

## Supplementary Figure legends

### Supplementary Figure 1. Testing reporter activity in cell lines engineered to express different miR-124 reporter constructs.

- a) Left: schematic illustration of the different reporter 3' UTR and the binding of miR-124 to it. The seed region of miR-124 are highlighted in gray  
Right: conservation of miR-124 binding sequence in Gria2 transcripts across mammals.
- b) RFP fluorescence in cells lines stably expressing a control reporter is not modified in GFP<sup>-</sup> cells (control for experiments in Fig 1b). n=3 independent experiments.
- c) Illustration of FACS sorting gating strategy for experiments in Fig 1c. Untransfected cells (left) were used to set up the sorting-windows.
- d) In transient transfection experiments, miR-124 levels in GFP<sup>+</sup> cells increased over time. No significant difference can be observed across reporter cell lines. n=3 independent experiments.

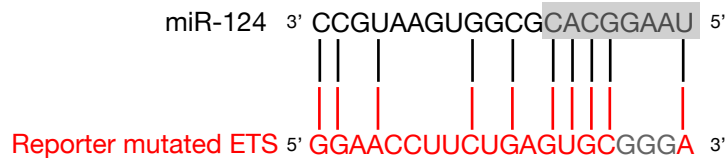
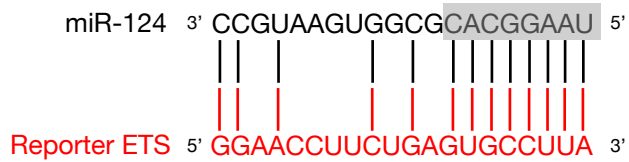
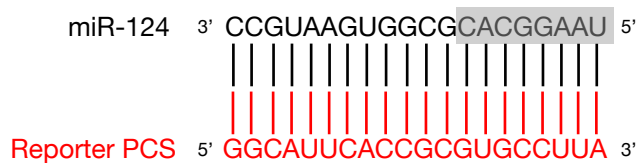
**Supplementary Figure 2. Control experiments for transient transfection of miR-124 reporters.**

- a) Quantification of miR-124 in HEK293T cells stably expressing this miRNA. As a reference, we used the levels of miR-124 in transient transfection experiments. Our stable cell lines display much lower levels of miR-124 (about 25% and 10% of the reference). n=2 independent experiments.
- b) In HEK293T cells (devoid of miR-124) no downregulation of any miR-124 reporter can be observed. n=2 independent experiments.
- c) Mutating 3 nucleotides in the seed region of the 1x ETS reporter abrogate the effect observed in HEK293T cells expressing low or moderate levels of miR-124. n=3 independent experiments.

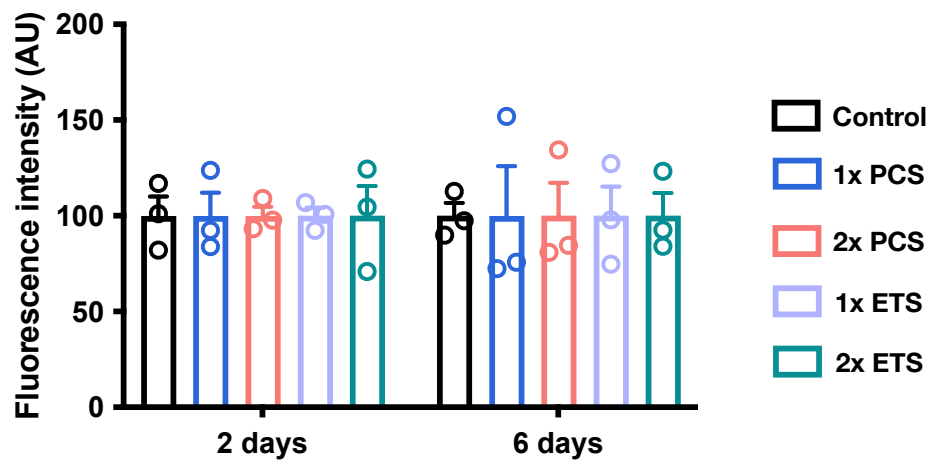
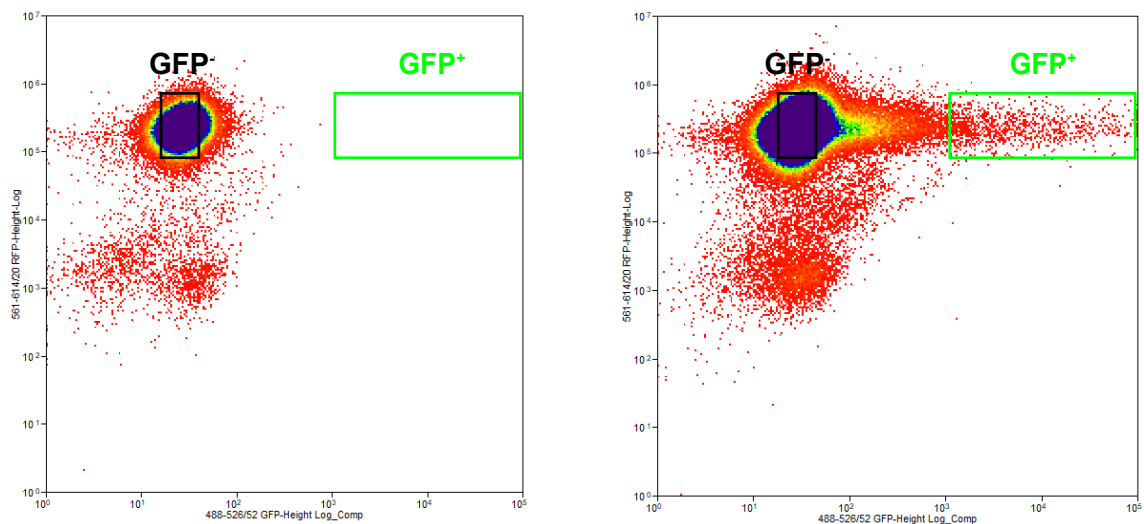
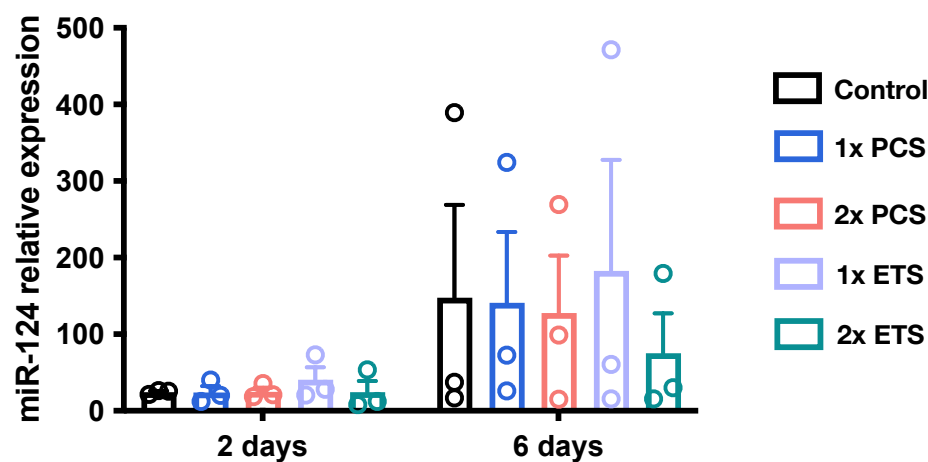
**Supplementary Figure 3. Testing reporter activity in primary neuronal cultures from cortex, cerebellum and hippocampus.**

- a) Proportion of RFP<sup>+</sup> and GFP<sup>+</sup> cells in primary neurons coming from different brain structures. (n=3 independent for hippocampus and cortex, n=4 for cerebellum).
- b) RFP and GFP levels in neuronal cultures transduced with AAV-RFP reporters. Whilst GFP fluorescence showed no change across conditions, RFP was strongly downregulated in neuronal cultures transduced with the miR-124 reporter. Two-way ANOVA, Dunnett post-hoc test (n=3 independent for hippocampus and cortex, n=4 for cerebellum).
- c) Gating strategy in primary cerebellar neurons infected with different AAV-RFP reporters for quantification of miR-124. Of note, in cultures infected with the AAV-RFP-miR-124, a leftward switch is observed in the fluorescence intensity.

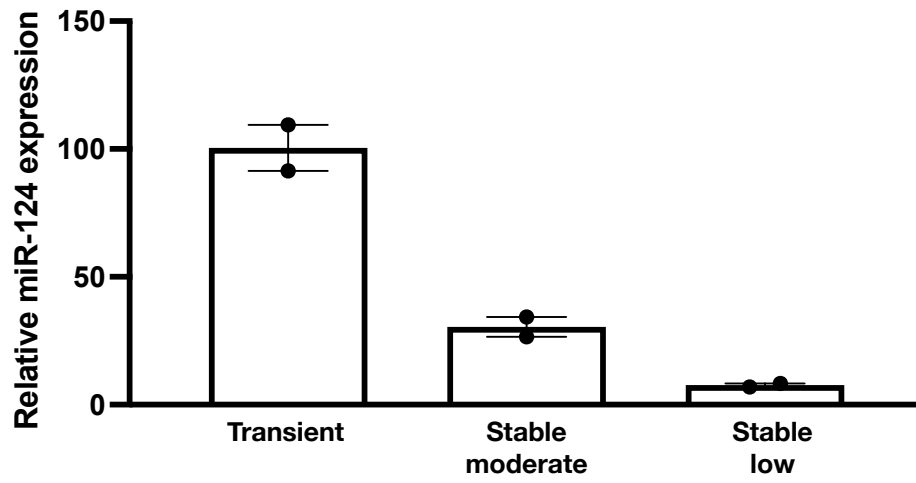
**Supplementary Figure 4. Schematic illustration of plasmid maps used in this studies.**

**a****miR-124 target sequence in Gria 2 transcript**

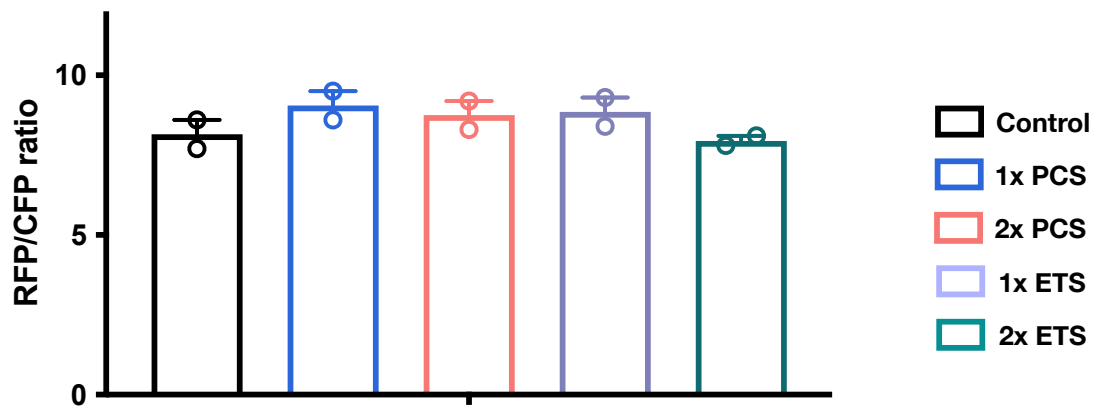
	150	160	170	180	190	200	210	220	230
Mouse	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Human	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Chimp	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Rhesus	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Squirrel	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Rat	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Rabbit	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Pig	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	AC	-	AAAGUGGU
Cow	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Cat	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Dog	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Brown bat	CG	U	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAUGGU
Elephant	A	-	GUGACUGGUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAUGGU
Opossum	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Macaw	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Chicken	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AAAGUGGU
Lizard	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	CAAGUGGU
X. tropicalis	A	-	GUGACUGAUCUCUGGAUUGAUAGAACCUCUUGAGGCCUACACAUGGU	-	UUUCUUGUGUUUUAUG	-	UC	-	AACGUGGU

**b****c****d**

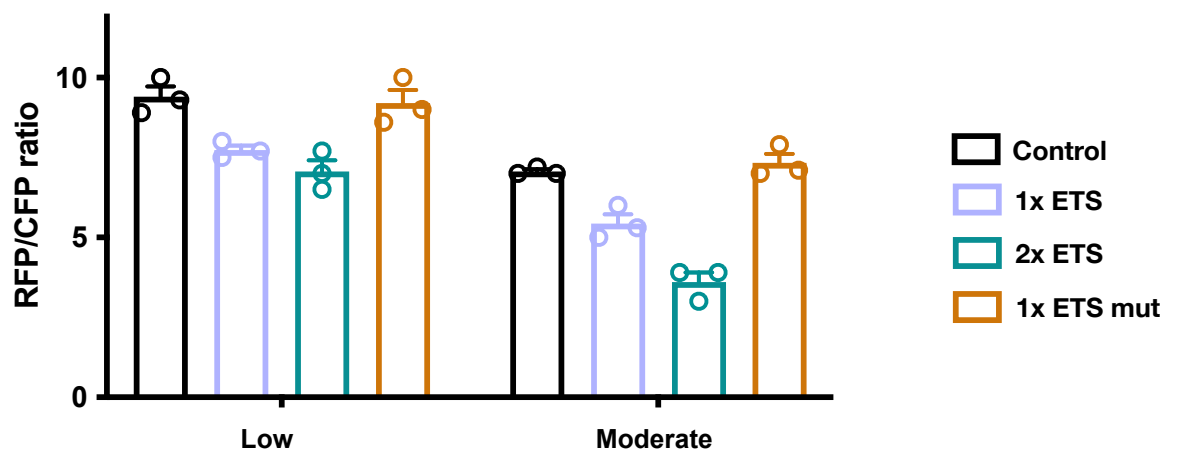
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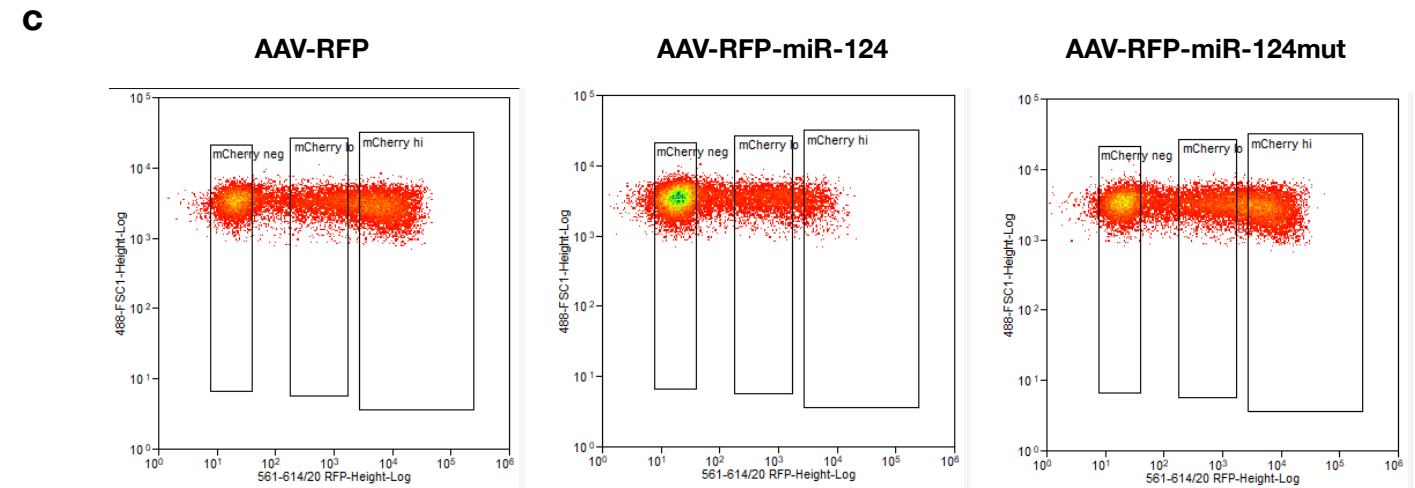
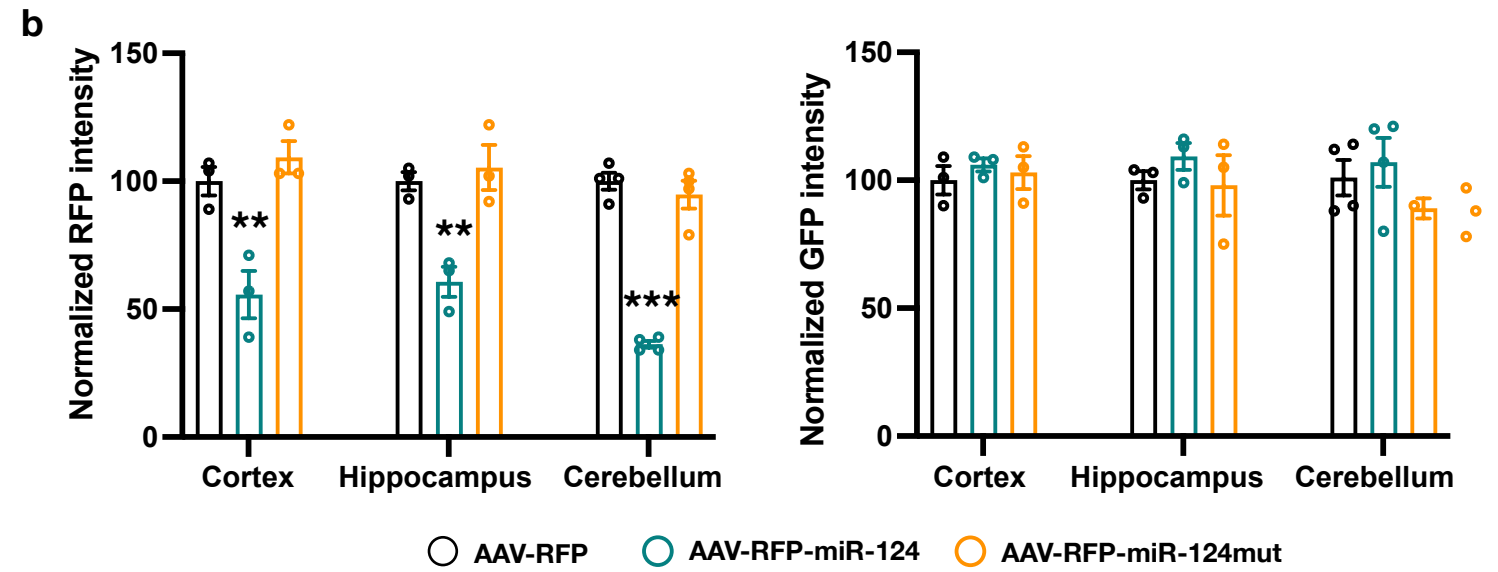
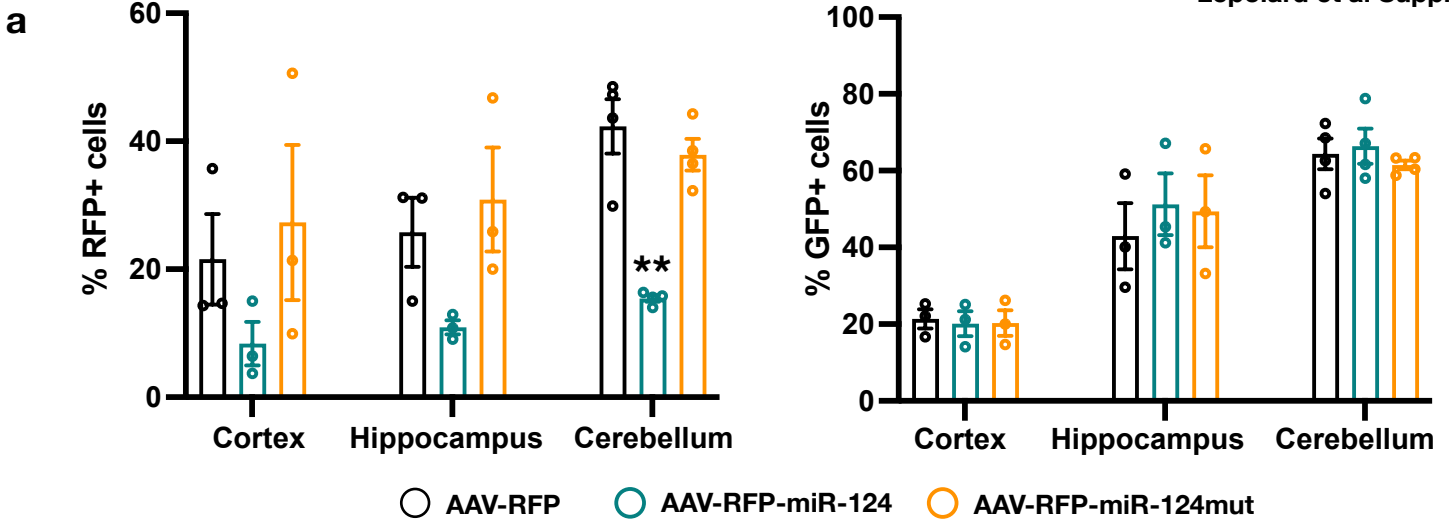


b



c





Lepolard et al Suppl Figure 4

