MiA Case Study 2

Data Analysis in R

Image of grazer Oxyrrhis marina and phytoplankton Dunaliella tertiolecta in culture

Load packages and data

```
library(readx1)
library(ggplot2)
library(dplyr)

data <- read.csv("CaseStudy2_ROI_Data.csv")</pre>
```

Prepare the data for plotting and analysis

- Normalize by exposure times
- Convert pixels to micrometers for area and lengths
 - Set conversion factor (µm/pixel) for 20X
 - Remember to square for area since each pixel will equal length times width of the value.

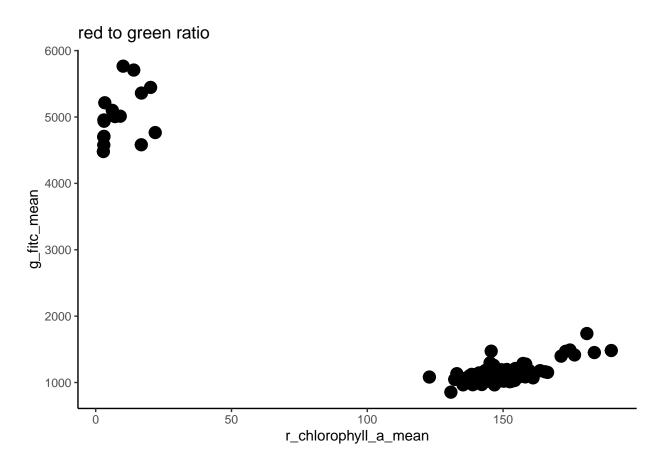
```
data$g_fitc_mean_exp <- data$g_fitc_mean/data$g_fitc_exp
data$b_dapi_mean <- data$b_dapi_mean/data$b_dapi_exp
data$r_chlorophyll_a_mean <- data$r_chlorophyll_a_mean/data$r_chlorophyll_a_exp

X20 = 0.211
data$area <- data$area*(X20^2)
data$major_length <- data$major_length*(X20)
data$minor_length <- data$minor_length*(X20)</pre>
```

Separate cell populations by fluorescence signal

• Create a scatter plot of red to green fluorescent signal ratio

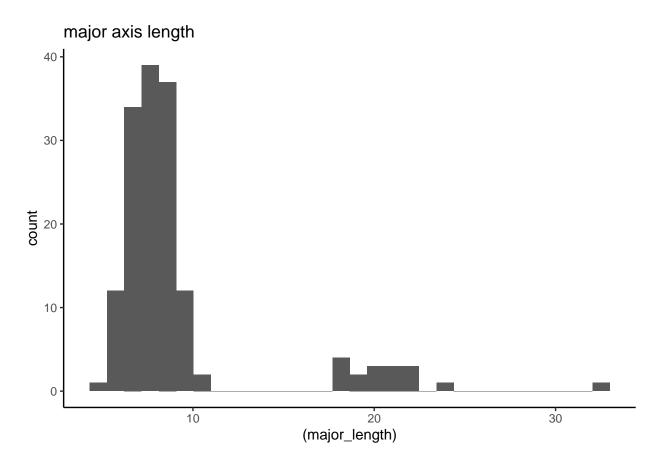
```
data %>%
   ggplot(aes(x = r_chlorophyll_a_mean,y = g_fitc_mean)) +
    geom_point(size =4) +
    theme_classic() +
     labs (title = "red to green ratio")
```



Separate cell populations by size

• Create a histogram of the major axis length

```
data %>%
    ggplot(aes(x = (major_length) )) +
    geom_histogram() +
    theme_classic() +
    labs (title = "major axis length")
```



Quantify cells in each size class using a cut-off value

- $\bullet\,$ Based on the histogram above, use cutoff value of 15 for major length
- Assign labels based on that value
- Count the number of each cell type