C14

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Load data

```
C14 <- readxl::read excel("TAN1810 Adriana samples complete final AGR 1.0.xlsx", sheet = 2)
C14$EXP <- as.character(C14$EXP)
C14
## # A tibble: 460 x 50
      Operator date julian_day Cycle `CTD#`
                                                STN 'Incub Time' EXP
                                                                        SAMPLE
##
      <chr>
               <dbl>
                          <dbl> <dbl> <chr>
                                              <dbl> <chr>
                                                                  <chr> <chr>
   1 Adriana 43398
                                                                        SUR
##
                            298
                                     1 U9103
                                                 15 T24
                                                                  1
   2 Adriana
               43398
                            298
                                     1 U9103
                                                 15 T24
                                                                        SUR
## 3 Adriana 43398
                            298
                                     1 U9103
                                                 15 T24
                                                                  1
                                                                        SUR
## 4 Adriana 43398
                            298
                                     1 U9103
                                                 15 T24
                                                                  1
                                                                        SUR
## 5 Adriana 43398
                            298
                                     1 U9103
                                                 15 T24
                                                                  1
                                                                        SUR
   6 Adriana 43398
                            298
                                     1 U9103
                                                 15 T24
                                                                  1
                                                                        SUR
##
  7 Adriana 43398
                            298
                                                                  1
                                                                        SUR
                                     1 U9103
                                                 15 T24
   8 Adriana 43398
                            298
                                     1 U9103
                                                 15 T24
                                                                        SUR
## 9 Adriana 43398
                            298
                                     1 U9103
                                                 15 T24
                                                                  1
                                                                        SUR
## 10 Adriana 43398
                            298
                                     1 U9103
                                                 15 T24
## # ... with 450 more rows, and 41 more variables: `Biosecurity code` <chr>,
       Lat_DD <dbl>, Long_DD <dbl>, `Karl Code` <dbl>, DEPTH <dbl>, `CTD Z
       code` <dbl>, `Vial code` <chr>, `Type (D for DNA and S for
## #
## #
       scintilation) \(`<chr>, \(`BOX\) NUMBER...18\(`<dbl>, \(`Sorting\) number\(`<dbl>,
## #
       `sorting population` <chr>, `Cells sorted` <dbl>, `Sample name` <chr>,
## #
       ...23 <dbl>, ...24 <chr>, ...25 <dbl>, `S#` <dbl>, `Count Time` <dbl>,
## #
       CPMA <dbl>, DPM1 <dbl>, SIS <dbl>, tSIE <dbl>, `A:2S%` <dbl>,
## #
       MESSAGES <1gl>, `BOX NUMBER...34` <dbl>, Tray <dbl>, Vial <dbl>, DIC <dbl>,
## #
       `SA (uCi/mL)` <dbl>, `NPP (mgC d-1; need to double check the units, seems a
## #
       bit too large for me) ` <dbl>, `SA in DPM (aproximation) ` <dbl>,
## #
       ...41 <lgl>, ...42 <lgl>, ...43 <lgl>, ...44 <lgl>, ...45 <lgl>,
       ...46 <chr>, ...47 <chr>, ...48 <chr>, ...49 <dbl>, ...50 <chr>
```

Load libraries, and narrow down the columns for the dataset.

```
library(dplyr)
library(tidyverse)
library(ggplot2)
# create a new dataset
C14_1 <- select(C14, julian_day, Cycle, STN, EXP, SAMPLE, Lat_DD, Long_DD, DEPTH, 'Vial code', 'sorting C14_1</pre>
```

```
## # A tibble: 460 x 12
##
                          STN EXP
                                     SAMPLE Lat_DD Long_DD DEPTH `Vial code`
      julian_day Cycle
##
           <dbl> <dbl> <dbl> <chr> <chr>
                                              <dbl>
                                                      <dbl> <dbl> <chr>
                                     SUR
                                              -44.6
                                                       175.
                                                                12 A
##
   1
             298
                      1
                            15 1
##
    2
             298
                      1
                            15 1
                                     SUR
                                              -44.6
                                                       175.
                                                                12 A
##
    3
             298
                           15 1
                                     SUR
                                              -44.6
                                                       175.
                                                                12 A
                      1
##
    4
             298
                                     SUR
                                              -44.6
                      1
                           15 1
                                                       175.
                                                                12 A
             298
                                     SUR
                                              -44.6
                                                                12 A
##
    5
                      1
                            15 1
                                                       175.
##
    6
             298
                      1
                           15 1
                                     SUR
                                              -44.6
                                                       175.
                                                                12 A
##
   7
                                     SUR
             298
                      1
                           15 1
                                              -44.6
                                                       175.
                                                                12 A
##
    8
             298
                      1
                           15 1
                                     SUR
                                              -44.6
                                                       175.
                                                                12 B
                                     SUR
                                                       175.
                                                                12 B
##
    9
             298
                            15 1
                                              -44.6
                      1
## 10
             298
                      1
                            15 1
                                     SUR.
                                              -44.6
                                                       175.
                                                                12 B
## # ... with 450 more rows, and 3 more variables: `sorting population` <chr>,
       `Cells sorted` <dbl>, DPM1 <dbl>
To include only essential columns. Can edit code to include other columns that are essential later.
# Tidy for essential columns
C14_DPM <- C14_1 %>%
  select ( EXP, SAMPLE, 'Vial code', 'sorting population', 'Cells sorted', DPM1)
C14_DPM
## # A tibble: 460 x 6
##
      EXP
            SAMPLE `Vial code` `sorting population` `Cells sorted`
                                                                        DPM1
##
      <chr> <chr> <chr>
                                 <chr>
                                                                 <dbl> <dbl>
    1 1
            SUR
                                                                  2000
##
                    Α
                                 Pico
                                                                           62
##
    2 1
            SUR
                    Α
                                 Pico
                                                                  4000
                                                                           93
##
   3 1
            SUR
                                 Pico
                                                                 10000
                                                                          179
                    Α
##
   4 1
            SUR
                                                                  2000
                                                                          75
                    Α
                                 Syn
            SUR
##
  5 1
                    Α
                                 Syn
                                                                  4000
                                                                          48
                                                                 10000
##
    6 1
            SUR
                    Α
                                 Syn
                                                                          140
   7 1
                                                                  1000
##
            SUR
                                                                          926
                    Α
                                 Nano
##
   8 1
            SUR
                    В
                                 Pico
                                                                  2000
                                                                           61
## 9 1
            SUR
                                                                  4000
                                                                          90
                    В
                                 Pico
                                                                 10000
## 10 1
            SUR
                                 Pico
                                                                          184
## # ... with 450 more rows
```

First, I focused on experiment 1 only to try 2 different methods I had in mind.

```
#filter for exp 1
C14_E1 <- filter(C14_DPM, EXP==1)
C14_E1
## # A tibble: 28 x 6
##
      EXP
            SAMPLE `Vial code` `sorting population` `Cells sorted`
                                                                       DPM1
##
      <chr> <chr> <chr>
                                <chr>
                                                                <dbl> <dbl>
   1 1
##
            SUR
                                                                 2000
                                                                         62
                    Α
                                Pico
   2 1
            SUR
                    Α
                                Pico
                                                                 4000
                                                                         93
##
  3 1
            SUR
                                Pico
                                                                10000
                                                                        179
                    Α
##
   4 1
            SUR
                   Α
                                Syn
                                                                 2000
                                                                         75
## 5 1
            SUR
                    Α
                                Syn
                                                                 4000
                                                                         48
## 6 1
            SUR
                                Syn
                                                                10000
                                                                        140
```

```
7 1
             SUR
                                                                     1000
                                                                            926
                                  Nano
##
    8 1
             SUR
                     В
                                                                     2000
                                                                             61
                                  Pico
   9 1
             SUR
                     В
                                  Pico
                                                                     4000
                                                                             90
## 10 1
             SUR
                     В
                                  Pico
                                                                    10000
                                                                            184
## # ... with 18 more rows
```

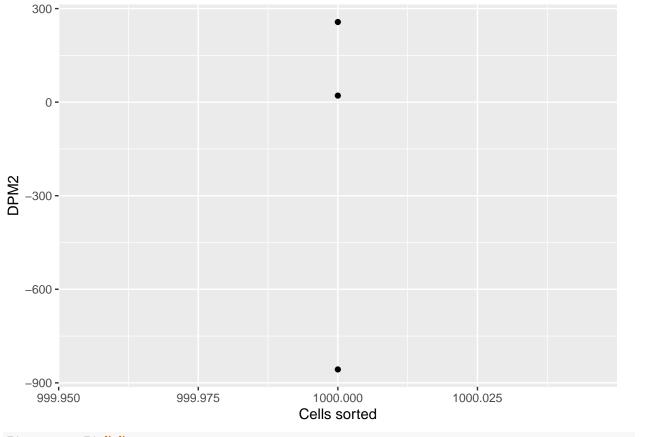
To make calculations easier, I first created a new dataset that would bring vial D to a new column called DPM_dark. I then create another column DPM2 = light - dark. DPM2 would be DPM data that I will use to calculate my PP.

Problem: this method only works for complete dataset, and I would need to repeat for every experiment and depth. How do I set conditions to make sure that DPM_dark moves to its corresponding population? I would like to have a code that will apply to the entire dataset, without needed to subset the data.

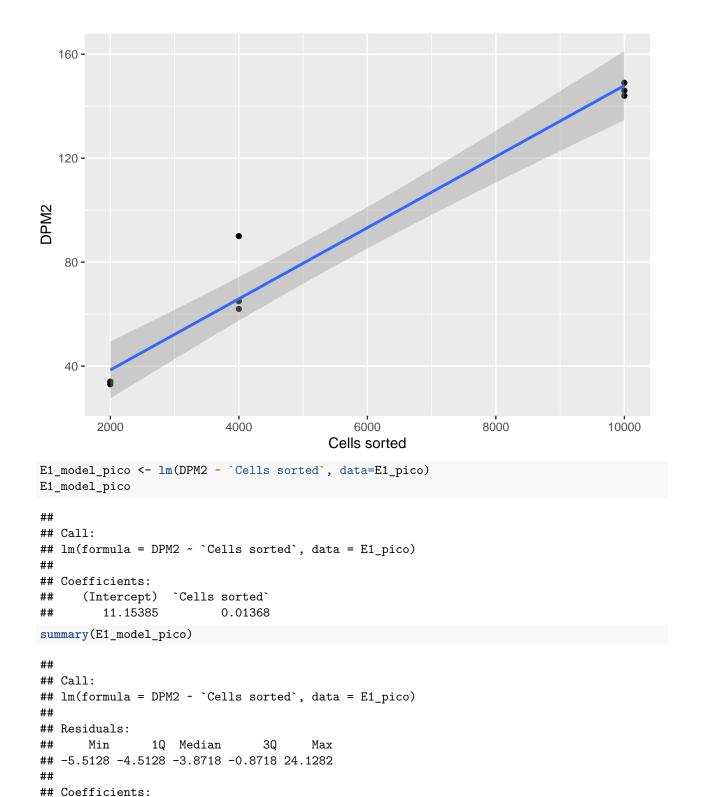
```
# create new column for D, repeating from A to C
# select values for DPM dark
subset1 <- subset(C14_E1, `Vial code` == "D", select = c("DPM1"))</pre>
subset1
## # A tibble: 7 x 1
##
      DPM1
##
     <dbl>
## 1
        28
## 2
        28
## 3
        35
## 4
        26
## 5
        30
## 6
        37
       905
# select for vials A to C
C14_E1 <- filter(C14_E1, `Vial code` != "D")
# create DPM dark column
C14_E1 <- mutate(C14_E1, DPM_dark = rep(subset1$DPM1, 3))
\# DPMD2 = DPM \ light - dark
C14_E1 <- mutate(C14_E1, DPM2 = DPM1 - DPM_dark)
C14_E1
## # A tibble: 21 x 8
##
      EXP
             SAMPLE `Vial code`
                                 `sorting popula~ `Cells sorted`
                                                                     DPM1 DPM_dark DPM2
##
      <chr> <chr> <chr>
                                 <chr>
                                                             <dbl> <dbl>
                                                                              <dbl> <dbl>
             SUR
                                                                                 28
##
    1 1
                    Α
                                 Pico
                                                              2000
                                                                       62
                                                                                       34
                                                                                 28
##
    2 1
             SUR
                    Α
                                 Pico
                                                              4000
                                                                       93
                                                                                       65
##
   3 1
             SUR
                                 Pico
                                                             10000
                                                                      179
                                                                                 35
                                                                                      144
                    Α
                                                                       75
                                                                                 26
                                                                                       49
##
   4 1
             SUR
                    A
                                 Syn
                                                              2000
##
   5 1
             SUR
                    Α
                                 Syn
                                                              4000
                                                                       48
                                                                                 30
                                                                                       18
##
   6 1
             SUR
                                                             10000
                                                                      140
                                                                                 37
                                                                                      103
                    Α
                                 Syn
   7 1
                                                                      926
                                                                                905
                                                                                       21
##
             SUR
                                                              1000
                    Α
                                 Nano
                                                                                 28
##
    8 1
             SUR
                    В
                                 Pico
                                                              2000
                                                                       61
                                                                                       33
## 9 1
             SUR
                    В
                                                              4000
                                                                                 28
                                                                                       62
                                 Pico
                                                                       90
## 10 1
             SUR
                                 Pico
                                                             10000
                                                                      184
                                                                                 35
                                                                                      149
```

IGNORE Method 1: Calculate coefficient when vials A to C combined (I have decided not to use this method as it reduces my number of replicates)

```
E1 <- C14 E1 %>%
  group_by(`sorting population`, `Cells sorted`, `Vial code`) %>%
  summarise(DPM2)
## # A tibble: 21 x 4
## # Groups: sorting population, Cells sorted [7]
      `sorting population` `Cells sorted` `Vial code` DPM2
##
                                   <dbl> <chr>
                                                     <dbl>
## 1 Nano
                                    1000 A
                                                        21
## 2 Nano
                                    1000 B
                                                       257
## 3 Nano
                                    1000 C
                                                      -857
## 4 Pico
                                    2000 A
                                                        34
## 5 Pico
                                    2000 B
                                                        33
## 6 Pico
                                    2000 C
                                                        34
                                    4000 A
## 7 Pico
                                                        65
## 8 Pico
                                    4000 B
                                                        62
## 9 Pico
                                    4000 C
                                                        90
## 10 Pico
                                   10000 A
                                                       144
## # ... with 11 more rows
E1_nano <- E1 %>%
ungroup() %>%
 filter(`sorting population` == "Nano")
E1_plot_nano <- ggplot(E1_nano, aes(x = `Cells sorted`, y = DPM2)) + geom_point()
E1_plot_nano
```



```
E1_pico <- E1 %>%
  ungroup() %>%
  filter(`sorting population` == "Pico")
E1_plot_pico <- ggplot(E1_pico, aes(x = `Cells sorted`, y = DPM2)) + geom_point() + geom_smooth(method = E1_plot_pico)</pre>
```



1.812

Estimate Std. Error t value Pr(>|t|)

1.115e+01 6.155e+00

`Cells sorted` 1.368e-02 9.732e-04 14.056 2.19e-06 ***

Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1

##

##

(Intercept)

```
## Residual standard error: 9.925 on 7 degrees of freedom
## Multiple R-squared: 0.9658, Adjusted R-squared: 0.9609
## F-statistic: 197.6 on 1 and 7 DF, p-value: 2.186e-06
E1_syn <- E1 %>%
  ungroup() %>%
  filter(`sorting population` == "Syn")
E1_plot_syn <- ggplot(E1_syn, aes(x = `Cells sorted`, y = DPM2)) + geom_point() + geom_smooth(method =
E1_plot_syn
  120 -
   90 -
   60 -
   30 -
                                              6000
        2000
                                                                 8000
                           4000
                                                                                    10000
                                          Cells sorted
E1_model_syn <- lm(DPM2 ~ `Cells sorted`, data=E1_syn)</pre>
E1_model_syn
##
## Call:
## lm(formula = DPM2 ~ `Cells sorted`, data = E1_syn)
## Coefficients:
                   `Cells sorted`
##
      (Intercept)
         8.358974
                         0.009141
##
summary(E1_model_syn)
##
## Call:
## lm(formula = DPM2 ~ `Cells sorted`, data = E1_syn)
##
## Residuals:
        Min
                                     ЗQ
##
                  1Q
                      Median
                                             Max
```

```
## -26.9231 -0.9231
                      1.2308
                               3.2308 22.3590
##
## Coefficients:
##
                 Estimate Std. Error t value Pr(>|t|)
## (Intercept)
                 8.358974
                            8.316608
                                       1.005 0.348323
## `Cells sorted` 0.009141
                            0.001315
                                       6.952 0.000221 ***
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 13.41 on 7 degrees of freedom
## Multiple R-squared: 0.8735, Adjusted R-squared: 0.8554
## F-statistic: 48.32 on 1 and 7 DF, p-value: 0.0002209
```

Method 2: calculate regression coefficient for each vial

This method is the one that I would like to use. The idea that I have is to group the data by sorting population (nano, pico, syn) and then by vial code. I would like to use a map function to create a new column that creates an LM function for each sorting population and vial, such that lm(DPM2~cells sorted), and then corresponding columns for the slope, p-value or each LM.

Problem: I took some examples online to try and fit to my data but the code doesnt work.

```
library(dplyr)
library(broom)
library(tsibble)
```

```
# This is the code example that I used. The link is https://community.rstudio.com/t/how-to-calculate-sl
df %>%
    group_nest(Source.Name, visbility, soundvolume) %>%
    mutate(model = map(data, ~lm(m ~ stim_ending_t, data = .x))) %>%
    mutate(slope = map_dbl(model, ~tidy(.x)$estimate[2]))

C14_E1_ts <- as.tibble(C14_E1)
C14_E1_ts
E1_trial <- C14_E1_ts %>%
    ungroup() %>%
    group_by(`sorting population`, `Vial code`) %>%
    mutate(model = map(data, ~lm(DPM2~`Cells sorted`, data = .))) %>%
    mutate(slope = map_dbl(model, ~tidy(.)$estimate[2]))
E1_trial
```

Daniel solution

A few things:

- Please read in detail R for data science chapter on grouping data: https://r4ds.had.co.nz/transform.html
- Please read in detail R for data science chapter on joining data: https://r4ds.had.co.nz/relational-
- You can also go back to my class here: https://vaulot.github.io/course-ntu-data-science-2020/R-session-04-data wrangling.html
- When you group data you must explict tell which operation you want to perform (e.g. n() for counting, sum(), mean ()). If not no need to group data
- Make your code general from the start so that to extend to all cases, no need to rewrite it.
- It is much better to rename immediately your varibales in a clean way

Read the data

```
C14_DPM <- readxl::read_excel("TAN1810 Adriana samples complete _final_AGR_1.0.xlsx", sheet = "Sheet1")
     select ( EXP, SAMPLE, `Vial code`, `sorting population`, `Cells sorted`, DPM1) %>%
                                                                                            # Can add mor
     rename(exp = EXP,
            sample = SAMPLE,
            vial = `Vial code`,
            population = `sorting population`,
            cells_sorted = `Cells sorted`,
            dpm = DPM1) \%
     filter(exp == 1) # You will just need to remove this line to make the code valid for all experime
C14_DPM
## # A tibble: 28 x 6
##
        exp sample vial population cells_sorted
                                                    dpm
##
      <dbl> <chr> <chr> <chr>
                                            <dbl> <dbl>
          1 SUR
##
   1
                   Α
                         Pico
                                             2000
                                                     62
          1 SUR
                                             4000
   2
                   Α
                         Pico
                                                     93
   3
          1 SUR
                         Pico
                                            10000
                                                    179
                   Α
```

```
##
##
##
   4
          1 SUR
                          Syn
                                              2000
                                                       75
                   Α
##
   5
          1 SUR
                    Α
                          Syn
                                              4000
                                                       48
##
   6
          1 SUR
                                             10000
                                                      140
                   Α
                          Syn
   7
##
          1 SUR
                          Nano
                                              1000
                                                      926
##
  8
          1 SUR
                    R
                          Pico
                                              2000
                                                       61
## 9
          1 SUR
                    В
                          Pico
                                              4000
                                                       90
## 10
          1 SUR
                    В
                                             10000
                                                      184
                          Pico
## # ... with 18 more rows
```

Substract the dark DPM

You need to make join based on the common variables

```
C14 DPM Dark <- C14 DPM %>%
 filter(vial == "D") %>%
  rename(dpm_dark = dpm) %>%
  select(-vial) # Remove the vial column
C14_DPM_Dark
## # A tibble: 7 x 5
##
       exp sample population cells_sorted dpm_dark
```

```
##
     <dbl> <chr>
                   <chr>
                                        <dbl>
                                                 <dbl>
## 1
         1 SUR
                   Pico
                                        2000
                                                     28
## 2
         1 SUR
                   Pico
                                        4000
                                                     28
## 3
         1 SUR
                                                     35
                   Pico
                                       10000
## 4
         1 SUR
                   Syn
                                        2000
                                                     26
## 5
         1 SUR
                                                     30
                   Syn
                                        4000
## 6
         1 SUR
                                       10000
                                                     37
                   Syn
## 7
         1 SUR
                                                    905
                   Nano
                                        1000
C14_DPM_corrected <- left_join(C14_DPM,C14_DPM_Dark ) %>%
  filter (vial != "D") %>%
  mutate(dpm_corrected = dpm - dpm_dark) %>%
  filter(dpm_corrected >= 0) # Remove negative values
C14_DPM_corrected
## # A tibble: 20 x 8
##
        exp sample vial population cells_sorted
                                                        dpm dpm_dark dpm_corrected
##
      <dbl> <chr>
                    <chr> <chr>
                                               <dbl>
                                                      <dbl>
                                                                <dbl>
                                                                               <dbl>
##
    1
           1 SUR
                           Pico
                                                2000
                                                         62
                                                                   28
                                                                                   34
                     Α
    2
##
           1 SUR
                     Α
                           Pico
                                                4000
                                                         93
                                                                   28
                                                                                   65
           1 SUR
##
    3
                                               10000
                                                        179
                                                                   35
                                                                                  144
                           Pico
                     Α
##
    4
           1 SUR
                     Α
                           Syn
                                                2000
                                                         75
                                                                   26
                                                                                   49
##
    5
           1 SUR
                     Α
                           Syn
                                                4000
                                                         48
                                                                   30
                                                                                   18
##
    6
           1 SUR
                                               10000
                                                        140
                                                                   37
                                                                                  103
                     Α
                           Syn
##
    7
           1 SUR
                                                1000
                                                        926
                                                                  905
                                                                                   21
                           Nano
                     Α
##
    8
           1 SUR
                                                2000
                                                                                   33
                     В
                           Pico
                                                         61
                                                                   28
##
   9
           1 SUR
                    В
                                                4000
                                                         90
                                                                   28
                                                                                  62
                           Pico
## 10
           1 SUR
                    В
                           Pico
                                               10000
                                                        184
                                                                   35
                                                                                  149
           1 SUR
                                                2000
                                                                   26
                                                                                   28
## 11
                     В
                           Syn
                                                         54
           1 SUR
                                                4000
                                                                                  42
## 12
                    В
                           Syn
                                                         72
                                                                   30
## 13
           1 SUR
                                               10000
                                                        138
                                                                   37
                                                                                  101
                     В
                           Syn
## 14
           1 SUR
                           Nano
                                                1000
                                                       1162
                                                                  905
                                                                                  257
                    В
## 15
           1 SUR
                     C
                           Pico
                                                2000
                                                         62
                                                                   28
                                                                                  34
## 16
           1 SUR
                     С
                           Pico
                                                4000
                                                        118
                                                                   28
                                                                                  90
## 17
           1 SUR
                     С
                           Pico
                                               10000
                                                        181
                                                                   35
                                                                                  146
## 18
           1 SUR
                     С
                                                2000
                                                         52
                                                                   26
                                                                                   26
                           Syn
           1 SUR
                     С
## 19
                           Syn
                                                4000
                                                         74
                                                                   30
                                                                                   44
```

Method 1 - Compute Im by grouping ABC together

Syn

Plots

20

1 SUR

Do plots for each group. One regression line based on the EXP, Sample and Populations

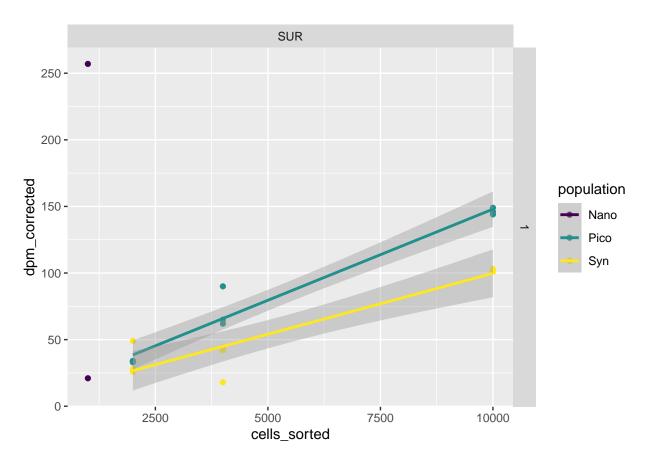
```
ggplot(data = C14_DPM_corrected, aes(x=cells_sorted, y=dpm_corrected, color=population)) +
geom_point() + stat_smooth(method="lm") +
facet_grid(rows=vars(exp), cols=vars(sample)) +
scale_color_viridis_d()
```

10000

140

37

103



Do linear model

```
y = ax + b
```

See: https://cran.r-project.org/web/packages/broom/vignettes/broom_and_dplyr.html

```
C14_DPM_model_1 <- C14_DPM_corrected %>%
  group_by(exp, sample, population) %>%
  tidyr::nest() %>%
  mutate(
    fit = purrr::map(data, ~ lm(dpm_corrected ~ cells_sorted, data = .x)),
    tidied = purrr::map(fit, tidy)
) %>%
  unnest(tidied)

C14_DPM_model_output_1 <- C14_DPM_model_1 %>%
  select(exp:population, term, estimate) %>%
  pivot_wider(names_from="term", values_from ="estimate") %>%
  rename (a = cells_sorted, b = `(Intercept)`)
C14_DPM_model_output_1
```

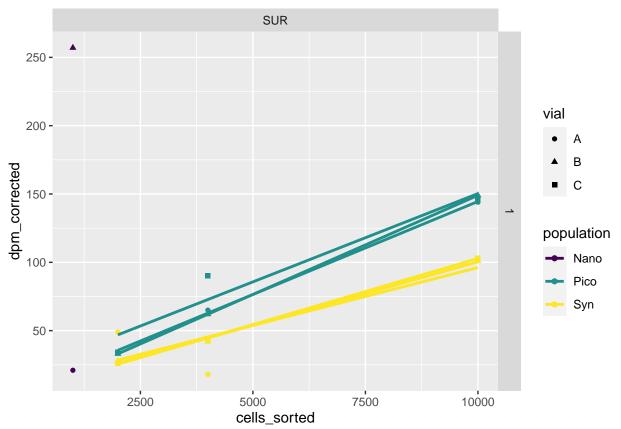
```
## # A tibble: 3 x 5
## # Groups:
               exp, sample, population [3]
##
       exp sample population
                                  b
                                           a
##
     <dbl> <chr> <chr>
                              <dbl>
                                       <dbl>
         1 SUR
                              11.2
                                     0.0137
## 1
                  Pico
## 2
         1 SUR
                               8.36 0.00914
                  Syn
```

Method 2 - Compute Im for ABC separately

Plots

Do plots for each group. One regression line based on the EXP, Sample and Populations

```
ggplot(data = C14_DPM_corrected, aes(x=cells_sorted, y=dpm_corrected, color=population, shape=vial)) +
geom_point() + stat_smooth(method="lm", se=FALSE) +
facet_grid(rows=vars(exp), cols=vars(sample)) +
scale_color_viridis_d()
```



Do linear model

```
y = ax + b
```

See: https://cran.r-project.org/web/packages/broom/vignettes/broom_and_dplyr.html

```
C14_DPM_model_2 <- C14_DPM_corrected %>%
  group_by(exp, sample, population, vial) %>%
  tidyr::nest() %>%
  mutate(
    fit = purrr::map(data, ~ lm(dpm_corrected ~ cells_sorted, data = .x)),
    tidied = purrr::map(fit, tidy)
) %>%
  unnest(tidied)

C14_DPM_model_output_2 <- C14_DPM_model_2 %>%
```

```
select(exp:population, term, estimate) %>%
 pivot_wider(names_from="term", values_from ="estimate" ) %>%
 rename (a = cells_sorted, b = `(Intercept)`)
C14_DPM_model_output_2
## # A tibble: 8 x 6
## # Groups: exp, sample, vial, population [8]
      exp sample vial population b
    <dbl> <chr> <chr> <chr> <
##
                                 <dbl>
                                         <dbl>
## 1
       1 SUR
                      Pico
                                 8.38 0.0136
                Α
## 2
       1 SUR
                Α
                      Syn
                                 11.5 0.00846
## 3
       1 SUR
                Α
                      Nano
                                 21
                                      NA
        1 SUR
                                       0.0145
## 4
                В
                      Pico
                                 4.
## 5
        1 SUR
                                  7.46 0.00929
                В
                      Syn
## 6
       1 SUR
                                257 NA
                В
                      Nano
## 7
        1 SUR
                C
                      Pico
                                 21.1 0.0129
                                  6.08 0.00967
## 8
        1 SUR
                C
                      Syn
```

Denise edits

I made some additional changes. I filtered out Nano population, so the data will only have Pico and Syn. I also removed the filter for experiment 1, so the code is the entire dataset. I only used method 2 (compute lm for ABC separately). This set of edits will continue until the computation of PP as I have added the DIC and SA values from Andres, joining tables based on station no and depth (m).

Final problems:

- From experiment 6: SUR: vial C: pico: 2000 cells sorted -> corrected DPM value = 1485, much higher than DPM of 4000 and 10000 cells counted.
- What should I do with negative slope values that result in negative pp values?

Read the data

C14_DPM

filter(population != "Nano")

```
library(dplyr)
library(tidyverse)
library(ggplot2)
library(broom)
library(tsibble)
C14_DPM <- readxl::read_excel("TAN1810 Adriana samples complete _final_AGR_1.0.xlsx", sheet = "Sheet1")
     select ( Cycle, EXP, STN, DEPTH, SAMPLE, `Vial code`, `sorting population`, `Cells sorted`, DPM1) '
  # Can add more variables latter
  # station and depth for adding updated DIC values
     rename(cycle = Cycle,
            exp = EXP,
            station = STN,
            depth = DEPTH,
            sample = SAMPLE,
            vial = `Vial code`,
            population = `sorting population`,
            cells_sorted = `Cells sorted`,
            dpm = DPM1) \%
```

```
## # A tibble: 385 x 9
##
                exp station depth sample vial population cells_sorted
                                                                                dpm
##
       <dbl> <dbl>
                      <dbl> <dbl> <chr>
                                            <chr> <chr>
                                                                       <dbl> <dbl>
                                12 SUR
##
    1
           1
                  1
                          15
                                            Α
                                                   Pico
                                                                        2000
                                                                                  62
##
    2
           1
                  1
                          15
                                12 SUR
                                            Α
                                                   Pico
                                                                        4000
                                                                                 93
##
    3
           1
                  1
                          15
                                 12 SUR
                                            Α
                                                   Pico
                                                                        10000
                                                                                179
##
    4
                          15
                                12 SUR
                                                   Syn
                                                                        2000
                                                                                 75
           1
                  1
                                            Α
##
    5
           1
                  1
                          15
                                12 SUR
                                                   Syn
                                                                        4000
                                                                                 48
                                            Α
##
                                12 SUR
                                                                       10000
                                                                                140
    6
           1
                          15
                                                   Syn
                  1
                                            Α
##
    7
           1
                  1
                          15
                                12 SUR
                                            В
                                                   Pico
                                                                        2000
                                                                                 61
##
    8
           1
                          15
                                12 SUR
                                            В
                                                   Pico
                                                                        4000
                                                                                 90
                  1
##
    9
                  1
                          15
                                12 SUR
                                            В
                                                   Pico
                                                                        10000
                                                                                184
           1
## 10
                                12 SUR
                                                                        2000
           1
                  1
                          15
                                            В
                                                   Syn
                                                                                 54
## # ... with 375 more rows
```

Substract the dark DPM

You need to make join based on the common variables

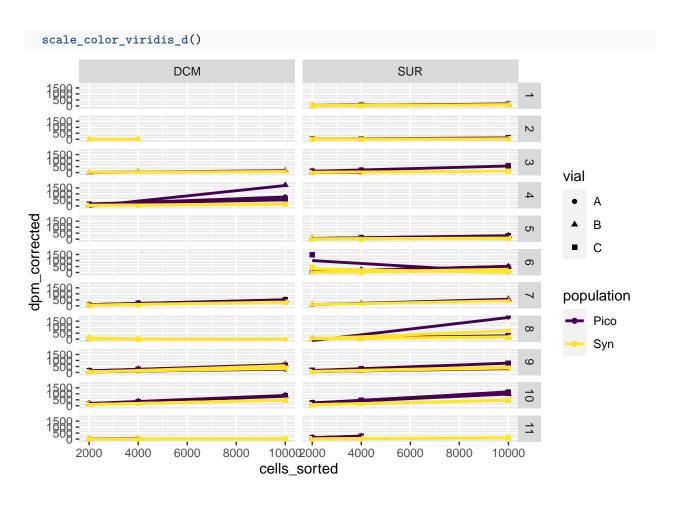
```
C14 DPM Dark <- C14 DPM %>%
  filter(vial == "D") %>%
  rename(dpm_dark = dpm) %>%
  select(-vial) # Remove the vial column
C14_DPM_Dark
## # A tibble: 103 x 8
##
               exp station depth sample population cells_sorted dpm_dark
      <dbl> <dbl>
##
                     <dbl> <dbl> <chr>
                                          <chr>>
                                                              <dbl>
                                                                       <dbl>
##
                               12 SUR
                                                               2000
                                                                          28
    1
           1
                 1
                         15
                                          Pico
                               12 SUR
                                                               4000
                                                                          28
##
    2
           1
                 1
                         15
                                          Pico
##
    3
           1
                 1
                         15
                               12 SUR
                                          Pico
                                                              10000
                                                                          35
##
    4
                               12 SUR
                                                                          26
           1
                 1
                        15
                                          Syn
                                                               2000
##
    5
                        15
                               12 SUR
                                                               4000
                                                                          30
           1
                 1
                                          Syn
    6
                               12 SUR
                                                              10000
                                                                          37
##
           1
                 1
                        15
                                          Syn
                                                                          25
##
    7
           1
                 2
                         24
                               12 SUR
                                          Pico
                                                               2000
##
    8
           1
                 2
                         24
                               12 SUR
                                          Pico
                                                               4000
                                                                          28
##
    9
           1
                 2
                         24
                               12 SUR
                                          Pico
                                                             10000
                                                                          29
## 10
           1
                 2
                         24
                               12 SUR
                                          Syn
                                                               2000
                                                                          29
## # ... with 93 more rows
C14_DPM_corrected <- left_join(C14_DPM,C14_DPM_Dark ) %>%
  filter (vial != "D") %>%
  mutate(dpm_corrected = dpm - dpm_dark) %>%
  filter(dpm_corrected >= 0) %>% # Remove negative values
  filter(!(exp == 2 & sample == "DCM" & vial == "A")) # I removed this row as there was only one value
C14_DPM_corrected
## # A tibble: 246 x 11
##
               exp station depth sample vial population cells_sorted
                                                                             dpm dpm_dark
##
      <dbl> <dbl>
                     <dbl> <dbl> <chr>
                                          <chr> <chr>
                                                                    <dbl> <dbl>
                                                                                    <dbl>
##
                               12 SUR
                                                                     2000
                                                                                       28
    1
           1
                 1
                         15
                                                Pico
                                                                              62
##
    2
           1
                 1
                         15
                               12 SUR
                                          Α
                                                Pico
                                                                     4000
                                                                              93
                                                                                       28
##
    3
           1
                 1
                        15
                               12 SUR
                                                Pico
                                                                    10000
                                                                             179
                                                                                       35
##
    4
           1
                 1
                        15
                               12 SUR
                                          Α
                                                Syn
                                                                     2000
                                                                             75
                                                                                       26
##
    5
           1
                 1
                         15
                               12 SUR
                                          Α
                                                Syn
                                                                     4000
                                                                              48
                                                                                       30
##
    6
                         15
                               12 SUR
                                                                    10000
                                                                             140
                                                                                       37
           1
                 1
                                                Syn
                                          Α
##
    7
           1
                 1
                        15
                               12 SUR
                                          В
                                                Pico
                                                                     2000
                                                                             61
                                                                                       28
                               12 SUR
                                                                                       28
##
    8
           1
                 1
                        15
                                          В
                                                Pico
                                                                     4000
                                                                             90
##
    9
           1
                 1
                         15
                               12 SUR
                                          В
                                                Pico
                                                                    10000
                                                                             184
                                                                                       35
## 10
           1
                         15
                               12 SUR
                                          В
                                                                     2000
                                                                             54
                                                                                       26
                 1
                                                Syn
## # ... with 236 more rows, and 1 more variable: dpm_corrected <dbl>
```

Method 2 - Compute Im for ABC separately

Plots

Do plots for each group. One regression line based on the EXP, Sample and Populations

```
ggplot(data = C14_DPM_corrected, aes(x=cells_sorted, y=dpm_corrected, color=population, shape=vial)) +
geom_point() + stat_smooth(method="lm", se=FALSE) +
facet_grid(rows=vars(exp), cols=vars(sample)) +
```



need to check exp 6: SUR: Pico: C: 2000 cells sorted. Value is too high. Slope value is negative.

Do linear model

```
y = ax + b
See: https://cran.r-project.org/web/packages/broom/vignettes/broom_and_dplyr.html
C14_DPM_model_3 <- C14_DPM_corrected %>%
group_by(cycle, exp, station, depth, sample, population, vial) %>%
tidyr::nest() %>%
mutate(
   fit = purrr::map(data, ~ lm(dpm_corrected ~ cells_sorted, data = .x)),
   tidied = purrr::map(fit, tidy)
) %>%
unnest(tidied)
C14_DPM_model_output_3 <- C14_DPM_model_3 %>%
select(exp:population, term, estimate) %>%
pivot_wider(names_from="term", values_from ="estimate") %>%
rename (slope = cells_sorted, intercept = `(Intercept)`)
C14_DPM_model_output_3
```

```
## # A tibble: 88 x 9
                cycle, exp, station, depth, sample, vial, population [88]
  # Groups:
               exp station depth sample vial population intercept
##
                      <dbl> <dbl> <chr>
                                          <chr> <chr>
##
      <dbl> <dbl>
                                                                 <dbl>
                                                                          <dbl>
##
    1
           1
                 1
                         15
                               12 SUR
                                                 Pico
                                                                 8.38 0.0136
##
    2
                         15
                               12 SUR
                                                                11.5
                                                                        0.00846
           1
                 1
                                                 Syn
    3
                               12 SUR
                                                                        0.0145
##
           1
                 1
                         15
                                                 Pico
                                                                 4.
                                                                 7.46 0.00929
##
    4
           1
                 1
                         15
                               12 SUR
                                          В
                                                 Syn
##
    5
           1
                 1
                         15
                               12 SUR
                                          C
                                                 Pico
                                                                21.1
                                                                        0.0129
                                          С
                                                                 6.08 0.00967
##
    6
           1
                 1
                         15
                               12 SUR
                                                 Syn
##
    7
           1
                 2
                         24
                               12 SUR
                                          Α
                                                 Pico
                                                                15.9
                                                                        0.00258
                 2
                         24
                               12 SUR
                                                                -3.38 0.00438
##
    8
           1
                                          Α
                                                 Syn
                 2
##
    9
           1
                         24
                               12 SUR
                                          C
                                                 Pico
                                                                -6.77 0.0120
                                                                 0.769 0.00298
## 10
           1
                 2
                         24
                               12 SUR
                                          C
                                                 Syn
## # ... with 78 more rows
```

Include DIC and SA values

Import DIC data from Andres. I will use this data to join with the DPM model output. The corrected data frame will have the DIC and SA values. The data will join based on the common columns: station and depth.

Create DIC table

```
DIC_data <- readxl::read_excel("Chla NPP Raw TAN1810 Sept.xlsx", sheet = "Compiled TAN1810 NPP data")
\# excel sheet not optimal for R, have to delete columns and make row 5 headers.
names(DIC_data) <- DIC_data[5,] # make row 5 header</pre>
DIC_data \leftarrow DIC_data [-c(1, 2, 3, 4, 5), c(1:34)]
DIC_data <- DIC_data [, -c(1, 27)] # remove repeated columns
colnames(DIC_data) # check
##
    [1] "Mean contol activity DPM"
                                              "Date"
    [3] "Time Start"
                                               "Time End"
##
##
    [5] "Time
               days"
                                              "Station"
    [7]
       "Site"
                                              "Latitude"
##
##
    [9] "Longitude"
                                              "Depth m"
## [11] "DATE"
                                              "TIME"
  [13]
        "P#"
                                              "PID"
##
## [15] "S#"
                                              "Count Time"
                                              "CPMA"
## [17] "A:2S%"
## [19] "SIS"
                                              "tSIE"
##
  [21] "DPM1"
                                               "DPM to full volume"
## [23] "Sample ID"
                                               "Comments"
                                              "DIC"
## [25] "Volume filtered of bottles 320mls"
## [27] "Additive Chla Total for comparison"
                                              Chla>0.2\r\mg/m3
  [29] "Chla>2\r\nmg/m3"
                                               Chla>20\r\n mg/m3
                                              "SA"
## [31] "Unique code or sample identifier"
# select for station, depth, DIC, SA. Change all columns to integer to join with DPM output.
DIC_data_corrected <- select(DIC_data, Station, `Depth m`, DIC, SA) %>%
  unique() %>% # delete repeated rows
  rename(station = Station,
         depth = `Depth m`) %>%
  filter(depth != "35/40?") %>% # filter out character to change to integer
  mutate_if(is.character, as.double) # change to double (same format as DPM dataset)
```

```
DIC_data_corrected
## # A tibble: 101 x 4
                      DIC
##
      station depth
                               SA
##
        <dbl> <dbl> <dbl>
                           <dbl>
##
           15
   1
                  5
                     25.9 21500.
##
    2
           15
                 12
                     25.9 21087.
           24
##
   3
                     25.9 21089.
                  5
##
           24
                     25.7 20958.
                 12
                     25.9 20453.
##
   5
           24
                 20
           24
                     25.9 20761.
##
    6
                 30
           24
##
  7
                 40
                     26.2 21436.
##
   8
           24
                 50
                     26.1 21132.
           39
                     25.7 22025.
##
  9
                  5
## 10
           39
                 12
                     25.8 18035.
## # ... with 91 more rows
Merge DIC/SA table and DPM output
# merge tables
final_cal <- left_join(C14_DPM_model_output_3, DIC_data_corrected)</pre>
final_cal
## # A tibble: 88 x 11
## # Groups:
               cycle, exp, station, depth, sample, vial, population [88]
##
      cycle
              exp station depth sample vial population intercept
                                                                      slope
                                                                              DIC
##
      <dbl> <dbl>
                    <dbl> <dbl> <chr> <chr> <chr> <
                                                              <dbl>
                                                                      <dbl> <dbl>
##
                       15
                              12 SUR
                                                              8.38 0.0136
                                                                             25.9
   1
          1
                1
                                        Α
                                              Pico
##
                1
                       15
                              12 SUR
                                              Syn
                                                            11.5
                                                                    0.00846
                                                                             25.9
                                        Α
##
                             12 SUR
                                                              4.
                                                                    0.0145
   3
          1
                       15
                                              Pico
                                                                             25.9
                1
                                        В
##
   4
          1
                1
                       15
                             12 SUR
                                        В
                                              Syn
                                                             7.46 0.00929
                                                                             25.9
##
                       15
   5
          1
                             12 SUR
                                        С
                                              Pico
                                                            21.1
                                                                    0.0129
                                                                             25.9
                1
##
   6
          1
                1
                       15
                             12 SUR
                                        С
                                                              6.08 0.00967
                                              Syn
   7
##
          1
                2
                       24
                             12 SUR
                                        Α
                                              Pico
                                                             15.9
                                                                    0.00258
                                                                             25.7
    8
          1
                2
                       24
                             12 SUR
                                                             -3.38 0.00438
##
                                        Α
                                              Syn
                                                                             25.7
  9
                2
##
          1
                       24
                              12 SUR
                                        C
                                                            -6.77 0.0120
                                                                             25.7
                                              Pico
```

Calculate PP value, based on Daniel's formula found here:

C

12 SUR

... with 78 more rows, and 1 more variable: SA <dbl>

2

24

1

10

```
https://vaulot.netlify.com/2018/05/20/compute-primary-production-based-on-single-cell-c14-uptake/
final_cal <- mutate(final_cal, pp = DIC*slope*(1/(SA*24))*10^9*1.05) %>%
    ungroup() %>%
    select( -c(station, depth))
#add missing rows for data analysis later
final_cal <- complete(final_cal, nesting(cycle, exp), sample, vial, population)
final_cal</pre>
```

Syn

0.769 0.00298

25.7

##	1	1	1	DCM	Α	Pico	NA	NA	NA	NA	NA
##	2	1	1	DCM	Α	Syn	NA	NA	NA	NA	NA
##	3	1	1	DCM	В	Pico	NA	NA	NA	NA	NA
##	4	1	1	DCM	В	Syn	NA	NA	NA	NA	NA
##	5	1	1	DCM	C	Pico	NA	NA	NA	NA	NA
##	6	1	1	DCM	C	Syn	NA	NA	NA	NA	NA
##	7	1	1	SUR	Α	Pico	8.38	0.0136	25.9	21087.	732.
##	8	1	1	SUR	Α	Syn	11.5	0.00846	25.9	21087.	455.
##	9	1	1	SUR	В	Pico	4.	0.0145	25.9	21087.	779.
##	10	1	1	SUR	В	Syn	7.46	0.00929	25.9	21087.	499.
##	#	with	122	more	rows						

Some notes from 14 April:

- For the purposes of Eleanor's class, I will use method 2 (compute lm values for ABC separately) for more replicates, but method 1 will be used for actual calculations later.
- No need to check p value of the slope.
- Will not set intercept to 0.