

Supplementary Information

Involvement of integrin $\alpha\beta3$ in thyroid hormone-induced dendritogenesis

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Running title: TH modulates integrin $\alpha\beta3$ -mediated dendritogenesis

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Supplementary Figure 1. List of siRNA target sequences (Table 1) and qRT-PCR primer sequences (Table 2).

Supplementary Figure 2. mRNA expression levels of TR or integrin $\alpha\beta 3$ after siRNA treatment. A. Changes in TR α or TR β mRNA expression levels in primary cerebellar culture after siRNA transfection. B. Changes in integrin αv or integrin $\beta 3$ mRNA expression levels in primary cerebellar culture after siRNA transfection. C. Changes in TR α or TR β mRNA expression levels in Neuro-2A cells after siRNA transfection. All experiments were repeated three times, using independent RNA preparations to confirm the consistency of the results. mRNA levels were normalized against GAPDH. Data are shown as the mean \pm SEM. **** p < 0.0001 indicates statistical significance according to Student's unpaired t-test. See Supplementary Table 1.

Supplementary Figure 3. Effect of TH on Purkinje cell dendrite arborization in primary cerebellar culture. Cerebellar cells were cultured for 17 days, followed by an immunocytochemistry analysis with Calbindin D-28K (green) and DAPI (blue). A. Representative photomicrographs showing the effects of TH on the morphology of Purkinje cells. Changes in the dendritic areas of Purkinje cells following T₃ (B), T₄ (C), or rT₃ (D) treatment. Dendritic areas were quantified using ImageJ Fiji software (NIH). Bars indicate 50 μ m. Data are shown as the mean \pm SEM. **** p < 0.0001 and ** p < 0.001 indicate statistical significance according to one-way ANOVA continued with post hoc Turkey's test. See Supplementary Table 1.

Supplementary Figure 4. Integrin $\alpha\beta3$ affect neurite outgrowth induced by TH in neuronal-derived Neuro-2A cells. Cells were induced to differentiate by serum starvation for one to three days, followed by immunofluorescence analysis with β -tubulin III (red), doublecortin (green), and DAPI (blue). Quantitative analysis of the effect of TH after exposure to integrin α siRNA (A), integrin $\beta3$ siRNA (B), integrin α and $\beta3$ siRNA (C), on neurite length in Neuro-2A cells. Neurite length was quantified using ImageJ Fiji software (NIH). Bars indicate 50 μ m. Data are shown as the mean \pm SEM. **** $p < 0.0001$, indicates statistical significance according to two-way or one-way ANOVA continued with post hoc Turkey's test compared to control. #### $p < 0.0001$ and ## $p < 0.01$, indicate statistical significance according to two-way ANOVA continued with post hoc Turkey's test. See Supplementary Table 1.

Supplementary Figure 5. mRNA levels of *Dio1*, *Dio2*, or *Dio3* after co-exposure with LM609. Changes in *Dio1* (A), *Dio2* (B), or *Dio3*(C), mRNA levels in primary cerebellar culture after co-exposure with LM609. All experiments were repeated three times, using independent RNA preparations to confirm the consistency of the results. mRNA levels were normalized against GAPDH. Data are shown as the mean \pm SEM. **** $p < 0.0001$, *** $p < 0.001$ and ** $p < 0.01$ * $p < 0.05$, indicates statistical significance according to two-way ANOVA continued with post hoc Turkey's test. See Supplementary Table 1.

Supplementary Table 1. Statistical Analysis Results.