

Tuberculosis Bulk RNA-seq Analysis

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1 Downloading and Formatting the Data

(download data from GSE, establish needed local directories, format data objects)

(describe following code)

```
library(tidyverse) # dplyr, tidyr, tibble, readr
library(readxl)
library(BioBase)
library(GEOquery)

dir.create(file.path("data_raw"), showWarnings = FALSE) # establish directory to raw data downloads

supplemental_dir <- file.path("data_raw", "GSE107994") # establish path to specific study directory
```

(describe following code)

```
if (!dir.exists(supplemental_dir) || length(list.files(supplemental_dir)) == 0) { # checks for supplemental files
  getGEOSuppFiles("GSE107994", baseDir = "data_raw") # accesses GEO for supplemental files (gene counts)
}

counts_file <- list.files( # grabs the specific file name for the raw gene counts data
  path = supplemental_dir,
  pattern = "Raw_counts.*Leicester.*\\.xlsx$",
  full.names = TRUE
)

counts <- readxl::read_xlsx(counts_file) # function to import .xlsx file as data table
```

(describe following code)

```
gse <- getGEO("GSE107994", GSEMatrix = TRUE) # grabs the series matrix from GEO  
phenotypes <- Biobase::pData(gse[[1]]) |> as_tibble() # formats GSE download as tabular sample metadata
```

(describe following code)

```
dir.create("data_processed", showWarnings = FALSE) # create data_processed directory folder if it does not exist  
readr::write_tsv(phenotypes, file.path("data_processed", "GSE107994_phenotypes.tsv")) # save phenotypes  
readr::write_tsv(counts, file.path("data_processed", "GSE107994_counts_raw.tsv")) # save counts locally
```

2 Inspecting the Data

(snapshot of counts and phenotypes tables,)

```
print(counts)
```

```
# A tibble: 58,051 x 178
  Genes    Gene_name Gene_biotype Leicester_with_progr~1 Leicester_with_progr~2
  <chr>    <chr>     <chr>          <dbl>           <dbl>
1 ENSG000~ TSPAN6   protein_cod~        1              16
2 ENSG000~ TNMD     protein_cod~       0              0
3 ENSG000~ DPM1     protein_cod~      215             263
4 ENSG000~ SCYL3    protein_cod~      233             333
5 ENSG000~ C1orf112  protein_cod~      54              57
6 ENSG000~ FGR      protein_cod~     21694            18130
7 ENSG000~ CFH      protein_cod~      46              44
8 ENSG000~ FUCA2    protein_cod~      604             428
9 ENSG000~ GCLC     protein_cod~      69              153
10 ENSG000~ NFYA    protein_cod~     310             420
# i 58,041 more rows
# i abbreviated names: 1: Leicester_with_progressor_longitudinal_Sample1,
#   2: Leicester_with_progressor_longitudinal_Sample2
# i 173 more variables: Leicester_with_progressor_longitudinal_Sample3 <dbl>,
#   Leicester_with_progressor_longitudinal_Sample4 <dbl>,
#   Leicester_with_progressor_longitudinal_Sample5 <dbl>,
#   Leicester_with_progressor_longitudinal_Sample6 <dbl>, ...
```

```
print(phenotypes)
```

```
# A tibble: 175 x 60
  title          geo_accession status submission_date last_update_date type
  <chr>          <chr>       <chr>    <chr>        <chr>        <chr>
1 Leicester_with_p~ GSM2886274  Publi~ Dec 12 2017 May 15 2019  SRA
2 Leicester_with_p~ GSM2886275  Publi~ Dec 12 2017 May 15 2019  SRA
3 Leicester_with_p~ GSM2886276  Publi~ Dec 12 2017 May 15 2019  SRA
4 Leicester_with_p~ GSM2886277  Publi~ Dec 12 2017 May 15 2019  SRA
5 Leicester_with_p~ GSM2886278  Publi~ Dec 12 2017 May 15 2019  SRA
6 Leicester_with_p~ GSM2886279  Publi~ Dec 12 2017 May 15 2019  SRA
7 Leicester_with_p~ GSM2886280  Publi~ Dec 12 2017 May 15 2019  SRA
8 Leicester_with_p~ GSM2886281  Publi~ Dec 12 2017 May 15 2019  SRA
9 Leicester_with_p~ GSM2886282  Publi~ Dec 12 2017 May 15 2019  SRA
10 Leicester_with_p~ GSM2886283  Publi~ Dec 12 2017 May 15 2019 SRA
# i 165 more rows
# i 54 more variables: channel_count <chr>, source_name_ch1 <chr>,
#   organism_ch1 <chr>, characteristics_ch1 <chr>, characteristics_ch1.1 <chr>,
#   characteristics_ch1.2 <chr>, characteristics_ch1.3 <chr>,
#   characteristics_ch1.4 <chr>, characteristics_ch1.5 <chr>,
#   characteristics_ch1.6 <chr>, characteristics_ch1.7 <chr>,
#   characteristics_ch1.8 <chr>, characteristics_ch1.9 <chr>, ...
```

3 Data Cleanup

(standardize raw gene counts, reformat to cases x genes)