

Figure 2f

```
library(readxl)
library(lubridate)

##
## Attaching package: 'lubridate'
## The following objects are masked from 'package:base':
##
##   date, intersect, setdiff, union
library(mgcv)

## Loading required package: nlme
## This is mgcv 1.8-34. For overview type 'help("mgcv-package")'.
library(ggplot2)
library(ggsignif)
library(readr)
library(emmeans)
library(tidyr)
library(plyr)
library(dplyr)

##
## Attaching package: 'dplyr'
## The following objects are masked from 'package:plyr':
##
##   arrange, count, desc, failwith, id, mutate, rename, summarise,
##   summarize
## The following object is masked from 'package:nlme':
##
##   collapse
## The following objects are masked from 'package:stats':
##
##   filter, lag
## The following objects are masked from 'package:base':
##
##   intersect, setdiff, setequal, union
#load in raw data
dat <- data.frame(read_csv("2f_data.csv"))

##
## -- Column specification -----
## cols(
##   `Repeat(Date)` = col_character(),
##   Algae = col_character(),
```

```
## Treatment = col_character(),
## Well_ID = col_character(),
## Larvae = col_character(),
## uptakes = col_double()
## )

head(dat)

## Repeat.Date. Algae Treatment Well_ID Larvae uptakes
## 1 I SSB01 No S1 1 0
## 2 I SSB01 No S1 2 0
## 3 I SSB01 No S1 3 0
## 4 I SSB01 No S1 4.1 0
## 5 I SSB01 No S1 4-2 0
## 6 I SSB01 No S1 5 1

#clean up raw data
dat$Algae <- factor(dat$Algae, levels= c("SSB01", "M. gaditana", "N. oculata", "C. velia", "Beads"))
dat$Well_ID <- factor(dat$Well_ID)
dat$Treatment <- factor(dat$Treatment)
dat$Repeat.Date. <- factor(as.character(dat$Repeat.Date.))
head(dat)

## Repeat.Date. Algae Treatment Well_ID Larvae uptakes
## 1 I SSB01 No S1 1 0
## 2 I SSB01 No S1 2 0
## 3 I SSB01 No S1 3 0
## 4 I SSB01 No S1 4.1 0
## 5 I SSB01 No S1 4-2 0
## 6 I SSB01 No S1 5 1

#model data with generalized linear mixed modes
mod <- gam(uptakes ~ Algae,data=dat,family=nb())
S <- summary(mod)$p.table
S

## Estimate Std. Error z value Pr(>|z|)
## (Intercept) -2.302585 0.5504790 -4.182876 2.878447e-05
## AlgaeM. gaditana 2.678636 0.6033763 4.439413 9.020476e-06
## AlgaeN. oculata 2.143520 0.6323722 3.389650 6.998197e-04
## AlgaeC. velia 1.500239 0.6731203 2.228782 2.582840e-02

#model pairwise comparison (Tukey)
mod.emm.s <- emmeans(mod, "Algae")
pairwise <- data.frame(pairs(mod.emm.s))
pairwise$label <- ifelse(pairwise$p.value < 0.001, "***",ifelse(pairwise$p.value < 0.01, "**",ifelse(pairwise$p.value < 0.05, "*",)))
pairwise <- pairwise %>% separate(contrast, c("start", "end"), " - ")
head(pairwise)

## start end estimate SE df t.ratio p.value label
## 1 SSB01 M. gaditana -2.6786363 0.6033763 145 -4.439413 0.0001038127 ***
## 2 SSB01 N. oculata -2.1435204 0.6323722 145 -3.389650 0.0049442507 **
## 3 SSB01 C. velia -1.5002386 0.6731203 145 -2.228782 0.1203578719
## 4 M. gaditana N. oculata 0.5351159 0.3973707 145 1.346642 0.5348885965
## 5 M. gaditana C. velia 1.1783977 0.4594559 145 2.564768 0.0545559748
## 6 N. oculata C. velia 0.6432818 0.4969220 145 1.294533 0.5678912990
```

```
write_csv(pairwise, "2f_pairwise.csv")
```

```
#clean up results for graph
result <- data.frame(mod.emm.s)
results <- data.frame(Algae=result$Algae)
#Transform data (linkfunction)
results$mean<- exp(result$emmean)
results$down <- exp(result$lower.CL)
results$up <- exp(result$upper.CL)
head(results)
```

```
##           Algae      mean      down      up
## 1           SSB01 0.1000000 0.03368898 0.2968330
## 2 M. gaditana 1.4565217 0.89382809 2.3734492
## 3 N. oculata 0.8529412 0.46106827 1.5778762
## 4 C. velia 0.4482759 0.20846399 0.9639614
```

```
write_csv(results, "2f_summary.csv")
```

```
# prepair for geom_signif
signi <- data.frame(start = factor(pairwise$start))
signi$end <- factor(pairwise$end)
signi$label <- factor(pairwise$label)
signi <- filter(signi, label != "")
signi$y <- seq(7.75, length.out=nrow(signi), by=-0.25)
signi
```

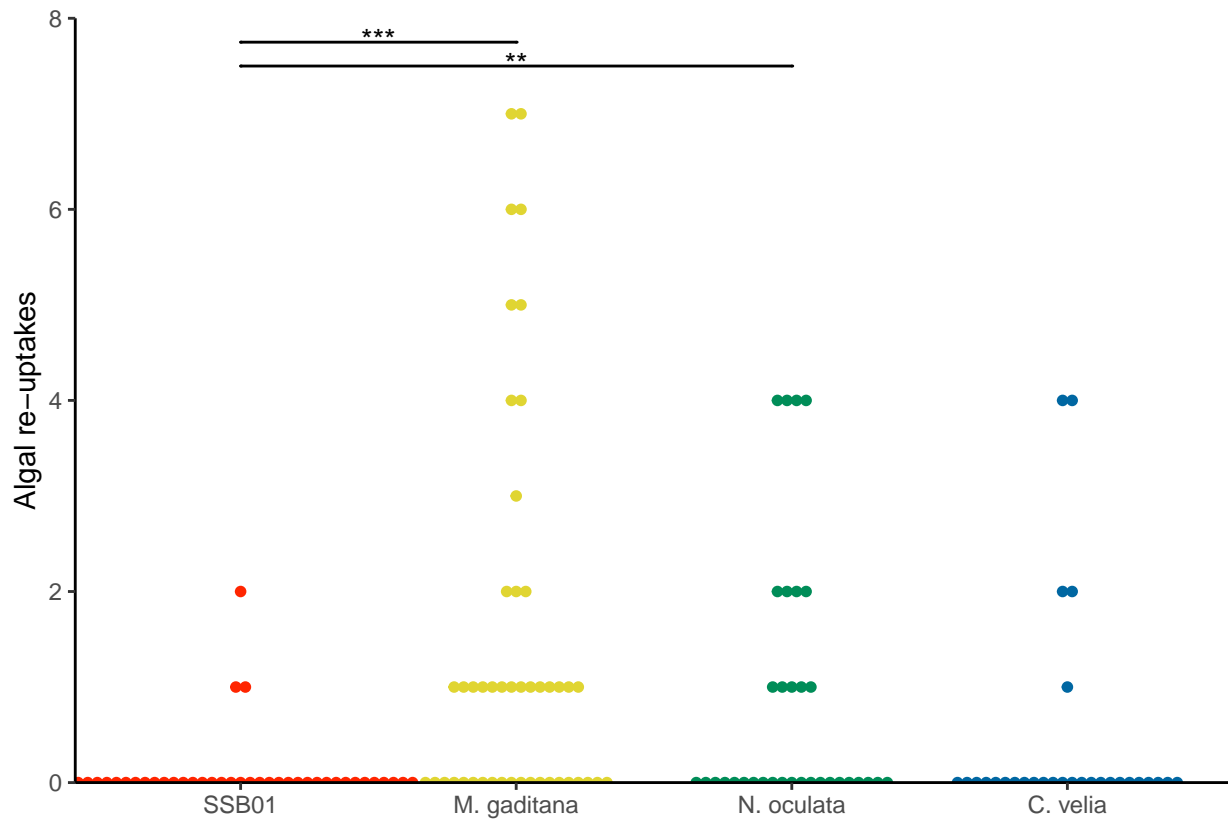
```
## start      end label      y
## 1 SSB01 M. gaditana *** 7.75
## 2 SSB01 N. oculata ** 7.50
```

```
#Plot graph
formatter60 <- function(x){
x/60
}
P1 <- ggplot() +
  geom_dotplot(data = dat, aes(x=Algae, y=uptakes, color = Algae, fill = Algae), binwidth=1,binaxis = "y",
  scale_color_manual(breaks = c("SSB01", "M. gaditana", "N. oculata", "C. velia"),
    values=c("#FF2500", "#E0D532", "#008D58", "#0067A2")) +
  scale_fill_manual(breaks = c("SSB01", "M. gaditana", "N. oculata", "C. velia"),
    values=c("#FF2500", "#E0D532", "#008D58", "#0067A2")) +
  theme_classic() +
  labs(x = "", y = "Algal re-uptakes") +
  geom_signif(data = signi , aes(xmin = start, xmax = end, annotations = label, y_position = y), tip_length=0.5,
  scale_y_continuous(expand = c(0, 0), limits = c(0,8)) # labels = formatter60, breaks=c(120,240,360,480))
```

```
## Warning: Ignoring unknown parameters: size
```

```
## Warning: Ignoring unknown aesthetics: xmin, xmax, annotations, y_position
```

```
plot(P1)
```



```
ggsave("Figure_2f.pdf",width=16,height=11,units="cm")
```

```
# get n count per treatment
ndat <- ddpoly(dat, c("Algae", "Treatment", "Repeat.Date."), summarise,
              N = length(Larvae))
n_Figure <- ddpoly(ndat, c("Algae", "Treatment"), summarise,
                  N = length(Repeat.Date.))
n_Figure
```

```
##      Algae Treatment N
## 1      SSB01      No 4
## 2 M. gaditana      No 4
## 3 N. oculata      No 4
## 4 C. velia      No 4
```

```
write_csv(n_Figure, "2f_n.csv")
```