

Figure-S2F.R

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```
# This Script Generates Figure S2F
# Script By: Eishani Kumar Sokolowski

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

# Loading libraries
library(dplyr)
```

```
##
## Attaching package: 'dplyr'
```

```
## The following objects are masked from 'package:stats':
##
##   filter, lag
```

```
## The following objects are masked from 'package:base':
##
##   intersect, setdiff, setequal, union
```

```
library(ggplot2)
library(cowplot)
library(ggpubr)
```

```
##
## Attaching package: 'ggpubr'
```

```
## The following object is masked from 'package:cowplot':
##
##   get_legend
```

```
library(tidyverse)
```

```
## — Attaching core tidyverse packages — tidyverse 2.0.0 —
## ✓ forcats   1.0.0   ✓ stringr   1.5.1
## ✓ lubridate 1.9.3   ✓ tibble    3.2.1
## ✓ purrr     1.0.2   ✓ tidyr     1.3.1
## ✓ readr     2.1.5
```

```
## — Conflicts ————— tidyverse_conflicts() —
## * dplyr::filter()    masks stats::filter()
## * dplyr::lag()       masks stats::lag()
## * lubridate::stamp() masks cowplot::stamp()
## i Use the conflicted package (<http://conflicted.r-lib.org/>) to force all conflicts
to become errors
```

```
library(plyr)
```

```
## -----
## You have loaded plyr after dplyr – this is likely to cause problems.
## If you need functions from both plyr and dplyr, please load plyr first, then dplyr:
## library(plyr); library(dplyr)
## -----
##
## Attaching package: 'plyr'
##
## The following object is masked from 'package:purrr':
##
##   compact
##
## The following object is masked from 'package:ggpubr':
##
##   mutate
##
## The following objects are masked from 'package:dplyr':
##
##   arrange, count, desc, failwith, id, mutate, rename, summarise,
##   summarize
```

```
library(scales)
```

```
##
## Attaching package: 'scales'
##
## The following object is masked from 'package:purrr':
##
##   discard
##
## The following object is masked from 'package:readr':
##
##   col_factor
```

```
library(reshape2)
```

```
##  
## Attaching package: 'reshape2'  
##  
## The following object is masked from 'package:tidyr':  
##  
##      smiths
```

```
#####
# Loading all the files
#####

# FC values
alpha.ERS <- read.csv("./alpha.all.genes_FDR_ERS_NO_MIN_PERCENT_FOR_BULK_DEGs_HEATMAP.csv")
alpha.INF <- read.csv("./alpha.all.genes_FDR_INF_NO_MIN_PERCENT_FOR_BULK_DEGs_HEATMAP.csv")
beta.ERS <- read.csv("./beta.all.genes_FDR_ERS_NO_MIN_PERCENT_FOR_BULK_DEGs_HEATMAP.csv")
beta.INF <- read.csv("./beta.all.genes_FDR_INF_NO_MIN_PERCENT_FOR_BULK_DEGs_HEATMAP.csv")

# Bulk DEGs
ERS.specific <- read.csv("./Common_Between_scrnaseq_and_bulk_ERS-specific_Downregulated_DEGs.csv")
INF.specific <- read.csv("./Common_Between_scrnaseq_and_bulk_INF-specific_Downregulated_DEGs.csv")
Shared <- read.csv("./Common_Between_scrnaseq_and_bulk_Shared_Downregulated_DEGs.csv")

#####
# Making a list of DEGs
#####

# Making a list of DEGs
DEGs <- rbind(Shared, INF.specific, ERS.specific)
DEGs <- DEGs[-c(1)]
DEGs <- unique(DEGs)
DEGs <- DEGs$Vstem.IntersectionSets...11...

#####
# Extracting FCs
#####

# Alpha - ERS
alpha.ERS <- alpha.ERS[-c(2,4:7)]
colnames(alpha.ERS) <- c("Genes", "Alpha_ERS_FC")

# Alpha - INF
alpha.INF <- alpha.INF[-c(2,4:7)]
colnames(alpha.INF) <- c("Genes", "Alpha_INF_FC")

# Beta - ERS
beta.ERS <- beta.ERS[-c(2,4:7)]
colnames(beta.ERS) <- c("Genes", "Beta_ERS_FC")

# Beta - INF
beta.INF <- beta.INF[-c(2,4:7)]
colnames(beta.INF) <- c("Genes", "Beta_INF_FC")

#####
```

```
# Combining FC dataframe
#####

# Making a complete FC dataframe
FC <- merge(alpha.ERS, alpha.INF)
FC <- merge(FC, beta.ERS)
FC <- merge(FC, beta.INF)

#####
# Finalizing Dataframe
#####

# Extracting DEGs
Bulk.DEGs.FC.Cell.Type <- FC[FC$Genes %in% DEGs,]
rownames(Bulk.DEGs.FC.Cell.Type) <- Bulk.DEGs.FC.Cell.Type$Genes

# Reshaping the dataframe
Bulk.DEGs.FC.Cell.Type <- melt(Bulk.DEGs.FC.Cell.Type)
```

```
## Using Genes as id variables
```

```

colnames(Bulk.DEGs.FC.Cell.Type) <- c("Genes", "Type", "LFC")

# Unlogging the LFC values
Bulk.DEGs.FC.Cell.Type$FC <- 2^(Bulk.DEGs.FC.Cell.Type$LFC)

# Scaling the fold change
Bulk.DEGs.FC.Cell.Type <- ddply(Bulk.DEGs.FC.Cell.Type, .(Genes), transform, Scaled_FC =
rescale(FC))

#####
# Plotting Heatmap
#####

# Making factors for Type
Bulk.DEGs.FC.Cell.Type$Type <- factor(Bulk.DEGs.FC.Cell.Type$Type, levels=c("Alpha_ERS_F
C",
                                     "Beta_ERS_F
C",
                                     "Alpha_INF_F
C",
                                     "Beta_INF_F
C"))

# Making factors for DEGs
Bulk.DEGs.FC.Cell.Type$Genes <- factor(Bulk.DEGs.FC.Cell.Type$Genes, levels=c(DEGs))

# Plotting
p <- ggplot(Bulk.DEGs.FC.Cell.Type, aes(Genes, Type, fill=Scaled_FC)) +
  geom_tile(color = "black", lwd = 0) + coord_flip() + theme_classic() +
  theme(axis.text.y = element_text(size=0)) +
  scale_fill_gradient2(low = "blue", mid = "white", high = "grey90", midpoint = 0.50, br
eaks = c(0,1)) +
  theme(axis.text.x = element_text(angle = 90, vjust = 0.60, hjust=1))

# View
ggarrange(p, nrow=1, ncol=2)

```

