lipid_v01232020

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Elise & Artemis Keister analysed abundance of lipid classes on the iron*heat cultures. this is the first pass at figs/stats.

visualize total lipids per samples. script updated 4/22/2020 to remove non total lipid content data. the file with the lipid classes analysis is in ~/summer 2018/culture_lipids

```
# set the working directory
setwd("~/Desktop/PhD/summer 2018/culture_lipids/")

# load 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(reshape2)
library(ggplot2)
library(ggpubr)
```

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

```
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'
```

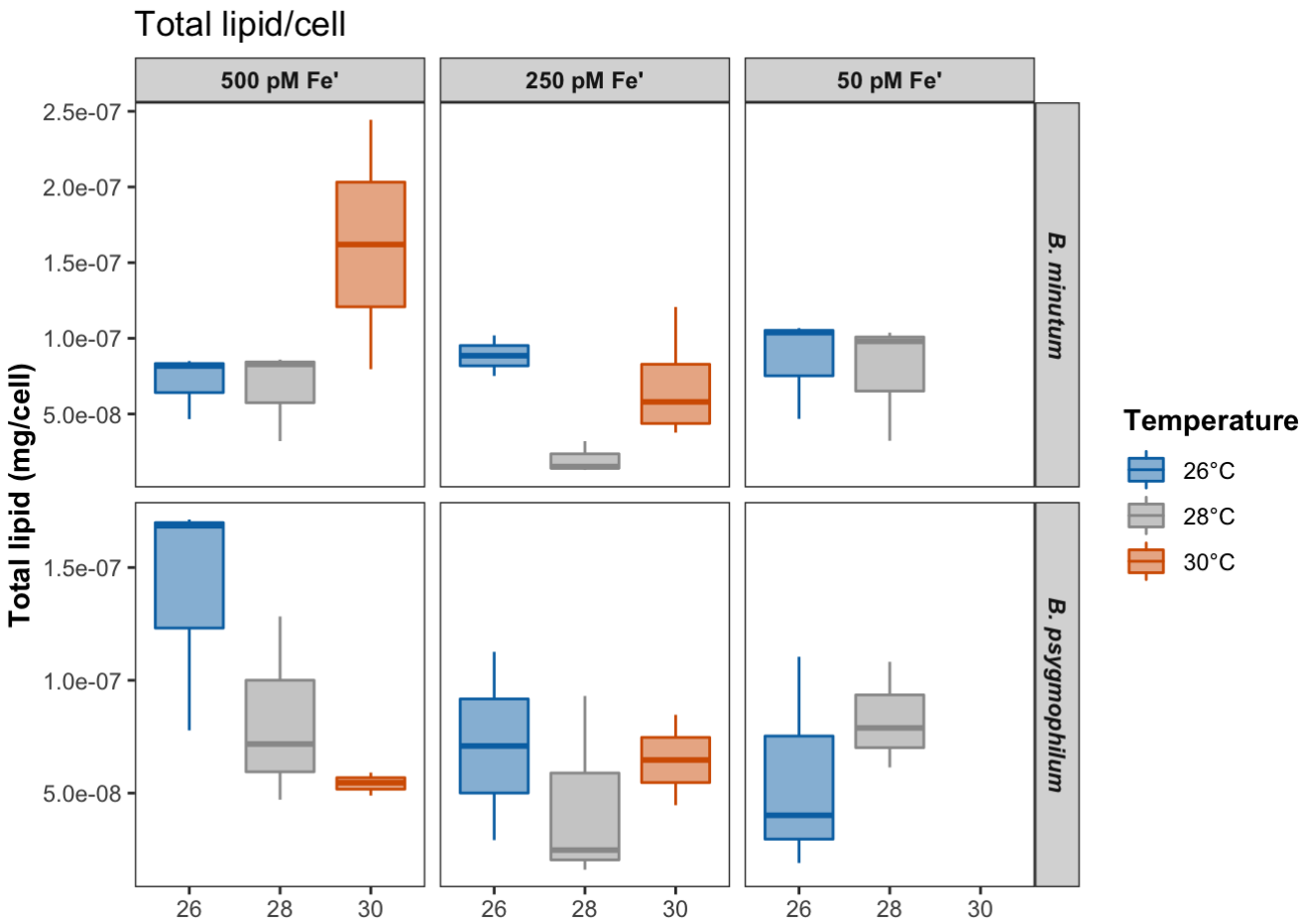
```
## The following object is masked from 'package:ggpubr':
##
##     get_legend
```

```
library(PMCMR)
```

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## PMCMR is superseded by PMCMRplus and will be no longer maintained.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

```
# read in the data
tot <- read_excel("~/Desktop/PhD/summer 2018/culture_lipids/HR_cultures_lipid quant.xlsx", sheet = "for_R_total")
tot <- as.data.frame(tot)

# color palette
cols <-c( "26" = "#0072B2", "28" = "#999999", "30" = "#D55E00")

# make ironconc and temp factors
tot$Temp <- as.factor(tot$Temp)
tot$Ironconc <- as.factor(tot$Ironconc)
tot$Species <- as.factor(tot$Species)
tot$treatment_ID <- as.factor(tot$treatment_ID)

# make levels fancy
tot$Species <- factor(tot$Species, levels = c("min", "psyg"))
levels(tot$Species) <- c("B. minutum", "B. psygmophilum")
tot$Ironconc <- factor(tot$Ironconc, levels =c("100", "50", "10"))
levels(tot$Ironconc) <- c("500 pM Fe'", "250 pM Fe'", "50 pM Fe'")

# box plot
tot1 <- ggplot(data=tot, aes(x=Temp, y=total_lipid_no_outlier, fill=Temp, color=Temp)) +
  geom_boxplot(alpha = 0.5, aes(colour = factor(Temp)), varwidth = FALSE, position = position_dodge(1, preserve =
"single")) +
  theme_bw() +
  labs(y="Total lipid (mg/cell)", x=element_blank(), title = "Total lipid/cell") +
  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"),
        axis.text.x = element_text(angle=0),
        axis.title = element_text(face = "bold"),
        legend.position = "right") +
  facet_grid(Species~Ironconc, scales = "free_y") +
  scale_color_manual(values = c("#0072B2","#999999","#D55E00"), name = "Temperature", breaks = c("26", "28", "30"
), labels=c("26°C", "28°C", "30°C")) +
  scale_fill_manual(values = c("#0072B2","#999999","#D55E00"), name = "Temperature", breaks = c("26", "28", "30"
), labels=c("26°C", "28°C", "30°C"))
tot1
```

```
## Warning: Removed 2 rows containing non-finite values (stat_boxplot).
```




```
save_plot("lipid_cell.pdf", tot1, base_aspect_ratio = 1.6)
```

```
## Warning: Removed 2 rows containing non-finite values (stat_boxplot).
```

```
### do the total lipid stats.

# between species
kruskal.test(tot$total_lipid_no_outlier, tot$Species)
```

```
##
##  Kruskal-Wallis rank sum test
##
## data:  tot$total_lipid_no_outlier and tot$Species
## Kruskal-Wallis chi-squared = 0.00464, df = 1, p-value = 0.9457
```

```
# within species
bm <- dplyr::filter(tot, Species %in% c("B. minutum"))
bp <- dplyr::filter(tot, Species %in% c("B. psygmophilum"))

# normality tests
shapiro.test(bm$total_lipid_no_outlier)
```

```
##
##  Shapiro-Wilk normality test
##
## data:  bm$total_lipid_no_outlier
## W = 0.83551, p-value = 0.001503
```

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

```
##
##  Shapiro-Wilk normality test
##
## data:  bp$total_lipid_no_outlier
## W = 0.93434, p-value = 0.1511
```

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

```
##
##  Kruskal-Wallis rank sum test
##
## data:  bm$total_lipid_no_outlier and bm$Temp
## Kruskal-Wallis chi-squared = 2.7452, df = 2, p-value = 0.2535
```

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

```
##
##  Kruskal-Wallis rank sum test
##
## data:  bm$total_lipid_no_outlier and bm$Ironconc
## Kruskal-Wallis chi-squared = 3.4218, df = 2, p-value = 0.1807
```

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

```
##
##  Kruskal-Wallis rank sum test
##
## data:  bp$total_lipid_no_outlier and bp$Temp
## Kruskal-Wallis chi-squared = 0.87141, df = 2, p-value = 0.6468
```

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

```
##
##  Kruskal-Wallis rank sum test
##
## data:  bp$total_lipid_no_outlier and bp$Ironconc
## Kruskal-Wallis chi-squared = 1.84, df = 2, p-value = 0.3985
```

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

```
##
##  Pairwise comparisons using Dunn's-test for multiple
##                          comparisons of independent samples
##
## data:  total_lipid_no_outlier by Ironconc
##
##            500 pM Fe' 250 pM Fe'
## 250 pM Fe' 0.40       -
## 50 pM Fe'  0.58       0.24
##
## P value adjustment method: holm
```

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

```
##
##  Pairwise comparisons using Dunn's-test for multiple
##                          comparisons of independent samples
##
## data:  total_lipid_no_outlier by Ironconc
##
##            500 pM Fe' 250 pM Fe'
## 250 pM Fe' 0.53       -
## 50 pM Fe'  0.97       0.97
##
## P value adjustment method: holm
```

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

```
##
##  Pairwise comparisons using Dunn's-test for multiple
##                           comparisons of independent samples
##
## data:  total_lipid_no_outlier by Temp
##
##    26   28
## 28 0.36 -
## 30 0.80 0.48
##
## P value adjustment method: holm
```

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

```
##
##  Pairwise comparisons using Dunn's-test for multiple
##                           comparisons of independent samples
##
## data:  total_lipid_no_outlier by Temp
##
##    26 28
## 28 1  -
## 30 1  1
##
## P value adjustment method: holm
```

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

```
##
##  Pairwise comparisons using Dunn's-test for multiple
##                          comparisons of independent samples
##
## data:  total_lipid_no_outlier by treatment_ID
##
##            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      1.00       1.00       -          -
## min-50-26  1.00      1.00      1.00       1.00       1.00       -
## min-50-28  0.26      0.76      1.00       1.00       0.32       0.19
## min-50-30  1.00      1.00      1.00       1.00       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  1.00
##
## P value adjustment method: holm
```

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

```
##
##  Pairwise comparisons using Dunn's-test for multiple
##                          comparisons of independent samples
##
## data:  total_lipid_no_outlier by treatment_ID
##
##             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 1.00       1.00       1.00        1.00        -
## psyg-50-26  1.00       1.00       1.00        1.00        1.00
## psyg-50-28  1.00       1.00       0.66        1.00        1.00
## psyg-50-30  1.00       1.00       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  1.00       1.00
##
## P value adjustment method: holm
```