

Expanded View Figures

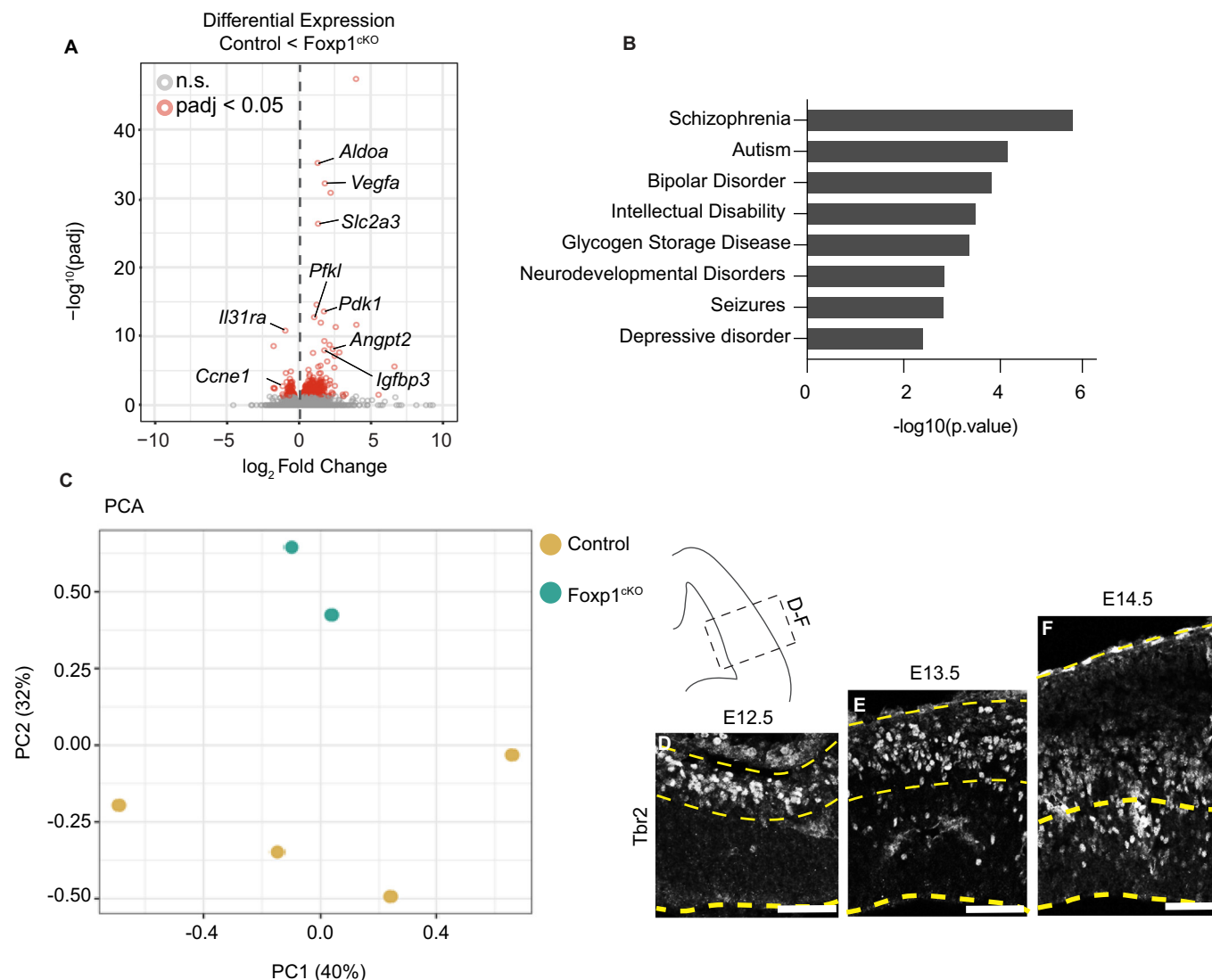
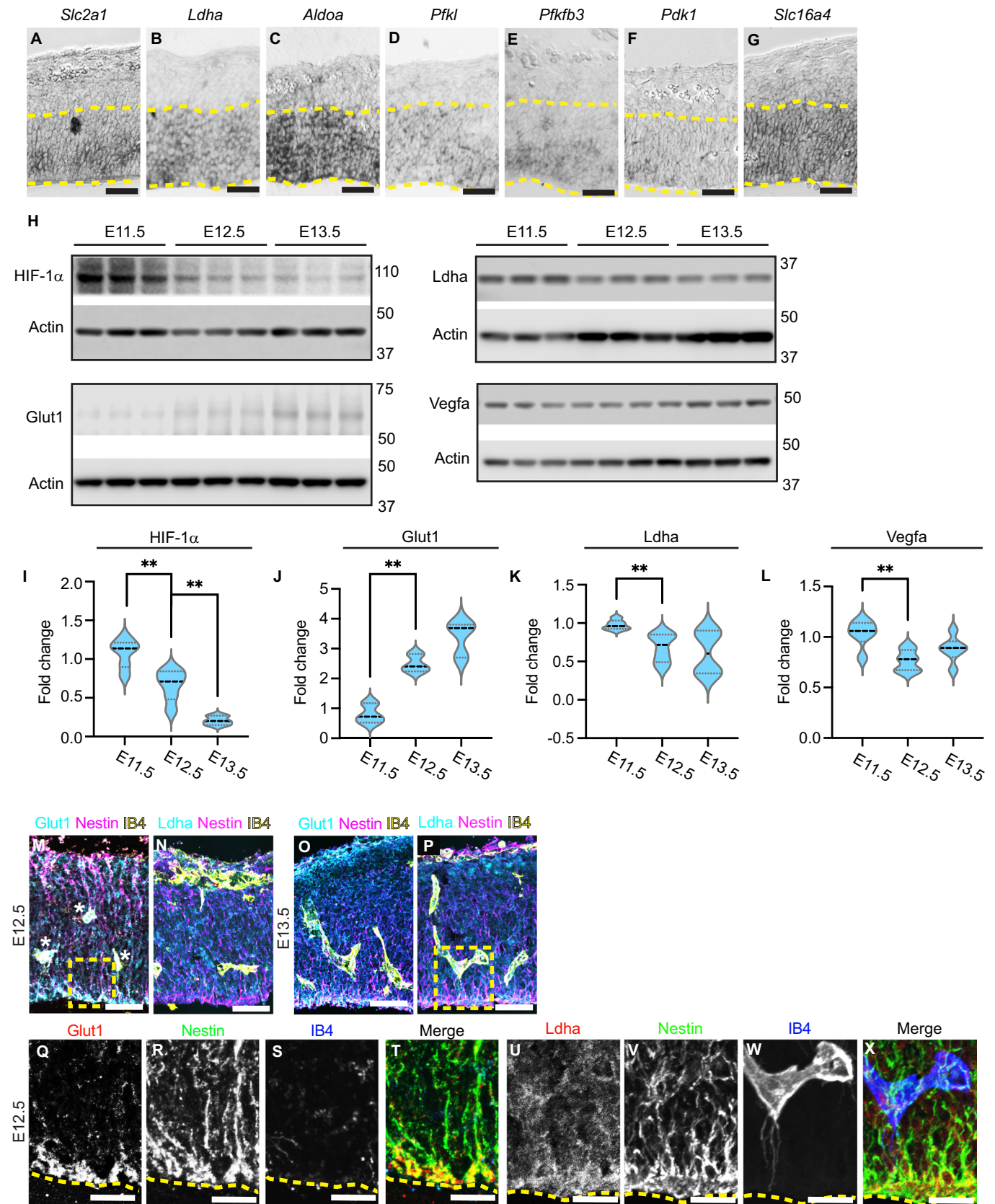


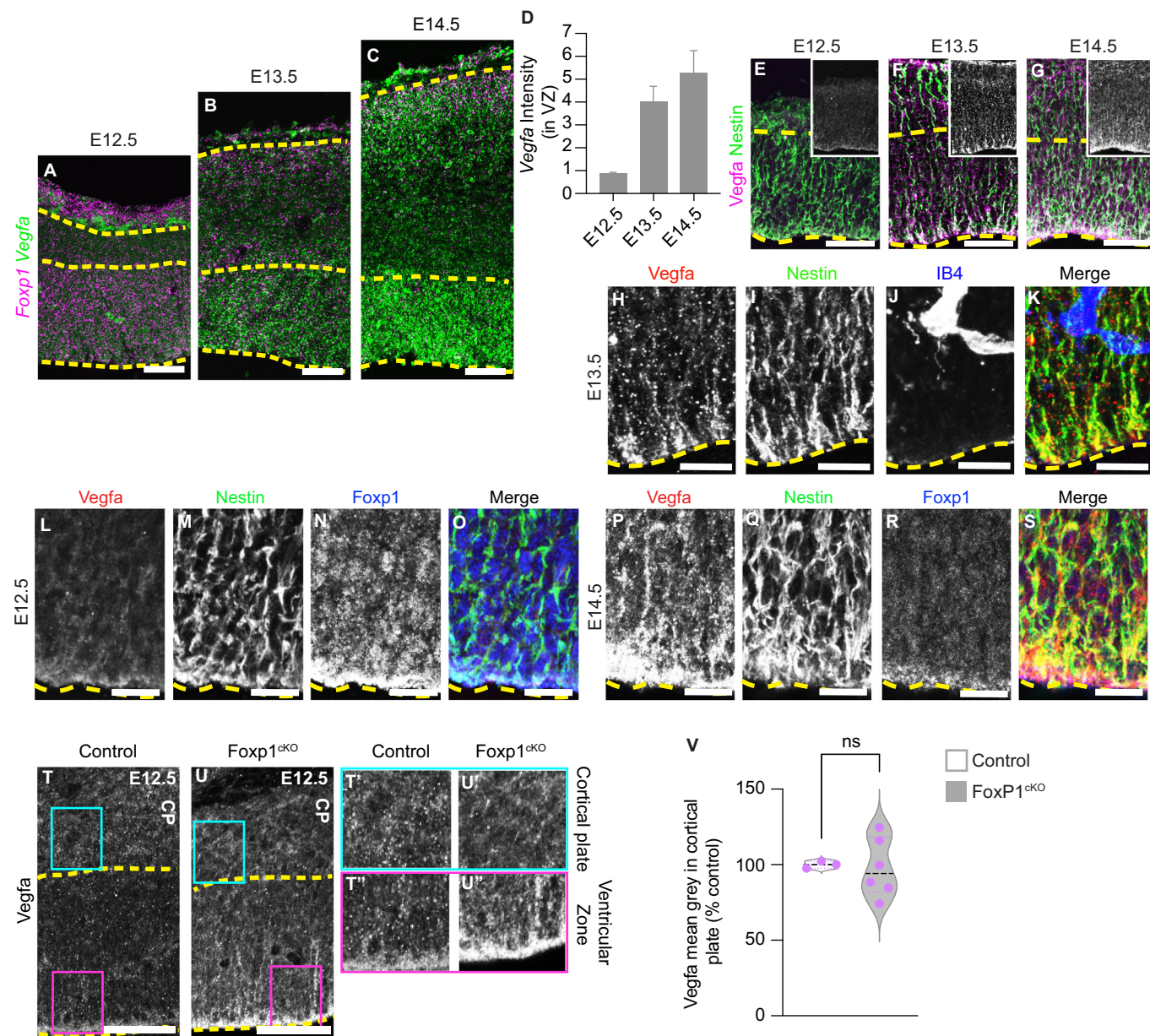
Figure EV1. RNA Seq analysis of Foxp1^{ckO} cortex.

(A) Volcano plot of gene expression changes in the absence of Foxp1 in E12.5 lateral cortex compared to control embryos. Gray circles denote non-significant gene changes (adjusted p -value > 0.05); red circles denote significantly differentially expressed genes (adjusted p -value < 0.05). (B) Human disorders associated with genes significantly misregulated in Foxp1^{ckO} mutants at E12.5. (C) The principal component analysis (PCA) of control and Foxp1^{ckO} mutants shows PC1 and PC2. (D-F) IHC for Tbr2⁺ intermediate progenitors in wild-type cortex at E12.5, E13.5, and E14.5. Schematic denotes area image in (D-F). Scale bars 50 μm. Data information: significance determined by ANOVA (A, B).



◀ **Figure EV2. HIF-1 α target gene expression in RG in the wild-type cortex.**

(A–G) Wild-type mRNA expression of glycolysis genes *Slc2a1*, *Ldha*, *Aldoa*, *Pfkfb3*, *Pdk1*, and *Slc16a4* in the wild-type lateral cortex at E12.5. (H) Western blot analysis of HIF-1 α , Glut1, Ldha, and Vegfa (with Beta Actin) in wild-type cortical lysates at E11.5, E12.5, E13.5. (I) Fold change of HIF-1 α levels normalized to Beta Actin between E11.5 and E13.5. (J) Fold change of Glut1 levels normalized to Beta Actin between E11.5 and E13.5. (K) Fold change of Ldha levels normalized to Beta Actin between E11.5 and E13.5. (L) Fold change of Vegfa levels normalized to Actin between E11.5 and E13.5. (M, N) IHC for Glut1 and Ldha with Isolectin B4, and Nestin at E12.5 in the wild-type cortex. (O, P) IHC for Glut1 or Ldha with Isolectin B4, and Nestin at E13.5 in the wild-type cortex. Boxed areas are magnified in (Q–X). (Q–T) High magnification image of IHC for Glut1 in Nestin+ RG at E12.5. Isolectin B4 labels blood vessels. (U–X) High magnification images of IHC for Ldha in Nestin+ RG at E13.5. Isolectin B4 labels blood vessels. Scale bars 50 μ m (A–K), 10 μ m (L–S). Data information: $N = 5$ –9 embryos per time point, 3–6 replicates. $p = 0.0039$ and 0.0073 (I). $N = 5$ –9 embryos per time point, 3 replicates. $p = 0.0028$ (J). $N = 5$ –9 embryos per time point, 6 replicates. $p = 0.0041$ (K). $N = 5$ –9 embryos per time point, 6 replicates. $p = 0.0017$ (L). All Student's t-tests. All data represented as mean $\pm$ SEM. Source data are available online for this figure.



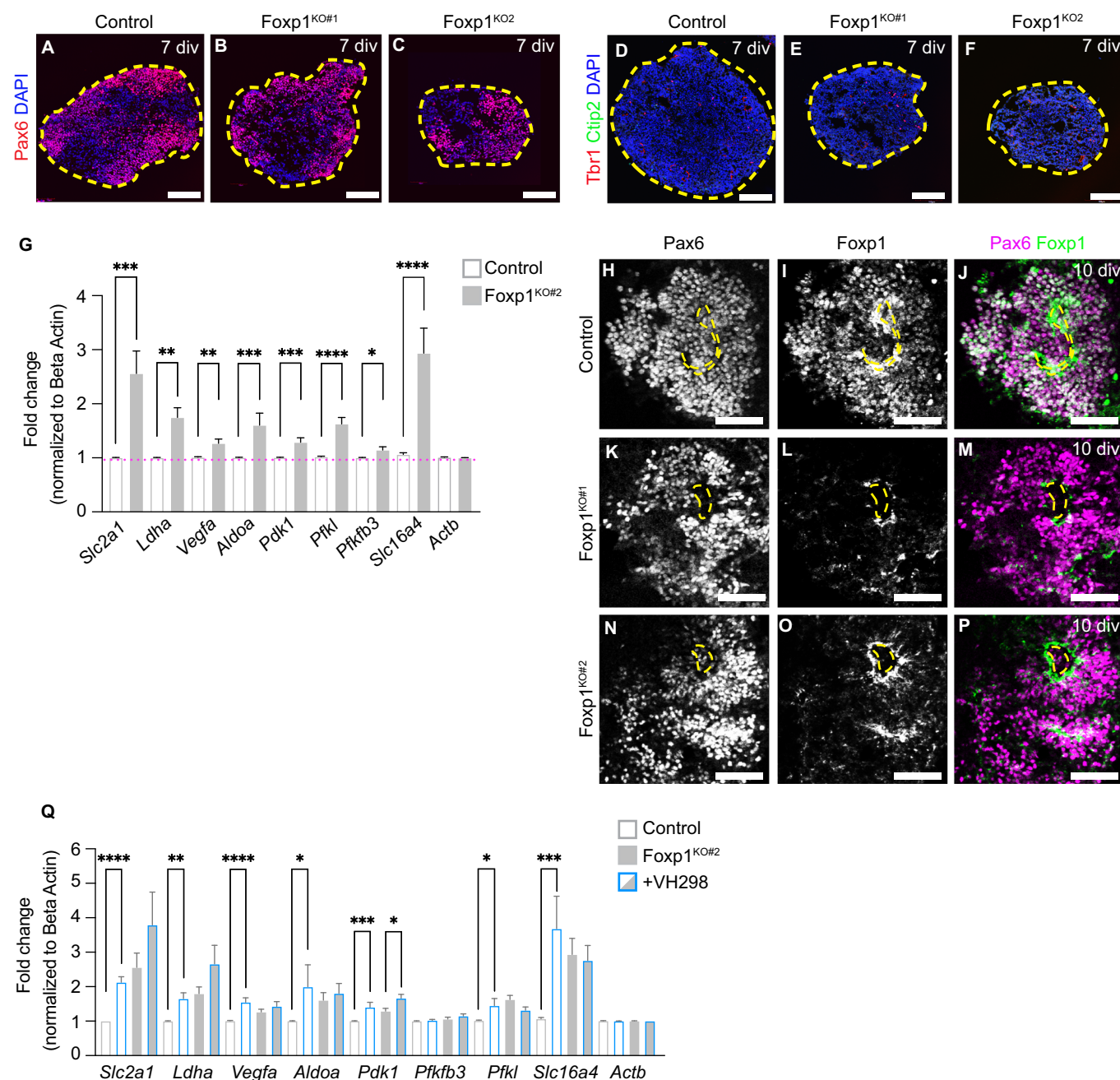


Figure EV4. Analysis of Foxp1-deficient cortical spheroids.

(A–C) IHC for Pax6 in control and Foxp1^{KO} spheroids at 7 days in vitro (div). (D–F) IHC for Ctip2 and Tbr1⁺ neurons in control and Foxp1^{KO} spheroids at 7 div. (G) qPCR analysis of HIF-1α target gene expression in control and Foxp1^{KO#2} spheroids at 10 div. (H–P) IHC for Foxp1 in Pax6⁺ NPCs in control and Foxp1^{KO} spheroids at 10 div. (Q) qPCR analysis of HIF-1α target gene expression in control and Foxp1^{KO#2} spheroids at 10 div. treated with DMSO or VH298. Scale bars 100 μm (A–F), 50 μm (H–P). Data information: *N* = 10–12 spheroids from 3 individual batches. *p* = 0.0007 (*Slc2a1*), 0.0108 (*Ldha*), 0.0031 (*Vegfa*), 0.0009 (*Aldoa*), 0.0002 (*Pdk1*), 0.0002 (*Pfkfb3*), 0.0226 (*Pfkfb3*), <0.0001 (*Slc16a4*). Student's *t*-test (G). *p* = <0.0001 (*Slc2a1*, control), 0.0013 (*Ldha*, control), <0.0001 (*Vegfa*, control), 0.0179 (*Aldoa*, control), 0.0002 (*Pdk1*, control), 0.0312 (*Pdk1*, Foxp1^{KO#2}), 0.0201 (*Pfkfb3*, control), 0.0003 (*Slc16a4*, control). Student's *t*-test (Q). All data represented as mean ± SEM.