

Supplementary figures for:

Defective thyroid hormone transport to the brain leads to astroglial alterations

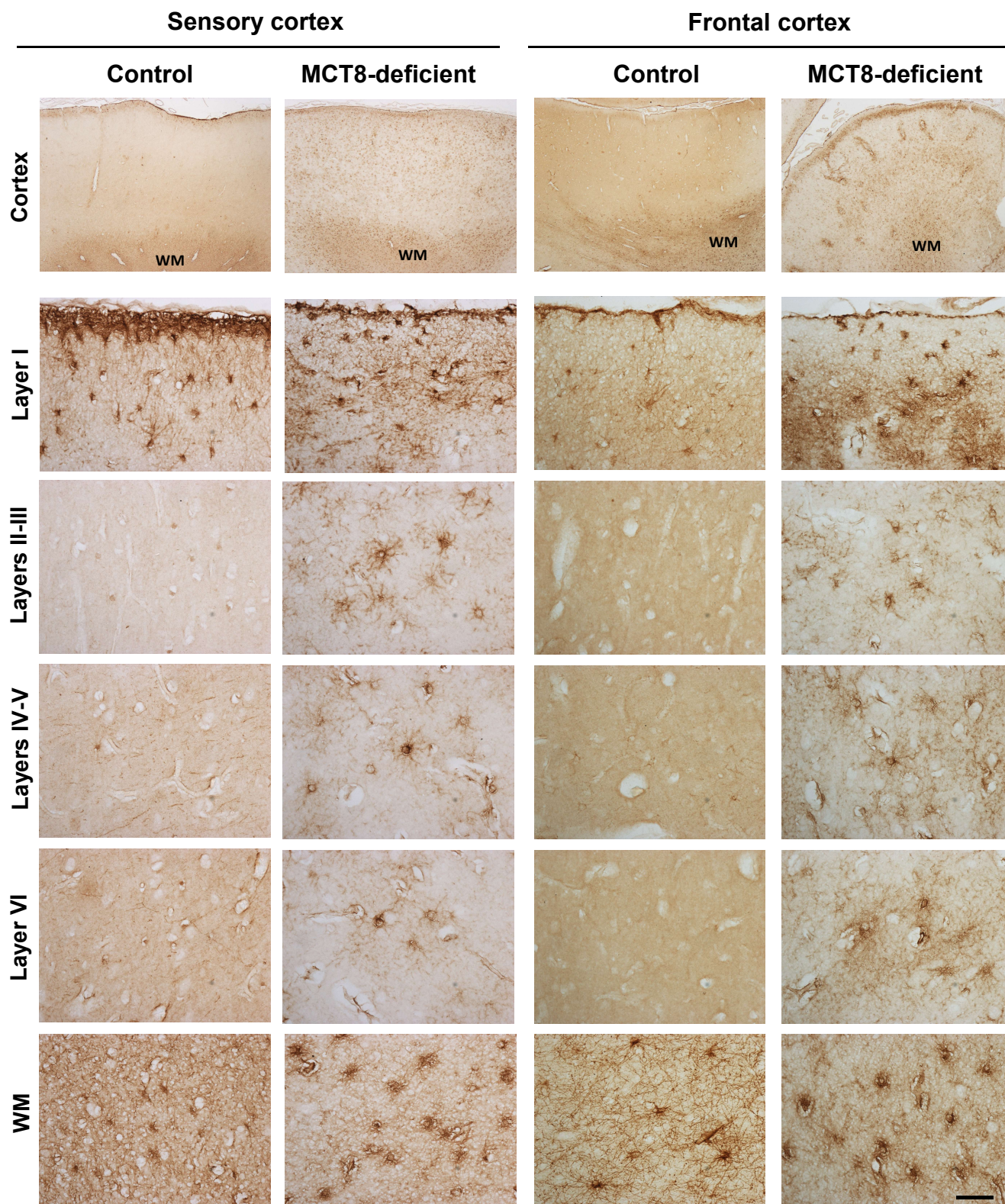
Marina Guillén-Yunta^{1*}, Ángel García-Aldea^{1*}, Víctor Valcárcel-Hernández¹, Ainara Sanz-Bógallo¹, Emma Muñoz-Moreno², Maria Gisele Matheus³, Carmen Grijota-Martínez^{1,4}, Soledad Báñez-López¹, Ana Montero-Pedrazuela¹, Ana Guadaño-Ferraz¹

1 Department of Neurological Diseases and Aging, Instituto de Investigaciones Biomédicas Sols-Morreale, Consejo Superior de Investigaciones Científicas (CSIC), Universidad Autónoma de Madrid (UAM), Madrid, Spain.

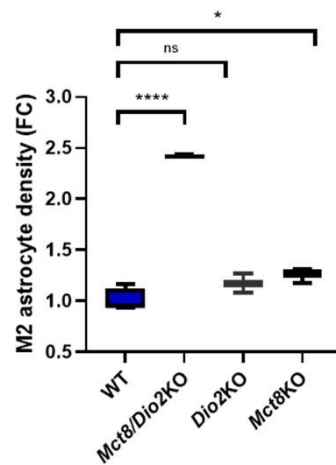
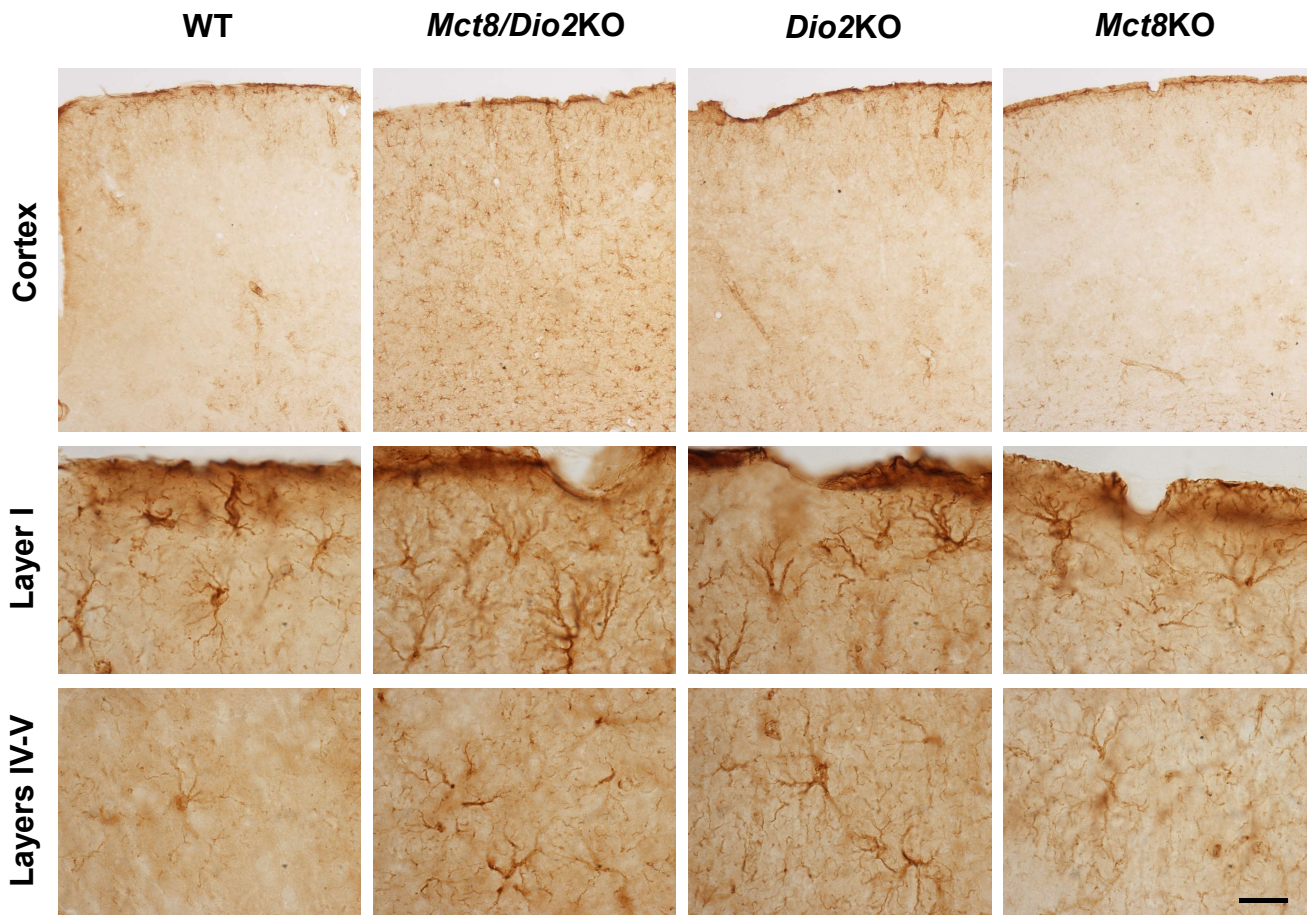
2 Magnetic Imaging Resonance Core Facility, Institut d'Investigacions Biomèdiques August Pi I Sunyer (IDIBAPS), Barcelona, Spain.

3 Department of Radiology and Radiological Science, Medical University of South Carolina, Charleston, SC, USA

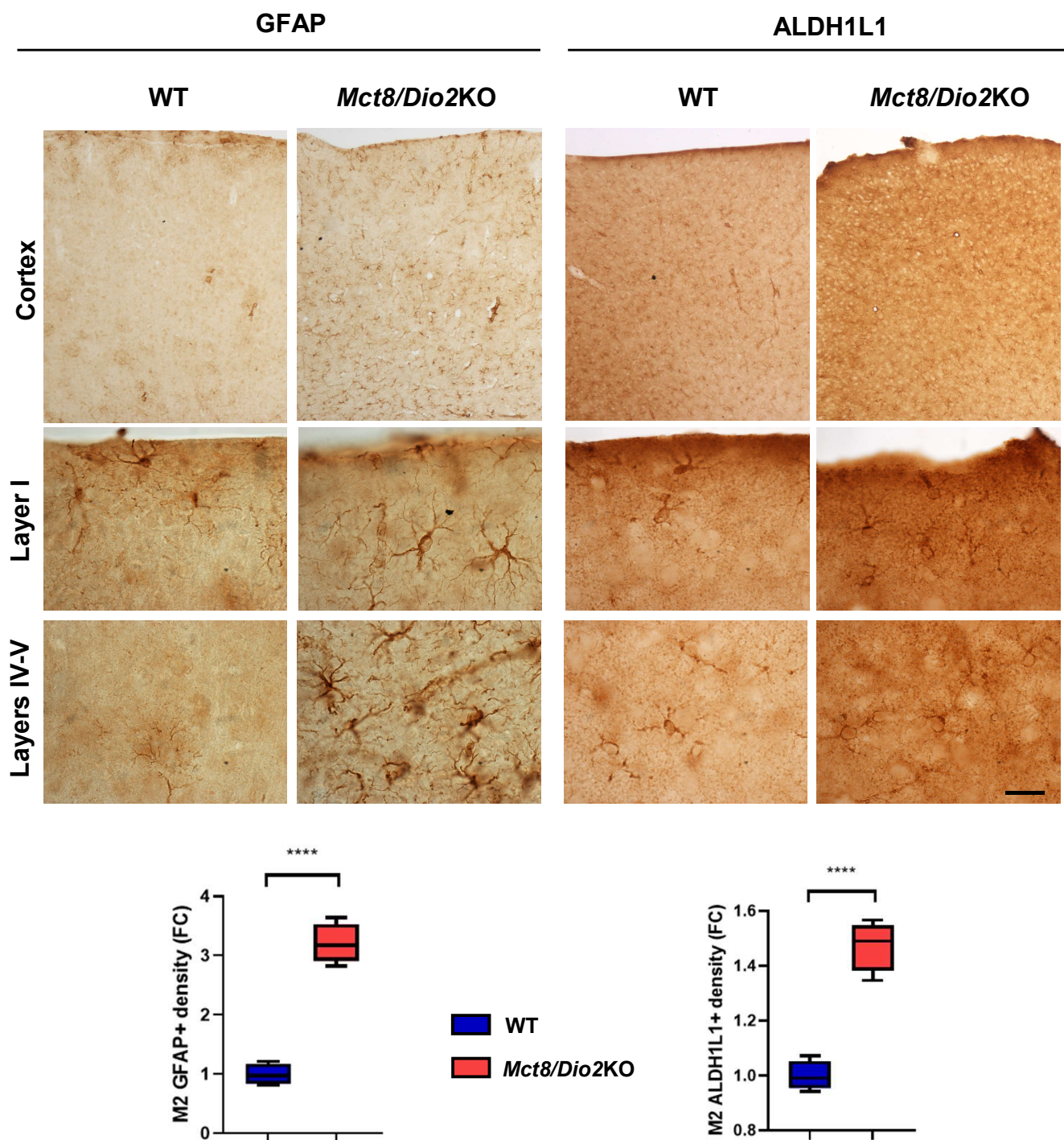
4 Department of Cell Biology, Faculty of Biology, Universidad Complutense de Madrid, Madrid, Spain



Supplementary Fig. 1 Expression of GFAP in the human sensory and frontal cortex. Expression of GFAP in the sensory cortex (left panels) and frontal cortex (right panels) of 12- and 10-year-old control subjects, respectively, and an 11-year-old MCT8-deficient subject. The upper panels show a panoramic view of the gyrus of the motor cortex, including all layers and the subcortical WM. Note the absence of staining in layers II-VI of the control samples and the widespread GFAP expression in the MCT8-deficient samples. The other panels show high-magnification images of different cortical layers and the subcortical WM. I-VI: layers of the motor cortex. WM: white matter. Scale bar: 1,000 μ m in upper panels; 50 μ m in other panels.



Supplementary Fig. 2 GFAP expression in the motor cortex of WT, *Mct8/Dio2KO*, *Dio2KO*, and *Mct8KO* mice. Analysis performed using 3-month-old WT, *Mct8/Dio2KO*, *Dio2KO* and *Mct8KO* mice (n=4 for each genotype). The upper panels show the general distribution pattern of GFAP⁺ astrocytes. Observe the increased expression of GFAP in astrocytes in the inner layers of the cortex in *Mct8/Dio2KO* and the intermediate phenotype of the single *Dio2KO* and *Mct8KO* mice. Higher magnification images show details in layer I and layers IV-V. Scale bar: 125 μ m in upper panels; 20 μ m in lower panels. Graph: quantitative analysis of the density of GFAP⁺ astrocytes in the M2 of animals of the four genotypes. * $p < 0.05$ and **** $p < 0.0001$ were determined by a two-tailed unpaired Student's t-test. M2: secondary motor cortex. FC: fold change.



Supplementary Fig. 3 GFAP and ALDH1L1 expression in the motor cortex of 6-month-old WT and *Mct8/Dio2KO* mice.

Analysis performed using WT (n=4) and *Mct8/Dio2KO* (n=4) mice. Left panels: GFAP expression. Right panels: ALDH1L1 expression. Upper panels show the general distribution pattern of GFAP+ and ALDH1L1+ astrocytes. Observe the different GFAP expression pattern in the inner layers of the cortex between both genotypes. Higher magnification images show details in layer I and layers IV-V. Scale bar: 125 μ m in upper panels; 20 μ m in lower panels. Graphs: quantitative analysis of the density of GFAP+ and ALDH1L1+ astrocytes in the M2 of WT (blue) and *Mct8/Dio2KO* (red) mice. ****p < 0.0001 was determined by a two-tailed unpaired Student's t-test. M2: secondary motor cortex. FC: fold change.