Differential Expression Analysis for bulk RNA-seq data
Vehicle 100 condition : AD vs CTRL

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Contents

1. Read the count data

In this section, we will read the clean count data from the synaptosomes_bulkRNA folder. We will read the data and merge them into a single table. The final table will be stored in `../dataresults/bulkRNA_counts_clean.csv`.
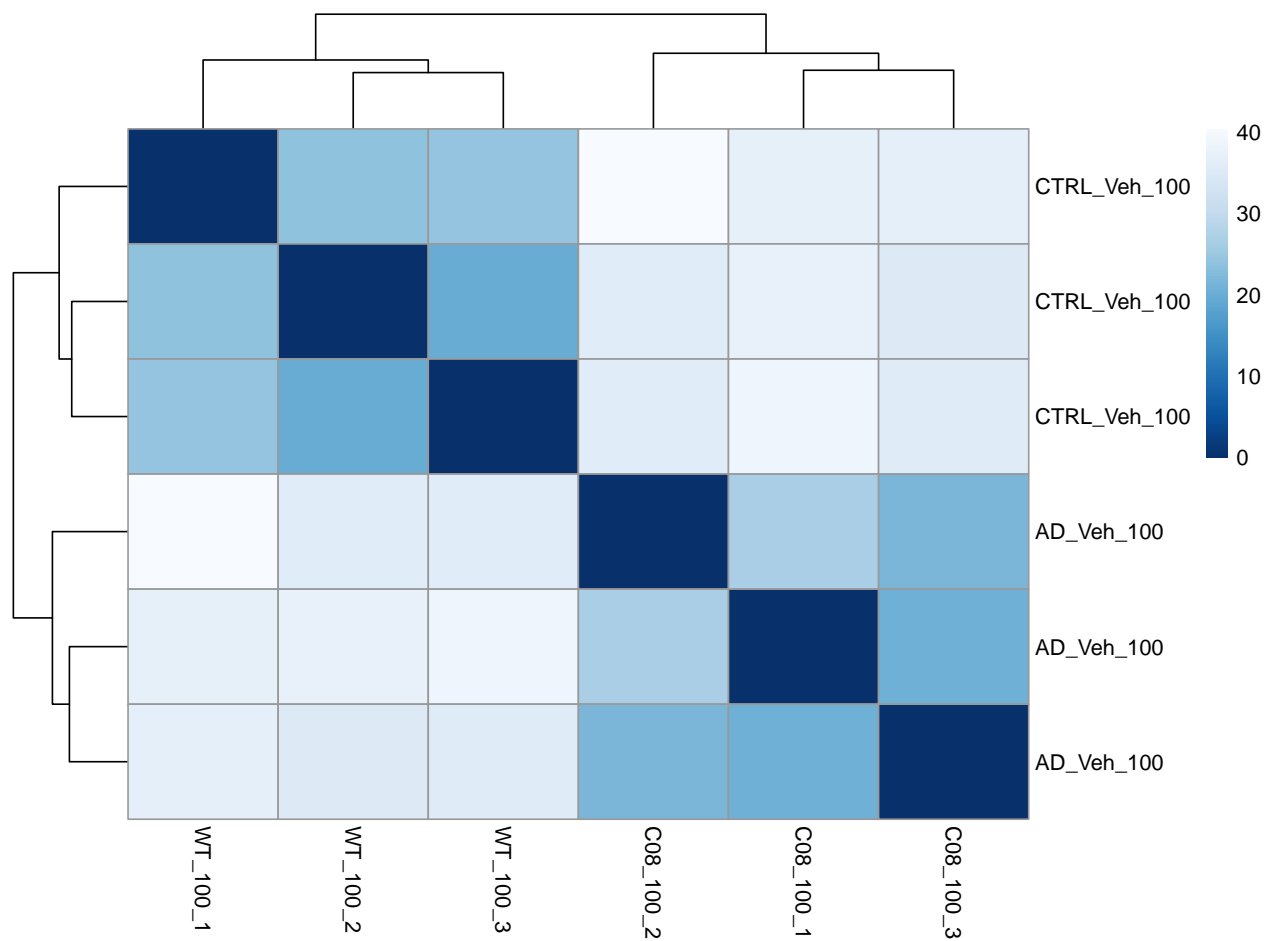
2. Differential expression analysis

In this section, we will perform differential expression analysis using DESeq2. We will compare the 22q vs Control in the vehicle condition. The results will be stored in `results/02-DEG-V_10/DESeq2_results.csv`.

```
## [1] "DEG analysis is done"
```
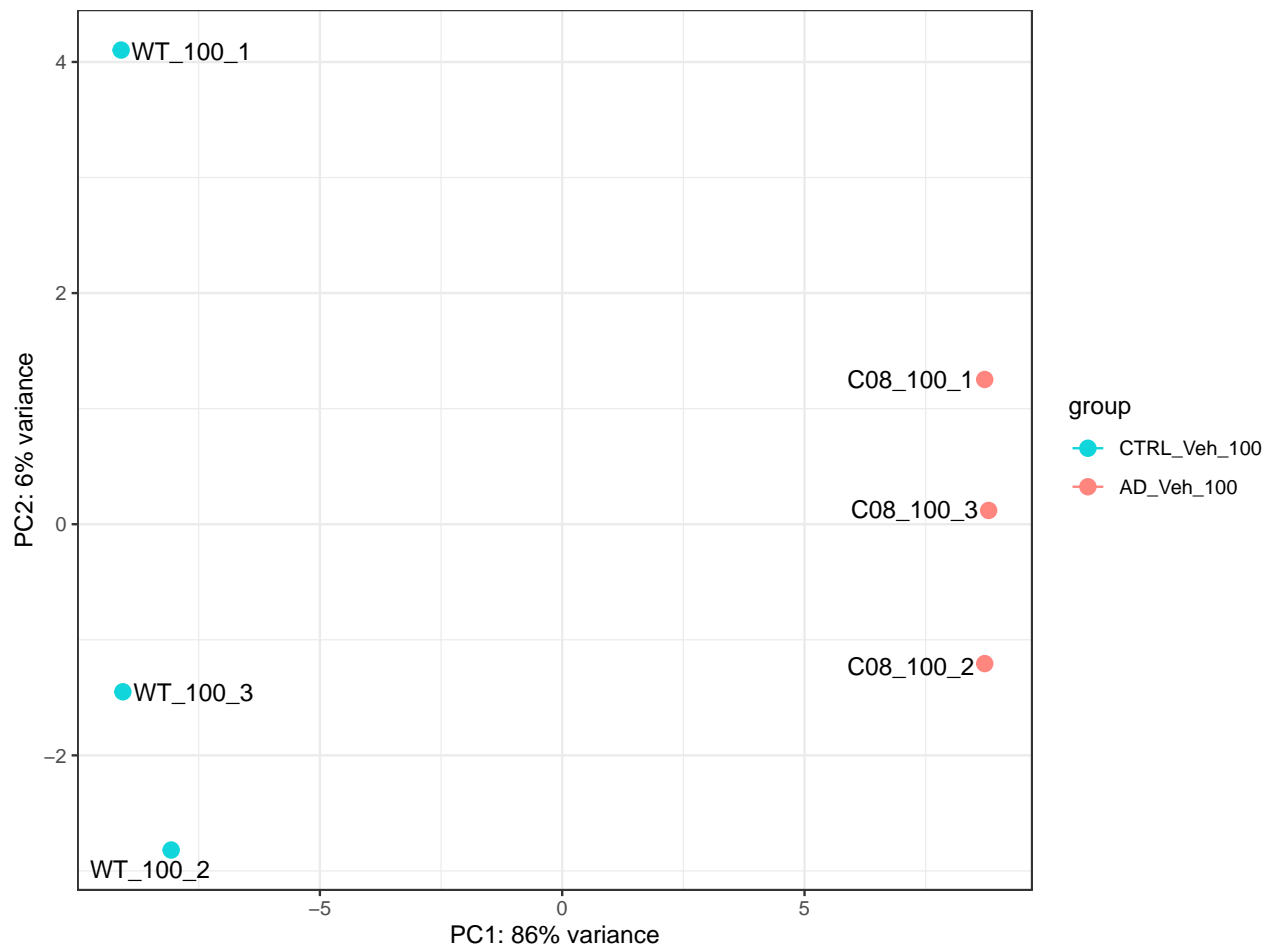
3. Visualization for reuslt

(1) Sample information


```
## [1] "Sample distance heatmap is done"
```

```
## [1] "PCA plot is done"
```

(2) DEG visualization - Volcano plot and Heatmap

```
## [1] "Volcano plot for 02_volcano_plot_log2fc_1"
```
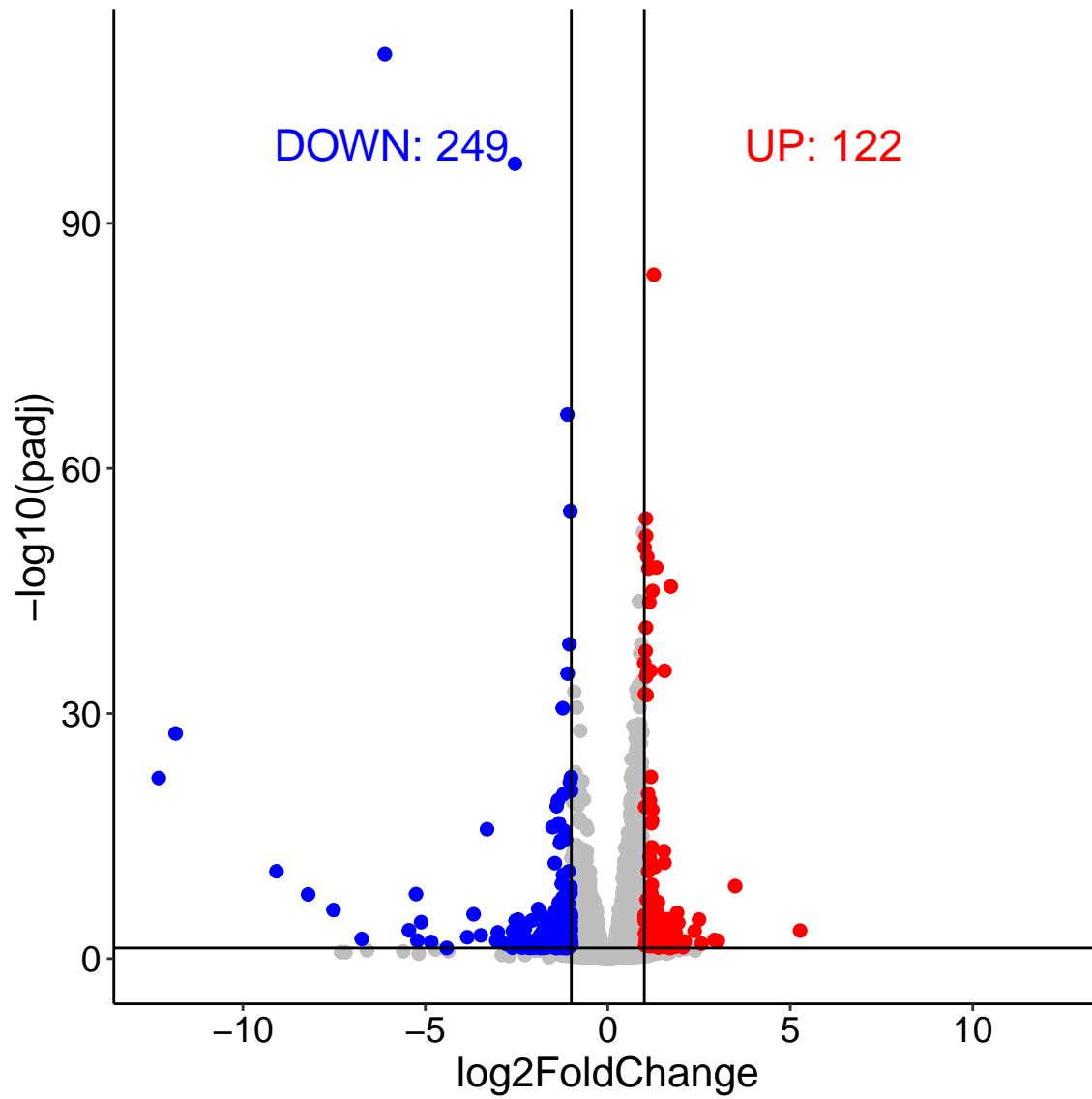


```
## [1] "Volcano plot for 03_volcano_plot_log2fc_1.5"
```
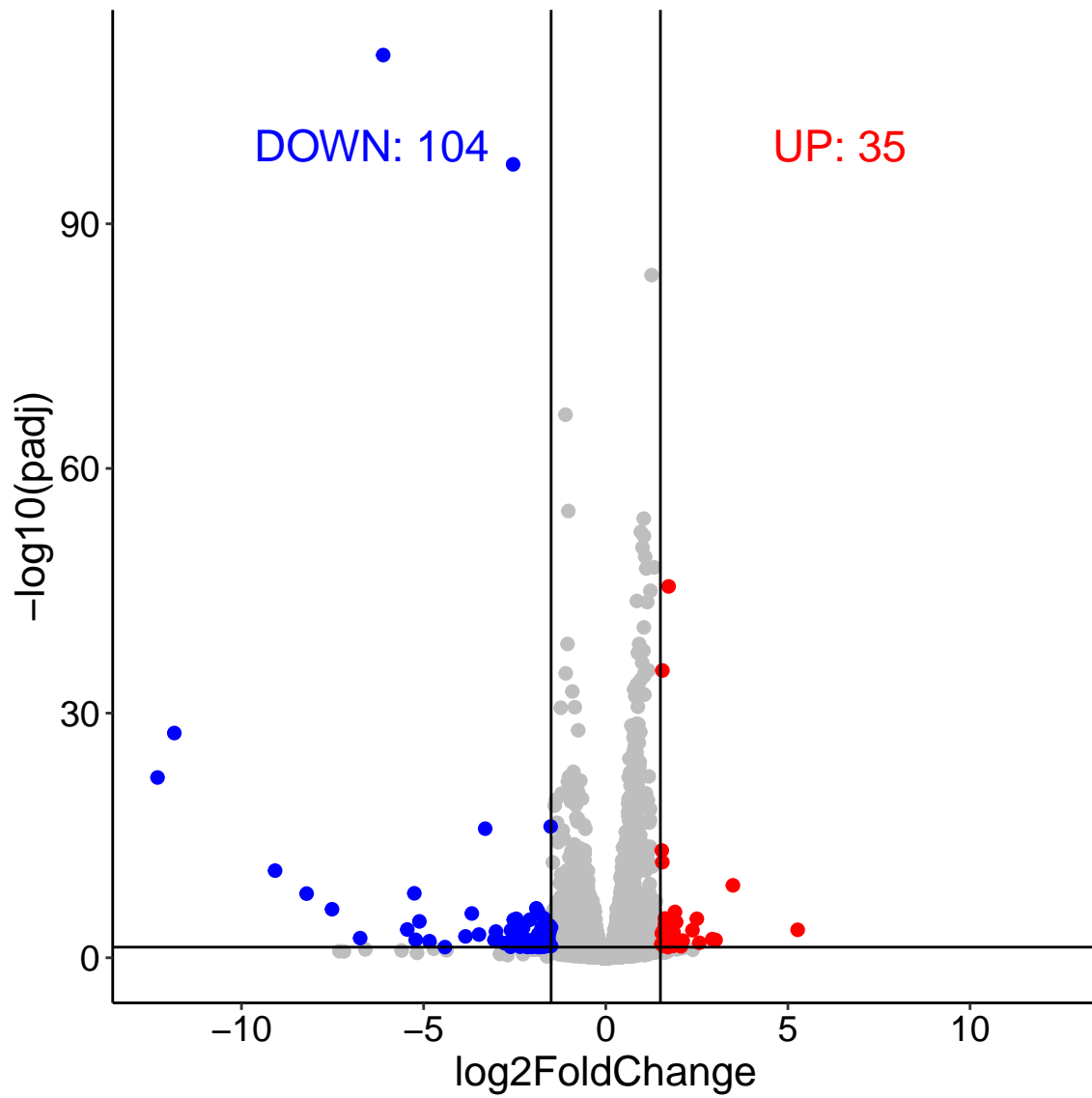
[1] "Volcano plot for 03_volcano_plot_log2fc_2"
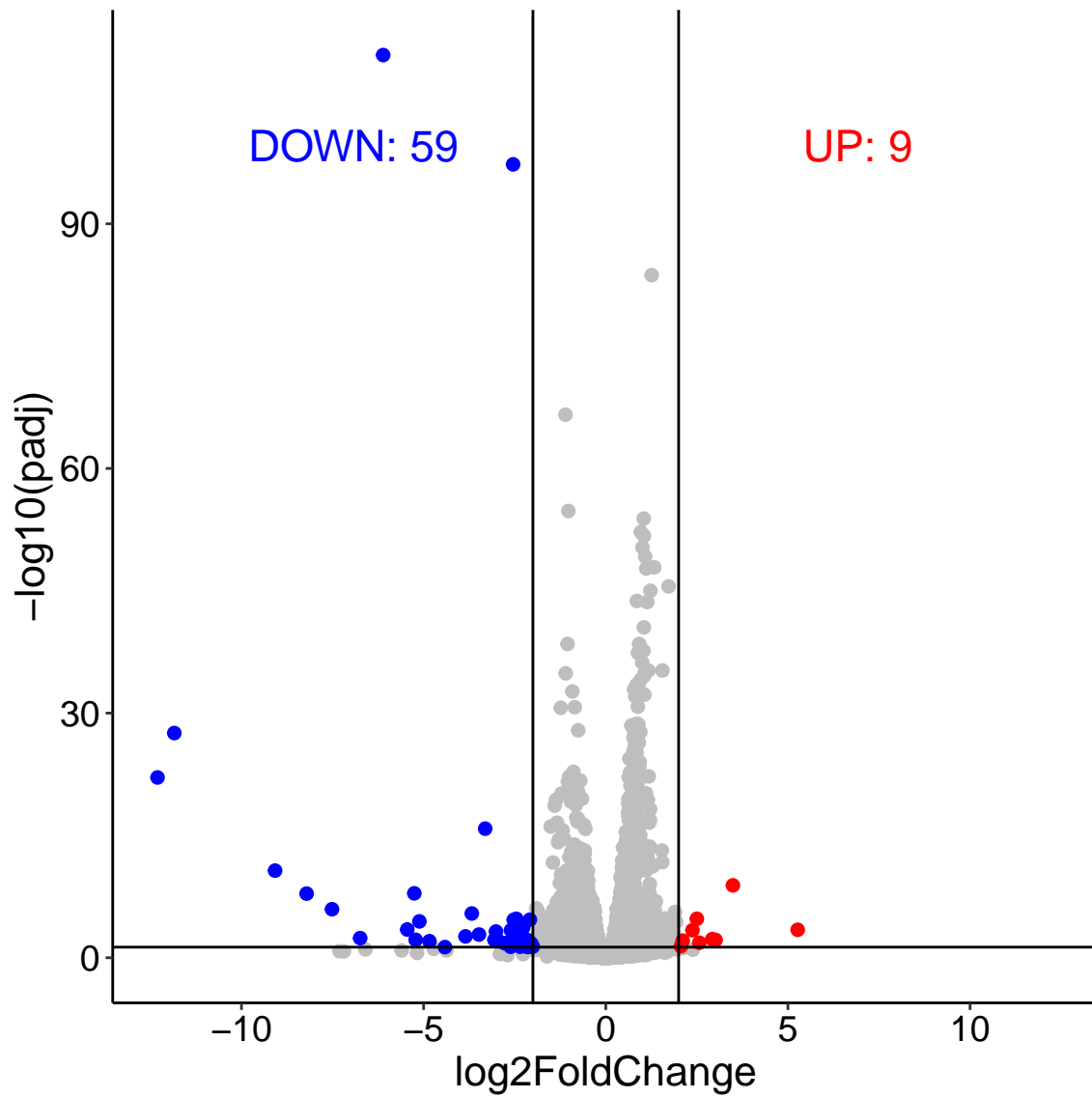
```
## [1] "Heatmap for 02_heatmap_log2fc_1 "
```
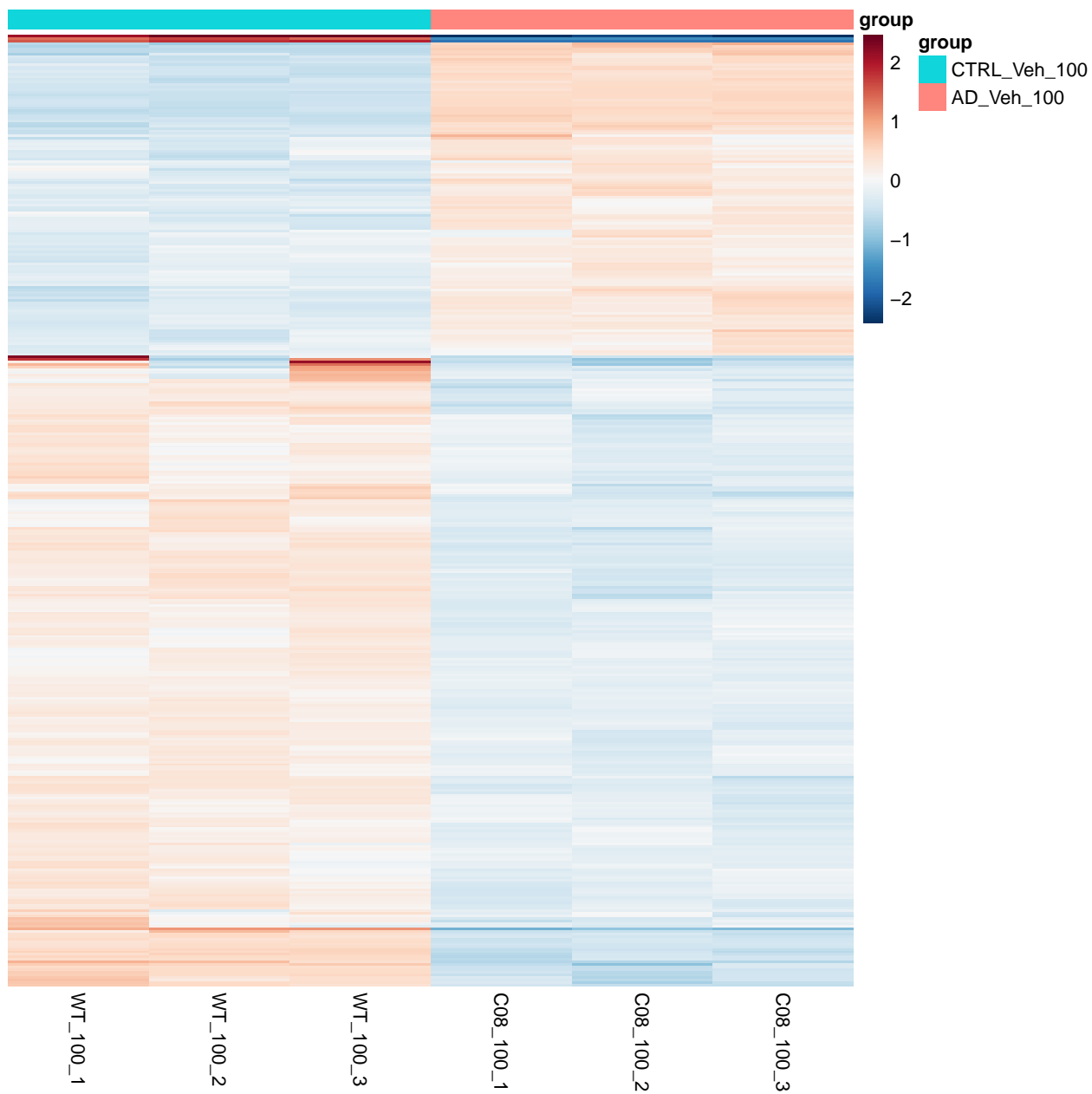
[1] "Heatmap for 02_heatmap_log2fc_1_row "
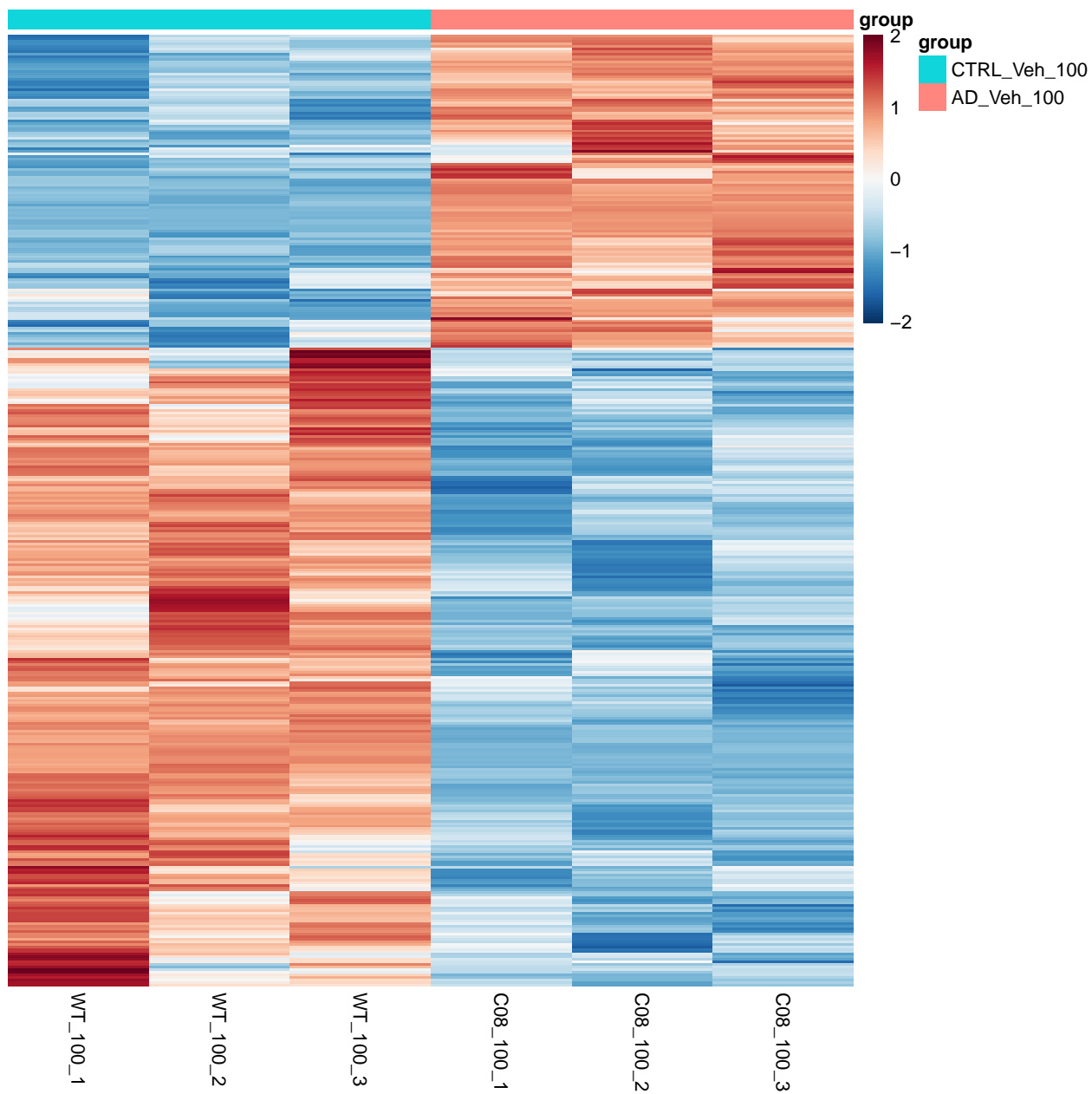
```
## [1] "Heatmap for 03_heatmap_log2fc_1.5 "
```
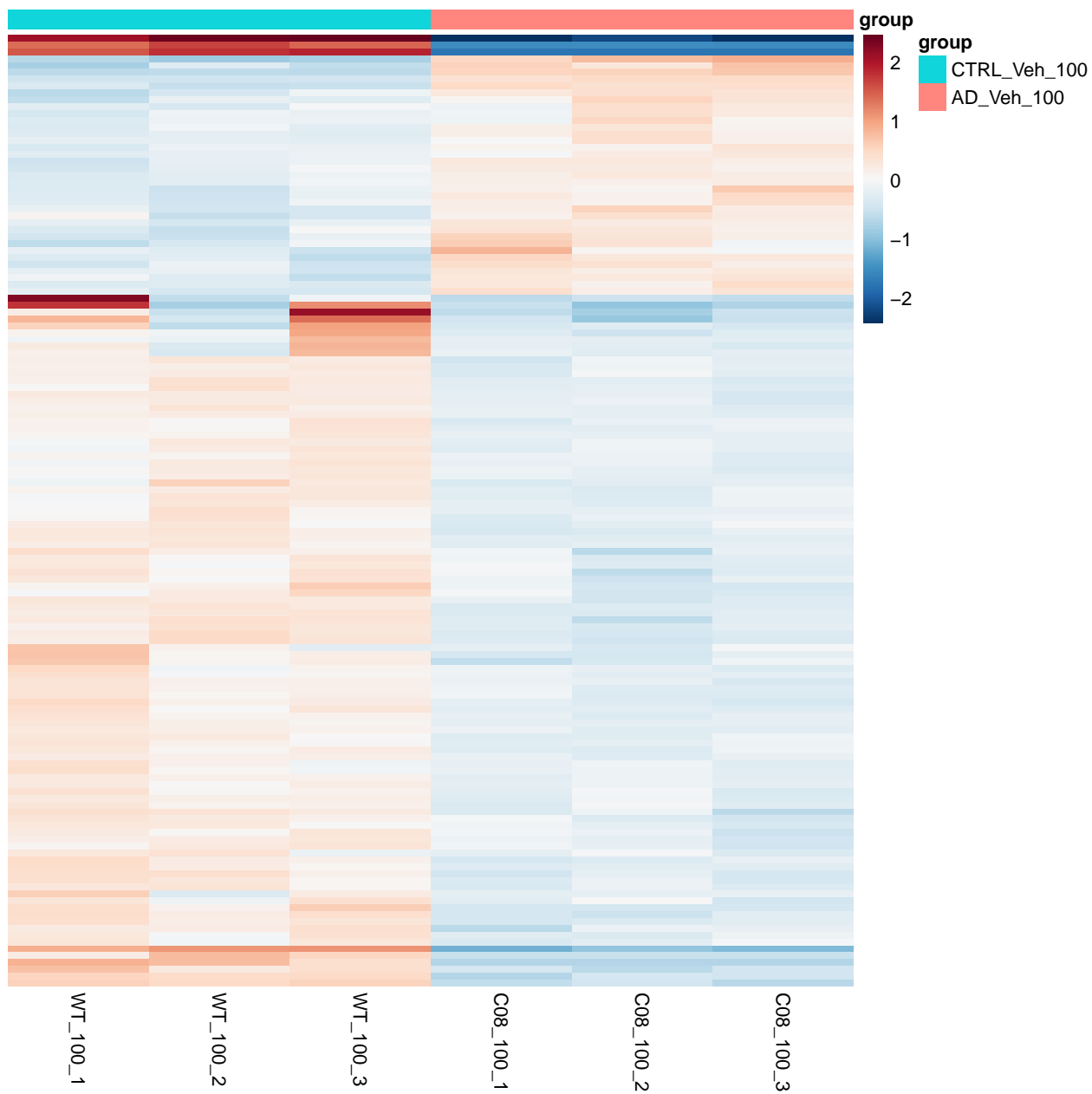
```
## [1] "Heatmap for 03_heatmap_log2fc_1.5_row "
```
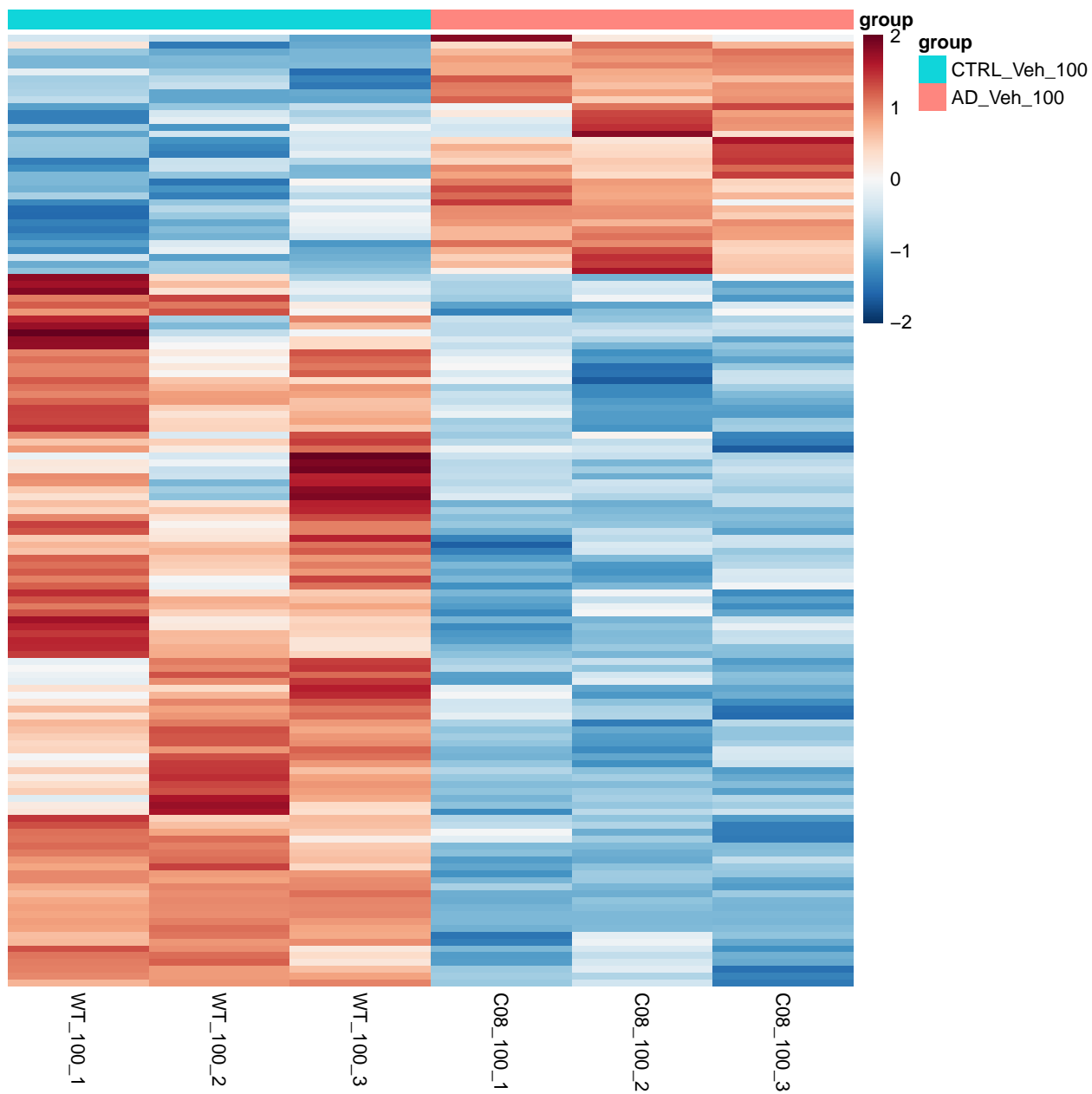
[1] "Heatmap for 03_heatmap_log2fc_2 "
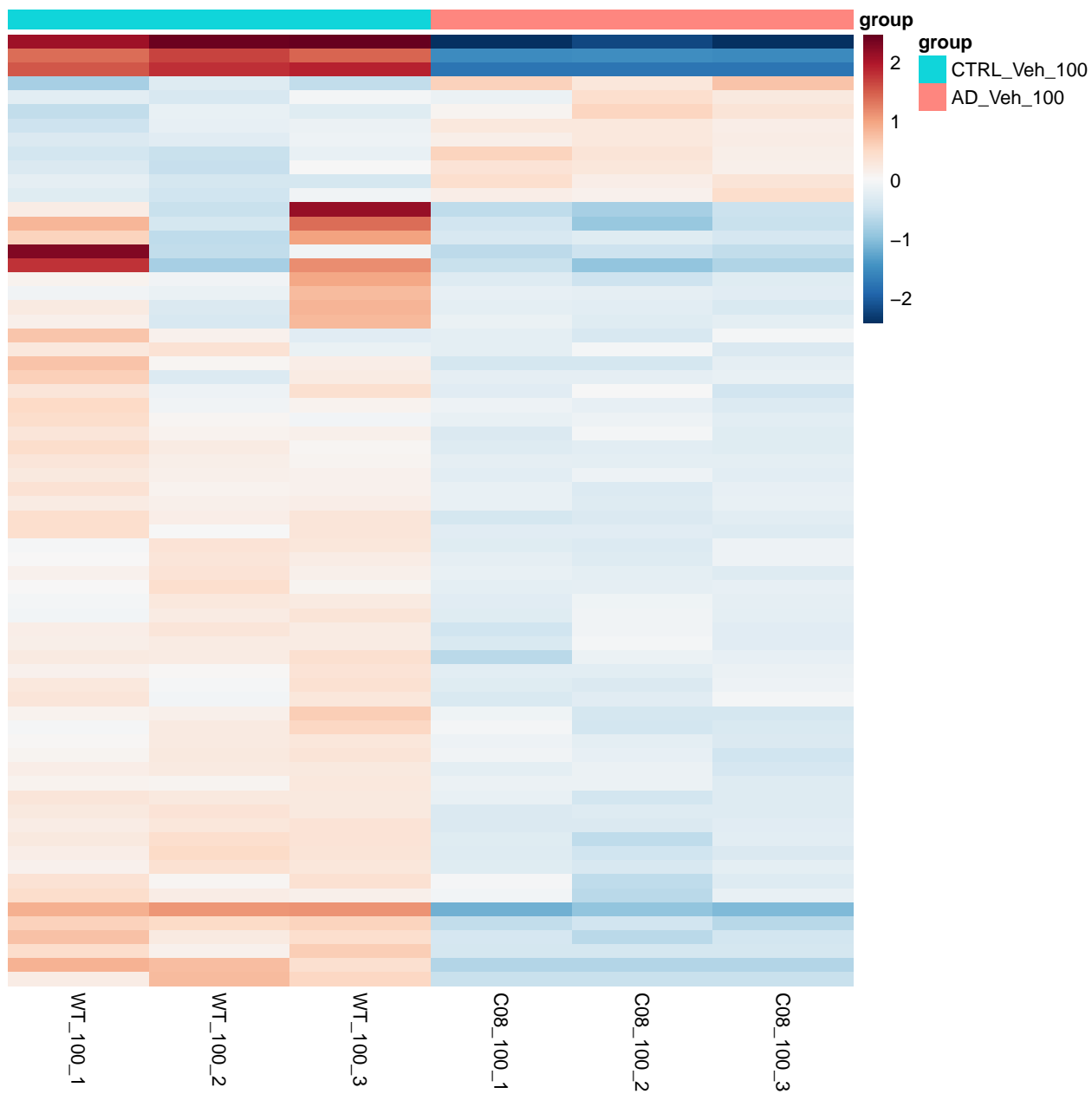
[1] "Heatmap for 03_heatmap_log2fc_2_row "
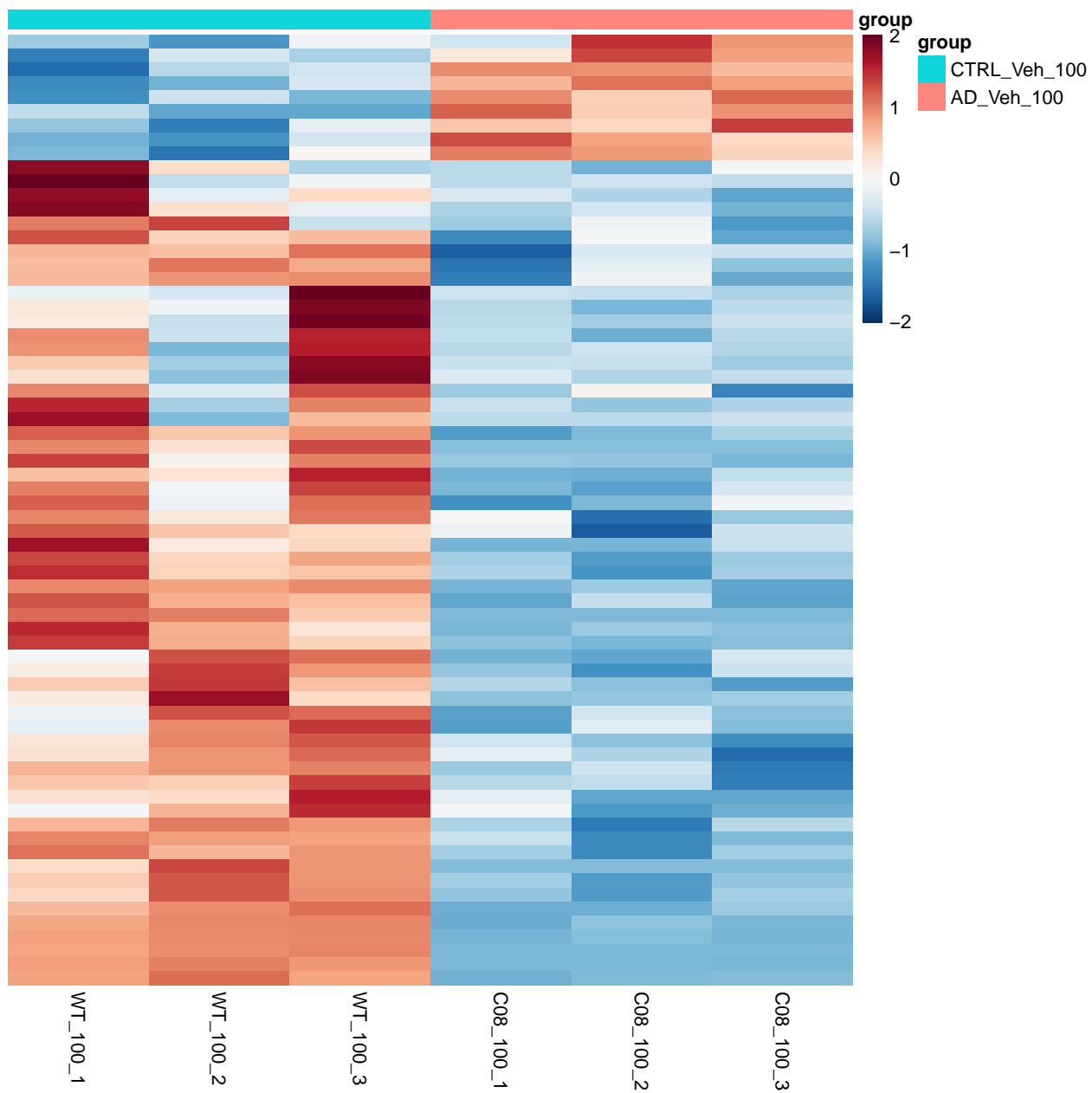
4. GSVA analysis

```
## No annotation package name available in the input data object.
## Attempting to directly match identifiers in data to gene sets.

## Estimating GSVA scores for 10531 gene sets.
## Estimating ECDFs with Gaussian kernels
##    |                                                              |
```
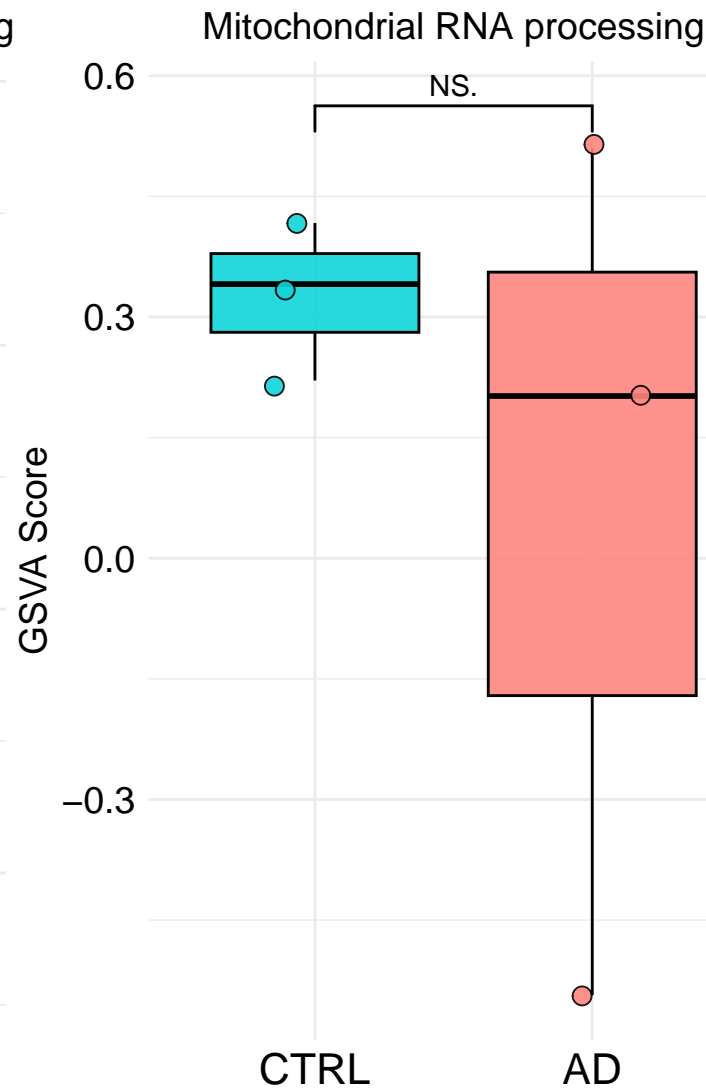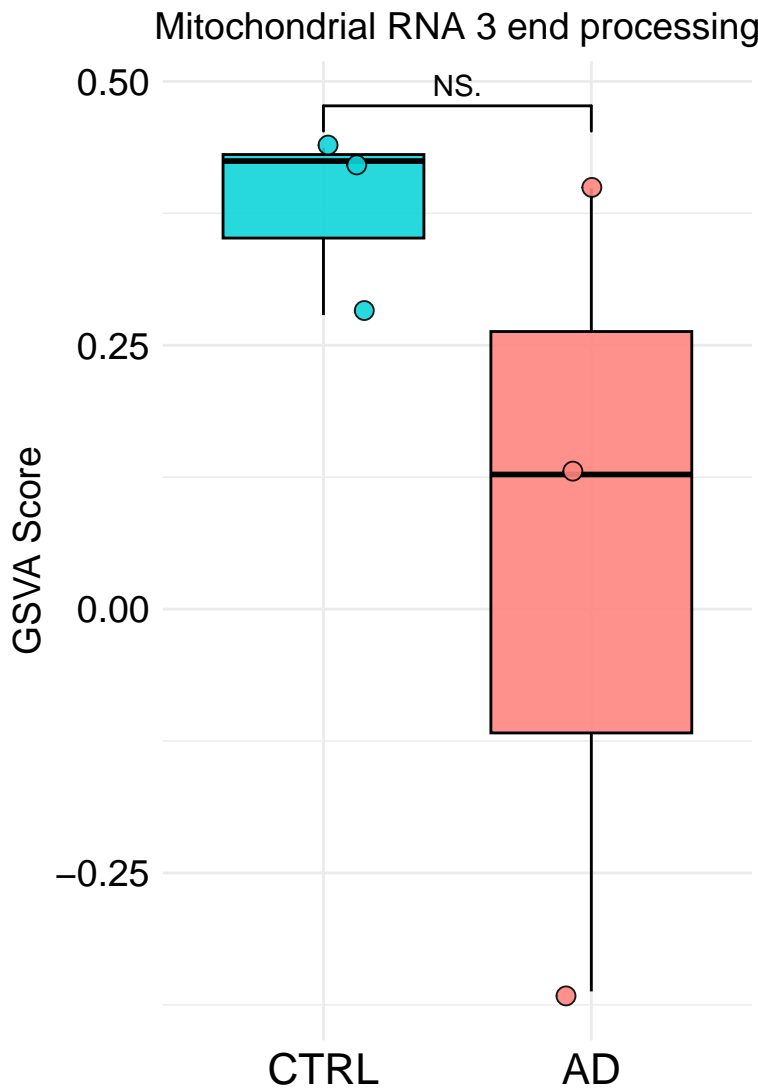
5. Pathway Enrichment Analysis

[1] "Enrichment analysis for 01-DEG_1.0_up "

Top 30 Pathways (Sorted by FDR)

[1] "Enrichment analysis for GOBP 01-DEG_1.0_up "
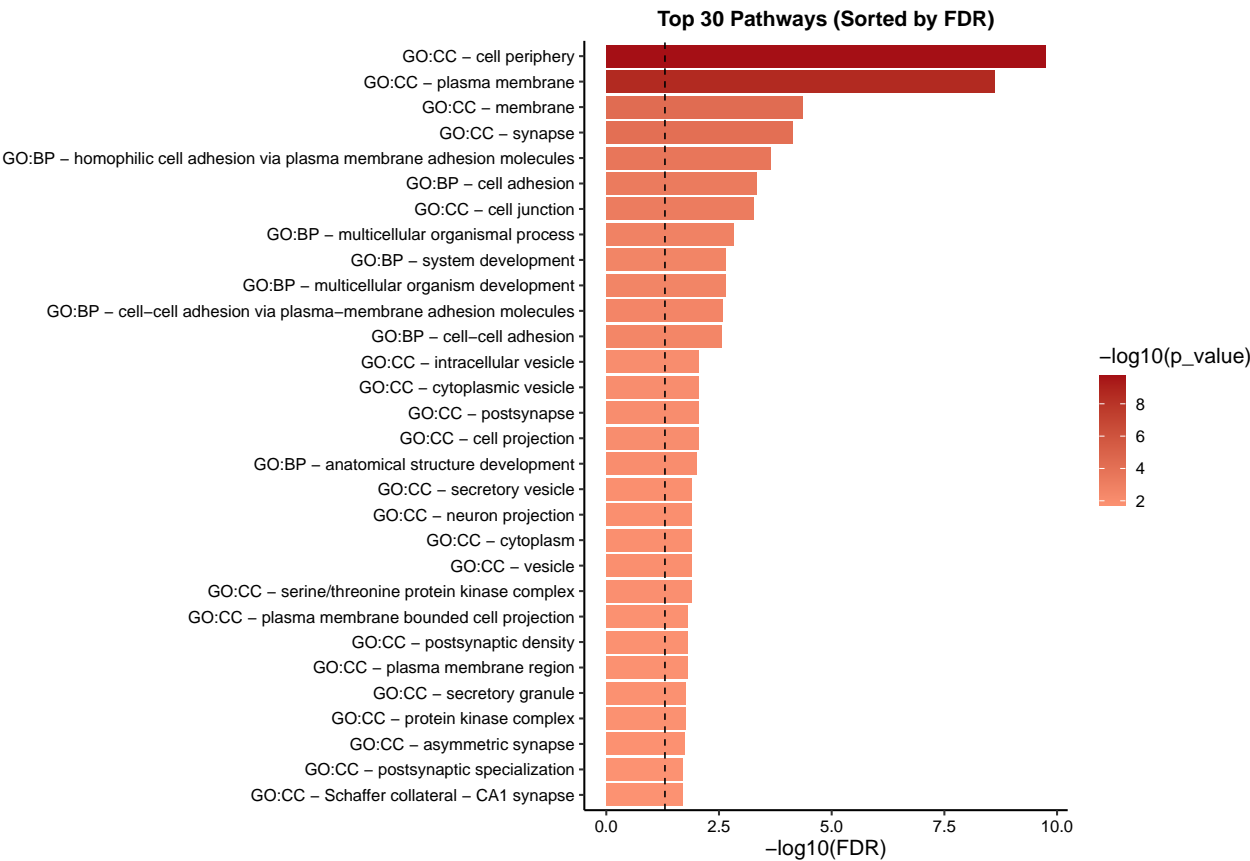
Top 30 Pathways for GOBP (Sorted by FDR)
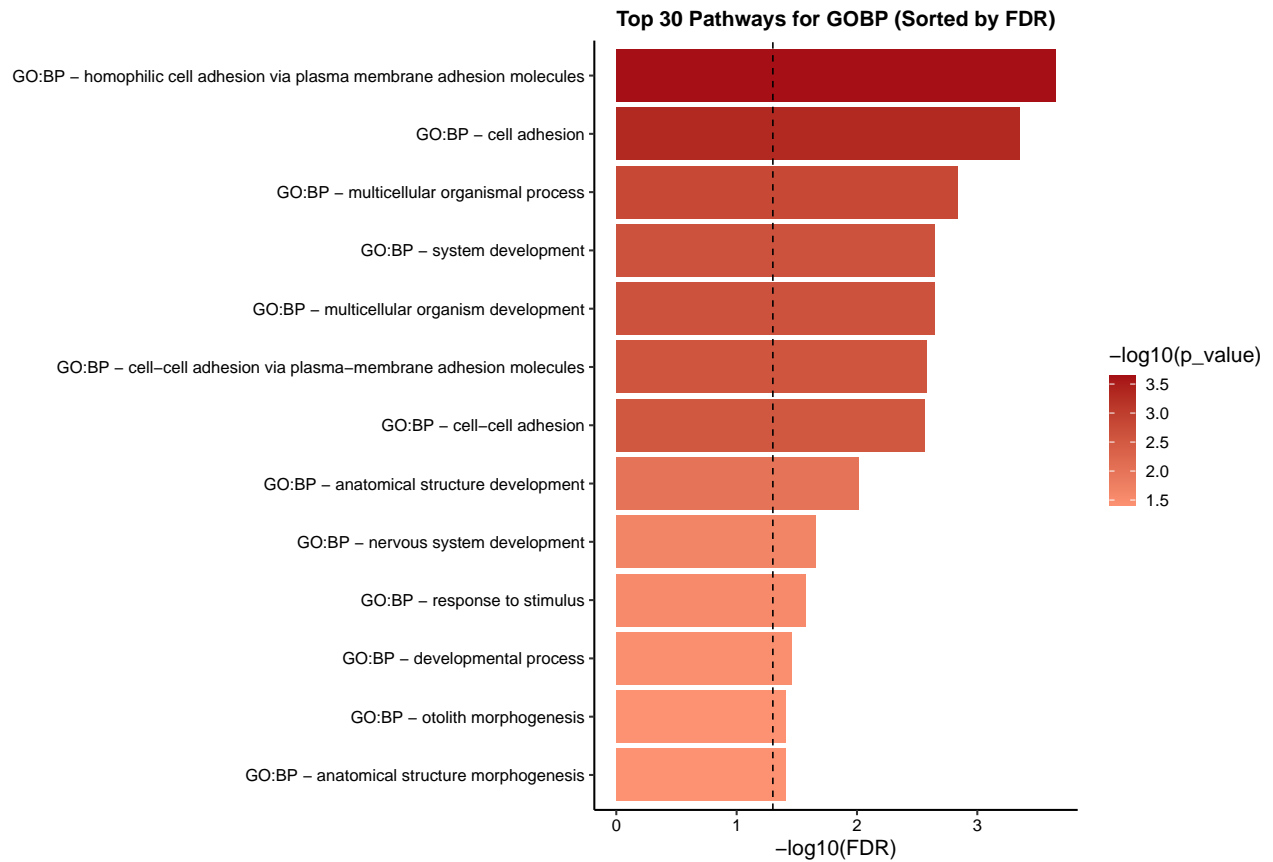
[1] "Enrichment analysis for 01-DEG_1.0_down "

Top 30 Pathways (Sorted by FDR)
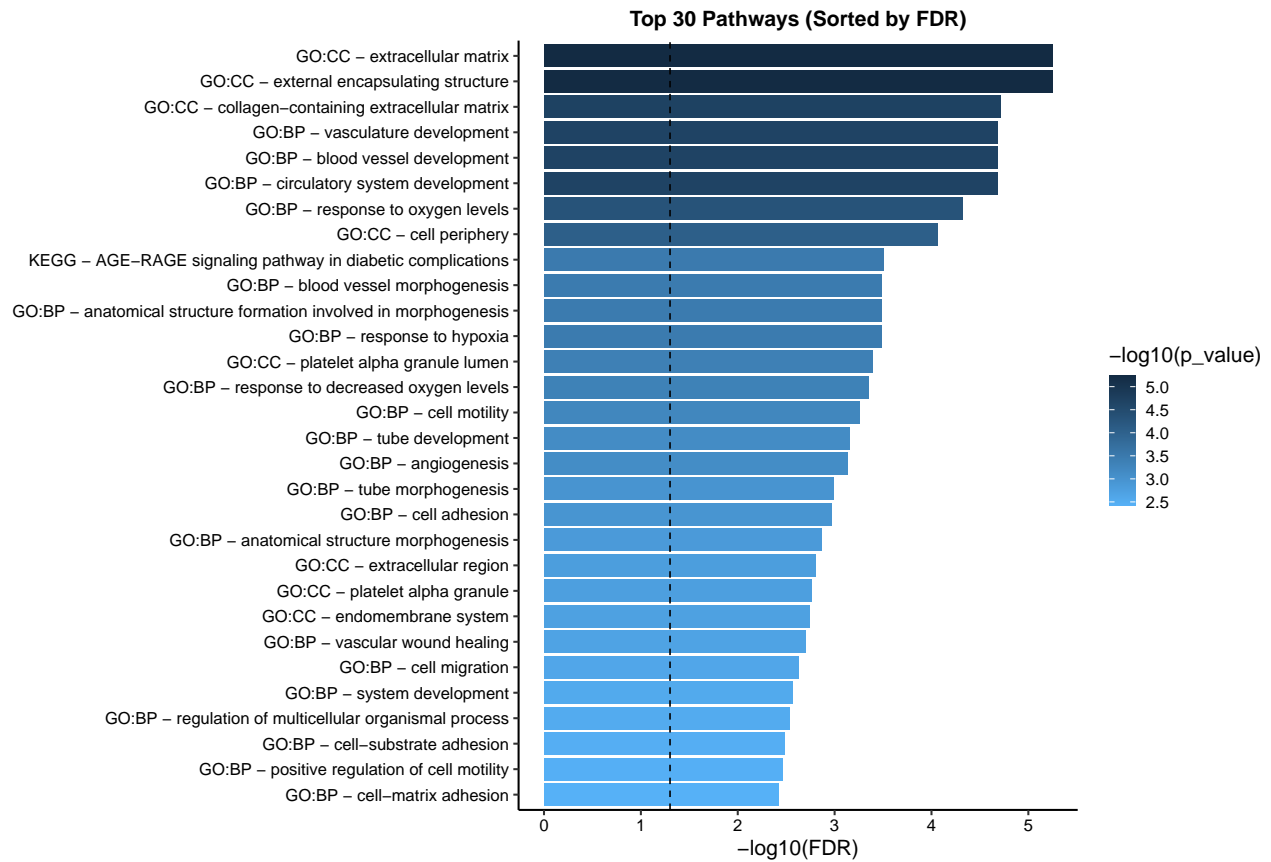
[1] "Enrichment analysis for GOBP 01-DEG_1.0_down "

Top 30 Pathways for GOBP (Sorted by FDR)

[1] "Enrichment analysis for 02-DEG_1.5_up "

Top 30 Pathways (Sorted by FDR)

CORUM – Sulphiredoxin–peroxiredoxin complex

CORUM – EIF3 complex (EIF3A, EIF3B, EIF3G, EIF3I, EIF3C)

CORUM – DLG5–SORBS3–CTNNB1 complex

CORUM – MLC1–Na,K–ATPase–Kir4.1–AQP4–TRPV4–syntrophin complex

CORUM – HEPACAM–MLC1–Na,K–ATPase–Kir4.1–AQP4–TRPV4 complex

CORUM – eIF3 complex (EIF3S6, EIF3S5, EIF3S4, EIF3S3, EIF3S6IP, EIF3S2, EIF3S9, EIF3S12, EIF3S10, EIF3S8, EIF3S1, EIF3S7)

CORUM – eIF3 complex (EIF3S6, EIF3S5, EIF3S4, EIF3S3, EIF3S6IP, EIF3S2, EIF3S9, EIF3S12, EIF3S10, EIF3S8, EIF3S1, EIF3S7, PCID1)

GO:CC – lamellar body

–log10(FDR)

–log10(p_value)

1.9
1.8
1.7
1.6

[1] "Enrichment analysis for GOBP 02-DEG_1.5_up "

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Top 30 Pathways for GOBP (Sorted by FDR)

−log10(FDR)

```
## [1] "Enrichment analysis for 02-DEG_1.5_down "
```

Top 30 Pathways (Sorted by FDR)

[1] "Enrichment analysis for GOBP 02-DEG_1.5_down "

Top 30 Pathways for GOBP (Sorted by FDR)

[1] "Enrichment analysis for O3-DEG_2_up "

Top 30 Pathways (Sorted by FDR)

CORUM − Sulphiredoxin−peroxiredoxin complex

CORUM − EIF3 complex (EIF3A, EIF3B, EIF3G, EIF3I, EIF3C)

CORUM − DLG5−SORBS3−CTNNB1 complex

CORUM − MLC1−Na,K−ATPase−Kir4.1−AQP4−TRPV4−syntrophin complex

CORUM − HEPACAM−MLC1−Na,K−ATPase−Kir4.1−AQP4−TRPV4 complex

CORUM − eIF3 complex (EIF3S6, EIF3S5, EIF3S4, EIF3S3, EIF3S6IP, EIF3S2, EIF3S9, EIF3S12, EIF3S10, EIF3S8, EIF3S1, EIF3S7)

CORUM − eIF3 complex (EIF3S6, EIF3S5, EIF3S4, EIF3S3, EIF3S6IP, EIF3S2, EIF3S9, EIF3S12, EIF3S10, EIF3S8, EIF3S1, EIF3S7, PCID1)

GO:CC − lamellar body

−log10(p_value)

1.9
1.8
1.7
1.6

0.0

−log10(FDR)

[1] "Enrichment analysis for GOBP O3-DEG_2_up "

Top 30 Pathways for GOBP (Sorted by FDR)

−log10(FDR)

1.275　　　　　1.300　　　　　1.325　　　　　1.350
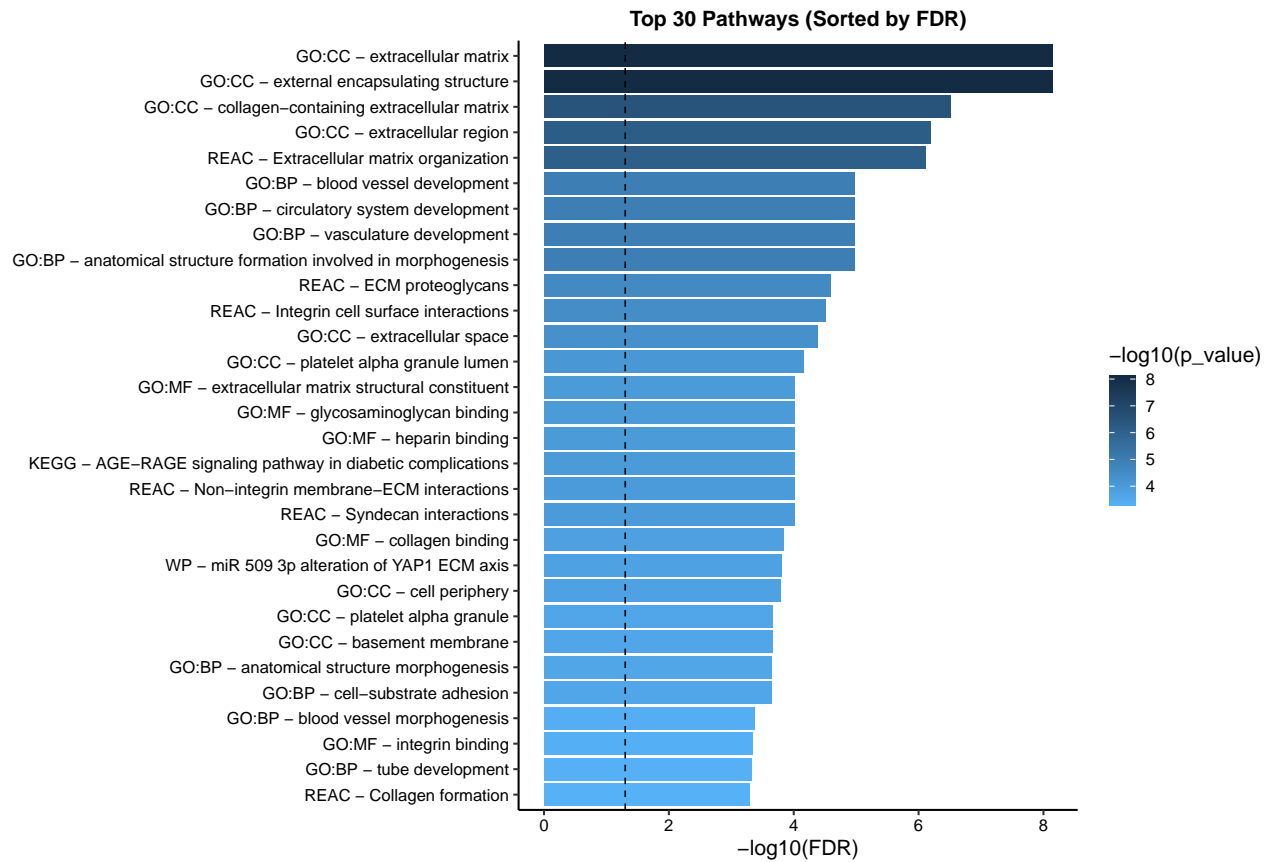
```
## [1] "Enrichment analysis for 03-DEG_2_down "
```

Top 30 Pathways (Sorted by FDR)
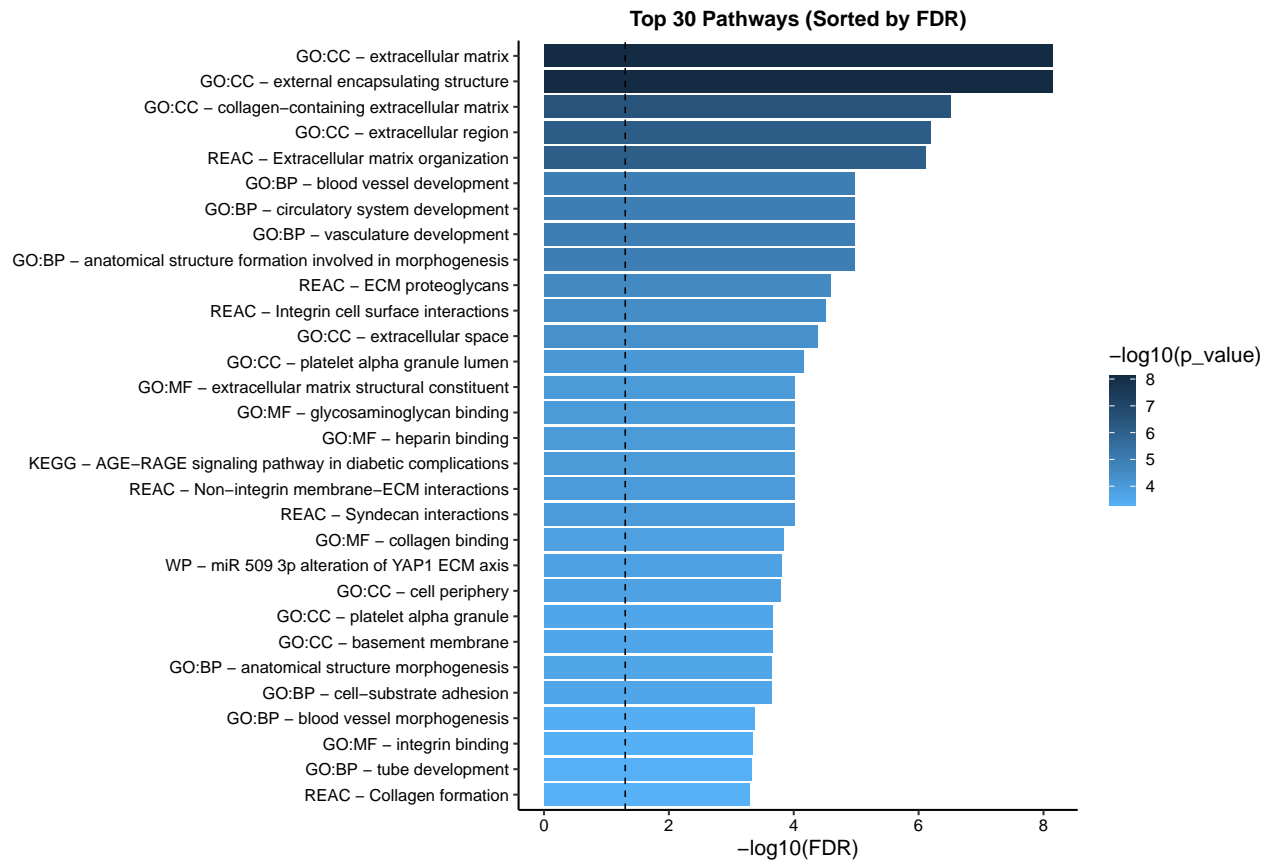

```
## [1] "Enrichment analysis for GOBP 03-DEG_2_down "
```

Top 30 Pathways for GOBP (Sorted by FDR)
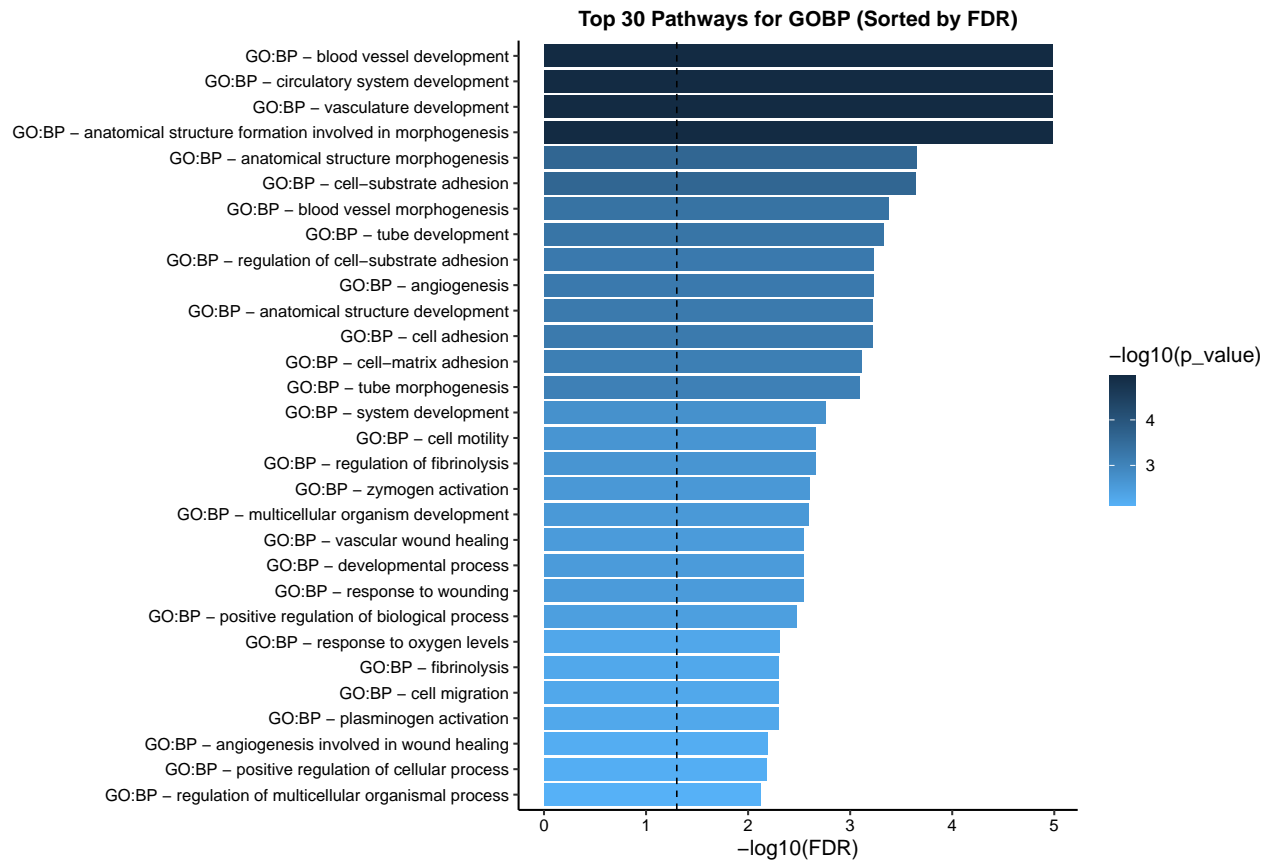
Session information

```
## R version 4.4.0 (2024-04-24)
## Platform: aarch64-apple-darwin20
## Running under: macOS Sonoma 14.3.1
##
## Matrix products: default
## BLAS:   /Library/Frameworks/R.framework/Versions/4.4-arm64/Resources/lib/libRblas.0.dylib
## LAPACK: /Library/Frameworks/R.framework/Versions/4.4-arm64/Resources/lib/libRlapack.dylib;  LAPACK v
##
## locale:
## [1] en_US.UTF-8/en_US.UTF-8/en_US.UTF-8/C/en_US.UTF-8/en_US.UTF-8
##
## time zone: America/New_York
## tzcode source: internal
##
## attached base packages:
## [1] parallel  stats4    stats     graphics  grDevices utils     datasets
## [8] methods   base
##
## other attached packages:
##  [1] GSEABase_1.66.0           graph_1.82.0
##  [3] annotate_1.82.0           XML_3.99-0.18
##  [5] extrafont_0.19            ggsignif_0.6.4
##  [7] patchwork_1.3.0           decoupleR_2.10.0
##  [9] GSVA_1.52.3               BiocParallel_1.38.0
## [11] edgeR_4.2.2               limma_3.60.6
## [13] GenomicFeatures_1.56.0    biomaRt_2.60.1
## [15] gprofiler2_0.2.3          RColorBrewer_1.1-3
## [17] data.table_1.16.4         org.Hs.eg.db_3.19.1
## [19] AnnotationDbi_1.66.0      clusterProfiler_4.12.6
## [21] ggfortify_0.4.17          pheatmap_1.0.12
## [23] EnhancedVolcano_1.22.0    ggrepel_0.9.6
## [25] apeglm_1.26.1             DESeq2_1.44.0
## [27] SummarizedExperiment_1.34.0 Biobase_2.64.0
## [29] MatrixGenerics_1.16.0     matrixStats_1.5.0
## [31] reshape2_1.4.4            Matrix_1.7-2
## [33] Signac_1.14.0             Seurat_5.2.1
## [35] SeuratObject_5.0.2        sp_2.2-0
## [37] rtracklayer_1.64.0        GenomicRanges_1.56.2
## [39] GenomeInfoDb_1.40.1       IRanges_2.38.1
## [41] S4Vectors_0.42.1          BiocGenerics_0.50.0
## [43] knitr_1.49                lubridate_1.9.4
## [45] forcats_1.0.0             stringr_1.5.1
## [47] dplyr_1.1.4               purrr_1.0.4
## [49] readr_2.1.5               tidyr_1.3.1
## [51] tibble_3.2.1              ggplot2_3.5.1
## [53] tidyverse_2.0.0
##
## loaded via a namespace (and not attached):
##    [1] SpatialExperiment_1.14.0   R.methodsS3_1.8.2
##    [3] progress_1.2.3             goftest_1.2-3
##    [5] HDF5Array_1.32.1           Biostrings_2.72.1
##    [7] vctrs_0.6.5                spatstat.random_3.3-2
```

```
##    [9] digest_0.6.37               png_0.1-8
##   [11] deldir_2.0-4                parallelly_1.42.0
##   [13] magick_2.8.5                MASS_7.3-64
##   [15] httpuv_1.6.15               qvalue_2.36.0
##   [17] withr_3.0.2                 xfun_0.51
##   [19] ggfun_0.1.8                 survival_3.8-3
##   [21] memoise_2.0.1               gson_0.1.0
##   [23] systemfonts_1.2.1           ragg_1.3.3
##   [25] tidytree_0.4.6              zoo_1.8-12
##   [27] pbapply_1.7-2               R.oo_1.27.0
##   [29] prettyunits_1.2.0           KEGGREST_1.44.1
##   [31] promises_1.3.2              httr_1.4.7
##   [33] restfulr_0.0.15             rhdf5filters_1.16.0
##   [35] globals_0.16.3              fitdistrplus_1.2-2
##   [37] rhdf5_2.48.0                rstudioapi_0.17.1
##   [39] UCSC.utils_1.0.0            miniUI_0.1.1.1
##   [41] generics_0.1.3              DOSE_3.30.5
##   [43] curl_6.2.1                  zlibbioc_1.50.0
##   [45] ScaledMatrix_1.12.0         ggraph_2.2.1
##   [47] polyclip_1.10-7             GenomeInfoDbData_1.2.12
##   [49] SparseArray_1.4.8           xtable_1.8-4
##   [51] evaluate_1.0.3              S4Arrays_1.4.1
##   [53] BiocFileCache_2.12.0        hms_1.1.3
##   [55] irlba_2.3.5.1               colorspace_2.1-1
##   [57] filelock_1.0.3              ROCR_1.0-11
##   [59] reticulate_1.40.0           spatstat.data_3.1-4
##   [61] magrittr_2.0.3              lmtest_0.9-40
##   [63] later_1.4.1                 viridis_0.6.5
##   [65] ggtree_3.12.0               lattice_0.22-6
##   [67] spatstat.geom_3.3-5         future.apply_1.11.3
##   [69] scattermore_1.2             shadowtext_0.1.4
##   [71] cowplot_1.1.3               RcppAnnoy_0.0.22
##   [73] pillar_1.10.1               nlme_3.1-167
##   [75] compiler_4.4.0              beachmat_2.20.0
##   [77] RSpectra_0.16-2             stringi_1.8.4
##   [79] tensor_1.5                  GenomicAlignments_1.40.0
##   [81] plyr_1.8.9                  crayon_1.5.3
##   [83] abind_1.4-8                 BiocIO_1.14.0
##   [85] gridGraphics_0.5-1          emdbook_1.3.13
##   [87] locfit_1.5-9.11             graphlayouts_1.2.2
##   [89] bit_4.5.0.1                 fastmatch_1.1-6
##   [91] textshaping_1.0.0           codetools_0.2-20
##   [93] BiocSingular_1.20.0         plotly_4.10.4
##   [95] mime_0.12                   splines_4.4.0
##   [97] Rcpp_1.0.14                 fastDummies_1.7.5
##   [99] sparseMatrixStats_1.16.0    dbplyr_2.5.0
## [101] Rttf2pt1_1.3.12             blob_1.2.4
## [103] here_1.0.1                  fs_1.6.5
## [105] listenv_0.9.1               ggplotify_0.1.2
## [107] statmod_1.5.0               tzdb_0.4.0
## [109] tweenr_2.0.3                pkgconfig_2.0.3
## [111] tools_4.4.0                 cachem_1.1.0
## [113] RSQLite_2.3.9               viridisLite_0.4.2
## [115] DBI_1.2.3                   numDeriv_2016.8-1.1
```

```
## [117] fastmap_1.2.0              rmarkdown_2.29
## [119] scales_1.3.0              grid_4.4.0
## [121] ica_1.0-3                 Rsamtools_2.20.0
## [123] coda_0.19-4.1             dotCall64_1.2
## [125] RANN_2.6.2                farver_2.1.2
## [127] tidygraph_1.3.1           scatterpie_0.2.4
## [129] yaml_2.3.10               cli_3.6.4
## [131] lifecycle_1.0.4           uwot_0.2.2
## [133] mvtnorm_1.3-3             timechange_0.3.0
## [135] gtable_0.3.6              rjson_0.2.23
## [137] ggridges_0.5.6            progressr_0.15.1
## [139] ape_5.8-1                 jsonlite_1.9.0
## [141] RcppHNSW_0.6.0            bitops_1.0-9
## [143] bit64_4.6.0-1             Rtsne_0.17
## [145] yulab.utils_0.2.0         spatstat.utils_3.1-2
## [147] bdsmatrix_1.3-7           GOSemSim_2.30.2
## [149] spatstat.univar_3.1-1     R.utils_2.12.3
## [151] lazyeval_0.2.2            shiny_1.10.0
## [153] htmltools_0.5.8.1         enrichplot_1.24.4
## [155] GO.db_3.19.1              sctransform_0.4.1
## [157] rappdirs_0.3.3            tinytex_0.55
## [159] glue_1.8.0                spam_2.11-1
## [161] httr2_1.1.0               XVector_0.44.0
## [163] RCurl_1.98-1.16           rprojroot_2.0.4
## [165] treeio_1.28.0             gridExtra_2.3
## [167] extrafontdb_1.0           igraph_2.1.4
## [169] R6_2.6.1                  SingleCellExperiment_1.26.0
## [171] labeling_0.4.3            RcppRoll_0.3.1
## [173] cluster_2.1.8             bbmle_1.0.25.1
## [175] Rhdf5lib_1.26.0           aplot_0.2.4
## [177] DelayedArray_0.30.1       tidyselect_1.2.1
## [179] ggforce_0.4.2             xml2_1.3.6
## [181] future_1.34.0             rsvd_1.0.5
## [183] munsell_0.5.1             KernSmooth_2.23-26
## [185] htmlwidgets_1.6.4         fgsea_1.30.0
## [187] rlang_1.1.5               spatstat.sparse_3.1-0
## [189] spatstat.explore_3.3-4
```