

Figure-1 A.R

sokole

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```
# This Script Generates Figure 1A
# Script By: Eishani Kumar Sokolowski
```

```
# Empty the environment & suppress warnings
rm(list = ls())
options(warn=-1)
```

```
# Loading the required libraries
library(tidyverse)
```

```
## — Attaching core tidyverse packages — tidyverse 2.0.0 —
## ✓ dplyr      1.1.4      ✓ readr      2.1.5
## ✓ forcats    1.0.0      ✓ stringr    1.5.1
## ✓ ggplot2     3.5.1      ✓ tibble     3.2.1
## ✓ lubridate  1.9.3      ✓ tidyr      1.3.1
## ✓ purrr      1.0.2
## — Conflicts — tidyverse_conflicts() —
## ✖ dplyr::filter() masks stats::filter()
## ✖ dplyr::lag()     masks stats::lag()
## i Use the conflicted package (<http://conflicted.r-lib.org/>) to force all conflicts to become errors
```

```
library(pheatmap)
library(Vennerable)
library(gplots)
```

```
##
## Attaching package: 'gplots'
##
## The following object is masked from 'package:stats':
##
##     lowess
```

```

# Load metadata
metadata <- read.csv("./Metadata_Heatmap.csv")
rownames(metadata) <- metadata$Islet_Number

# Load normalized expression matrices
my.data.1 <- read.csv("./normalized.with.ERCC.counts.for.heatmap.ER.Stress.csv")
my.data.2 <- read.csv("./normalized.with.ERCC.counts.for.heatmap.Inflammation.csv")
my.data <- merge(my.data.1,my.data.2, by="X")
rownames(my.data) <- my.data[,1]
my.data <- my.data[-c(1)]

# Load DEGs
ERS.specific <- read.csv("./Upregulated_only_in_ER_Stress_DEGs.csv")
ERS.specific <- ERS.specific[-c(2:7)]

INF.specific <- read.csv("./Upregulated_only_in_Inflammation_DEGs.csv")
INF.specific <- INF.specific[-c(2:7)]

Shared <- read.csv("./Upregulated_common_between_ER_Stress_Inflammation_DEGs.csv")
Shared <- Shared[-c(2:7)]

# Making annotation dataframes
metadata.annotation <- metadata
metadata.annotation <- metadata.annotation[order(metadata.annotation$Treatment),]
metadata.annotation <- metadata.annotation[-c(1:7)]
metadata.annotation.names <- rownames(metadata.annotation)
matrix.annotation <- rbind(ERS.specific,INF.specific,Shared)
rownames(matrix.annotation) <- metadata.annotation$X
matrix.annotation <- matrix.annotation[-c(1)]

# Making the heatmap matrix
genes.list <- rownames(matrix.annotation)
matrix <- my.data[(genes.list),]
matrix.ordered <- matrix %>% select(metadata.annotation.names)
matrix.ordered <- as.matrix.data.frame(matrix.ordered)
matrix.ordered <- na.omit(matrix.ordered)
matrix.ordered[is.na(matrix.ordered)] <- 0

# Specify colors
ann_colors = list(
  Treatment = c(A_DMS0="gray", B_Thapsigargin="forestgreen",C_Untreated="grey50",D_Cytokines="darkorange"),
  Regulation = c(Upregulated_ERS_specific = "forestgreen",
                  Upregulated_INF_specific="darkorange",
                  Upregulated_ERS_and_INF_Shared = "coral4")
)

# Make heatmap
pheatmap(matrix.ordered,
          scale = "row",
          clustering_distance_rows = "correlation",
          treeheight_row = 0,

```

```
treeheight_col = 0,  
annotation_col = metadata.annotation,  
cluster_row = F,  
cluster_cols = F,  
fontsize_row = 0.01,  
fontsize_col = 0.01,  
annotation_colors = ann_colors,  
color=bluered(1000))
```

