setwd( )

if (!require("BiocManager", quietly = TRUE))

install.packages("BiocManager")

BiocManager::install("ComplexHeatmap")

library(ComplexHeatmap)

library(tidyverse)

library(RColorBrewer)

library(circlize)

library(ggfortify)

library(ggplot2)

donors <- c("Donor1", "Donor2", "Donor3", "Donor5", "Donor6","Donor7")

donor\_colors <- setNames(RColorBrewer::brewer.pal(6, "Set2"), donors)

##Making Heatmap

#RNASeq Data= flupanel

#loading in data

flupanel <- read.csv('RNASeqPanelDEG.csv', header = TRUE, sep = ",")

flu\_meta <- read.csv('Meta\_data.csv',stringsAsFactors=FALSE,header=TRUE)

head(flupanel)

head(flu\_meta)

#col\_fun = colorRamp2(c(-6, 0, 6), c("blue", "white", "red"))

#col\_fun(seq(-5, 0, 5))

#making data matrix, removing column of SampleIDs and making them row names

flu\_df = subset(flupanel, select = -c(SampleID))

flu\_matrix <- as.matrix(flu\_df)

rownames(flu\_matrix) <- flupanel$SampleID

flu\_matrix

ComplexHeatmap::Heatmap(flu\_matrix) #Couldnt get any heatmaps to work-automatically generated colors etc

#Heatmap(flu\_matrix, name = "Fluidigm", col = cm.colors(256))

flu\_scaled <- scale(flu\_matrix) #Try to hashtag out, want to see if can do without scaling

Heatmap(flu\_scaled)

#transposing heatmap so cytokines are rows

flu\_transposed <- t(flu\_scaled)

head(flu\_transposed)

Heatmap(flu\_transposed)

Heatmap(flu\_transposed,

#cluster\_columns = FALSE,

column\_title = "RNA-Seq DEGs",

column\_title\_gp = gpar(fontsize = 20, fontface = "bold"),

#heatmap\_legend\_param = list(title = "Z-score"),

heatmap\_legend\_param = list(

title = "Z-score", at = c(-4, 0, 4),

labels = c("-4", "0", "4")),

column\_names\_gp = gpar(fontsize = 9),

top\_annotation = HeatmapAnnotation(

Condition = flu\_meta$Condition,

#Donor = pro\_meta$Donor,

Material = flu\_meta$Material,

Burn\_Type = flu\_meta$Burn\_Type,

Sex = flu\_meta$Sex,

col = list(Condition = c("Cntrl\_Veh"="green",

"SM\_20ug" = "paleturquoise4",

"FL\_20ug" = "sienna3"),

Donor = donor\_colors ,

Material = c( "Plastic" = "darkslategrey","PBS" = "deepskyblue3"),

Burn\_Type = c("Flaming" = "firebrick4", "Smoldering" = "chocolate2","None" = "deepskyblue2"),

Sex = c("Female" = "lightpink", "Male" = "lightblue")),

annotation\_legend\_param = list(Condition = list(labels = c("Cntrl\_Veh","SM\_20ug","FL\_20ug"),

at = c("Cntrl\_Veh","SM\_20ug","FL\_20ug")))))