

Scoring cells singscore

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Loading package

```
suppressMessages(library(dplyr))
suppressMessages(library(patchwork))
suppressMessages(library(ggplot2))
suppressMessages(library(readxl))
suppressMessages(library(singscore))
suppressMessages(library(pheatmap))
```

Loading the normalised count matrix

```
Norm_exprMat <- read_excel("../Data_DSPDA/Q3 - publication - V2.xlsx", sheet =
"TargetCountMatrix")
Norm_exprMat <- as.data.frame(Norm_exprMat)
rownames(Norm_exprMat) <- Norm_exprMat$TargetName
Norm_exprMat$TargetName <- NULL
```

Score sample with singscore

Get score Motro

```
Motro_DE <- read.csv("myeloids_markers_top20.csv")

Motro_DE <- Motro_DE[Motro_DE$cluster == 1,]
Top20_Motro <- Motro_DE$gene

rankData <- rankGenes(Norm_exprMat)
scoredf <- simpleScore(rankData, upSet = Top20_Motro)

## Warning in checkGenes(upSet, rownames(rankData)): 2 genes missing: Ly6a,
## AW112010

Motro <- scoredf$TotalScore
```

Get score AM

```
myeloid.markers <- read.csv("myeloids_markers_top20.csv")

myeloid.markers_AM <- myeloid.markers[myeloid.markers$cluster == 5,]$gene

# Ear2 and Kcnq1ot1 are not in the expression data
myeloid.markers_AM <- myeloid.markers_AM[-c(7,13)]

rankData <- rankGenes(Norm_exprMat)

scoredf <- simpleScore(rankData, upSet = myeloid.markers_AM)
AM <- scoredf$TotalScore

cell_singscore <- data.frame(Motro, AM)
```

Get score neutro

```
myeloid.markers_neut <- myeloid.markers[myeloid.markers$cluster == 6,]$gene

# Stfa2l1 and Cstdc4 are not in the expression data
myeloid.markers_neut <- myeloid.markers_neut[-c(5,8)]

rankData <- rankGenes(Norm_exprMat)

scoredf <- simpleScore(rankData, upSet = myeloid.markers_neut)
cell_singscore <- cbind(cell_singscore, Neutro = scoredf$TotalScore)
```

Get score cd64

```
myeloid.markers_cd64 <- myeloid.markers[myeloid.markers$cluster == 4,]$gene

# Ly6c2 and Ifi204 are not in the expression data
myeloid.markers_cd64 <- myeloid.markers_cd64[-c(2,12)]
```

```
rankData <- rankGenes(Norm_exprMat)
scoredf <- simpleScore(rankData, upSet = myeloid.markers_cd64)

cell_singscore <- cbind(cell_singscore, cd64 = scoredf$TotalScore)
```

Get score CD206- IM

```
myeloid.markers_IM <- myeloid.markers[myeloid.markers$cluster == 0,]$gene

rankData <- rankGenes(Norm_exprMat)
scoredf <- simpleScore(rankData, upSet = myeloid.markers_IM)

cell_singscore <- cbind(cell_singscore, "CD206- IM" = scoredf$TotalScore)
```

Get score CD206+ IM

```
myeloid.markers_CD206 <- myeloid.markers[myeloid.markers$cluster == 7,]$gene

rankData <- rankGenes(Norm_exprMat)

scoredf <- simpleScore(rankData, upSet = myeloid.markers_CD206)

cell_singscore <- cbind(cell_singscore, "CD206+ IM" = scoredf$TotalScore)
```

Write csv file for singscore

```
rownames(cell_singscore) <- colnames(Norm_exprMat)

#write.csv(cell_singscore, "cell_singscore.csv")
```

plotting score in pheatmap

```
annotation_sample <- read_excel("../Data_DSPDA/Q3 - publication - V2.xlsx", sheet =
"SegmentProperties")
singscores <- cell_singscore
singscores <- merge(singscores, annotation_sample[,c(5,25)], by.x = "row.names", by.y =
"SegmentDisplayName")
singscores <- singscores[order(singscores$Group),]
singscores$Group<- NULL

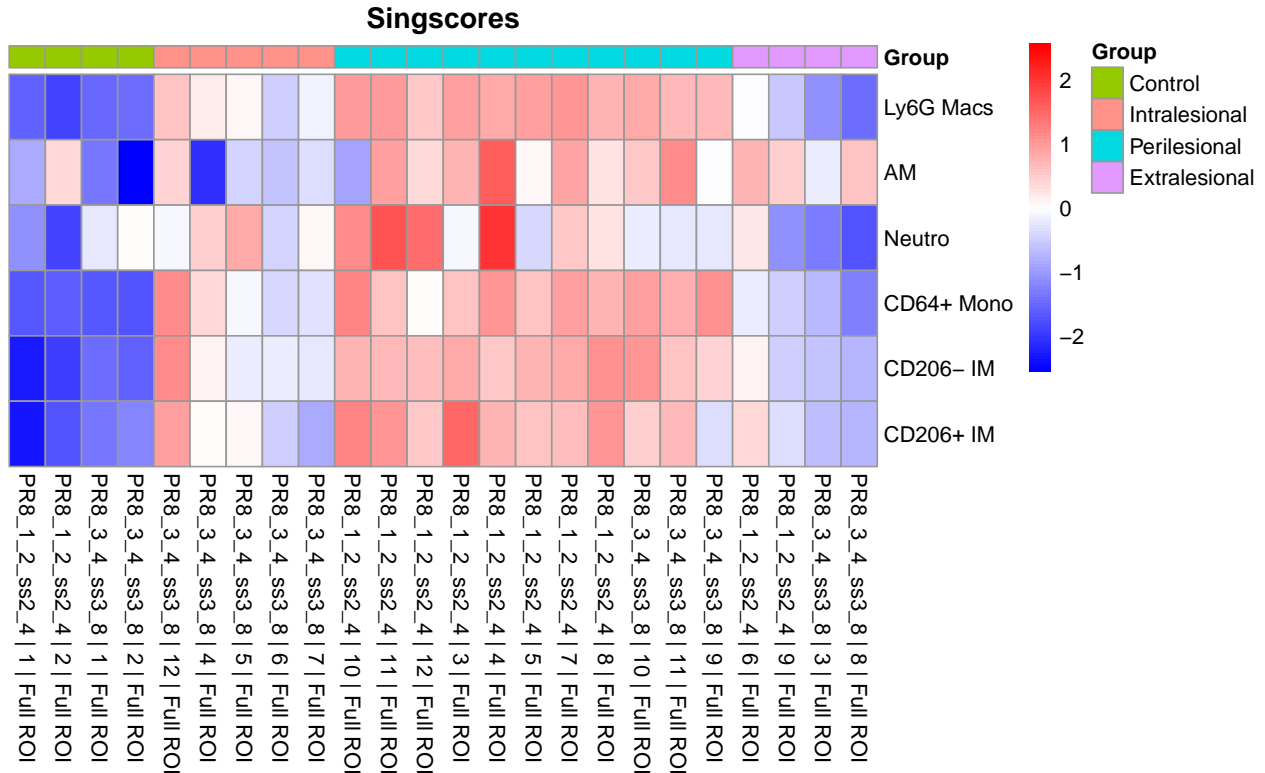
rownames(singscores) <- singscores$Row.names
singscores$Row.names <- NULL

df <- as.data.frame(annotation_sample[,c(5,25)])
df$Group <- as.factor(df$Group)
levels(df$Group) <- c("Control", "Intralesional", "Perilesional", "Extralesional")
rownames(df)<-df$SegmentDisplayName
df$SegmentDisplayName <- NULL
```

```

pheatmap_singscore<- pheatmap(t(singscores), cluster_rows=FALSE, show_rownames=T,
  cluster_cols=F, scale = "row", annotation_col=df, main = "Singscores",
  labels_row = c("Ly6G Macs", "AM", "Neutro", "CD64+ Mono", "CD206- IM", "CD206+
  IM" ), color = colorRampPalette(c("blue", "white", "red"))(100))

```



```
sessionInfo()
```

```

## R version 4.3.1 (2023-06-16)
## Platform: x86_64-pc-linux-gnu (64-bit)
## Running under: Ubuntu 22.04.3 LTS
##
## Matrix products: default
## BLAS: /usr/lib/x86_64-linux-gnu/openblas-pthread/libblas.so.3
## LAPACK: /usr/lib/x86_64-linux-gnu/openblas-pthread/libopenblaspr0.3.20.so; LAPACK version 3.10.0
##
## locale:
## [1] LC_CTYPE=en_US.UTF-8 LC_NUMERIC=C
## [3] LC_TIME=fr_BE.UTF-8 LC_COLLATE=en_US.UTF-8
## [5] LC_MONETARY=fr_BE.UTF-8 LC_MESSAGES=en_US.UTF-8
## [7] LC_PAPER=fr_BE.UTF-8 LC_NAME=C
## [9] LC_ADDRESS=C LC_TELEPHONE=C
## [11] LC_MEASUREMENT=fr_BE.UTF-8 LC_IDENTIFICATION=C
##
## time zone: Europe/Brussels
## tzcode source: system (glibc)
##
## attached base packages:

```

```

## [1] stats      graphics  grDevices  utils      datasets  methods   base
##
## other attached packages:
## [1] pheatmap_1.0.12  singscore_1.20.0 readxl_1.4.2    ggplot2_3.4.2
## [5] patchwork_1.1.2  dplyr_1.1.2
##
## loaded via a namespace (and not attached):
## [1] KEGGREST_1.40.0      SummarizedExperiment_1.30.2
## [3] gtable_0.3.3         xfun_0.39
## [5] lattice_0.21-8       Biobase_2.60.0
## [7] vctrs_0.6.3          tools_4.3.1
## [9] bitops_1.0-7         generics_0.1.3
## [11] stats4_4.3.1         tibble_3.2.1
## [13] fansi_1.0.4          AnnotationDbi_1.62.1
## [15] RSQLite_2.3.1        highr_0.10
## [17] blob_1.2.4           pkgconfig_2.0.3
## [19] Matrix_1.6-0         RColorBrewer_1.1-3
## [21] S4Vectors_0.38.1     graph_1.78.0
## [23] lifecycle_1.0.3      GenomeInfoDbData_1.2.10
## [25] farver_2.1.1         stringr_1.5.0
## [27] compiler_4.3.1       Biostrings_2.68.1
## [29] munsell_0.5.0         GenomeInfoDb_1.36.0
## [31] htmltools_0.5.5      RCurl_1.98-1.12
## [33] yaml_2.3.7           tidyr_1.3.0
## [35] pillar_1.9.0         crayon_1.5.2
## [37] limma_3.56.2         DelayedArray_0.26.3
## [39] cachem_1.0.8         locfit_1.5-9.8
## [41] tidyselect_1.2.0     digest_0.6.33
## [43] stringi_1.7.12       reshape2_1.4.4
## [45] purrr_1.0.1          fastmap_1.1.1
## [47] grid_4.3.1           colorspace_2.1-0
## [49] cli_3.6.1            magrittr_2.0.3
## [51] S4Arrays_1.0.4       XML_3.99-0.14
## [53] utf8_1.2.3           GSEABase_1.62.0
## [55] edgeR_3.42.4         withr_2.5.0
## [57] scales_1.2.1         bit64_4.0.5
## [59] rmarkdown_2.23       XVector_0.40.0
## [61] httr_1.4.6           matrixStats_1.0.0
## [63] bit_4.0.5            cellranger_1.1.0
## [65] png_0.1-8            memoise_2.0.1
## [67] evaluate_0.21        knitr_1.43
## [69] GenomicRanges_1.52.0 IRanges_2.34.0
## [71] rlang_1.1.1          Rcpp_1.0.11
## [73] xtable_1.8-4         glue_1.6.2
## [75] DBI_1.1.3            BiocGenerics_0.46.0
## [77] rstudioapi_0.14      annotate_1.78.0
## [79] plyr_1.8.8           R6_2.5.1
## [81] MatrixGenerics_1.12.2 zlibbioc_1.46.0

```