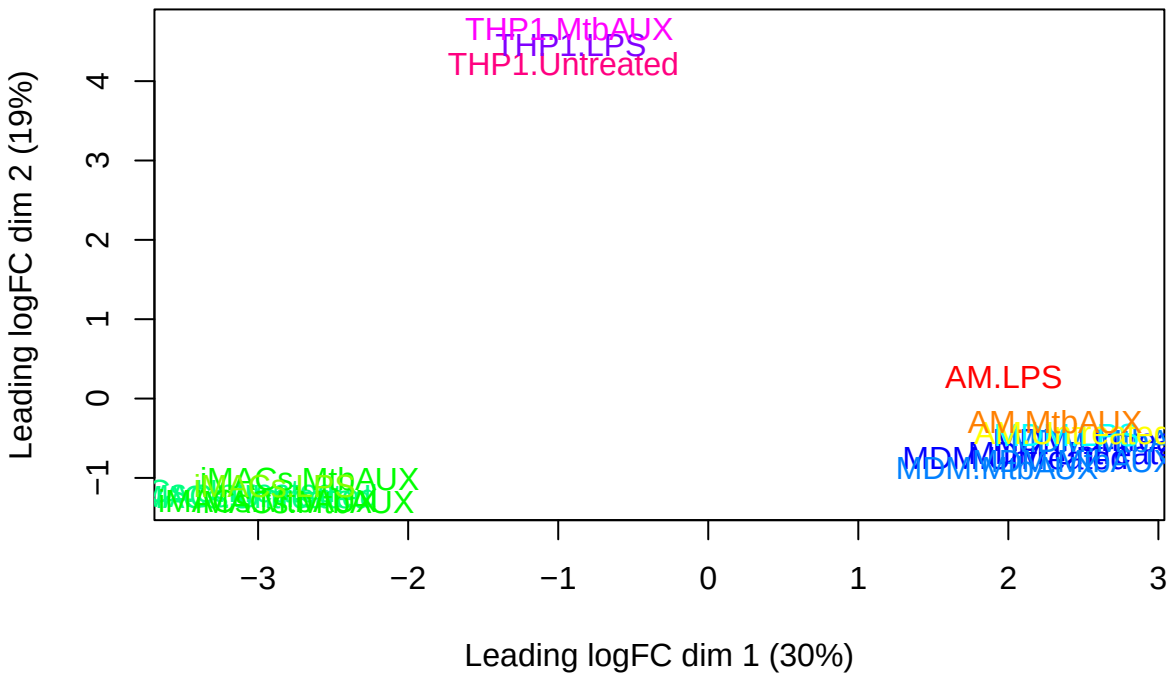
deg_analysis

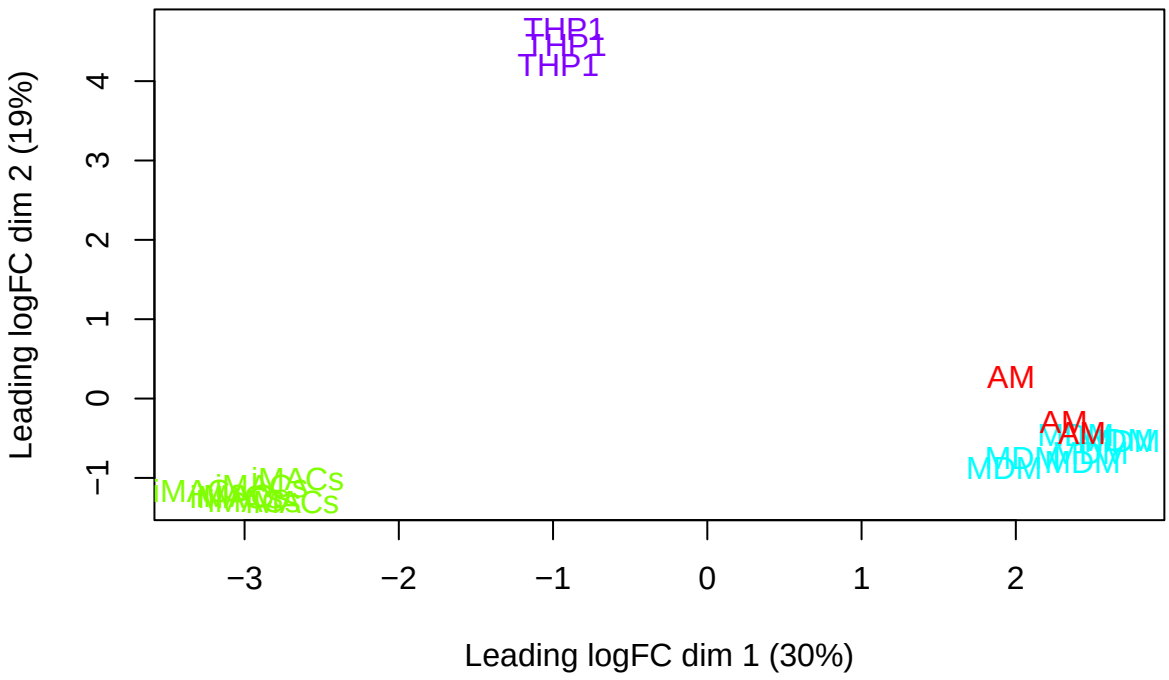
```
##
## Attaching package: 'dplyr'

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

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

## -- Attaching packages --------------------------------------- tidyverse 1.3.1 --

## v ggplot2 3.3.5     v purrr   0.3.4
## v tibble  3.1.6     v stringr 1.4.0
## v tidyr   1.2.0     v forcats 0.5.1
## v readr   2.1.2

## -- Conflicts ------------------------------------------ tidyverse_conflicts() --
## x dplyr::filter() masks stats::filter()
## x dplyr::lag()    masks stats::lag()

## Loading required package: viridisLite

##
## Attaching package: 'magrittr'

## The following object is masked from 'package:purrr':
##
##     set_names

## The following object is masked from 'package:tidyr':
##
##     extract

##   |                                                                              |
```

```
cpm <- cpm(dge_object2)
lcpm <- cpm(dge_object2, log = TRUE, prior.count =1)
```

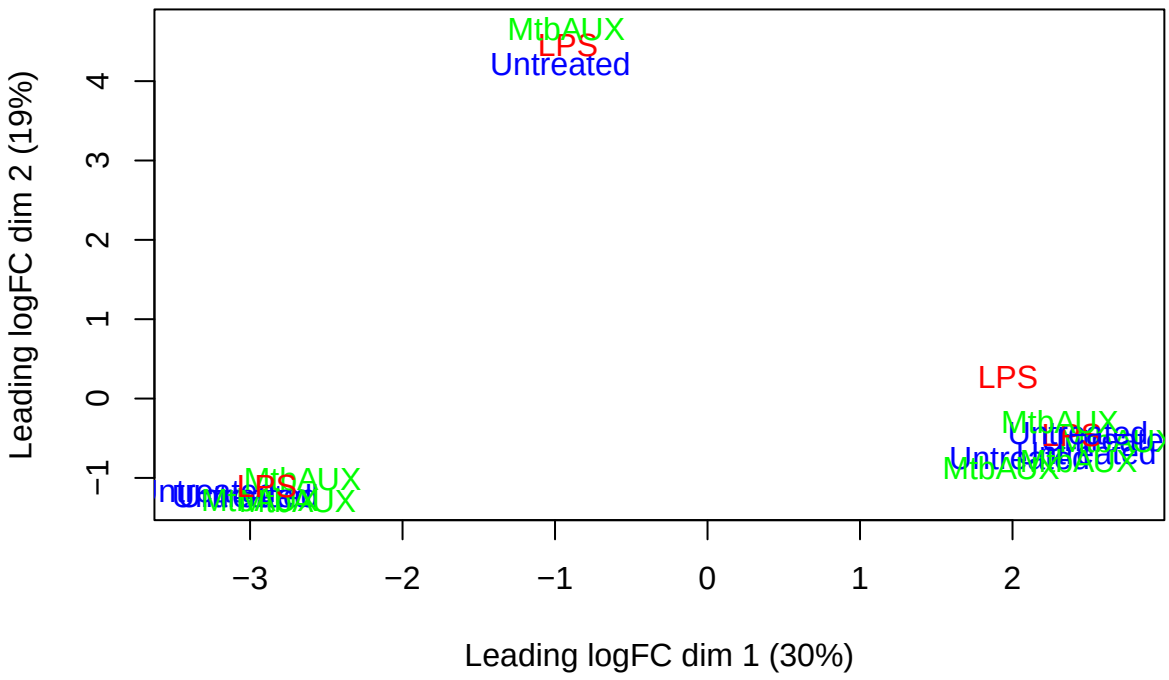
```
col.group <- group
levels(col.group) <- rainbow(12)
col.group <- as.character(col.group)
plotMDS(lcpm, labels = group, col = col.group)
```




```
col.cell <- factor(sample.info$Sample_Group)
levels(col.cell) <- rainbow(4)
col.cell <- as.character(col.cell)
plotMDS(lcpm,
        labels = factor(sample.info$Sample_Group),
        col = col.cell)
```




```
col.treatment <- factor(sample.info$Treatment)
levels(col.treatment) <- rainbow(3)
col.treatment <- as.character(col.treatment)
plotMDS(lcpm,
        labels = factor(sample.info$Treatment),
        col = col.treatment)
```




```
fit12 <- lmFit(lcpm, mm)
```

```
contr.matrix2 <- makeContrasts(
   # Within cell mtb vs untreated
   AM.MtbAUXvsAM.Untreated = AM.MtbAUX - AM.Untreated,
   iMACs.MtbAUXvsiMACs.Untreated = iMACs.MtbAUX - iMACs.Untreated,
   MDM.MtbAUXvsMDM.Untreated = MDM.MtbAUX - MDM.Untreated,
   THP1.MtbAUXvsTHP1.Untreated = THP1.MtbAUX - THP1.Untreated,
   # Within cell LPS vs untreated
   AM.LPSvsAM.Untreated = AM.LPS - AM.Untreated,
   iMACs.LPSvsiMACs.Untreated = iMACs.LPS - iMACs.Untreated,
   MDM.LPSvsMDM.Untreated = MDM.LPS - MDM.Untreated,
   THP1.LPSvsTHP1.Untreated = THP1.LPS - THP1.Untreated,

   #between cell MTB
   # iMACs.MtbAUXvsAM.MtbAUX = iMACs.MtbAUX - AM.MtbAUX,
   # MDM.MtbAUXvsAM.MtbAUX = MDM.MtbAUX - AM.MtbAUX,
   # THP1.MtbAUXvsAM.MtbAUX = THP1.MtbAUX - AM.MtbAUX,

   levels = colnames(mm)
)

contr.matrix2
```

```
##                   Contrasts
```

```
## Levels            AM.MtbAUXvsAM.Untreated iMACs.MtbAUXvsiMACs.Untreated
##   AM.LPS                                0                               0
##   AM.MtbAUX                             1                               0
##   AM.Untreated                         -1                               0
##   iMACs.LPS                             0                               0
##   iMACs.MtbAUX                          0                               1
##   iMACs.Untreated                       0                              -1
##   MDM.LPS                               0                               0
##   MDM.MtbAUX                            0                               0
##   MDM.Untreated                         0                               0
##   THP1.LPS                              0                               0
##   THP1.MtbAUX                           0                               0
##   THP1.Untreated                        0                               0
##                 Contrasts
## Levels            MDM.MtbAUXvsMDM.Untreated THP1.MtbAUXvsTHP1.Untreated
##   AM.LPS                                0                               0
##   AM.MtbAUX                             0                               0
##   AM.Untreated                          0                               0
##   iMACs.LPS                             0                               0
##   iMACs.MtbAUX                          0                               0
##   iMACs.Untreated                       0                               0
##   MDM.LPS                               0                               0
##   MDM.MtbAUX                            1                               0
##   MDM.Untreated                        -1                               0
##   THP1.LPS                              0                               0
##   THP1.MtbAUX                           0                               1
##   THP1.Untreated                        0                              -1
##                 Contrasts
## Levels            AM.LPSvsAM.Untreated iMACs.LPSvsiMACs.Untreated
##   AM.LPS                             1                          0
##   AM.MtbAUX                          0                          0
##   AM.Untreated                      -1                          0
##   iMACs.LPS                          0                          1
##   iMACs.MtbAUX                       0                          0
##   iMACs.Untreated                    0                         -1
##   MDM.LPS                            0                          0
##   MDM.MtbAUX                         0                          0
##   MDM.Untreated                      0                          0
##   THP1.LPS                           0                          0
##   THP1.MtbAUX                        0                          0
##   THP1.Untreated                     0                          0
##                 Contrasts
## Levels            MDM.LPSvsMDM.Untreated THP1.LPSvsTHP1.Untreated
##   AM.LPS                             0                          0
##   AM.MtbAUX                          0                          0
##   AM.Untreated                       0                          0
##   iMACs.LPS                          0                          0
##   iMACs.MtbAUX                       0                          0
##   iMACs.Untreated                    0                          0
##   MDM.LPS                            1                          0
##   MDM.MtbAUX                         0                          0
##   MDM.Untreated                     -1                          0
##   THP1.LPS                           0                          1
##   THP1.MtbAUX                        0                          0
```

```
##   THP1.Untreated                        0                         -1
```

```
tmp12 <- contrasts.fit(fit12, contr.matrix2)

efit12 <- eBayes(tmp12, trend = TRUE)

#topTable(efit12, coef = ncol(mm))

#summary(decideTests(efit12))


topTreat(efit12, coef = 1)
```

```
##                     logFC   AveExpr         t      P.Value    adj.P.Val
## ENSG00000236060  5.469944 -3.774130  27.45398 1.187031e-10 2.150543e-06
## ENSG00000231259  5.053420 -3.794956  25.50820 2.430096e-10 2.201303e-06
## ENSG00000288623  4.291892 -3.575266  20.40449 2.119995e-09 1.280265e-05
## ENSG00000288053 -3.765416 -3.677376 -18.40616 5.726089e-09 2.593489e-05
## ENSG00000185304  4.505500 -2.617705  16.70025 1.455325e-08 4.038974e-05
## ENSG00000273299 -3.411485 -3.678514 -16.68125 1.471260e-08 4.038974e-05
## ENSG00000288645  3.351039 -3.721440  16.57874 1.560568e-08 4.038974e-05
## ENSG00000258483  2.868345 -3.708451  14.14046 7.079333e-08 1.603203e-04
## ENSG00000251357  2.843478 -3.623293  13.69619 9.566972e-08 1.925832e-04
## ENSG00000258465  6.167526 -2.387441  12.09650 3.060450e-07 5.544617e-04
##                         B
## ENSG00000236060 7.331665
## ENSG00000231259 7.217670
## ENSG00000288623 6.776514
## ENSG00000288053 6.517105
## ENSG00000185304 6.235801
## ENSG00000273299 6.232287
## ENSG00000288645 6.213180
## ENSG00000258483 5.666201
## ENSG00000251357 5.543705
## ENSG00000258465 5.026263
```

```
# Function to create data frames with all results using the contrast matrix for naming
result_dfs <- function(con_mat) {
   contrast_names <- colnames(con_mat)
   results_list <- list()
   for (i in seq(from = 1, to = length(contrast_names))) {
      name <- print(contrast_names[i], quote = FALSE)

      top <- topTreat(efit12, coef = i, n = Inf)
      top <- gene.info %>% inner_join(rownames_to_column(top, "Gene_ID"), by ="Gene_ID")
      top <- data.frame(top)
      results_list[[contrast_names[i]]] <- assign(name, top)
      rownames(results_list[[contrast_names[i]]])<- top$Gene_ID
   }
   results_list
}

results<- result_dfs(contr.matrix2)
```

```
## [1] AM.MtbAUXvsAM.Untreated
## [1] iMACs.MtbAUXvsiMACs.Untreated
## [1] MDM.MtbAUXvsMDM.Untreated
## [1] THP1.MtbAUXvsTHP1.Untreated
## [1] AM.LPSvsAM.Untreated
## [1] iMACs.LPSvsiMACs.Untreated
## [1] MDM.LPSvsMDM.Untreated
## [1] THP1.LPSvsTHP1.Untreated
```

```
saveRDS(results, file = "results_list.RData")
```