

# chla\_per\_cell

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8/9/2020

```
# chlorophyll a per cell

# set the working directory
setwd("~/Desktop/PhD/chapters/chapter 4 - temp treatments/for_coauthors_v1/data_code/")
# load the packages
library(readxl)
library(dplyr)
```

```
##
## 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
```

```
library(ggplot2)
library(ggpubr)
```

```
## Loading required package: magrittr
```

```
library(PMCMR)
```

```
## PMCMR is superseded by PMCMRplus and will be no longer maintained. You may wish to install PMCMRplus instead.
```

```
library(cowplot)
```

```
##  
## *****
```

```
## Note: As of version 1.0.0, cowplot does not change the
```

```
## default ggplot2 theme anymore. To recover the previous
```

```
## behavior, execute:  
## theme_set(theme_cowplot())
```

```
## *****
```

```
##  
## Attaching package: 'cowplot'
```

```
## The following object is masked from 'package:ggpubr':  
##  
## get_legend
```

```
library(data.table) # for function `fread`
```

```
##  
## Attaching package: 'data.table'
```

```
## The following objects are masked from 'package:dplyr':  
##  
## between, first, last
```

```
library(broom)      # for function `tidy`  
library(reshape2)
```

```
##  
## Attaching package: 'reshape2'
```

```
## The following objects are masked from 'package:data.table':  
##  
##      dcast, melt
```

```
library(tidyr)
```

```
##  
## Attaching package: 'tidyr'
```

```
## The following object is masked from 'package:reshape2':  
##  
##      smiths
```

```
## The following object is masked from 'package:magrittr':  
##  
##      extract
```

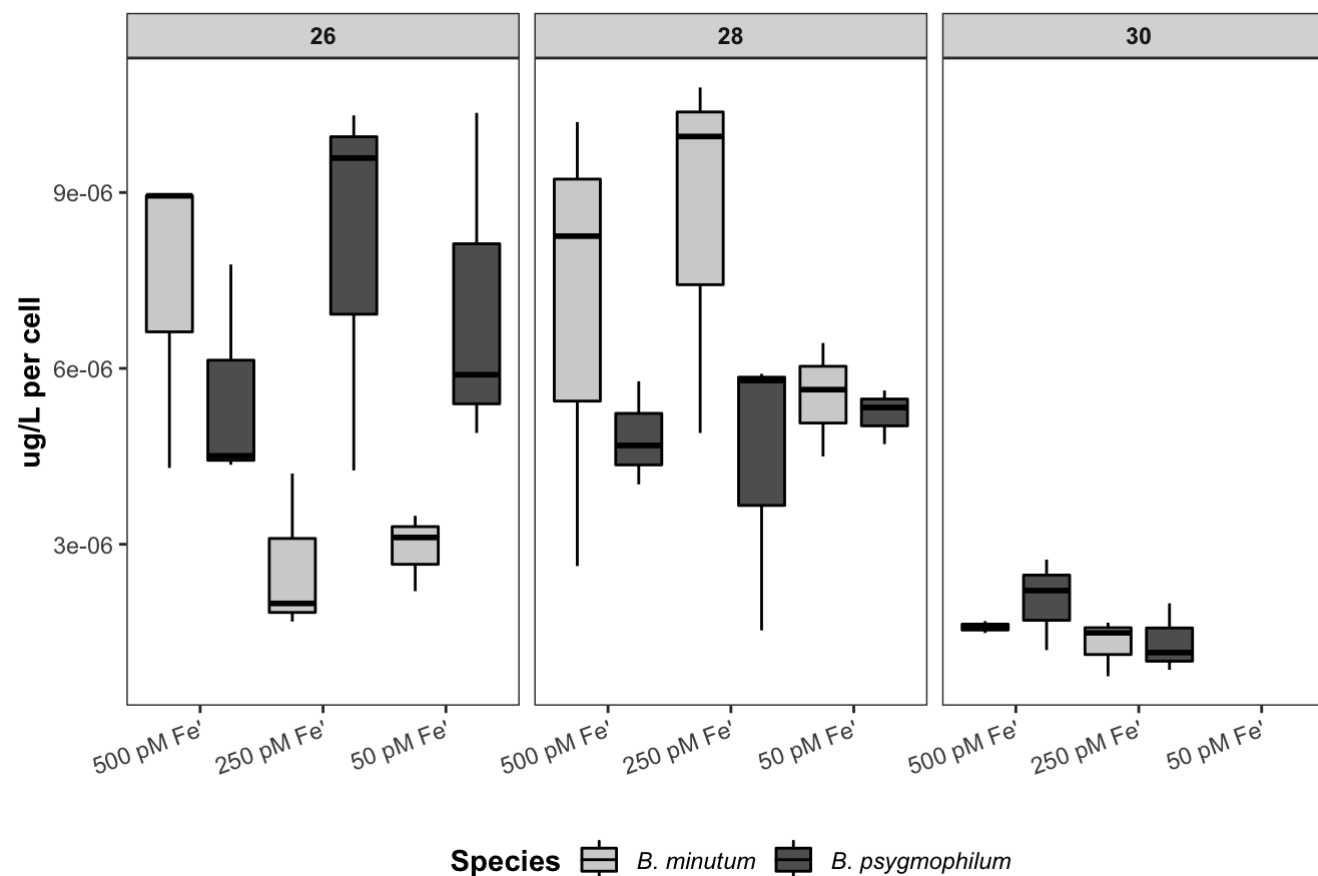
```

pig <- read_excel("~/Desktop/PhD/chapters/chapter 4 - temp treatments/for_coauthors_v1/data_code/hplc_fire_result
s_git.xlsx", sheet = "data")
# load the cell vol data back in
volused <- read_excel("~/Desktop/PhD/chapters/chapter 4 - temp treatments/for_coauthors_v1/data_code/cell_densit
y.xlsx", sheet = "vol")
# calculate the number of cells used
volused$cellspig <- (volused$CellDensity*volused$mLpigment)
# merge the data
all <- merge(pig, volused, by = "sampleID")
all$Chlorophyll_A_CELL <- (all$Chlorophyll_A/all$cellspig)
all$Temp <- as.factor(all$Temp)
all$Ironconc <- as.factor(all$Ironconc)
all$treatmentID.x <- as.factor(all$treatmentID.x)
all$species.x <- factor(all$species.x, levels = c("min", "psyg"))
levels(all$species.x) <- c("B. minutum", "B. psygmophilum")
all$Ironconc <- factor(all$Ironconc, levels = c("100", "50", "10"))
levels(all$Ironconc) <- c("500 pM Fe'", "250 pM Fe'", "50 pM Fe'")
# make a plot
# box plot with species next to one another
chlal <- ggplot(data=all, aes(x=Ironconc, y=Chlorophyll_A_CELL, fill=species.x, color= "black")) +
  geom_boxplot(alpha = 0.7, color = "black", varwidth = FALSE, position = position_dodge(1, preserve = "single"))
+
  theme_bw() +
  labs(y="ug/L per cell", x=element_blank(), title = "Chlorophyll a content") +
  theme(panel.grid = element_blank(),
        strip.text.x = element_text(face = "bold"),
        strip.text.y = element_text(face = "bold.italic"),
        legend.title = element_text(face = "bold"),
        legend.text = element_text(face = "italic"),
        axis.text.x = element_text(angle=20, vjust=1, hjust=1),
        axis.title = element_text(face = "bold"),
        legend.position = "bottom") +
  facet_grid(~Temp, scales = "fixed") +
  scale_fill_manual(values = c("grey", "black"), breaks = c("B. minutum", "B. psygmophilum"), name = "Species")
chlal

```

```
## Warning: Removed 7 rows containing non-finite values (stat_boxplot).
```

## Chlorophyll a content



```
#save_plot("FigS2_chla_v2.pdf", chla1 ,base_aspect_ratio = 1.6)
# illustrator edits: edit y-axis to have proper greek letters & superscripts; make sure "a" in title is italicized
## by species
kruskal.test(all$Chlorophyll_A_CELL, all$species.x)
```

```
##
## Kruskal-Wallis rank sum test
##
## data: all$Chlorophyll_A_CELL and all$species.x
## Kruskal-Wallis chi-squared = 0.2192, df = 1, p-value = 0.6396
```

```
# make objects for min and psyg
bm <- all %>% filter(species.x %in% c("B. minutum"))
bp <- all %>% filter(species.x %in% c("B. psygmophilum"))
# normality tests
shapiro.test(bm$Chlorophyll_A_CELL)
```

```
##
##  Shapiro-Wilk normality test
##
## data:  bm$Chlorophyll_A_CELL
## W = 0.88521, p-value = 0.01268
```

```
shapiro.test(bp$Chlorophyll_A_CELL)
```

```
##
##  Shapiro-Wilk normality test
##
## data:  bp$Chlorophyll_A_CELL
## W = 0.92655, p-value = 0.08163
```

```
# KW test
kruskal.test(bm$Chlorophyll_A_CELL, bm$Temp)
```

```
##
##  Kruskal-Wallis rank sum test
##
## data:  bm$Chlorophyll_A_CELL and bm$Temp
## Kruskal-Wallis chi-squared = 13.775, df = 2, p-value = 0.001021
```

```
kruskal.test(bm$Chlorophyll_A_CELL, bm$Ironconc)
```

```
##  
## Kruskal-Wallis rank sum test  
##  
## data:  bm$Chlorophyll_A_CELL and bm$Ironconc  
## Kruskal-Wallis chi-squared = 1.2153, df = 2, p-value = 0.5446
```

```
kruskal.test(bp$Chlorophyll_A_CELL, bp$Temp)
```

```
##  
## Kruskal-Wallis rank sum test  
##  
## data:  bp$Chlorophyll_A_CELL and bp$Temp  
## Kruskal-Wallis chi-squared = 12.56, df = 2, p-value = 0.001873
```

```
kruskal.test(bp$Chlorophyll_A_CELL, bp$Ironconc)
```

```
##  
## Kruskal-Wallis rank sum test  
##  
## data:  bp$Chlorophyll_A_CELL and bp$Ironconc  
## Kruskal-Wallis chi-squared = 3.0944, df = 2, p-value = 0.2128
```

```
# post hoc by iron  
posthoc.kruskal.dunn.test(Chlorophyll_A_CELL~Ironconc, data = bm, p.adjust.methods = "fdr")
```

```
##
## Pairwise comparisons using Dunn's-test for multiple
## comparisons of independent samples
##
## data: Chlorophyll_A_CELL by Ironconc
##
##           500 pM Fe' 250 pM Fe'
## 250 pM Fe' 0.86      -
## 50 pM Fe'  0.95      0.95
##
## P value adjustment method: holm
```

```
posthoc.kruskal.dunn.test(Chlorophyll_A_CELL~Ironconc, data = bp, p.adjust.methods = "fdr")
```

```
##
## Pairwise comparisons using Dunn's-test for multiple
## comparisons of independent samples
##
## data: Chlorophyll_A_CELL by Ironconc
##
##           500 pM Fe' 250 pM Fe'
## 250 pM Fe' 0.76      -
## 50 pM Fe'  0.28      0.31
##
## P value adjustment method: holm
```

```
# post hoc by temp
posthoc.kruskal.dunn.test(Chlorophyll_A_CELL~Temp, data = bm, p.adjust.methods = "fdr")
```



```
##
## Pairwise comparisons using Dunn's-test for multiple
## comparisons of independent samples
##
## data: Chlorophyll_A_CELL by Temp
##
##      26      28
## 28 0.08228 -
## 30 0.05043 0.00063
##
## P value adjustment method: holm
```

```
posthoc.kruskal.dunn.test(Chlorophyll_A_CELL~Temp, data = bp, p.adjust.methods = "fdr")
```

```
##
## Pairwise comparisons using Dunn's-test for multiple
## comparisons of independent samples
##
## data: Chlorophyll_A_CELL by Temp
##
##      26      28
## 28 0.3173 -
## 30 0.0015 0.0190
##
## P value adjustment method: holm
```

```
# post hoc by treatment ID
posthoc.kruskal.dunn.test(Chlorophyll_A_CELL~treatmentID.x, data = bm, p.adjust.methods = "fdr")
```

```
##
## Pairwise comparisons using Dunn's-test for multiple
## comparisons of independent samples
##
## data: Chlorophyll_A_CELL by treatmentID.x
##
##           min-10-26 min-10-28 min-100-26 min-100-28 min-100-30 min-50-26
## min-10-28  1.00      -          -          -          -          -
## min-100-26 1.00      1.00      -          -          -          -
## min-100-28 1.00      1.00      1.00      -          -          -
## min-100-30 1.00      1.00      0.72      0.97      -          -
## min-50-26  1.00      1.00      1.00      1.00      1.00      -
## min-50-28  1.00      1.00      1.00      1.00      0.30      0.77
## min-50-30  1.00      0.45      0.22      0.34      1.00      1.00
##           min-50-28
## min-10-28  -
## min-100-26 -
## min-100-28 -
## min-100-30 -
## min-50-26  -
## min-50-28  -
## min-50-30  0.06
##
## P value adjustment method: holm
```

```
posthoc.kruskal.dunn.test(Chlorophyll_A_CELL~treatmentID.x, data = bp, p.adjust.methods = "fdr")
```

```
##
## Pairwise comparisons using Dunn's-test for multiple
## comparisons of independent samples
##
## data: Chlorophyll_A_CELL by treatmentID.x
##
##          psyg-10-26 psyg-10-28 psyg-100-26 psyg-100-28 psyg-100-30
## psyg-10-28  1.00      -          -          -          -
## psyg-100-26 1.00      1.00      -          -          -
## psyg-100-28 1.00      1.00      1.00      -          -
## psyg-100-30 0.47      1.00      1.00      1.00      -
## psyg-50-26  1.00      1.00      1.00      1.00      0.71
## psyg-50-28  1.00      1.00      1.00      1.00      1.00
## psyg-50-30  0.13      0.90      1.00      1.00      1.00
##          psyg-50-26 psyg-50-28
## psyg-10-28  -          -
## psyg-100-26 -          -
## psyg-100-28 -          -
## psyg-100-30 -          -
## psyg-50-26  -          -
## psyg-50-28  1.00      -
## psyg-50-30  0.21      1.00
##
## P value adjustment method: holm
```