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Fig. S12 Cross-species comparison of cell identities between inner tepals and stamens of *N. colorata* and *P. aphrodite*.

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Fig. S16 The spatial distribution of genes related to gibberellin signaling pathway (Fig. 4E) at various developmental stages of water lily floral buds.

Fig. S17 The spatial distribution of genes related to ethylene signaling pathway (Fig. 4E) at various developmental stages of water lily floral buds.

Fig. S18 Spatial expression of genes related to vegetative morphogenesis at different developmental stages of water lily floral buds, as presented in Fig. 5E.

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Table. S12 Annotation of marker genes of *N. colorata* five major tissues gained at different evolutionary nodes.

Table. S13 GO enrichment related to plant organ establishment.

Table. S14 In situ hybridization probes of target genes.

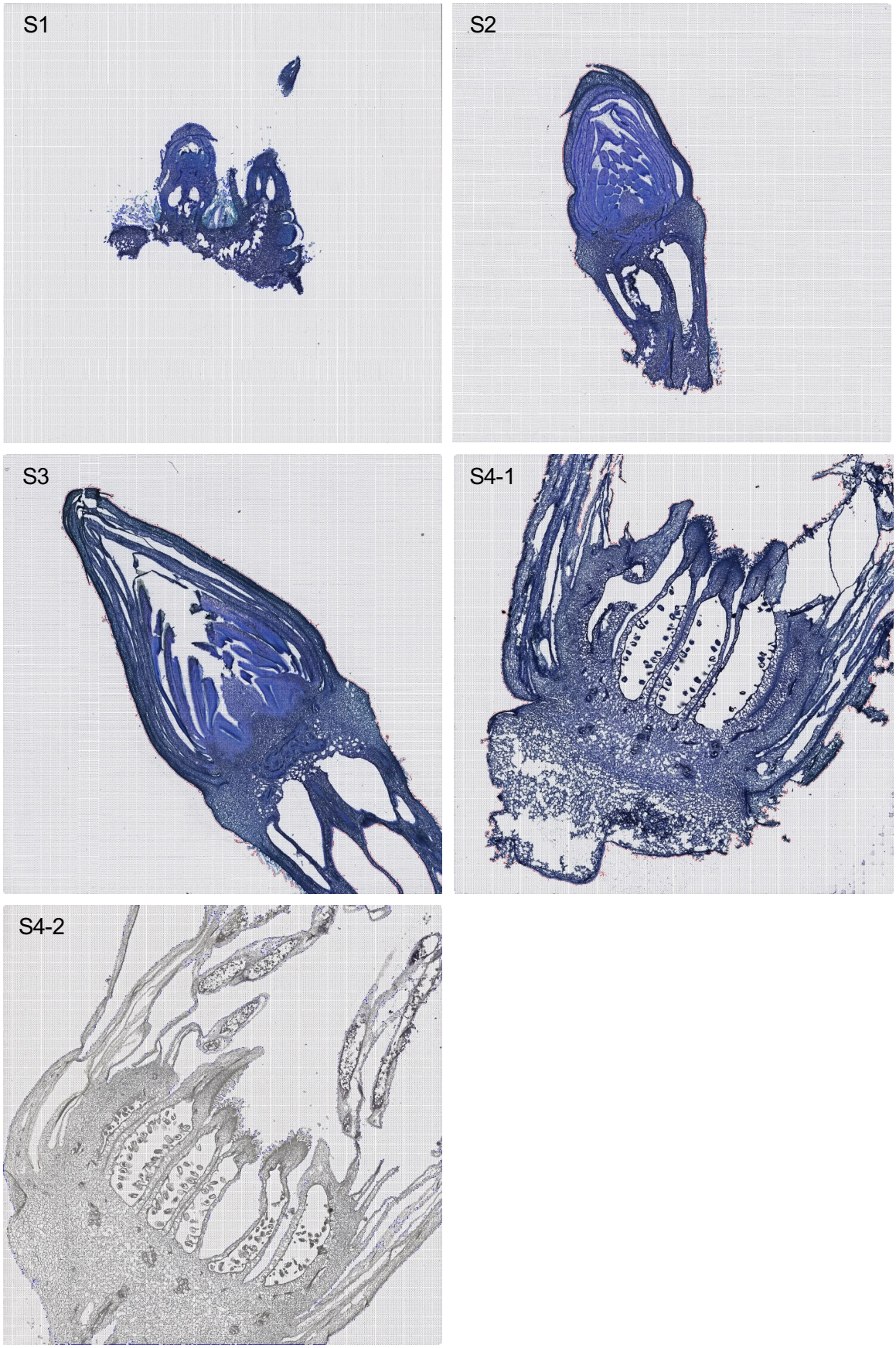


Fig. S1 Display of longitudinal section images of water lily flowers at developmental stages S1 to S4.

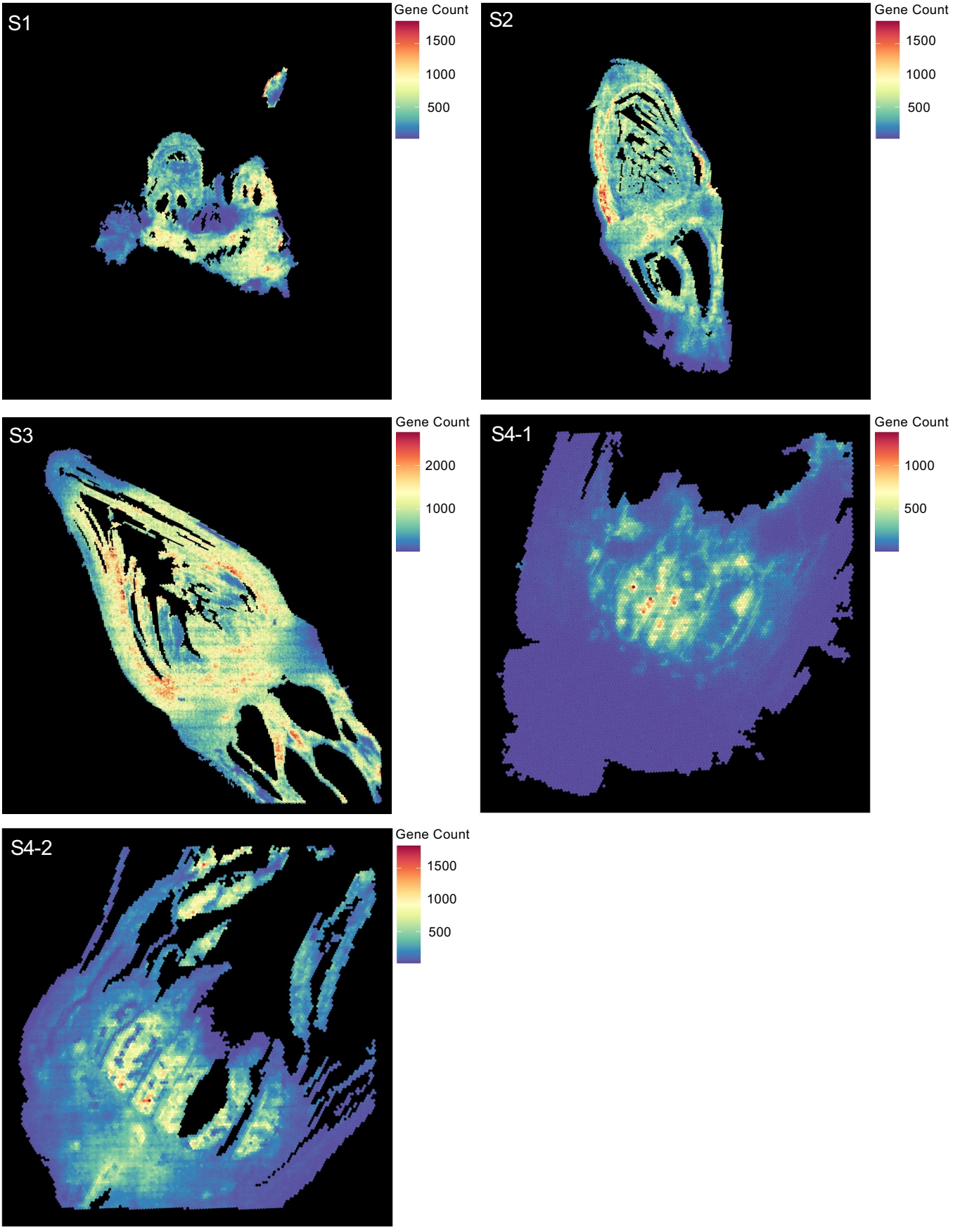
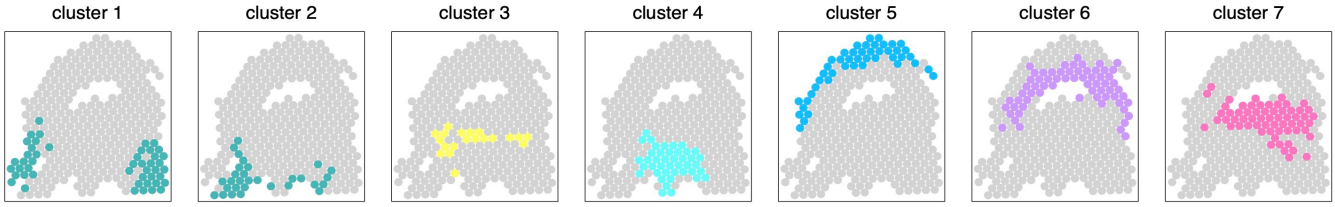


Fig. S2 Display of gene count per spot on the spatial transcriptome datasets of water lily flowers at developmental stages S1 to S4.

7 clusters for S1



18 clusters for S2

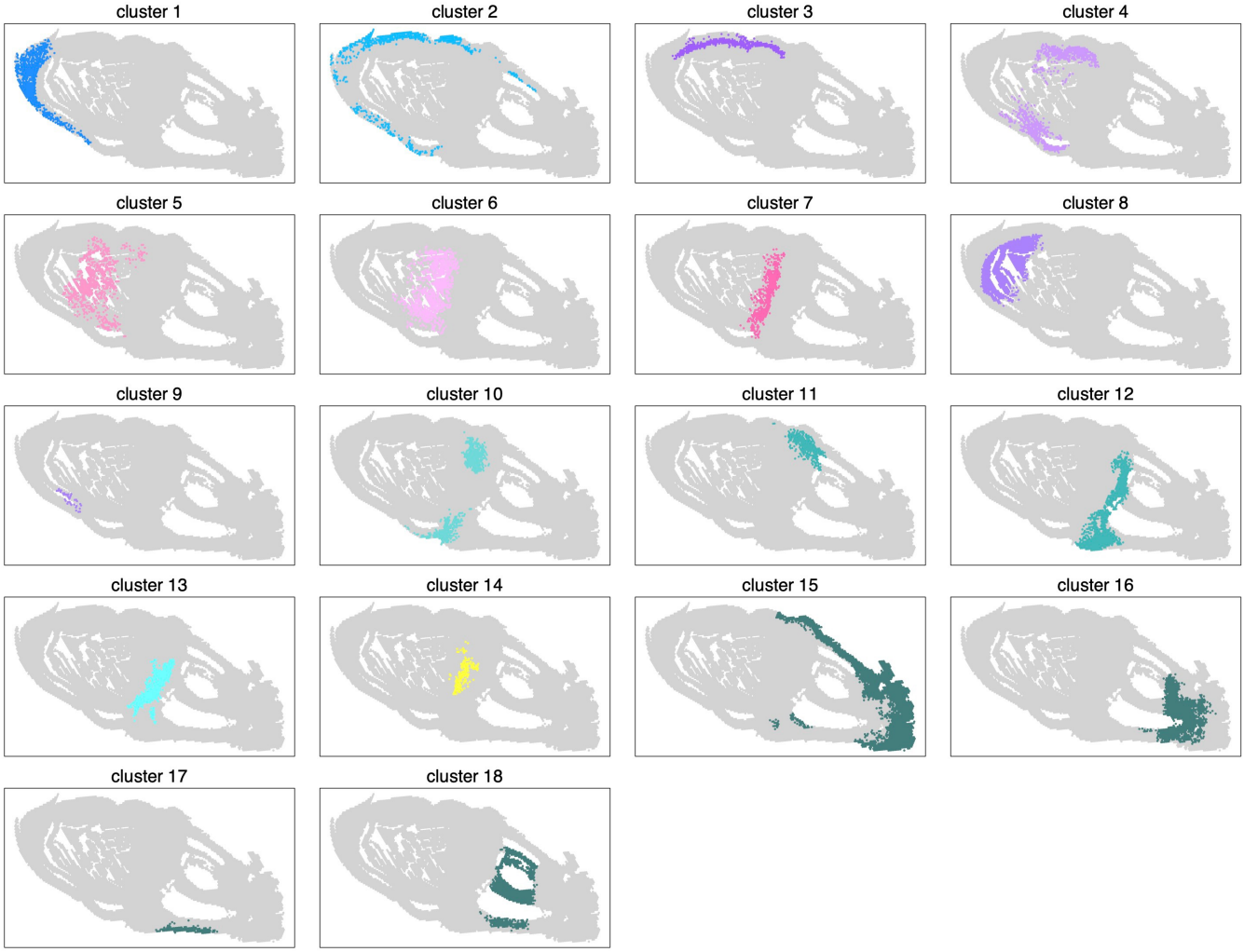


Fig. S3 Illustration of recognized cell types on the spatial transcriptome datasets in stage S1 and stage S2 water lily. Each group is colored according to those in Fig. 1A.



Fig. S4 Illustration of recognized cell types on the spatial transcriptome dataset in stage S3 water lily. Each group is colored according to those in Fig. 1A.


cluster 37
cluster 38
cluster 39
cluster 40
cluster 41
cluster 42
cluster 43
cluster 44
cluster 45
cluster 46
cluster 47
cluster 48
cluster 49
cluster 50
cluster 51
cluster 52
cluster 53
cluster 54
cluster 55
cluster 56
cluster 57
cluster 58
cluster 59
cluster 60
cluster 61
cluster 62
cluster 63
cluster 64
cluster 65
cluster 66
cluster 67
cluster 68
cluster 69
cluster 70

Continued

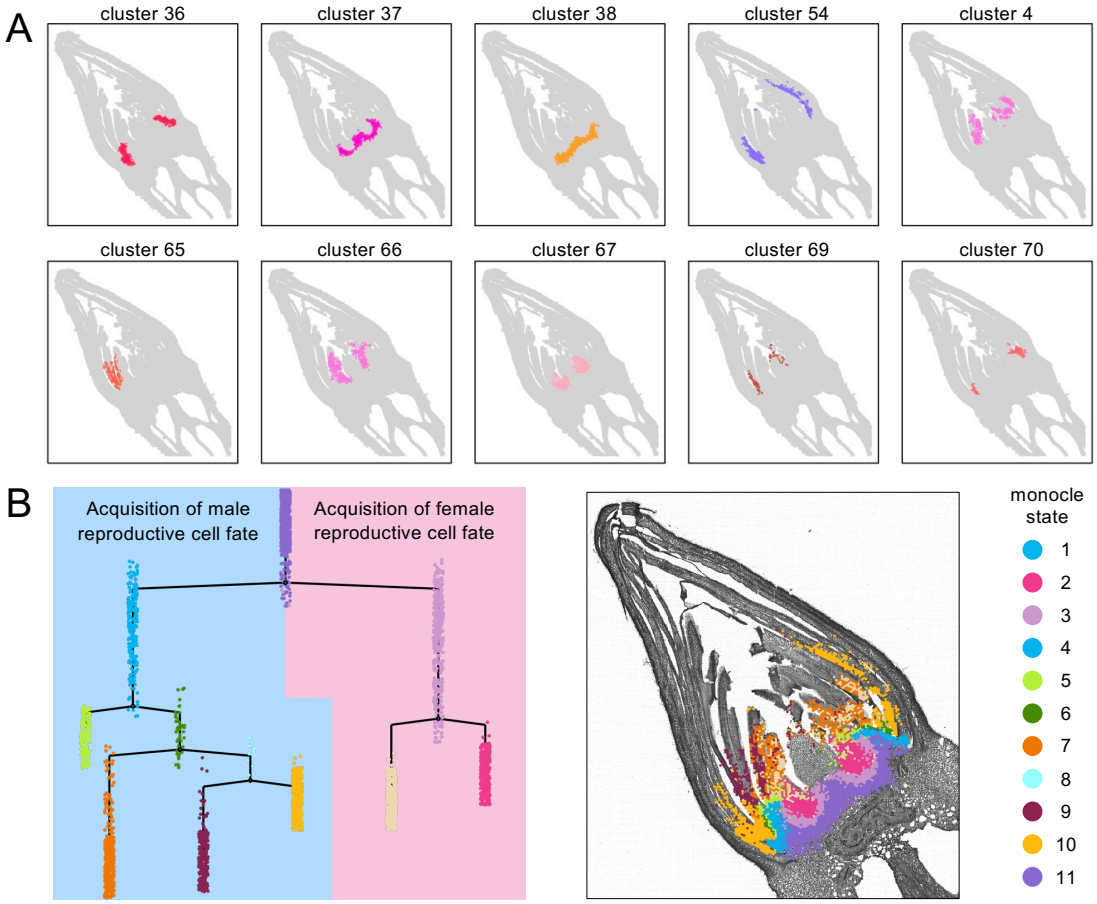


Fig. S5 Reconstructing of differentiation path in floral basal meristem, stamen primordia, carpal primordia and basal tissue of developing stamens and inner tepals. (A) Cell clusters chosen for trajectory analysis. (B) Distribution of cell types across monocle-inferred trajectory and tissue section.

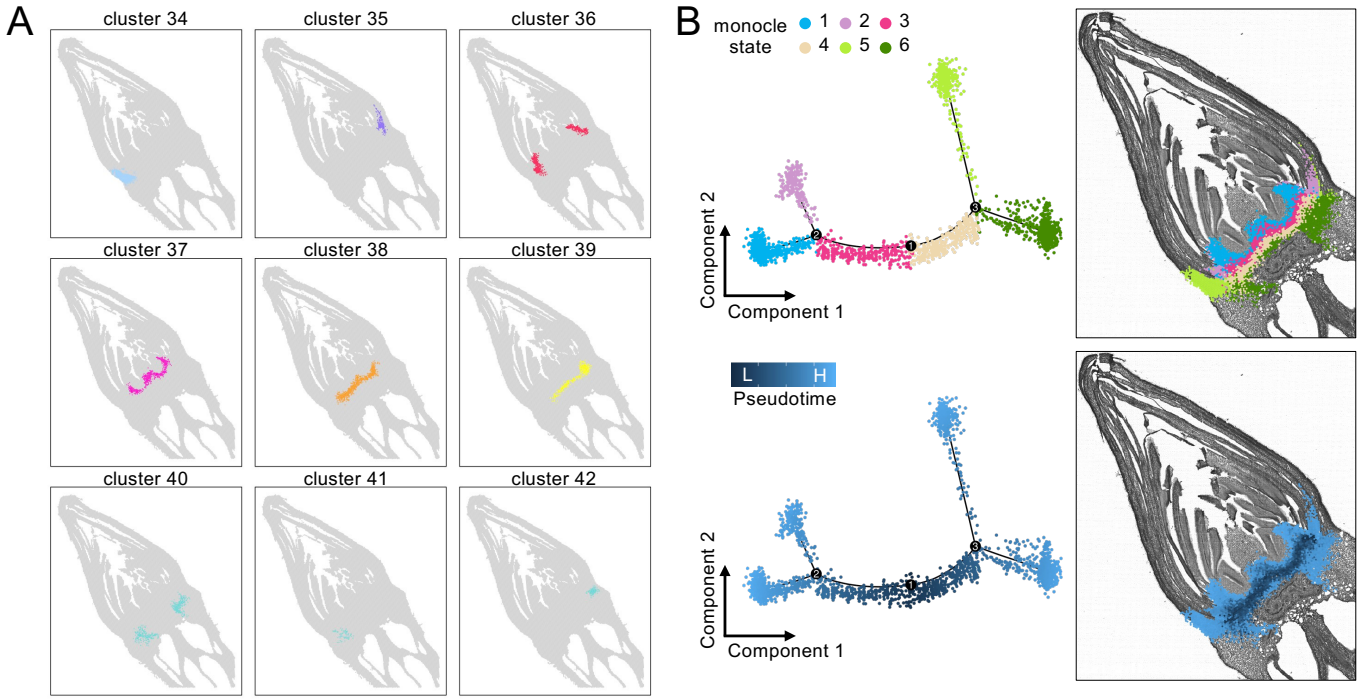


Fig. S6 A framework for reconstructing differentiation in floral and vegetative meristem. (A) Cell clusters chosen for trajectory analysis. (B) Distribution of cell types and pseudotime across monocle-inferred trajectory and tissue section.

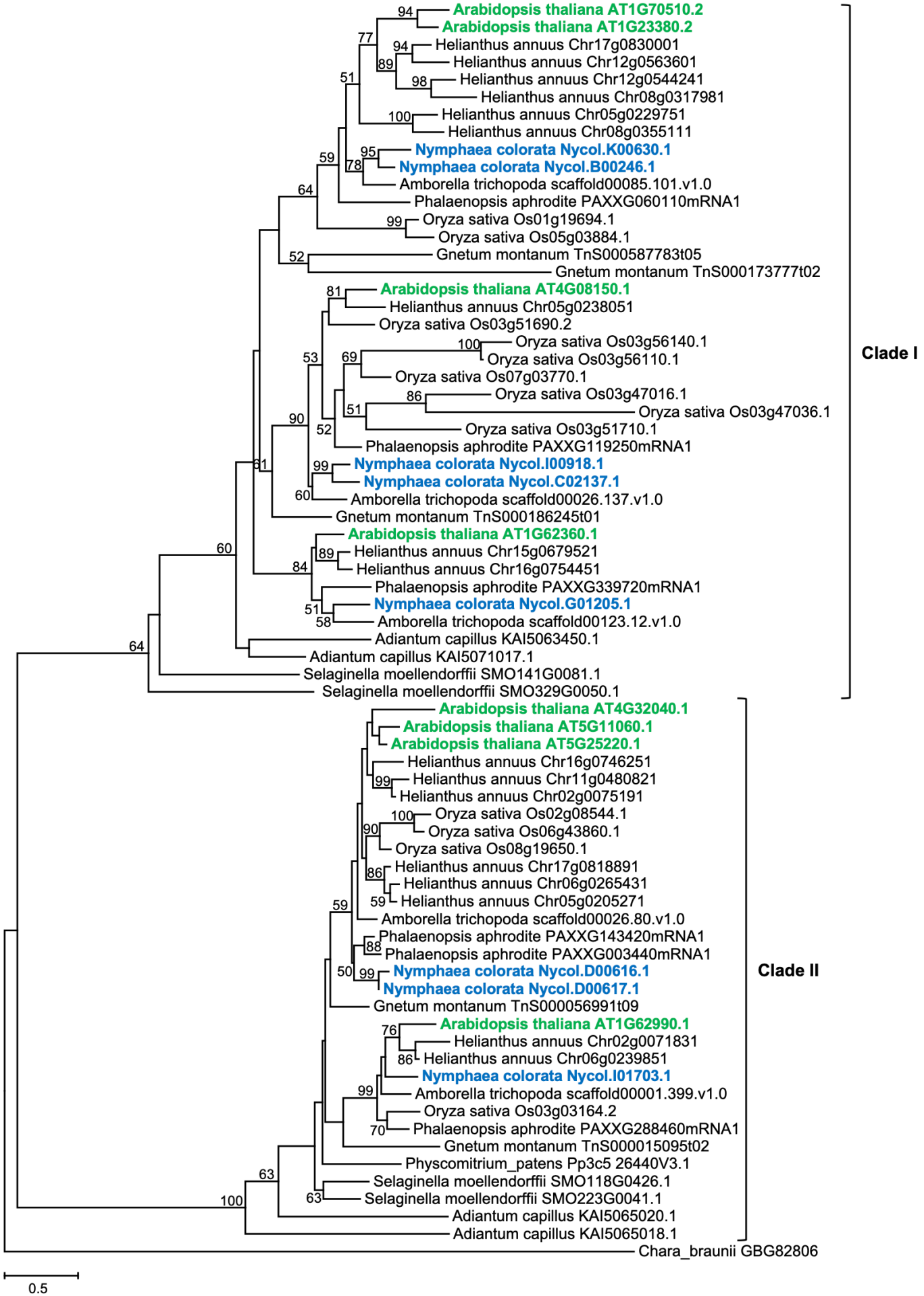


Fig. S7 The maximum likelihood tree of *KNOX* homologs of 12 seed plants. Bootstrap values are shown for each node.

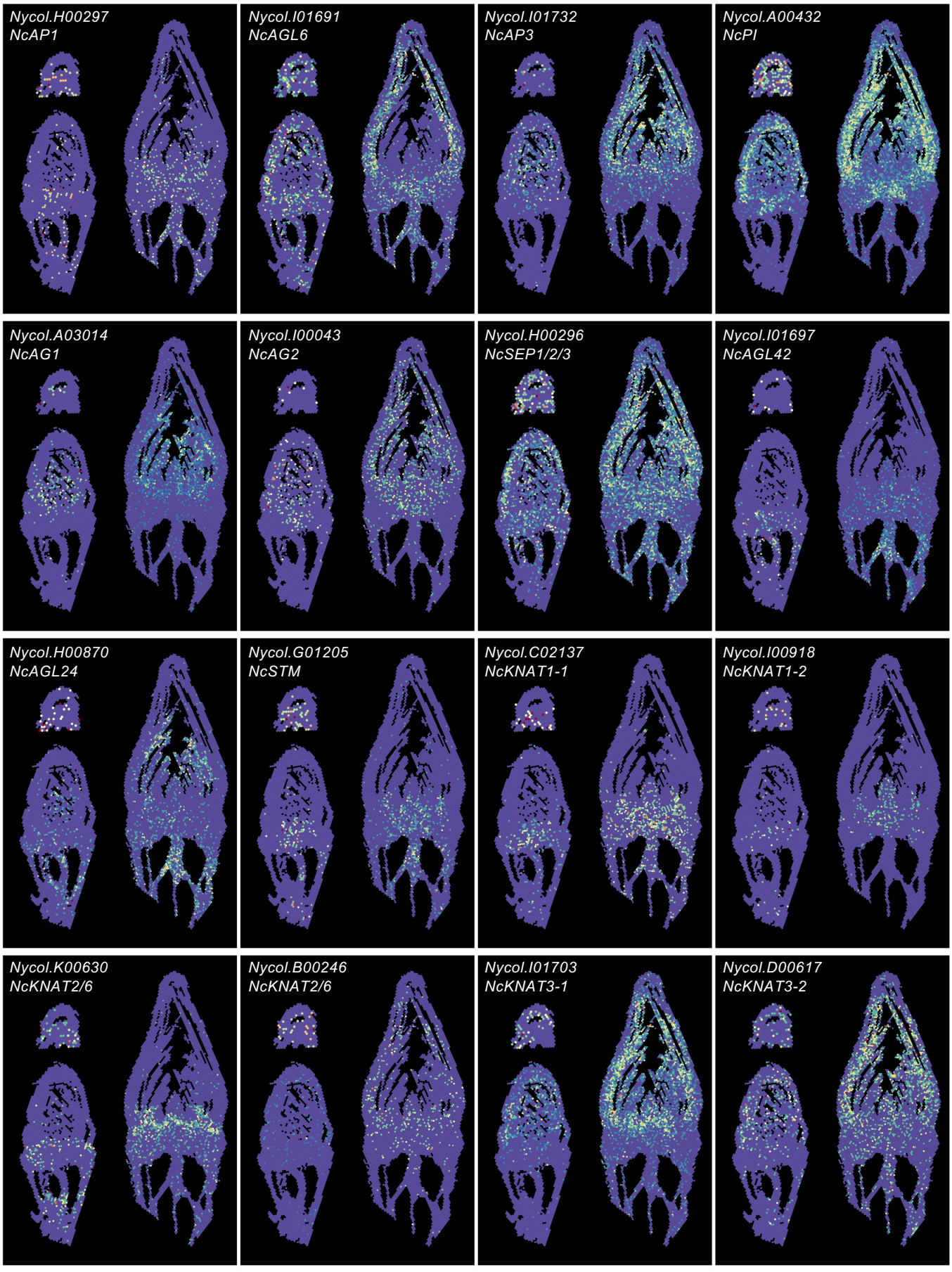
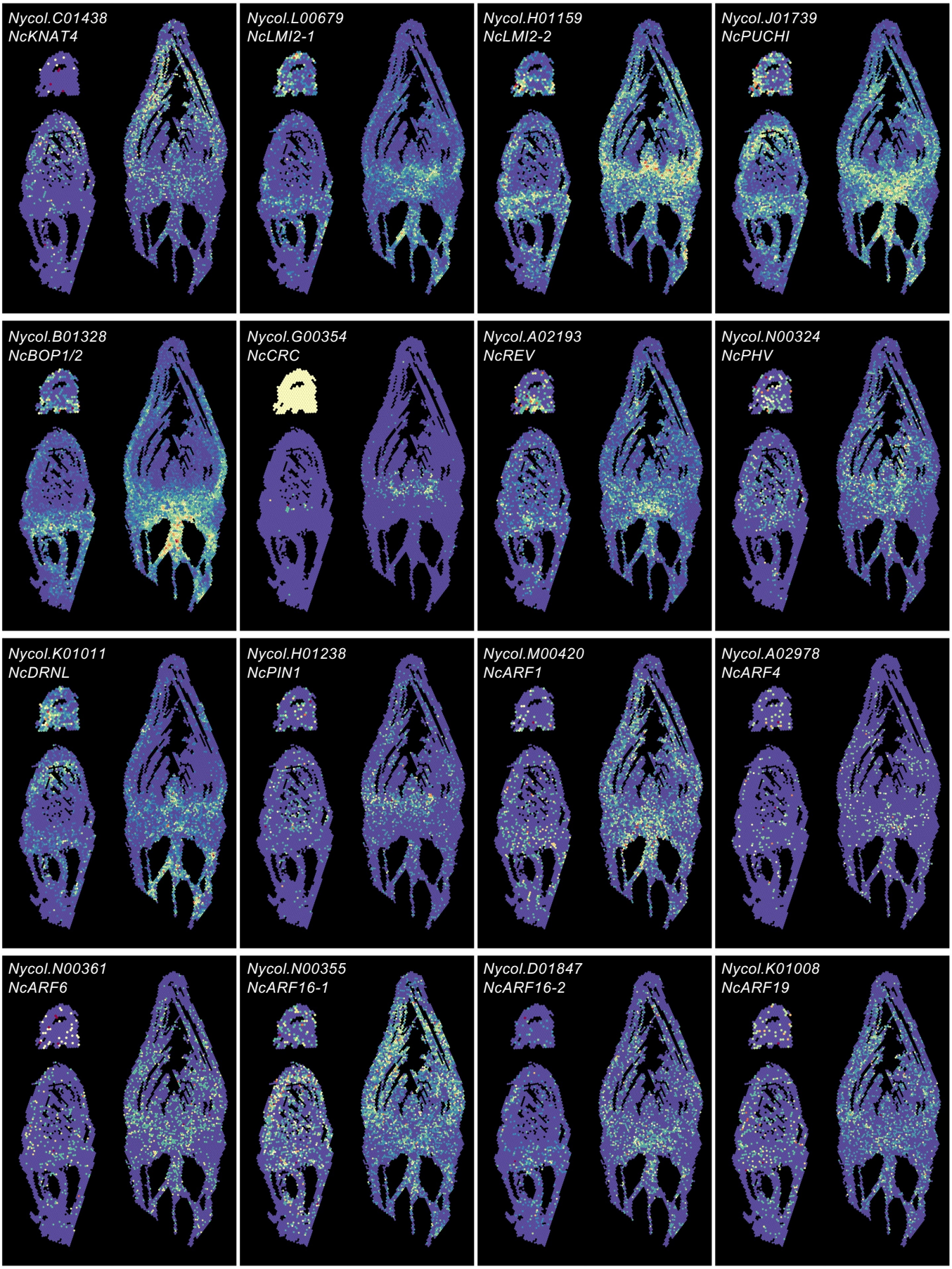
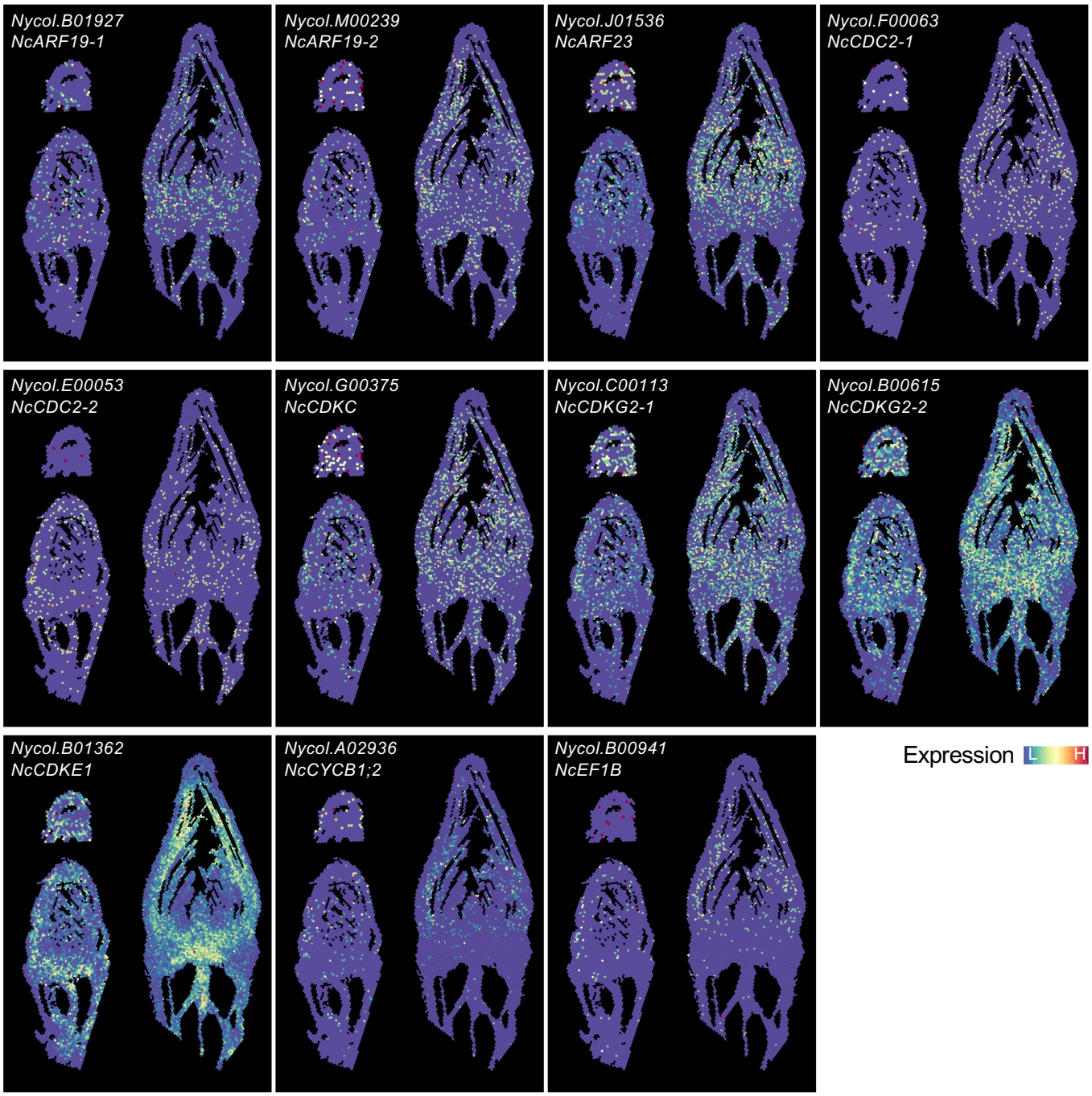


Fig. S8 Spatial expression of genes associated with meristem maintenance and differentiation, as presented in Fig. 2D.


Nycol.C01438
NcKNAT4
Nycol.L00679
NcLMI2-1
Nycol.H01159
NcLMI2-2
Nycol.J01739
NcPUCHI
Nycol.B01328
NcBOP1/2
Nycol.G00354
NcCRC
Nycol.A02193
NcREV
Nycol.N00324
NcPHV
Nycol.K01011
NcDRNL
Nycol.H01238
NcPIN1
Nycol.M00420
NcARF1
Nycol.A02978
NcARF4
Nycol.N00361
NcARF6
Nycol.N00355
NcARF16-1
Nycol.D01847
NcARF16-2
Nycol.K01008
NcARF19
Expression
L
H

Continued


Nycol.B01927
NcARF19-1
Nycol.M00239
NcARF19-2
Nycol.J01536
NcARF23
Nycol.F00063
NcCDC2-1
Nycol.E00053
NcCDC2-2
Nycol.G00375
NcCDKC
Nycol.C00113
NcCDKG2-1
Nycol.B00615
NcCDKG2-2
Nycol.B01362
NcCDKE1
Nycol.A02936
NcCYCB1;2
Nycol.B00941
NcEF1B
Expression
L
H

Continued

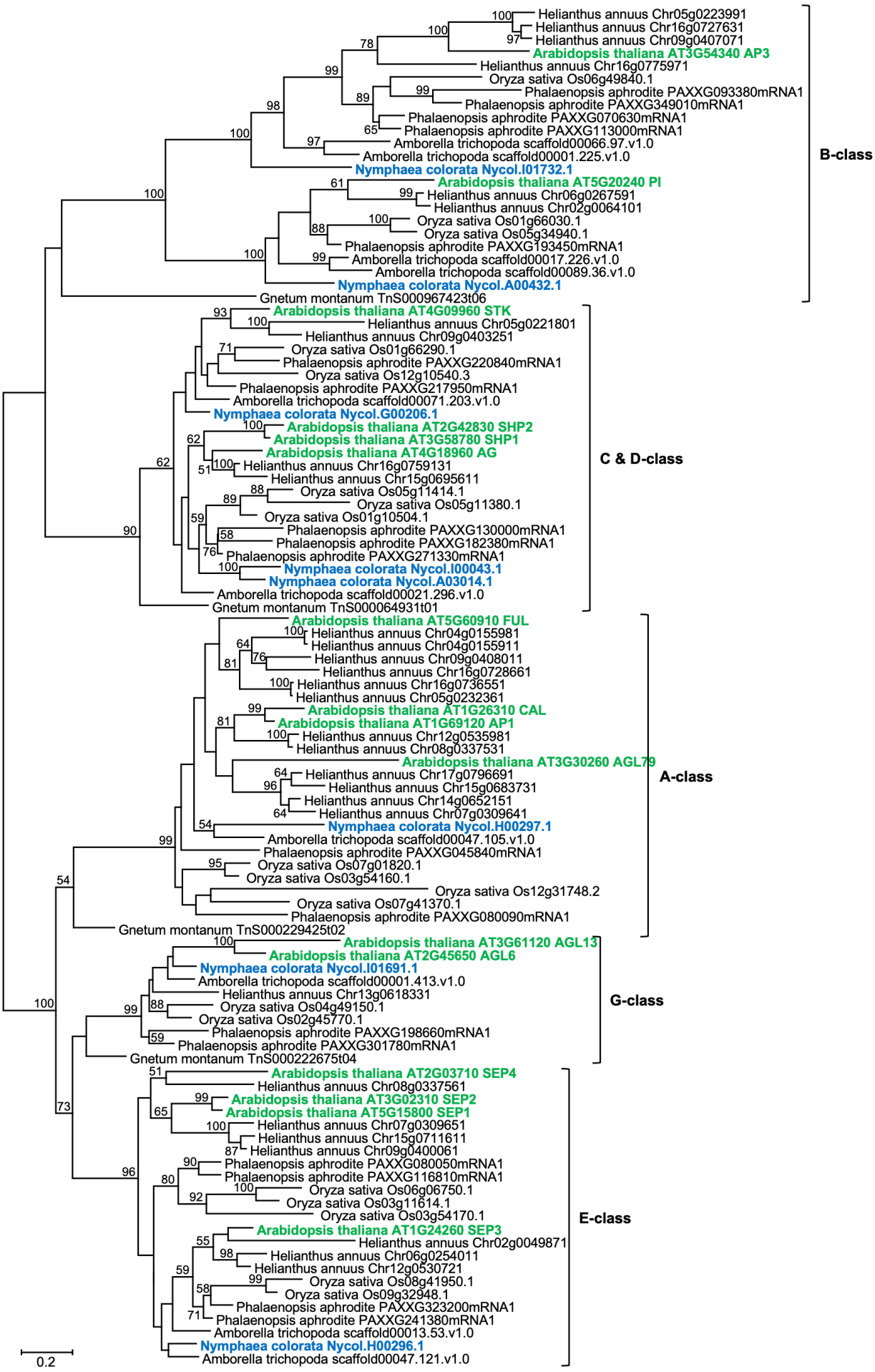


Fig. S9 The maximum likelihood tree of MADS-box homologs of 12 seed plants. Bootstrap values are shown for each node.

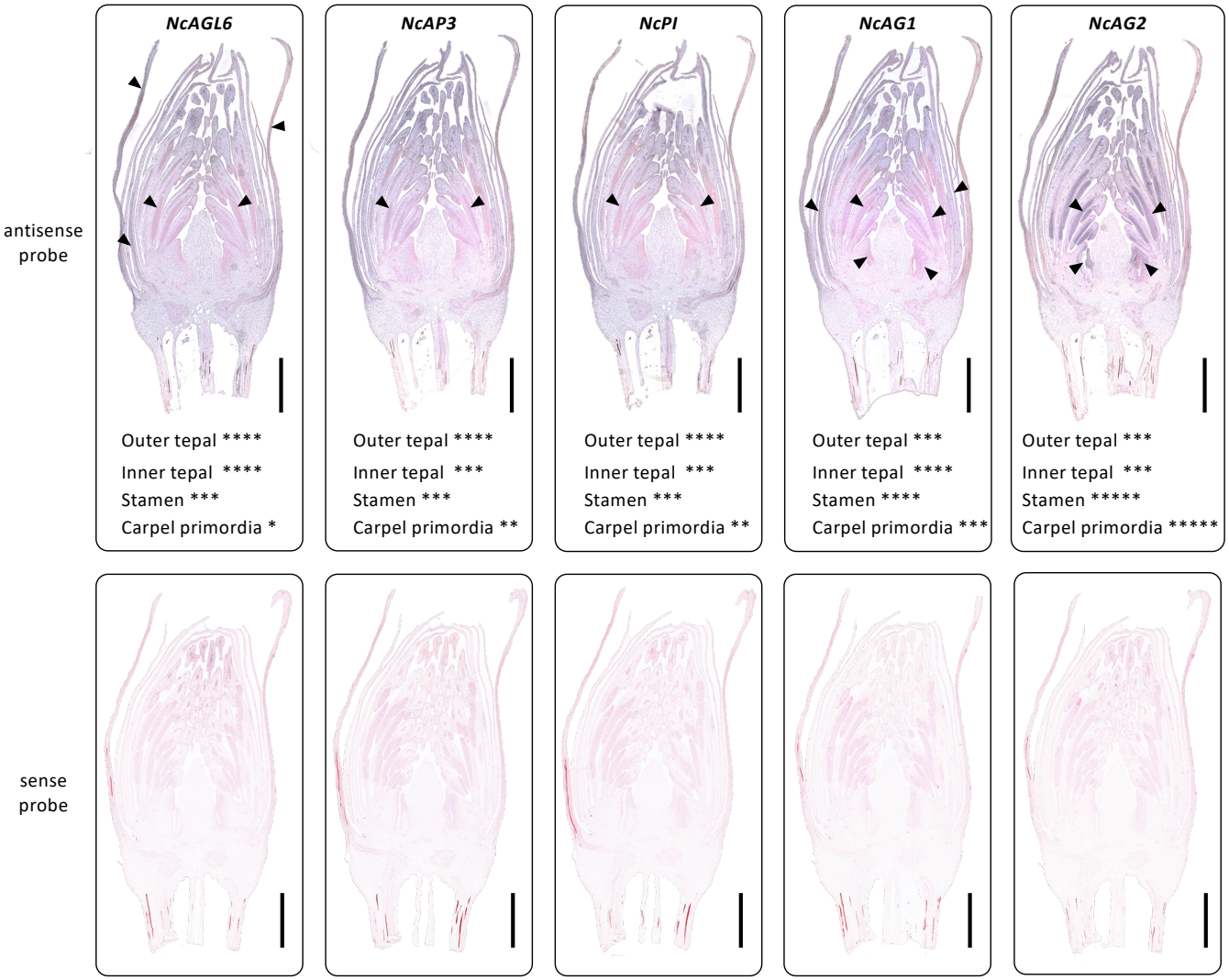


Fig. S10 In situ hybridization of *NcAGL6*, *NcPI*, *NcAP3*, *NcAG1*, *NcAG2*. Signals appeared as blue to purple with NBT/BCIP substrates. The expression intensity of genes in flower organs is marked with asterisks. Sense probes of the five MADS-box genes were used as negative controls. Scale bars = 1mm.

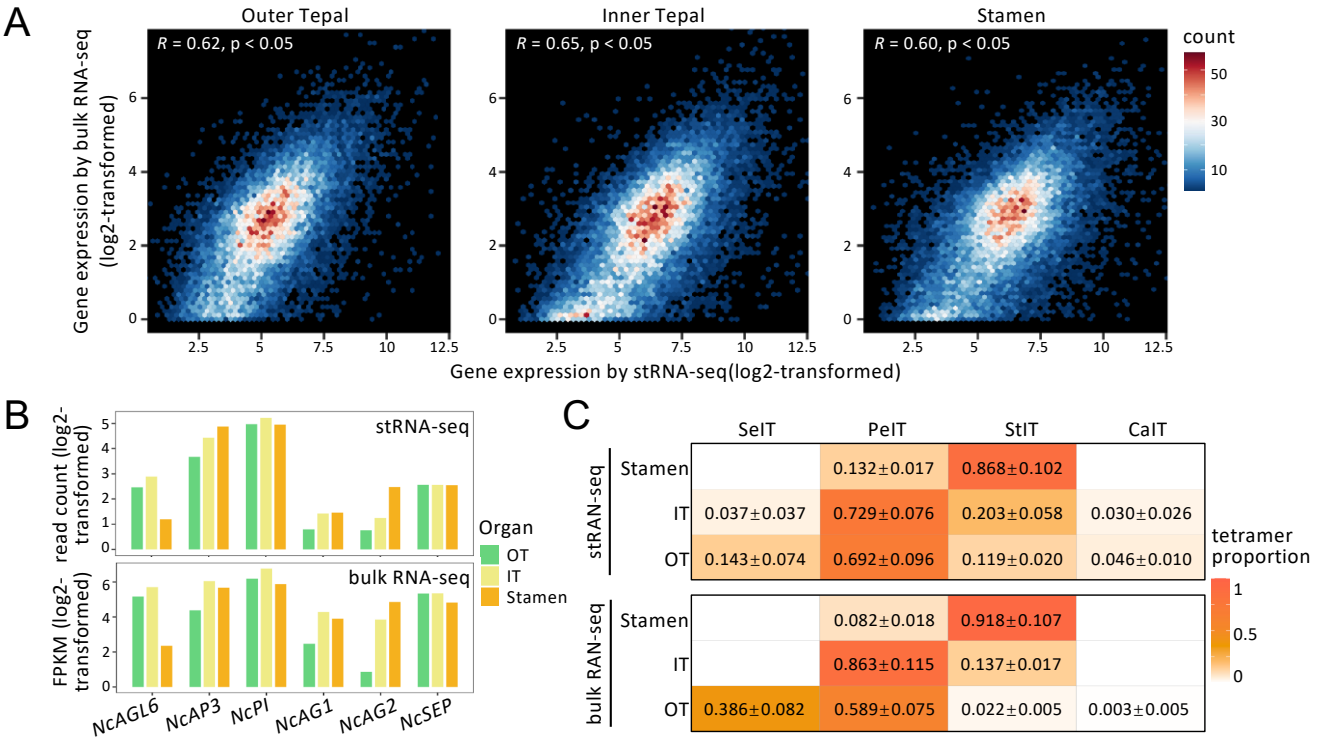


Fig. S11 Comparison of gene expression in outer tepal (OT), inner tepal (IT), and stamen of *N.colorata* detected by bulk RNA sequencing and spatial transcriptome sequencing. (A) Correlation comparison of the different sequencing datasets across the three organs. Among the two datasets, the outer tepal shares 16,493 genes, the inner tepal shares 17,231 genes, and the stamen shares 17,298 genes, with spearman correlation coefficients of 0.62, 0.65, and 0.60, respectively. (B) FPKM values (Log2(FPKM + 1)) from bulk RNA sequencing versus read count (Log2(scale count + 1)) from spatial transcriptomics for six MADS-box genes in three organs. (C) Heatmap showing the normalized relative contribution of MADS-box tetramers among floral organs from the two sequencing datasets.

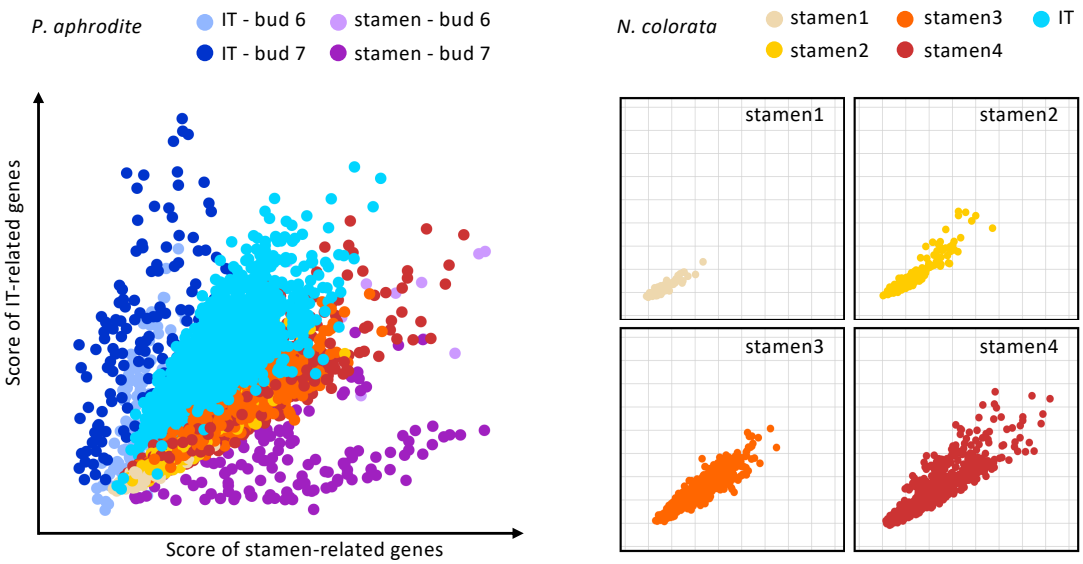


Fig. S12 Cross-species comparison of cell identities between inner tepals and stamens of *N. colorata* and *P. aphrodite*. Scatter plot shows expression scores of specific gene sets for stamen-specific (x-axis) and inner tepal- specific (y-axis) gene sets identified from floral buds of *P. aphrodite* and *N. colorata*.

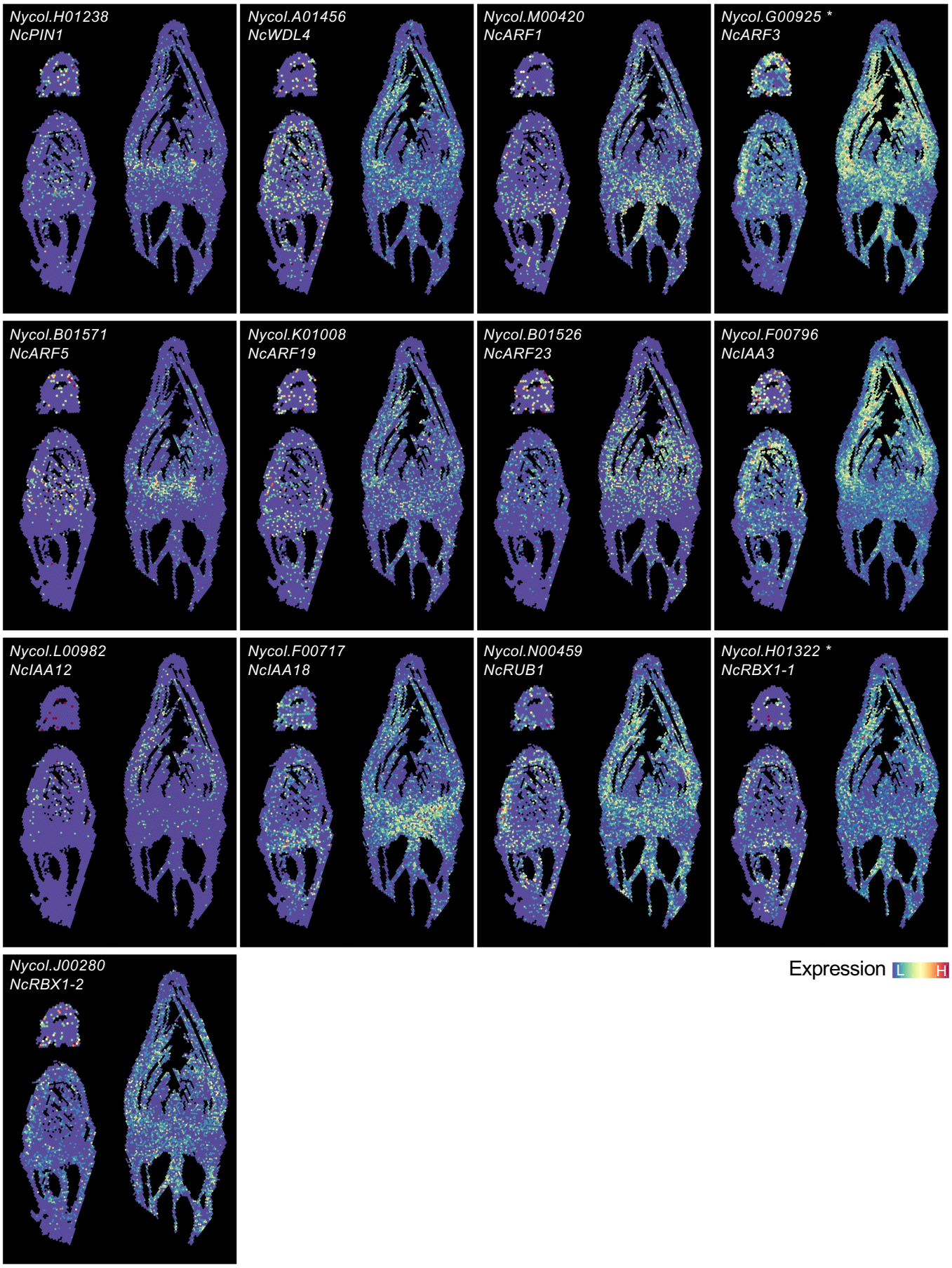


Fig. S13 Spatial expression of genes related to auxin signaling pathway (Fig. 4E) at different developmental stages of water lily floral buds. Genes marked with an asterisk were used to plot Fig. 4F.

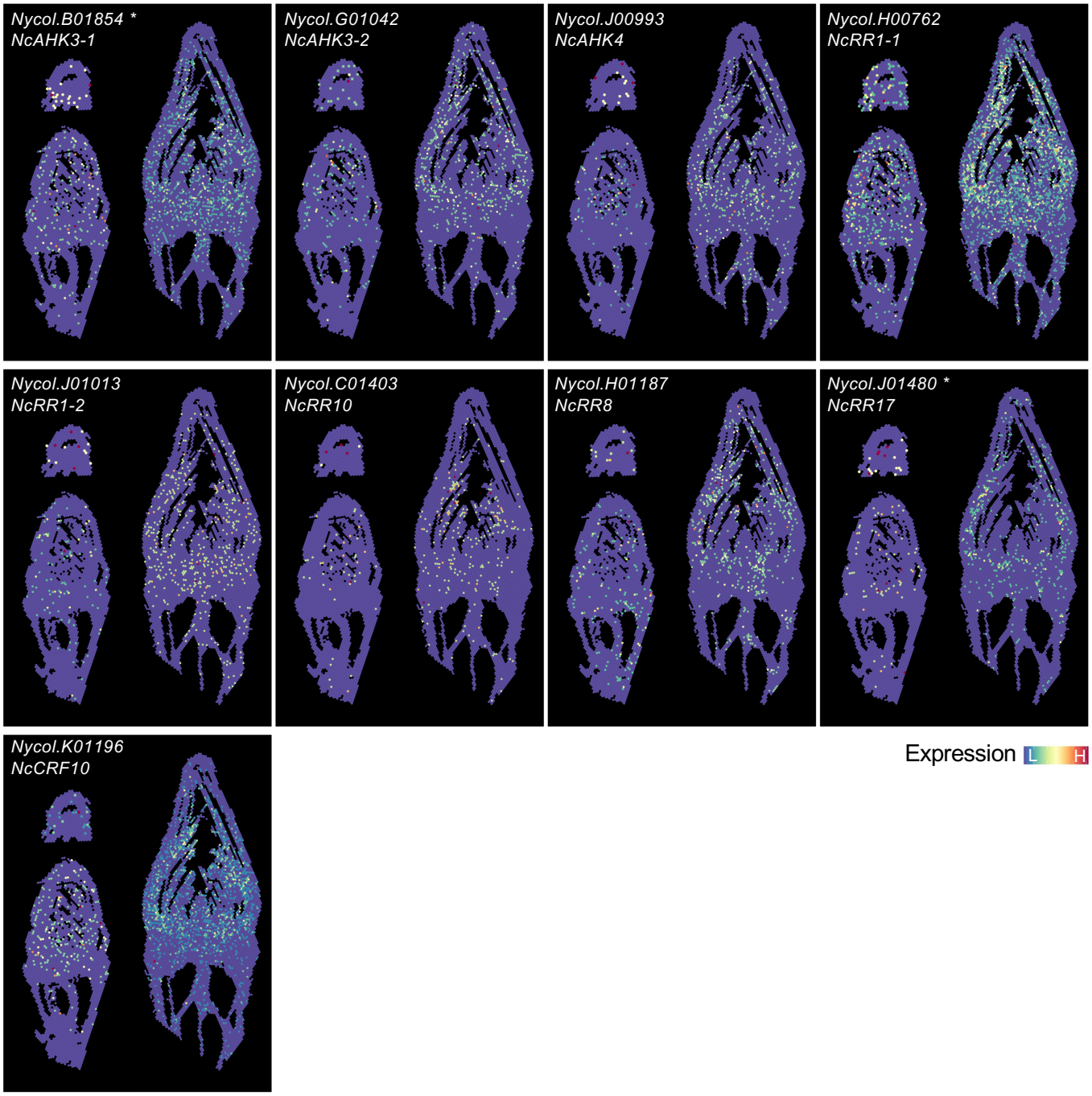


Fig. S14 Spatial expression of genes related to cytokinin signaling pathway (Fig. 4E) at different developmental stages of water lily floral buds. Genes marked with an asterisk were used to plot Fig. 4F.

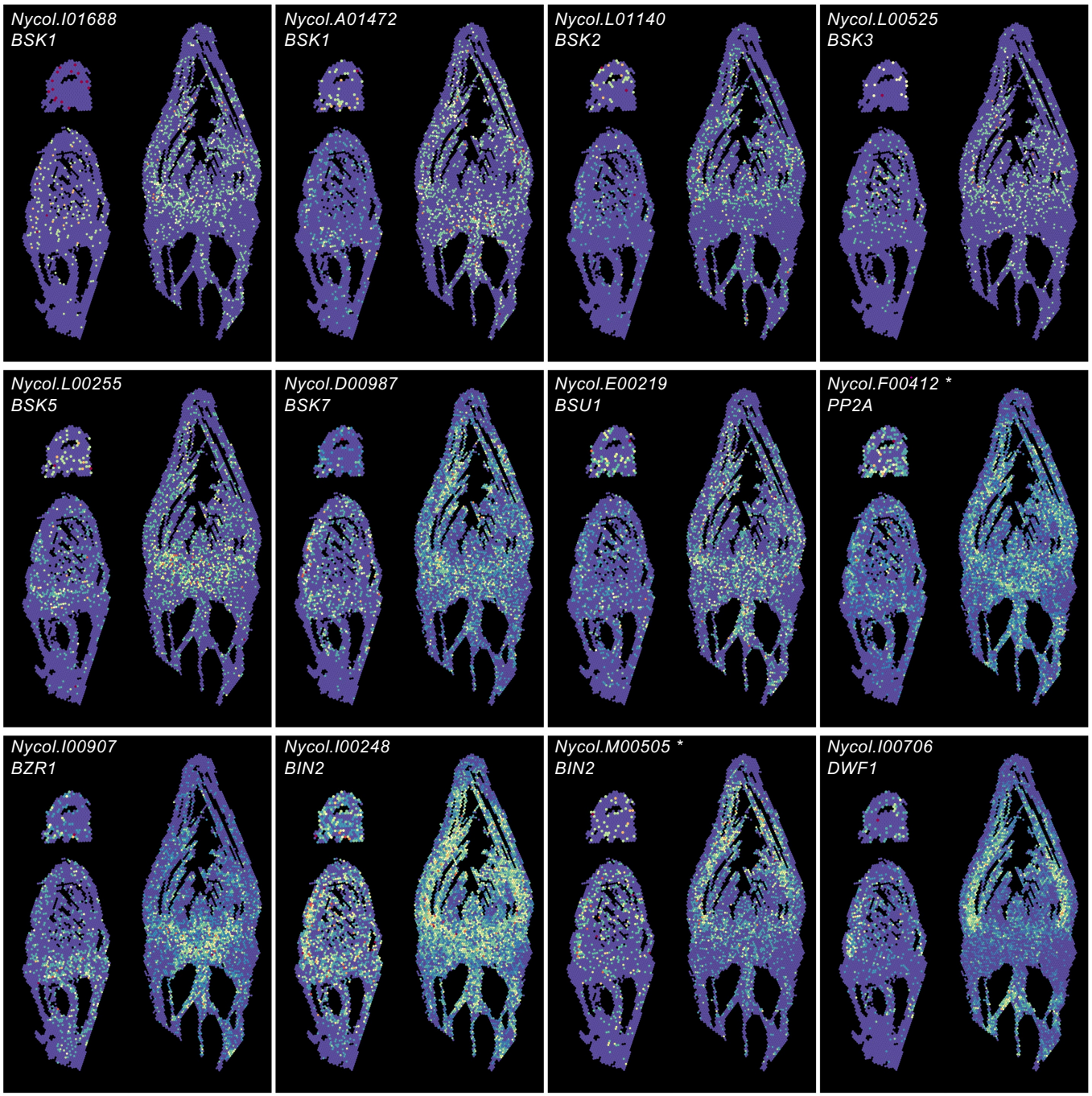


Fig. S15 Spatial expression of genes related to brassinosteroid signaling pathway (Fig. 4E) at different developmental stages of water lily floral buds. Genes marked with an asterisk were used to plot Fig. 4F.

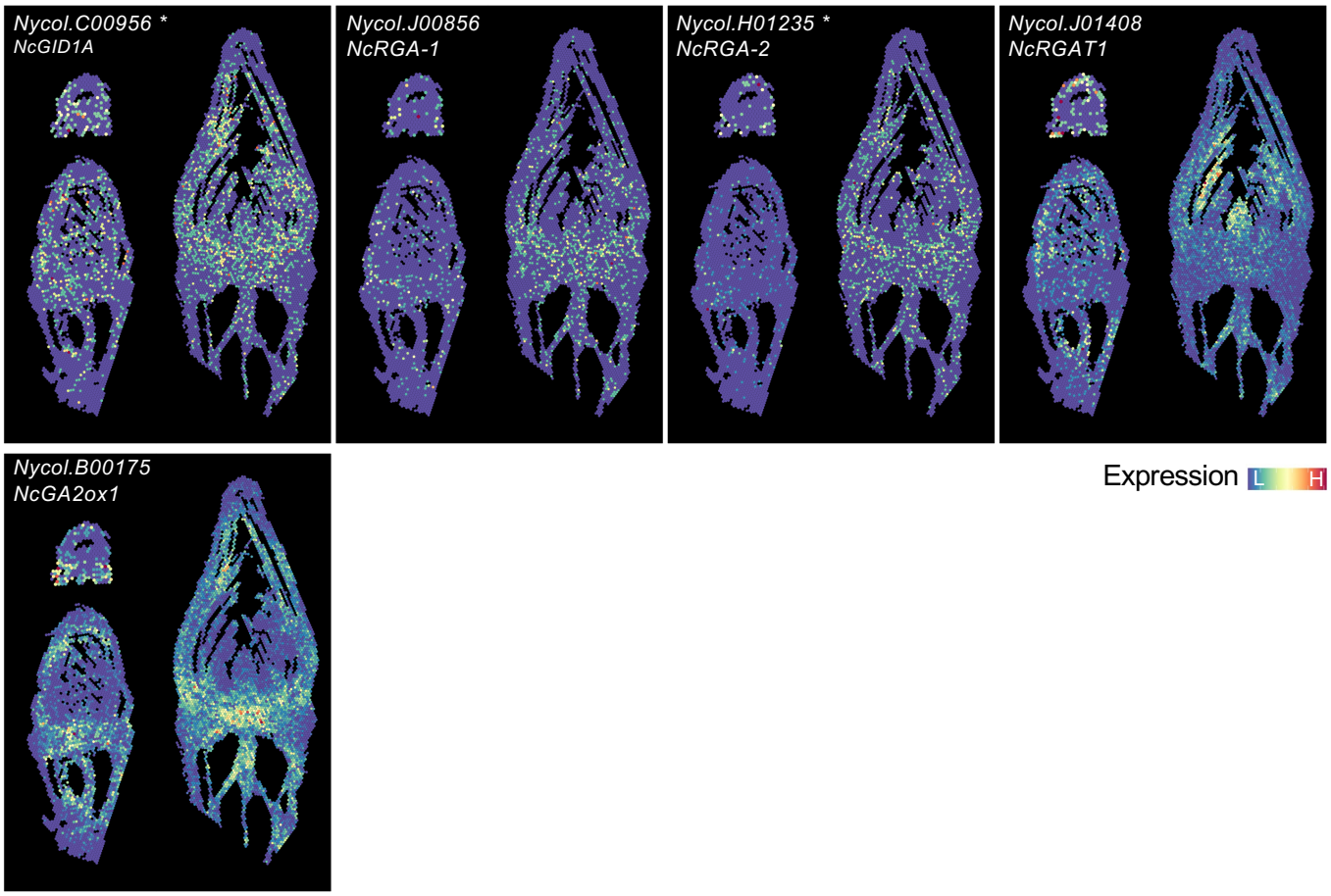


Fig. S16 Spatial expression of genes related to gibberellin signaling pathway (Fig. 4E) at different developmental stages of water lily floral buds. Genes marked with an asterisk were used to plot Fig. 4F.

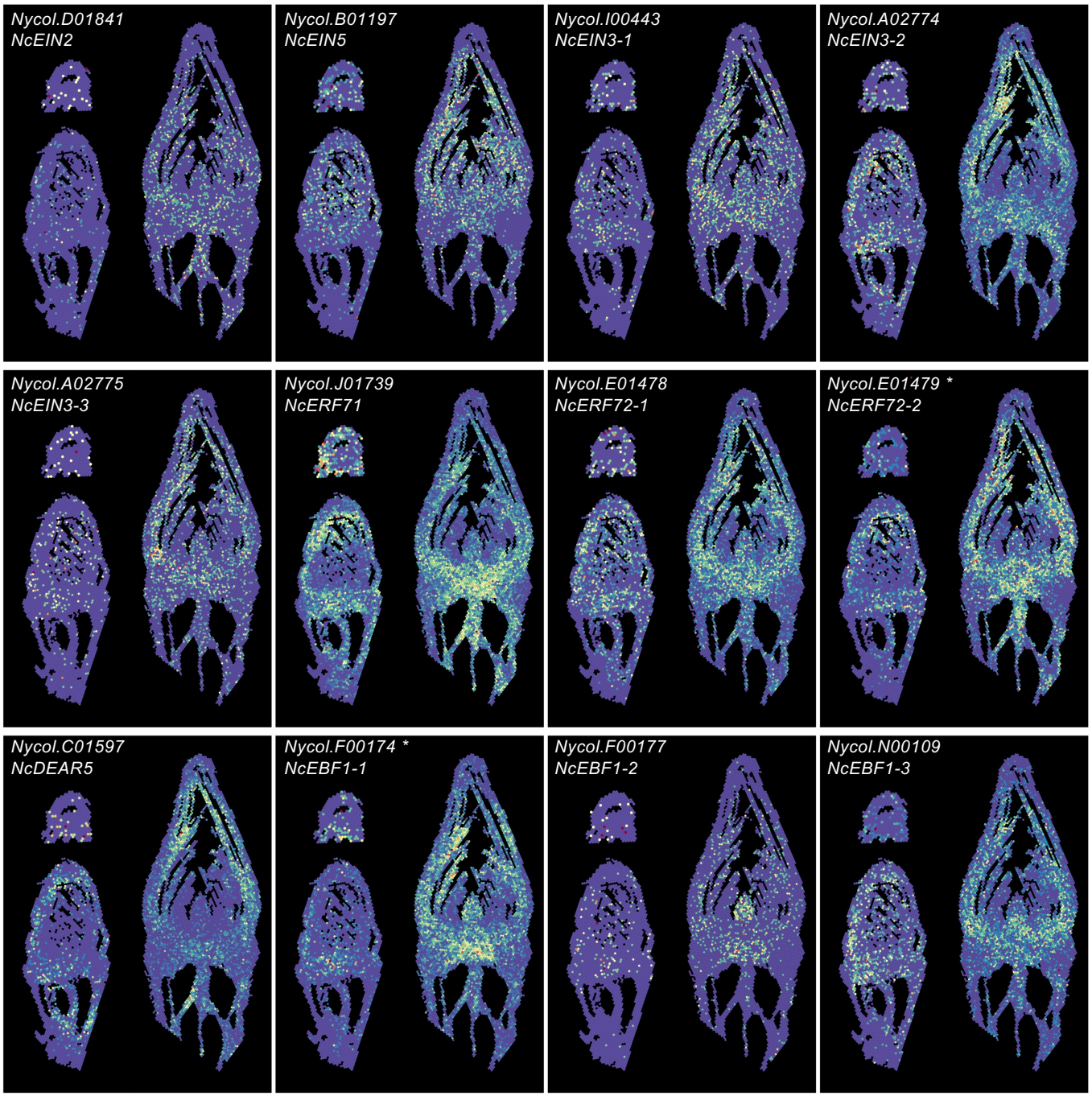


Fig. S17 Spatial expression of genes related to ethylene signaling pathway (Fig. 4E) at different developmental stages of water lily floral buds. Genes marked with an asterisk were used to plot Fig. 4F.

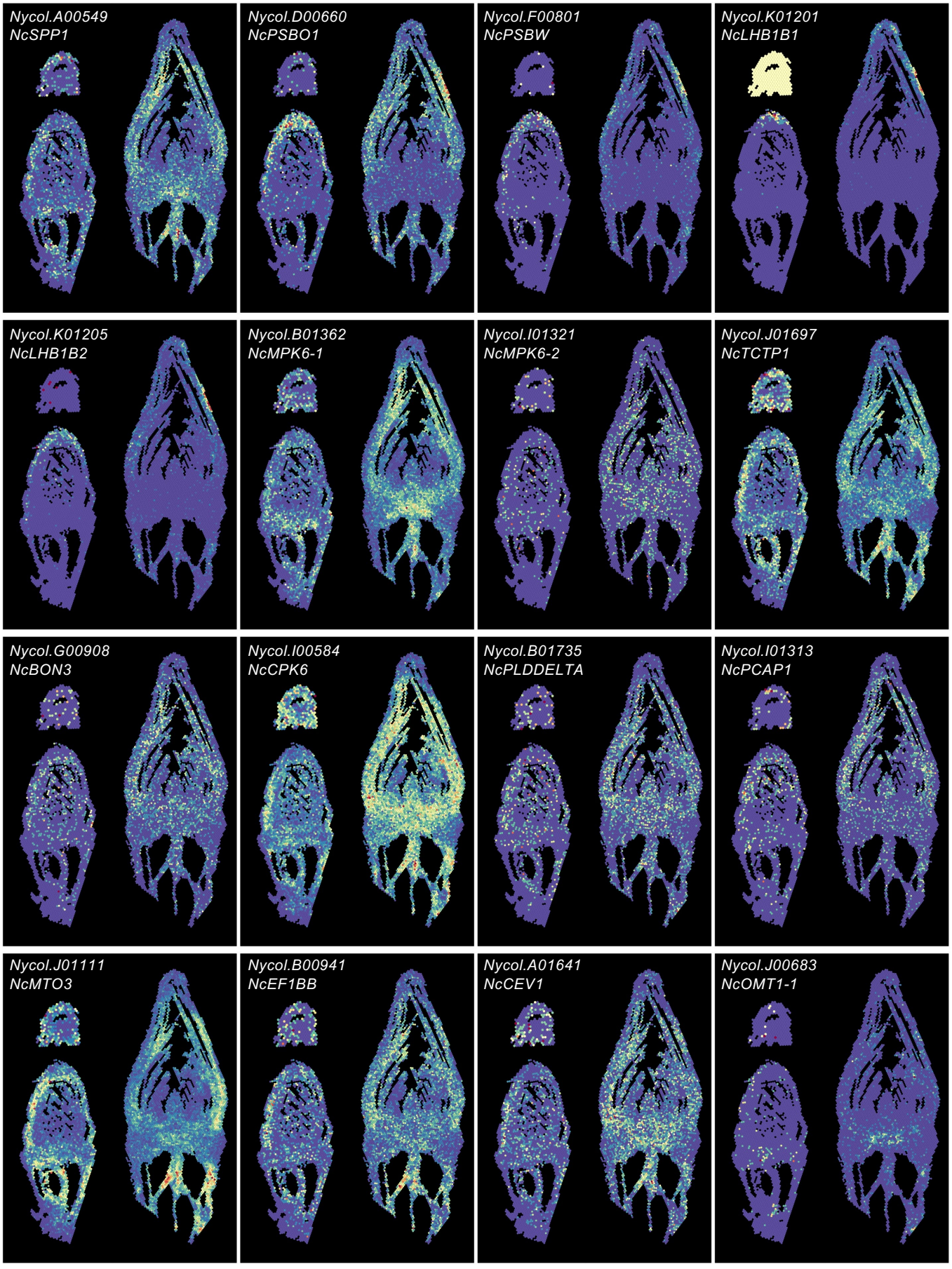
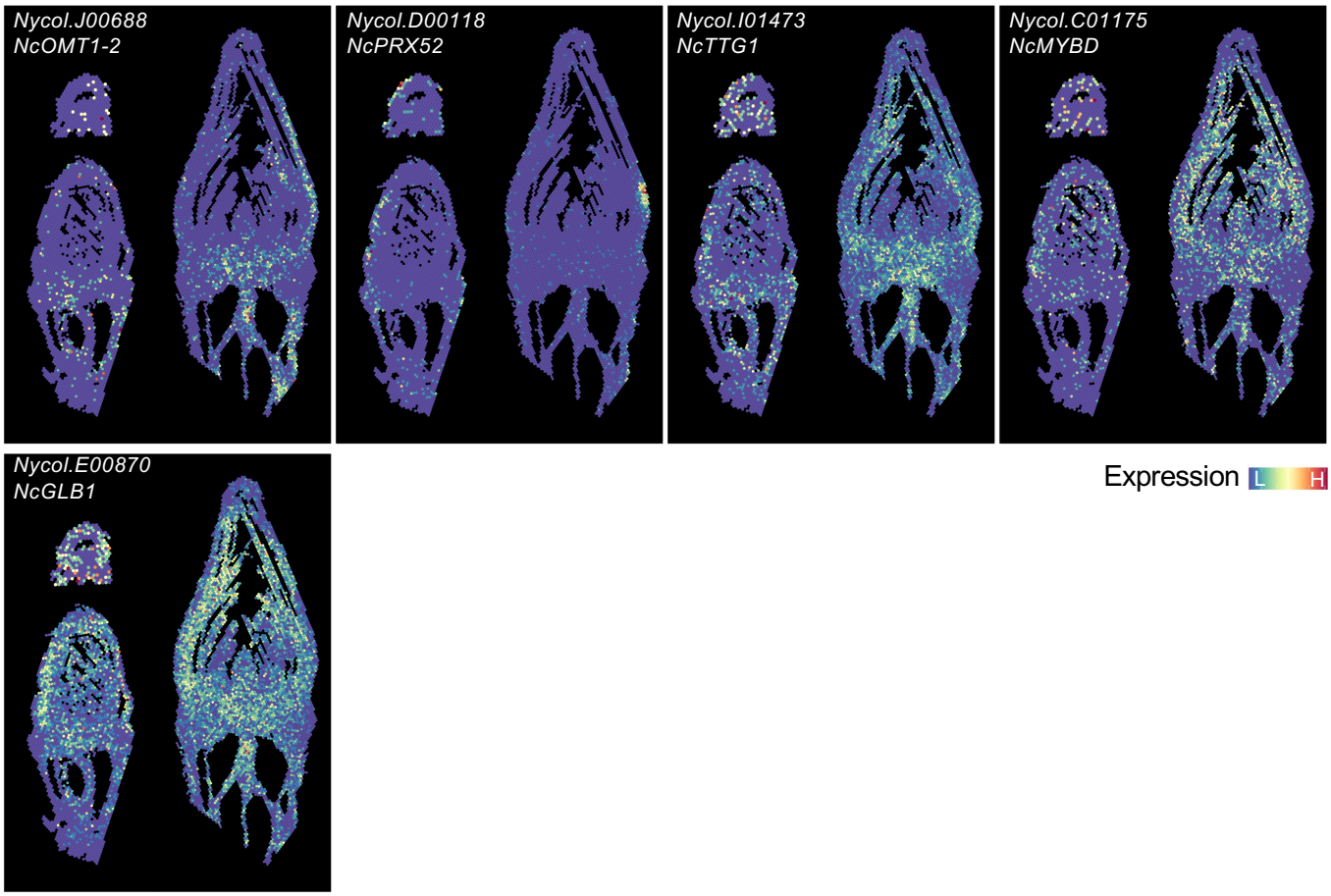


Fig. S18 Spatial expression of genes related to vegetative morphogenesis at different developmental stages of water lily floral buds, as presented in Fig. 5E.


Nycol.J00688
NcOMT1-2
Nycol.D00118
NcPRX52
Nycol.I01473
NcTTG1
Nycol.C01175
NcMYBD
Nycol.E00870
NcGLB1
Expression
L
H

Continued