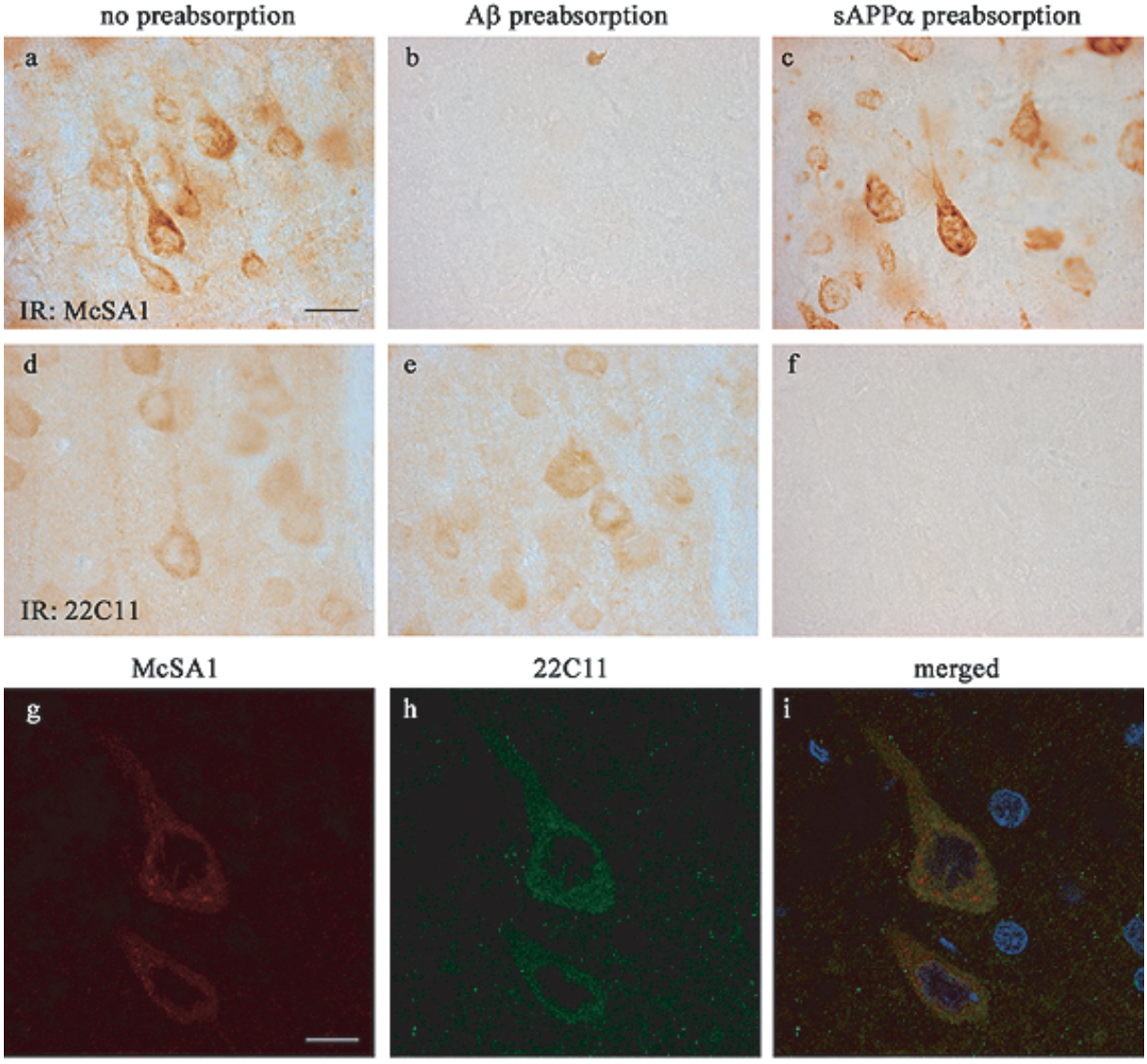
[2]Present affiliation: Sleep Research Centre, Université de Montreal, Faculty of Medicine, Sacré-Coeur Hospital, Montreal, QC, Canada.

*Correspondence to: A. Claudio Cuello, 3655 Promenade Sir William Osler, H3G 1Y6, Montreal, QC, Canada. Tel.: +1 514 398 3618; Fax: +1 514 398 8317; E-mail: claudio.cuello@mcgill.ca.

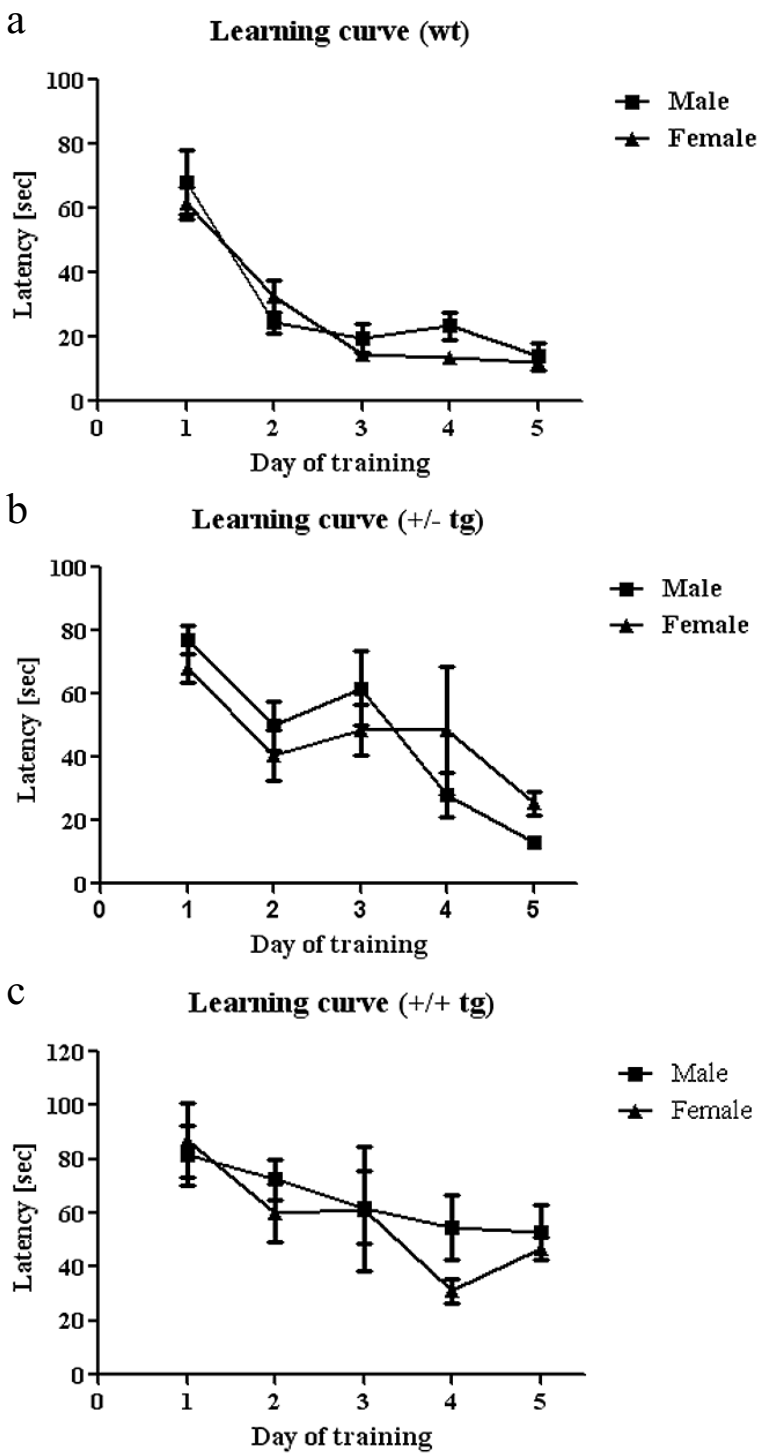
ISSN 1387-2877/10/$27.50



Supplemental Fig. 1. Topographical distribution of Aβ immunoreactivity in McGill-R-Thy1-APP Tg rats. McSA1 immunoreactivity was used to detect Aβ accumulation throughout homozygous Tg rat brain before (3 months, panels a to f) and after (20 months, panels g to h) plaque deposition. At the earlier stages of intracellular Aβ accumulation, hippocampal staining was found to be homogeneous at different levels of the structure (a and b). In the cortex, the strong immunoreactivity detected at the level of the entorhinal cortex (Ent) was also present in the piriform cortex (Pir, panel e). Intensely stained neurons were also detected in the amygdala (A) and in the retrosplenial cortex (c and f). After the plaque deposition is well established, both diffused and dense core amyloid aggregates are detected in most of the areas of hippocampus and cortex. In particular, in the hippocampus, where the plaque deposition starts, plaques were distributed from the subiculum throughout the entire structure (panel g). Fibrillar, thioflavine S-positive plaques were also detected in different areas of the cerebral cortex (g and h), with a trend of accumulation of big dense Aβ deposits in the parietal cortex, in more rostral sections (panel i). Scale bar in panel a = 500 μm.



Supplemental Fig. 2. Aβ-specificity of McSA1. McSA1 specificity in 3 month-old tg rats was investigated by pre-absorption with synthetic Aβ or sAβPPα (Sigma). Intracellular staining with McSA1 (1:4000, panel a) was completely abolished by pre-treatment for 3 hours with 2.5 μg/mL of Aβ_{1-42} (panel b), whereas 50 μg/mL of sAβPPα did not have any noticeable effect (panel c). On the other hand, 22C11-immunoreactivity observed inside pyramidal neurons (1:2000, panel d) was completely blocked by pre-absorption with 140ng/mL of sAβPPα (panel e), while equimolar (7ng/mL) amounts of Aβ_{1-42} did not affect the antibody for immuno-staining (panel f). Finally, panels g to i illustrate the double-labeling with McSA1 and 22C11 by confocal microscopy immuno-fluorescence. McSA1 IR (1:8000 followed by rhodamine conjugated secondary antibody 1:200, red signal) was distributed throughout the cell soma, and particularly concentrated in granule-like bodies while the more diffused 22C11 IR (1:500 followed by biothinylated secondary antibody 1:200 and Alexa488 streptavidine 1:200, green signal) did not overlap with that of McSA1. Nuclei were visualized with DAPI (1:1000 in PBS). Scale bar in panel a = μm, scale bar in panel g = 10 μm.



Supplemental Fig. 3. Learning performance of male and female McGill-R-Thy1-APP rats in the MWM. The learning phase during the 5 consecutive days of training in the MWM task is described for male and female 13 month-old rats. Wt (a), $+/-$ Tg (b) and $+/+$ Tg (c) all showed no gender-difference in their ability to learn the escape task ($p > 0.05$, two-way ANOVA). Moreover, the particular behavior of $+/-$ Tg animals on day 3, showing a slight increase of the latency (not significantly different from the previous day), was observed in both male and female animals, indicative of labile memory.