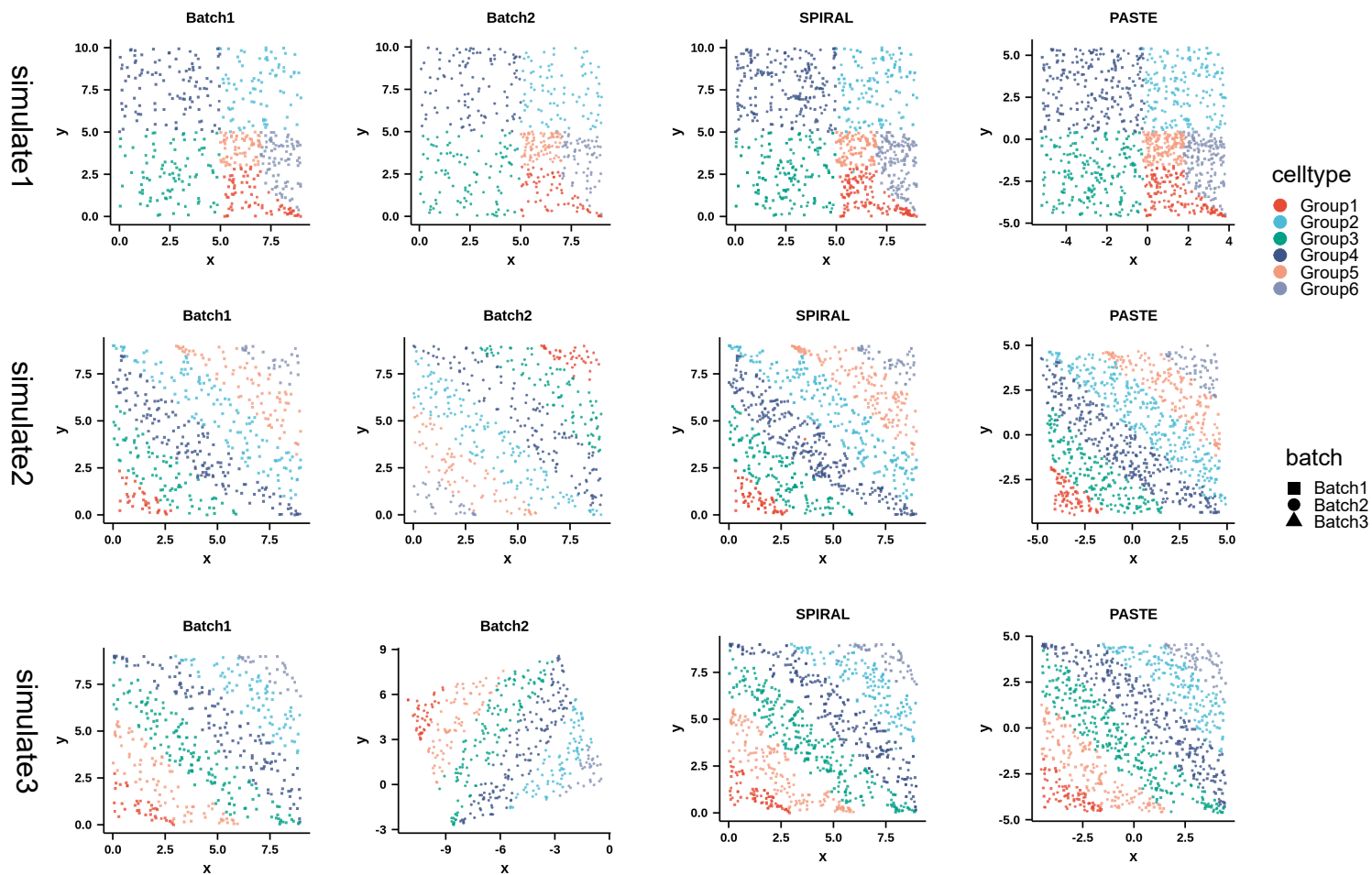
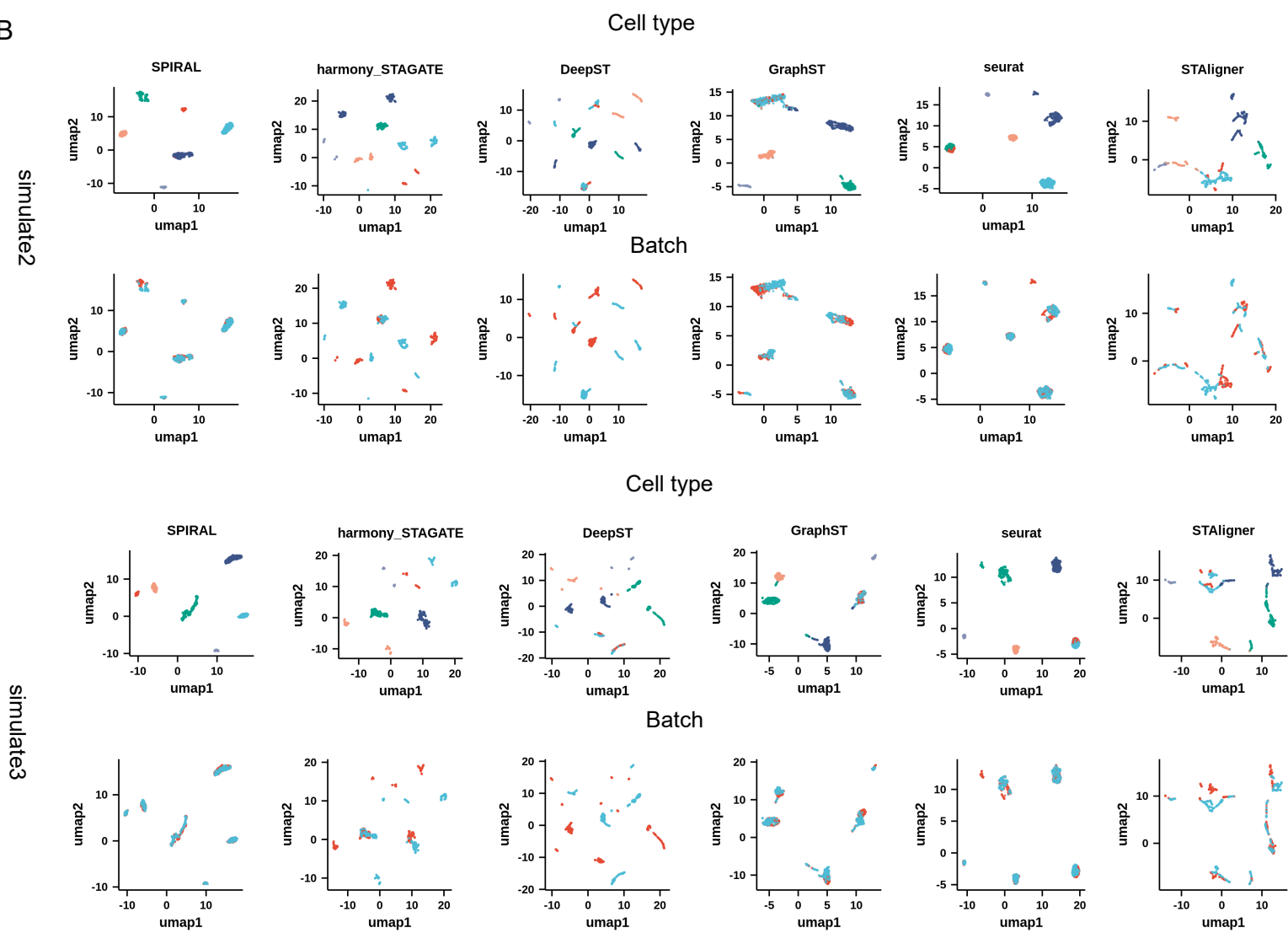


Figure S1

A



B



C

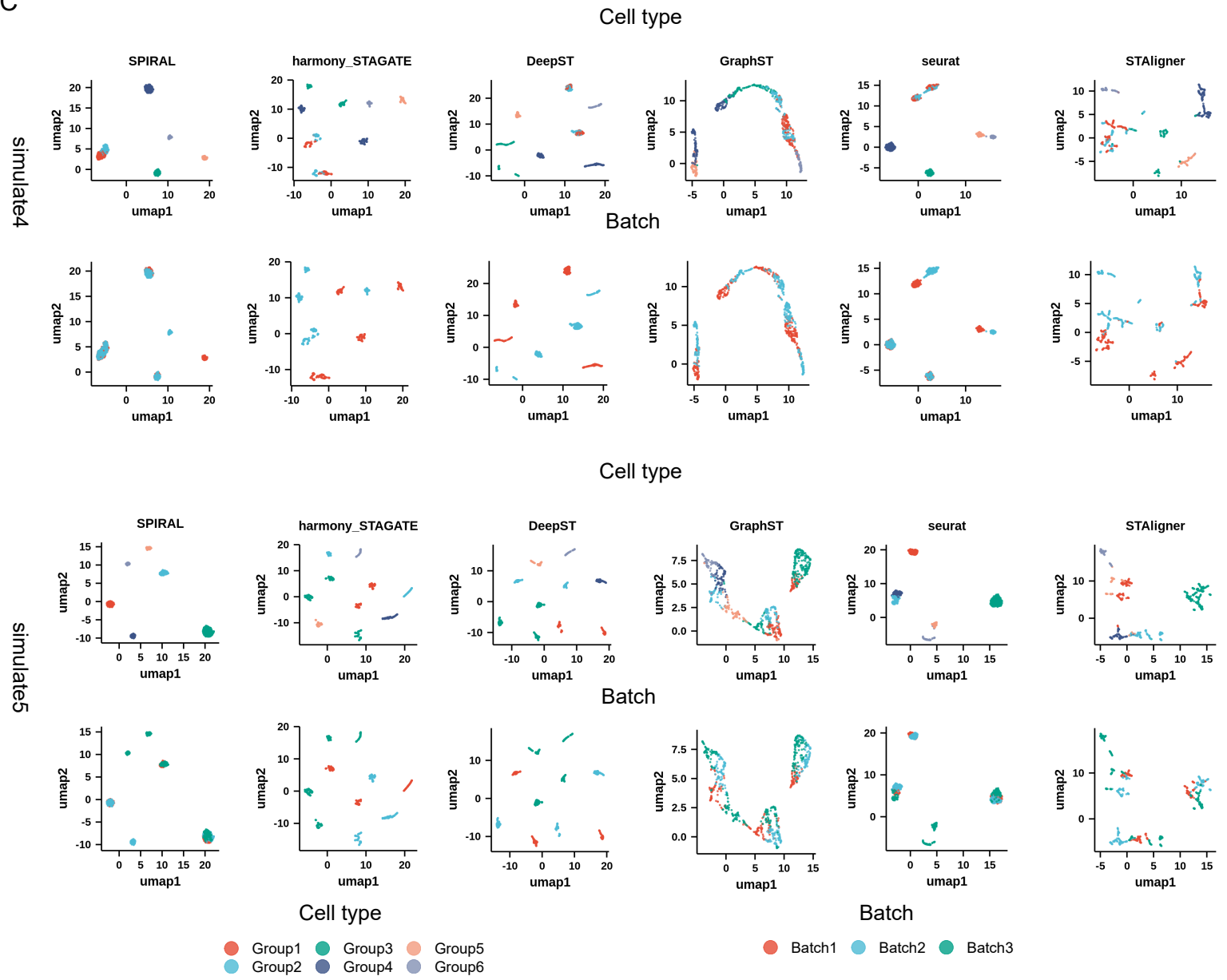
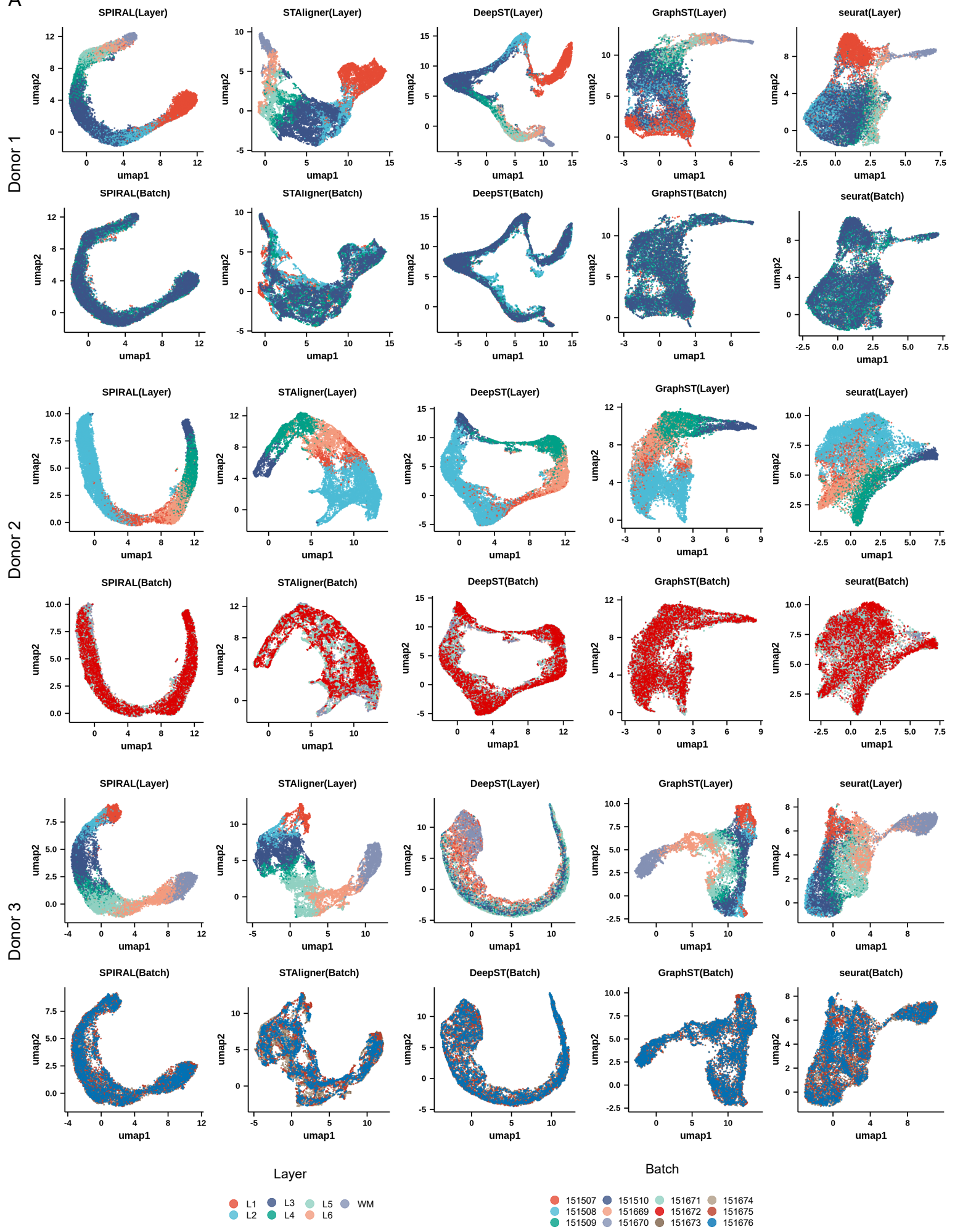
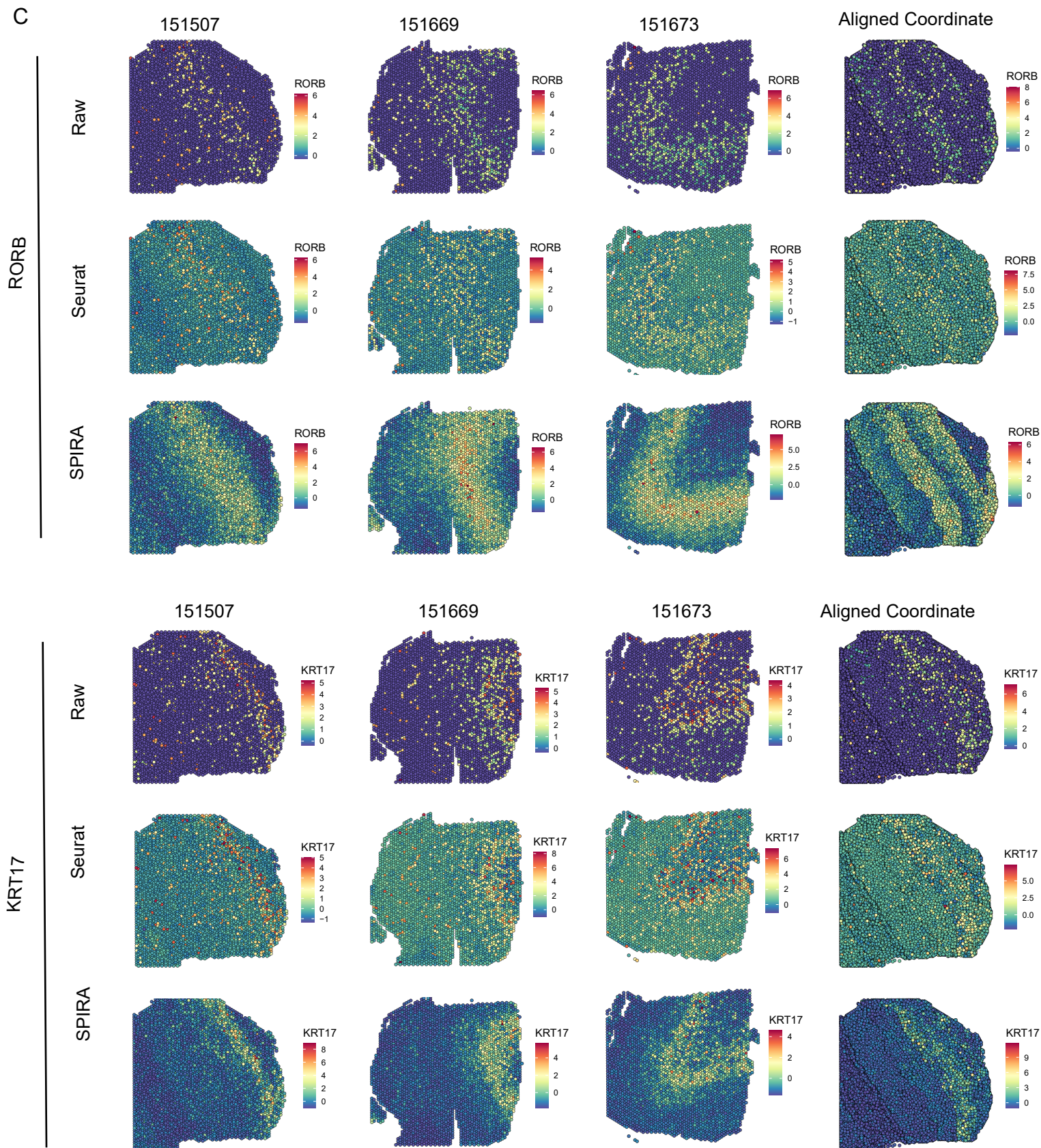
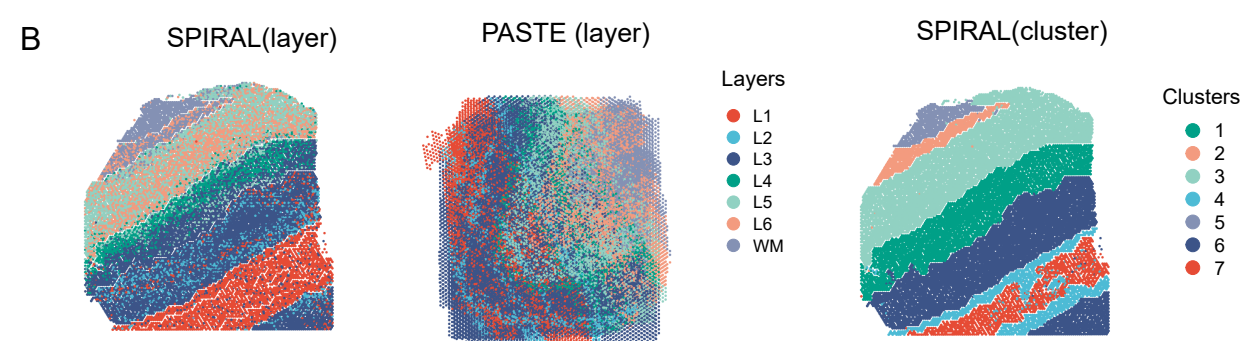


Figure S1: Simulating. **A** spatial arrangements of cell types on original and aligned coordinates by SPIRAL and PASTE for simulate 1-3 datasets. Colors represent cell types and shapes represent batches. **B-C** UMAP visualizations of simulate2-3 (B) and simulate4-5 (C) datasets, each dataset occupies two rows, in which colors represent cell types (upper) and batches (bottom).

Figure S2

A





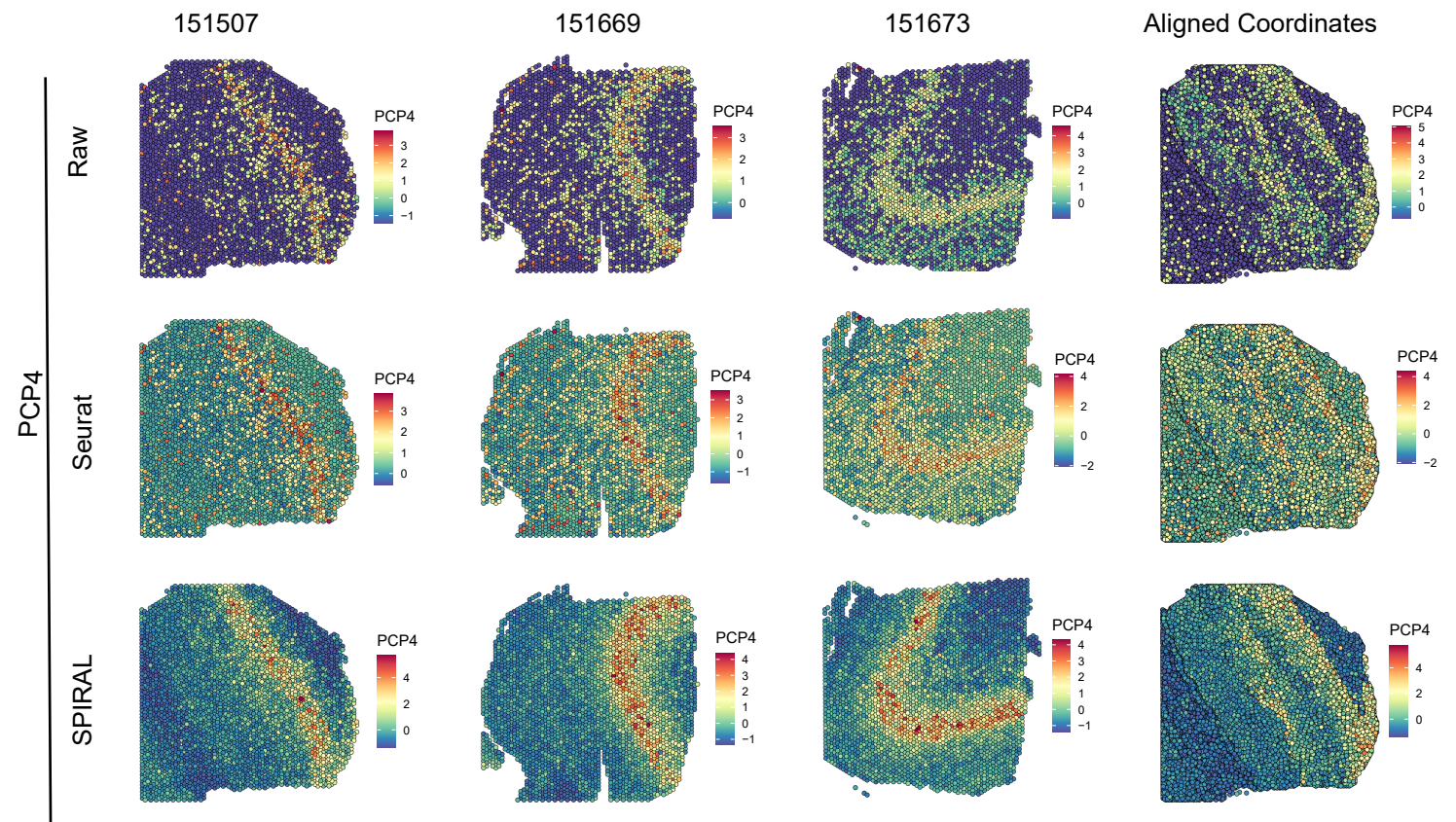
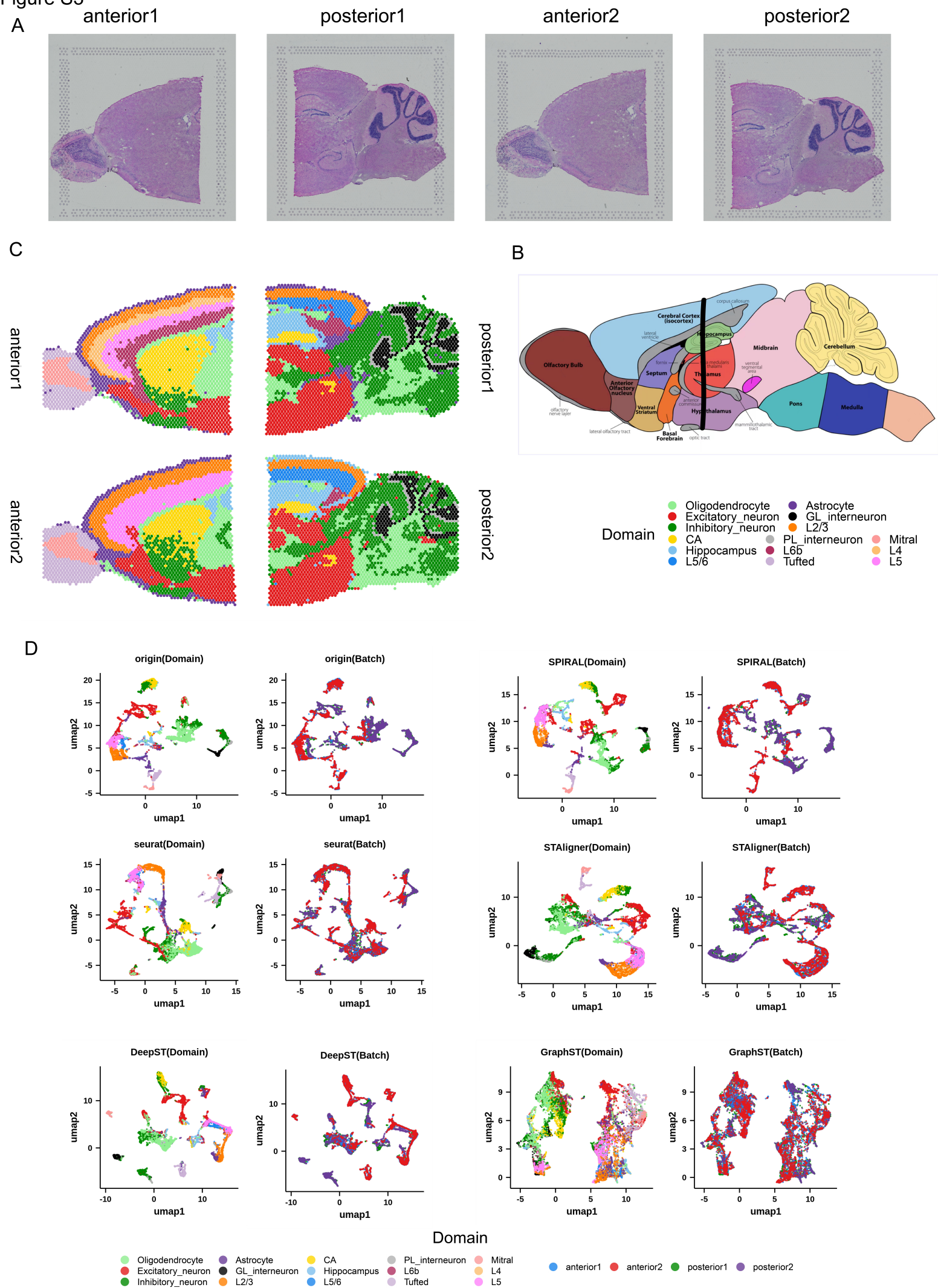


Figure S2: DLPFC. **A** UMAP visualizations of four-section integrations for each donor (donor1 in upper, donor2 in middle, donor3 in bottom). **B** Spatial arrangements of annotated layers on aligned coordinates by SPIRAL and PASTE (left, middle) and spatial distributions of clusters (mclust; right) on SPIRAL-aligned coordinates. **C** Spatial distributions of raw, Seurat and SPIRAL-integrated domain-marker gene expressions, RORB, KRT17 and PCP4 in 151507, 151669, 151673 and SPIRAL-aligned coordinates.

Figure S3



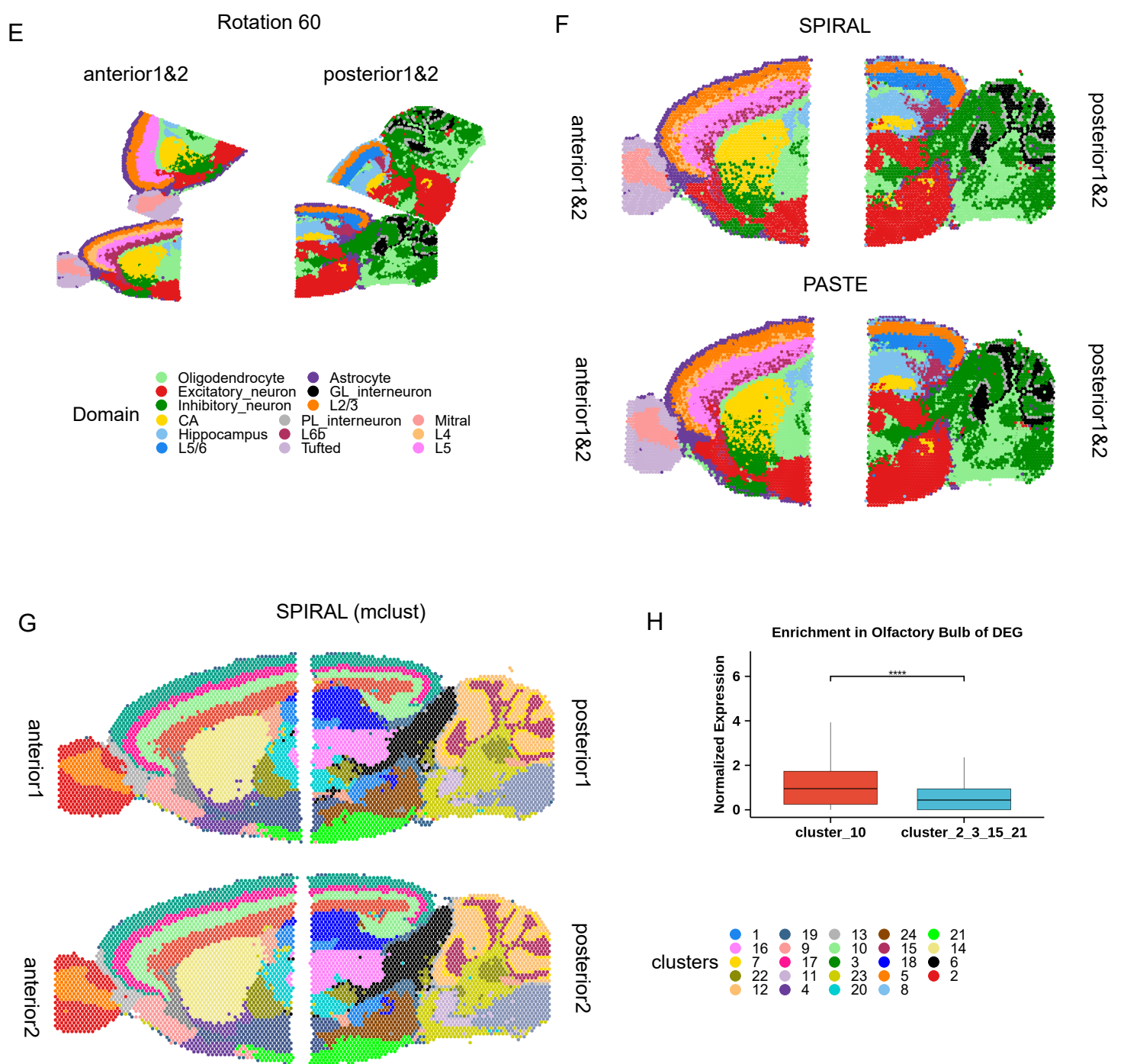
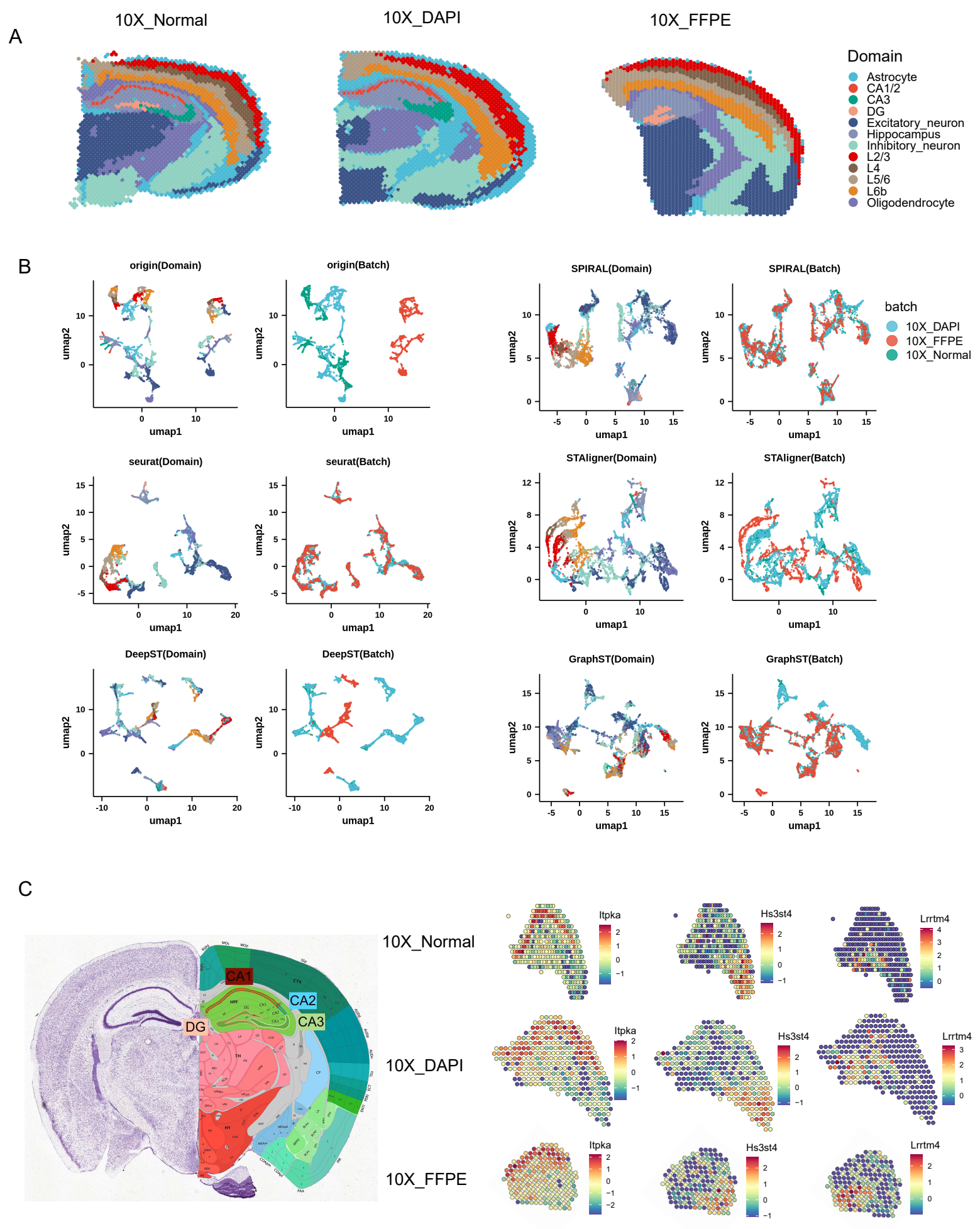


Figure S3: Sagittal mouse brain. **A** H&E staining images of both anterior and posterior mouse brains. **B** The anatomy diagram of sagittal mouse brain, where black line divides mouse brain into anterior and posterior parts. **C** spatial visualizations of annotated domain labels for four samples. **D** UMAP visualizations of raw and integrated embeddings by five methods in integrating four samples. Each method occupies two columns, where domains are colored in left and batches are colored in right. **E** Spatial arrangements of annotated domains on samples with one of replicates were rotated 60 degree. **F** Spatial arrangements of annotated domains on aligned coordinates of anterior1&2 (left) and posterior1&2 (right) by SPIRAL (upper) and PASTE (bottom). **G** Spatial disposition of mclust-derived clusters by SPIRAL on four samples. **H** Comparison of the expressions of DEGs from cluster10 and DEGs from cluster 2,3,15,21 in mouse olfactory bulb domains.

Figure S4



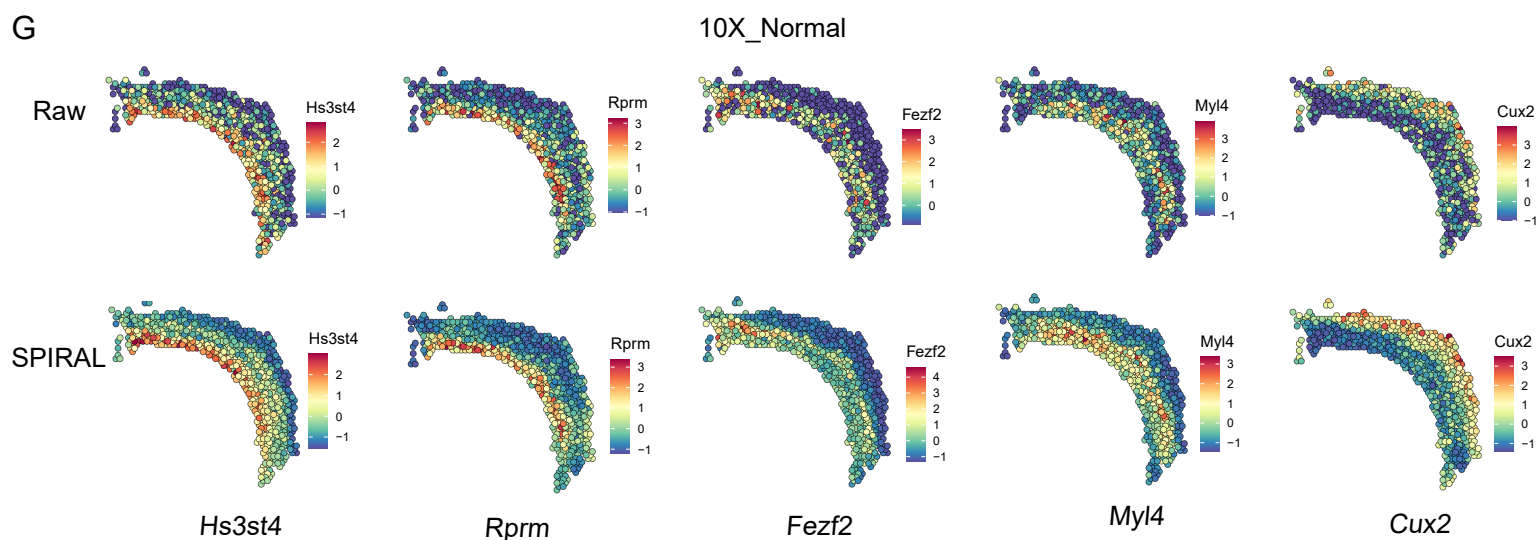
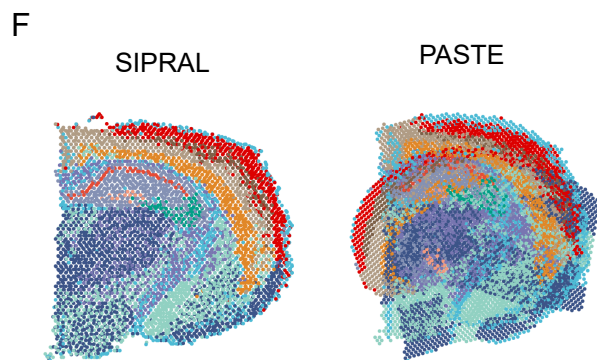
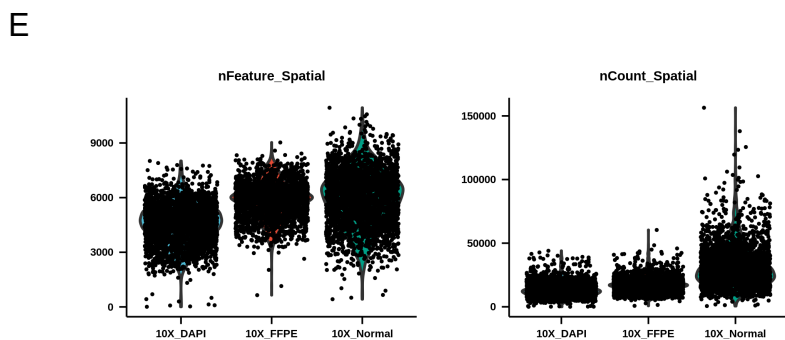
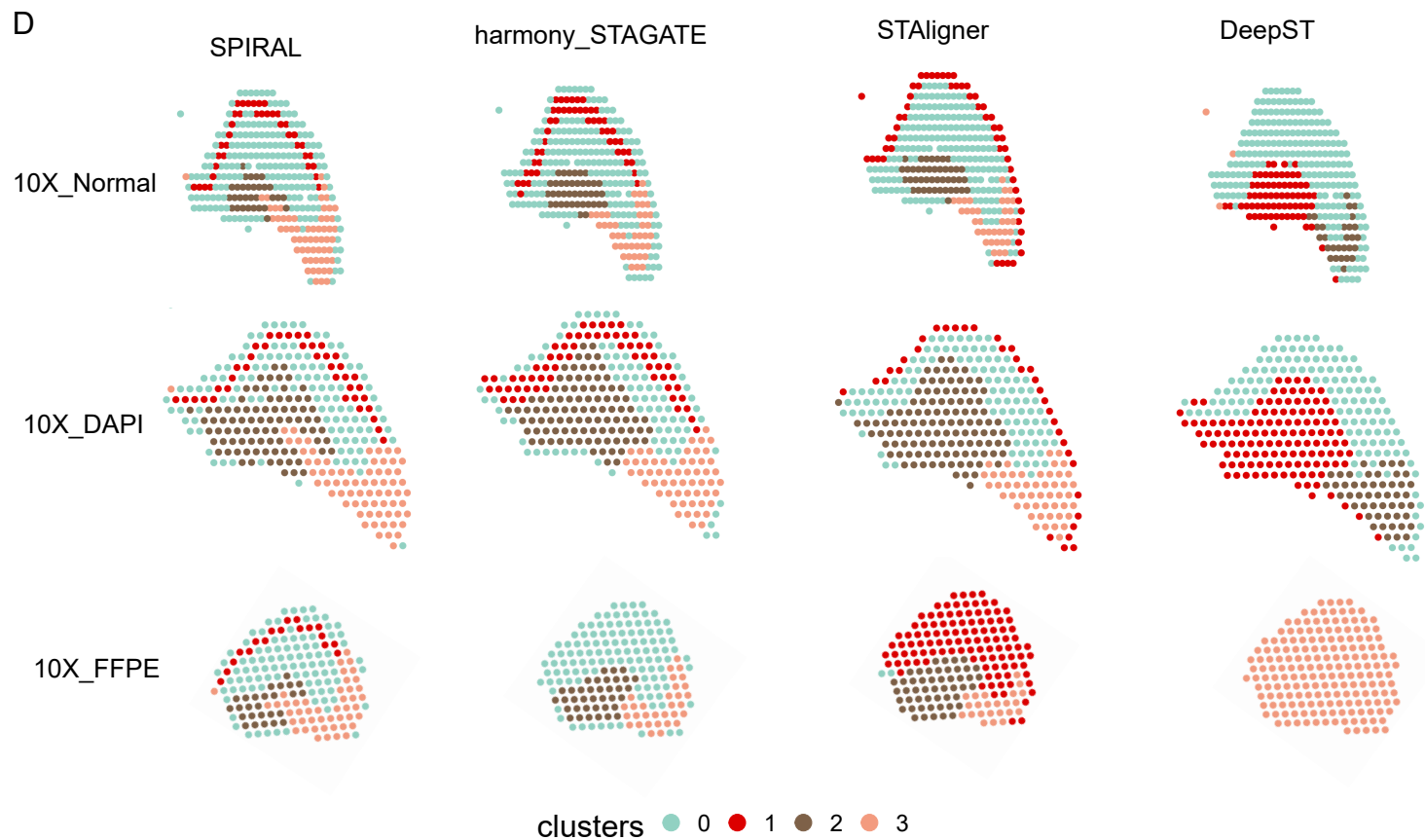
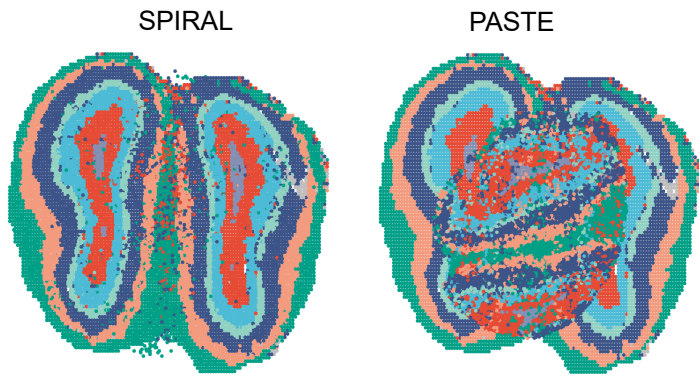


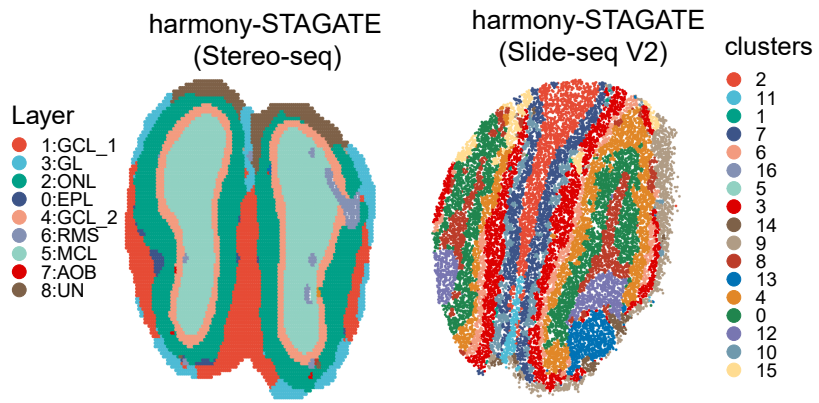
Figure S4: Coronal mouse brain. **A** Annotations of domains generated by marker genes and Allen mouse brain atlas in three coordinates. **B** UMAP visualizations of integrations of embeddings from origin and five methods. **C** The anatomy diagram of coronal mouse brain for hippocampus structures (left) and the distributions of the corresponding gene expressions (right). **D** The arrangements of derived clusters by for spatial based methods on the region of hippocampus. **E** Statistics of sequence depth of three protocols. **F** Spatial arrangements of annotated domains on aligned coordinates by SPIRAL (left) and PASTE (right). **G** Spatial patterns of domain-marker gene expressions from raw data and SPIRAL integrated data in the cortex region of 10X Normal data.

Figure S5

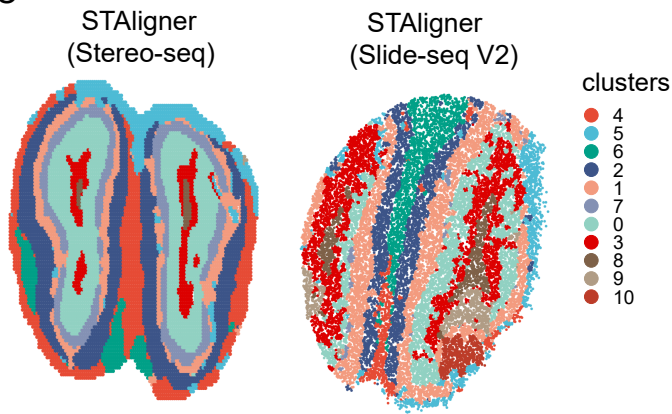
A



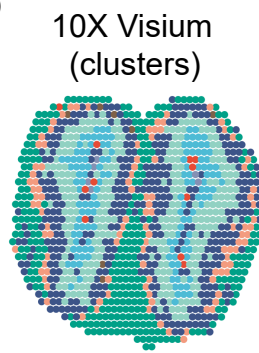
B



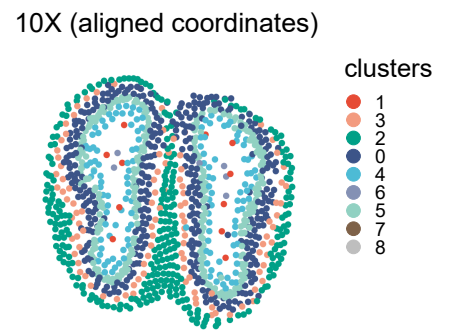
C



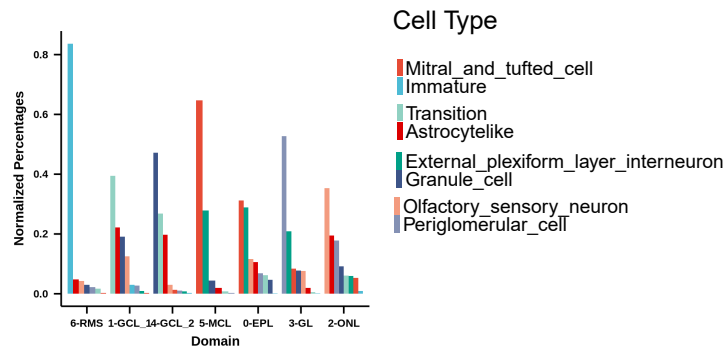
D



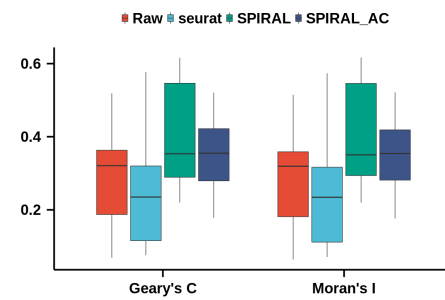
E



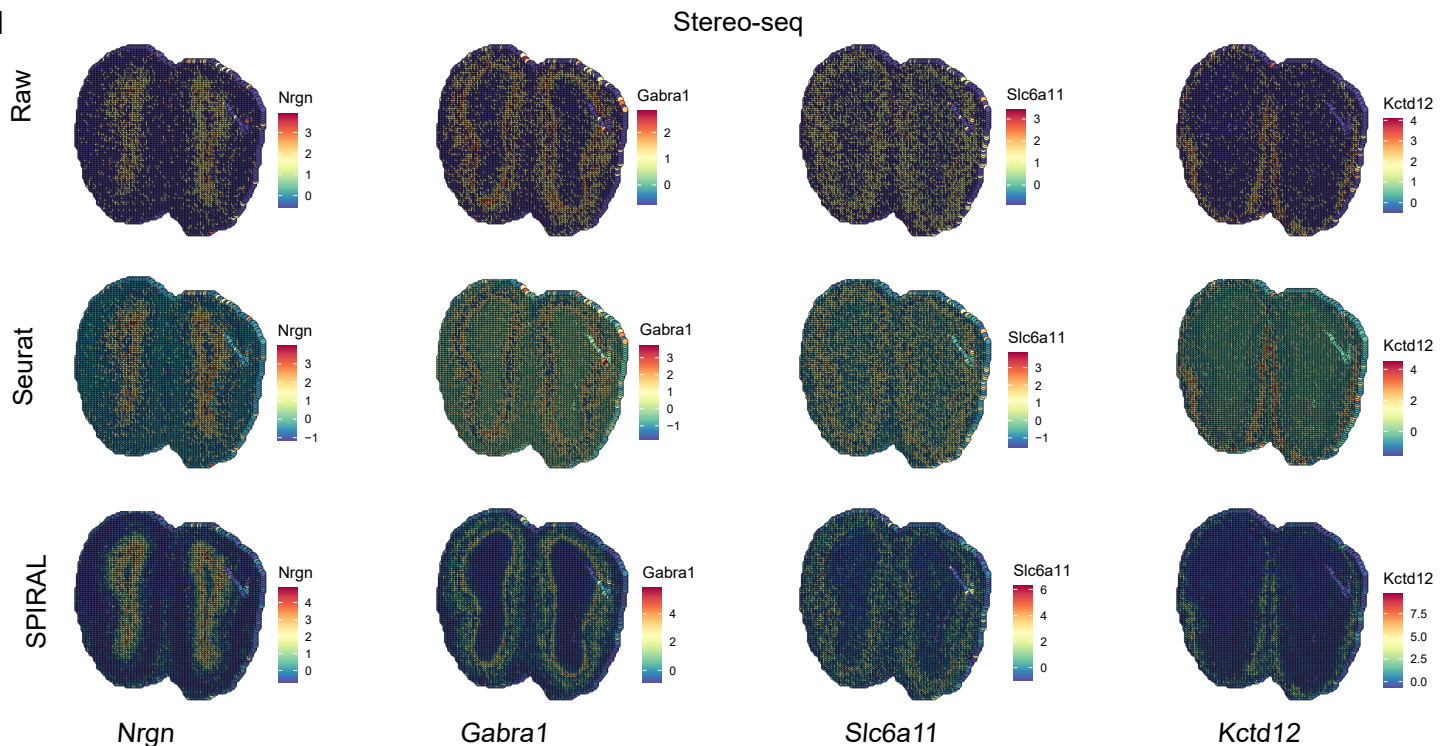
F Cell type Assignment



G Spatial autocorrelation



H



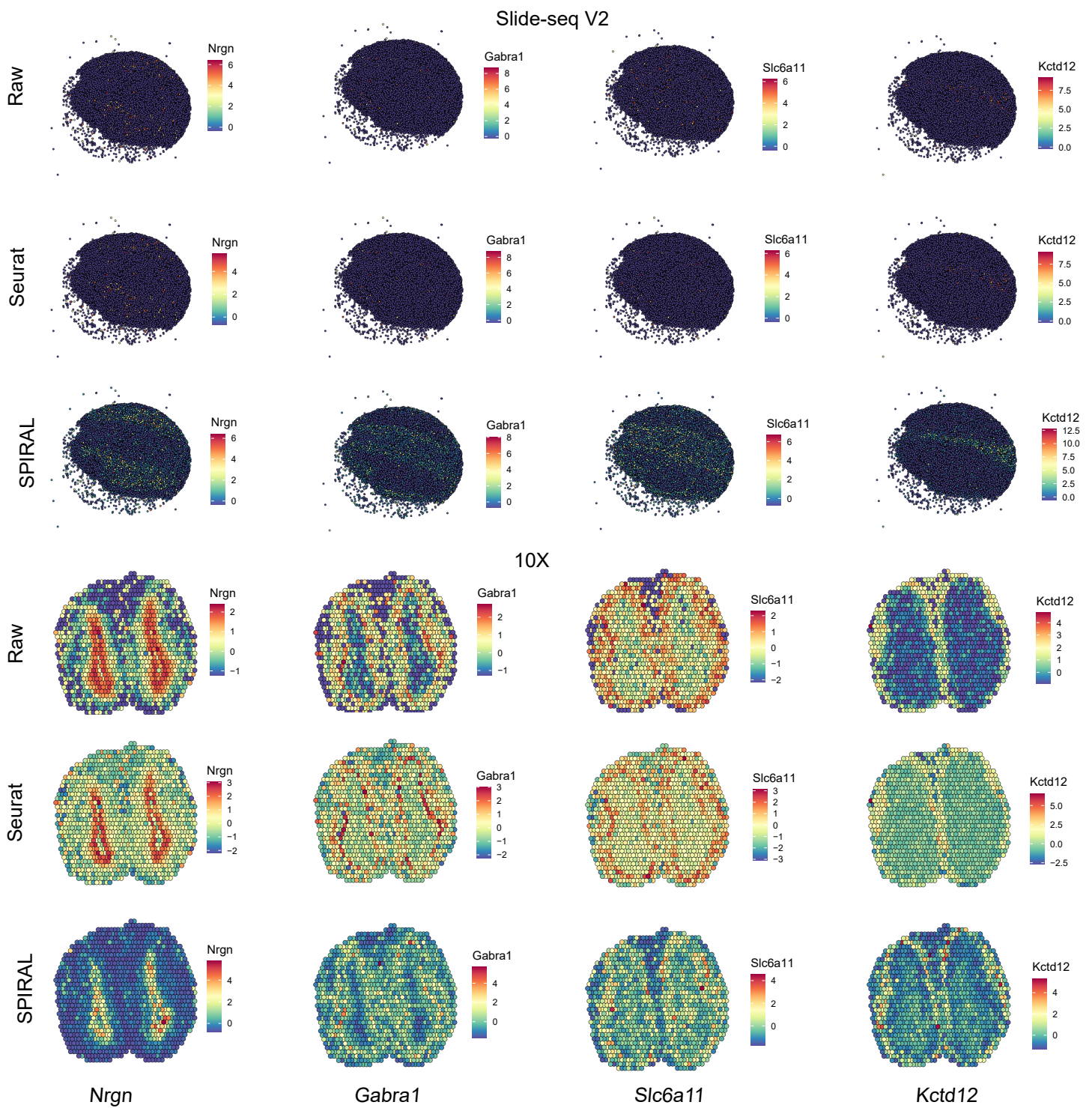


Figure S5: Mouse olfactory bulb. **A** Spatial distributions of clusters derived by SPIRAL in aligned coordinates by SPIRAL (left) and PASTE (right). **B&C** Spatial distributions of Louvain-derived clusters on the embeddings of harmony_STAGATE (B) and STAligner (C). **D** Spatial distributions of predicted clusters of 10X Visium data using model trained by Stereo seq data and Slide V2 data. **E** Spatial distributions of predicted clusters on aligned coordinates by SPIRAL. **F** The percentages of cell types in each cluster. **G** Spatial autocorrelations of raw and integrated gene expressions in original and aligned coordinates. **H** Spatial distributions of raw, SPIRAL-integrated and Seurat-integrated layer-marker expressions, Nrgn, Gabra1, Slc6a11 and Kctd12 on three samples. Each method occupies one row.