



**Supplemental Figure S3. Effects of ADAR1 knockdown in H9 cells upon motor neuron differentiation.**

(A) Repression of neuron-specific gene expression in ADAR1 KD H9 cells upon MN differentiation. Heatmaps showing the hierarchical clustering of expression profiles of different expressed genes (DEGs) as measured by RNA-seq. Pairwise distances were measured by PCC. Different gene clusters were labeled besides the gene tree with

different colors. The data shown in the maps were logarithm transformed expression values.

(B) ADAR1 KD hESCs exhibited clusters of repressed neuronal gene expression upon MN differentiation. A representative cluster (#11) containing 109 genes was significantly repressed in ADAR1 KD H9 cells after the induction of NE (d10) ( $P = 0.08$  for d0,  $4.8e-10$  for d10,  $8.0e-9$  for d 17 or  $8.7e-7$ for d35, pairwise t-test).

(C) Cluster 11 enriches motor neuron specific functions.

(D) Verification of RNA-seq using RT-qPCR analysis. Error bars represent  $\pm$  SD in triplicate experiments.