



**S13 Fig: RNA-sequencing analysis of *Phf6*-deleted and control E15.5 cortex and E16.5 cultured cortical neurons**

(A) Multidimensional scaling (MDS) plot of N = 5 *Phf6*<sup>-/-</sup> vs. 3 *Phf6*<sup>+/-</sup> E15.5 cortices samples used for RNA-sequencing analysis, showing clustering of samples after adjustment for litter-effect; color-coded by genotype.

(B) MDS plot of N = 4 *Phf6*<sup>lox/Y</sup>; *Nes-cre*<sup>Tg/+</sup> vs. 3 *Phf6*<sup>+/-</sup>; *Nes-cre*<sup>Tg/+</sup> E16.5 cortical neuron isolates used for RNA-sequencing analysis showing clustering of samples after adjustment for litter-effect; color-coded by genotype.

(C,D) Mean-difference plot for *Phf6*<sup>-/-</sup> vs. *Phf6*<sup>+/-</sup> E15.5 cortex RNA-sequencing data showing log<sub>2</sub>-fold change for each gene over average log<sub>2</sub> expression (C). Mean-difference

plot for *Phf6*<sup>lox/Y</sup>;*Nes-cre*<sup>Tg/+</sup> vs. *Phf6*<sup>+Y</sup>;*Nes-cre*<sup>Tg/+</sup> E16.5 cultured cortical neuron RNA-sequencing data (D). Significantly up- and downregulated genes in the *Phf6* deleted samples relative to controls are coloured red and blue respectively. NS = not significant.

(E) Log<sub>2</sub> fold-change in RNA levels of ZIC family transcription factor genes in E15.5 *Phf6*<sup>-Y</sup> vs. *Phf6*<sup>+Y</sup> cortex.

(F) Log<sub>2</sub> fold-change in RNA levels of the *Usp18* gene in *Phf6*<sup>lox/Y</sup>;*Nes-cre*<sup>Tg/+</sup> vs. *Phf6*<sup>+Y</sup>;*Nes-cre*<sup>Tg/+</sup> E16.5 cortical neurons.

RNA sequencing data were analysed as described in the methods section.