Supplement 4

Jan Vicente

5/20/2020

```
library(knitr)
opts_chunk$set(tidy.opts=list(width.cutoff=60),tidy=TRUE)
```

Weight consumption (WC)

```
WC<-read_excel("Table S1.xls") #import Table S1
WC_df<-as.data.frame(WC)
WC_df$species <- as.factor(WC_df$species)
#ANOVA using linear model using function "anova"
WC.lme.base1<-lm(wc~species, data=WC_df)
WC_lme_base1_aov<-aov(WC.lme.base1)
```

WC:fitting a random effect

```
#Fitting a random effect
WC_lme_ran<-lme(wc ~  species, random=~ 1|trial, data=WC_df, method = "REML")
```

WC:comparing models

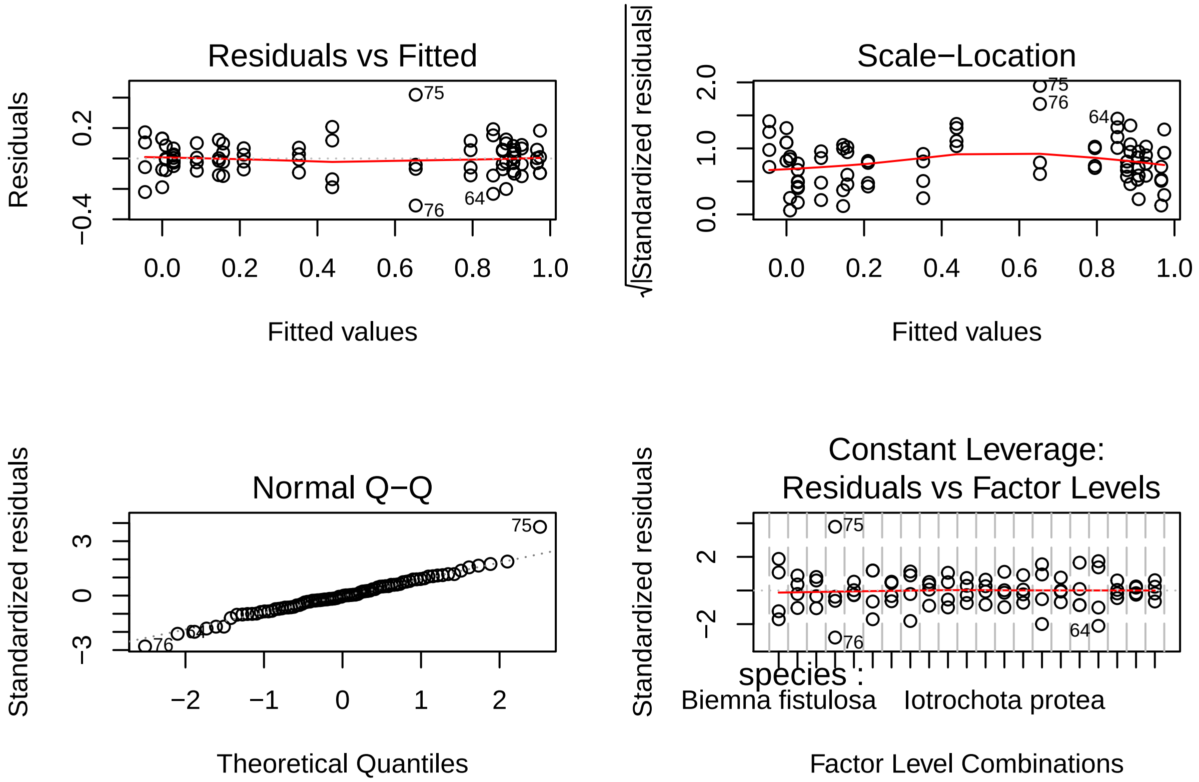
```
#compare model
testmodel<-anova(WC_lme_ran,
    WC.lme.base1)
testmodel
```

```
##              Model df       AIC      BIC   logLik   Test      L.Ratio p-value
## WC_lme_ran       1 23 -5.458549 43.83355 25.72927
## WC.lme.base1     2 22 -7.458549 39.69042 25.72927 1 vs 2 3.055028e-09       1
```

```
#best model is WC.lme.base1
```

WC:Test Homogoeneity of variance

```r
layout(matrix(c(1,2,3,4),2,2))
plot(WC_lme_base1_aov) #Several outliers are spotted.
```



##WC:Test HOV

```r
with(WC_df,leveneTest(wc~species)) #Does not pass HOV
```

```
## Levene's Test for Homogeneity of Variance (center = median)
##       Df F value  Pr(>F)
## group 20  2.0963 0.01373 *
##       63
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

WC:#remove only row 64 (Mycale parishi_trial4) from MC_df

```r
WC_df_no_64<-WC_df[-c(64), ]
WC_df_no_64.lme.base1<-lm(wc~species, data=WC_df_no_64)
WC_df_no_64.lme.base1_aov<-aov(WC_df_no_64.lme.base1)
summary(WC_df_no_64.lme.base1_aov)
```

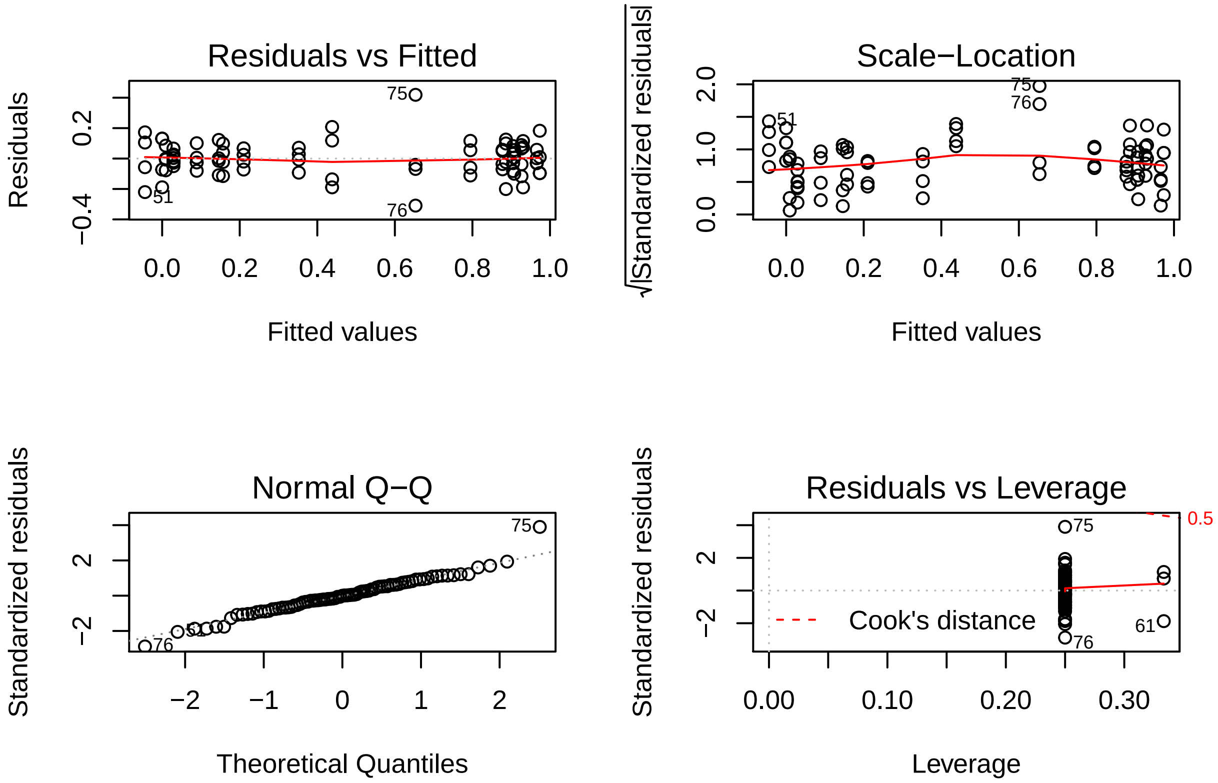
```
##               Df Sum Sq Mean Sq F value Pr(>F)
## species      20 12.960  0.6480   42.09 <2e-16 ***
## Residuals    62  0.955  0.0154
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
with(WC_df_no_64,leveneTest(wc~species))
```

```
## Levene's Test for Homogeneity of Variance (center = median)
##       Df F value  Pr(>F)
## group 20  1.5632 0.09241 .
##       62
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

WC:#visually look for HOV

```
#HOV assumption visual
layout(matrix(c(1,2,3,4),2,2))
plot(WC_df_no_64.lme.base1)
```



```
#Lot more equality of variance of the residulas
#qqplot looks better and variance seems alot more stable
#Lets test levene's test to see if HOV passes
```

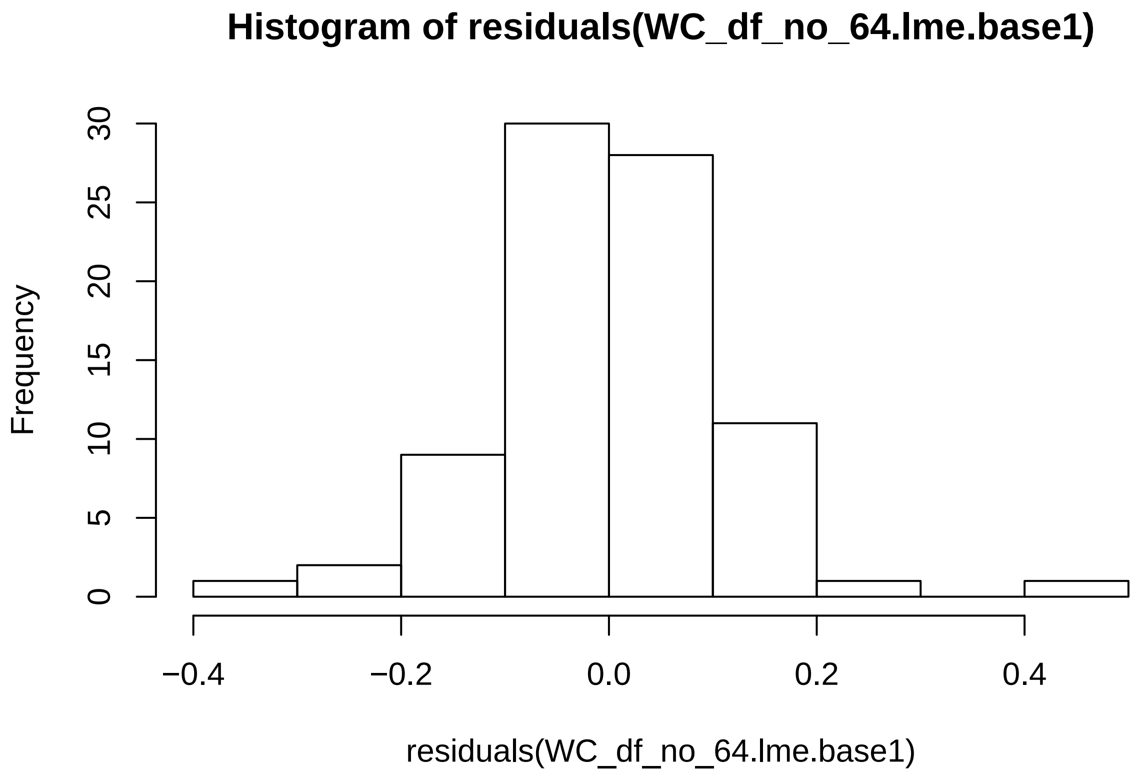
WC:Levene’s test HOV

```
#HOV assumption-Levene's test
with(WC_df_no_64,leveneTest(wc~species))
```

```
## Levene's Test for Homogeneity of Variance (center = median)
##       Df F value  Pr(>F)
## group 20  1.5632 0.09241 .
##       62
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

WC:histogram

```
hist(residuals(WC_df_no_64.lme.base1))
```



##WC:Anderson Darlington Normality test

```
#Anderson Darlington
ad.test(residuals(WC_df_no_64.lme.base1))
```

```
##
```

```
##  Anderson-Darling normality test
##
## data:  residuals(WC_df_no_64.lme.base1)
## A = 0.42987, p-value = 0.3016
```

WC:Camer-von Mises normality test

```
#Cramer-von Mises
cvm.test(residuals(WC_df_no_64.lme.base1))
```

```
##
##  Cramer-von Mises normality test
##
## data:  residuals(WC_df_no_64.lme.base1)
## W = 0.04989, p-value = 0.5077
```

WC:Lilliefors (Kolmogorov-Smirnov) normality test

```
lillie.test(residuals(WC_df_no_64.lme.base1))
```

```
##
##  Lilliefors (Kolmogorov-Smirnov) normality test
##
## data:  residuals(WC_df_no_64.lme.base1)
## D = 0.064016, p-value = 0.5486
```

WC:Pearson chi-square normality test

```
pearson.test(residuals(WC_df_no_64.lme.base1))
```

```
##
##  Pearson chi-square normality test
##
## data:  residuals(WC_df_no_64.lme.base1)
## P = 4.4699, p-value = 0.8779
```

pearson.test(residuals(WC_anova2)) Pearson chi-square normality test

data: residuals(WC_anova2) P = 2.2857, p-value = 0.9861

WC:ANOVA results of model (WC_df_no_64.lme.base1_aov)

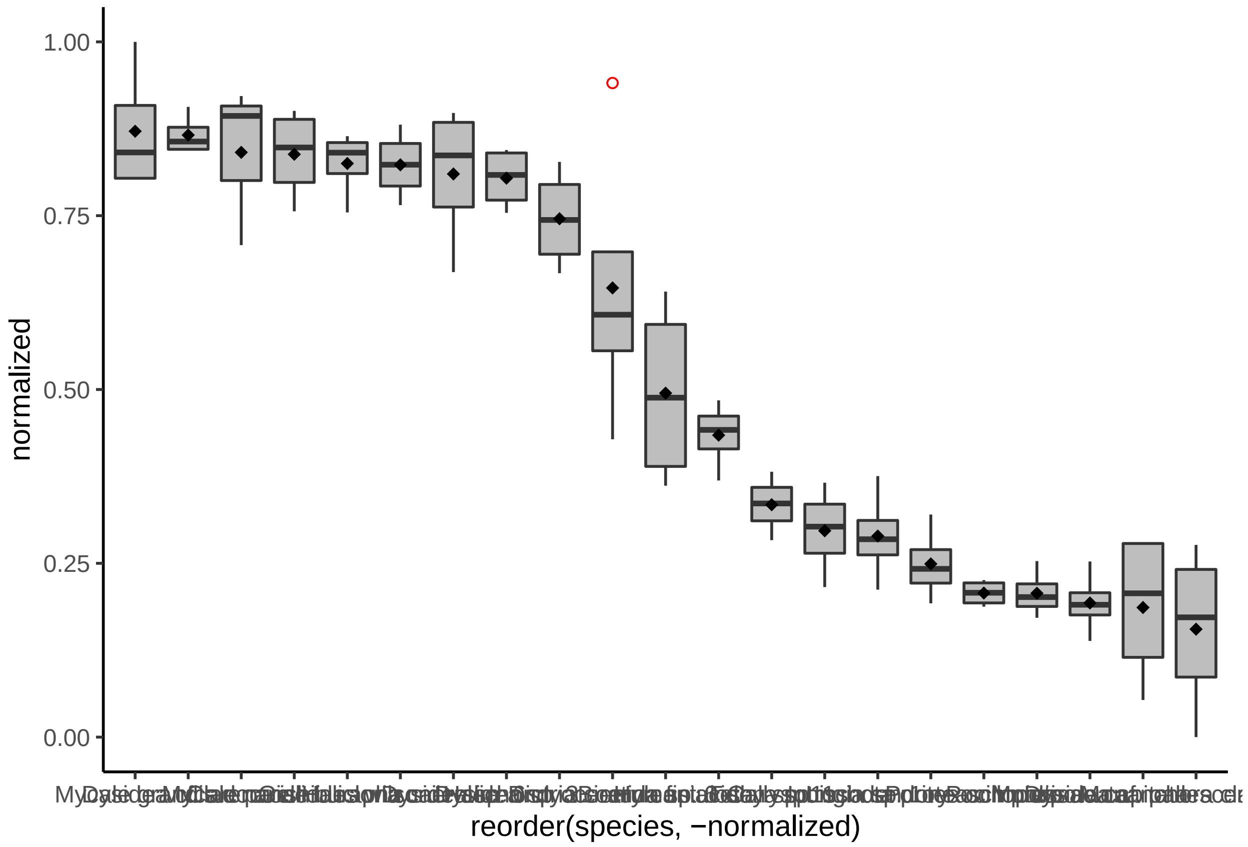
```
summary(WC_df_no_64.lme.base1_aov)
```

```
##              Df Sum Sq Mean Sq F value Pr(>F)    
## species      20 12.960  0.6480   42.09 <2e-16 ***
## Residuals    62  0.955  0.0154                   
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

WC:Tukey's post hoc analysis

```
#post-hoc analysis
WC_Tukey<-TukeyHSD(WC_df_no_64.lme.base1_aov)
WC_Tukey <- TukeyHSD(WC_df_no_64.lme.base1_aov)
# the [1:1] locates the part of the output to be exported
WC_Tukey_df<-as.data.frame(WC_Tukey[1:1])
# convert row name into first column
WC_Tukey_df<- cbind(rownames(WC_Tukey_df), data.frame(WC_Tukey_df, row.names=NULL))
write_csv(WC_Tukey_df, "WC_Tukey_df.csv")
#plot
Fig.2a <- ggplot(WC_df_no_64, aes(x=reorder(species,-normalized), y=normalized)) +
  stat_summary(fun=mean, geom="point", shape=23, size=1)+
  geom_boxplot(outlier.colour = "red", outlier.shape = 1, fill="grey")+
  stat_summary(fun=mean, geom="point", shape=18, size=2, color="black", fill="black")+
  theme_classic()

Fig.2a
```



##WC:Heatmap plot to generate Fig. 2b

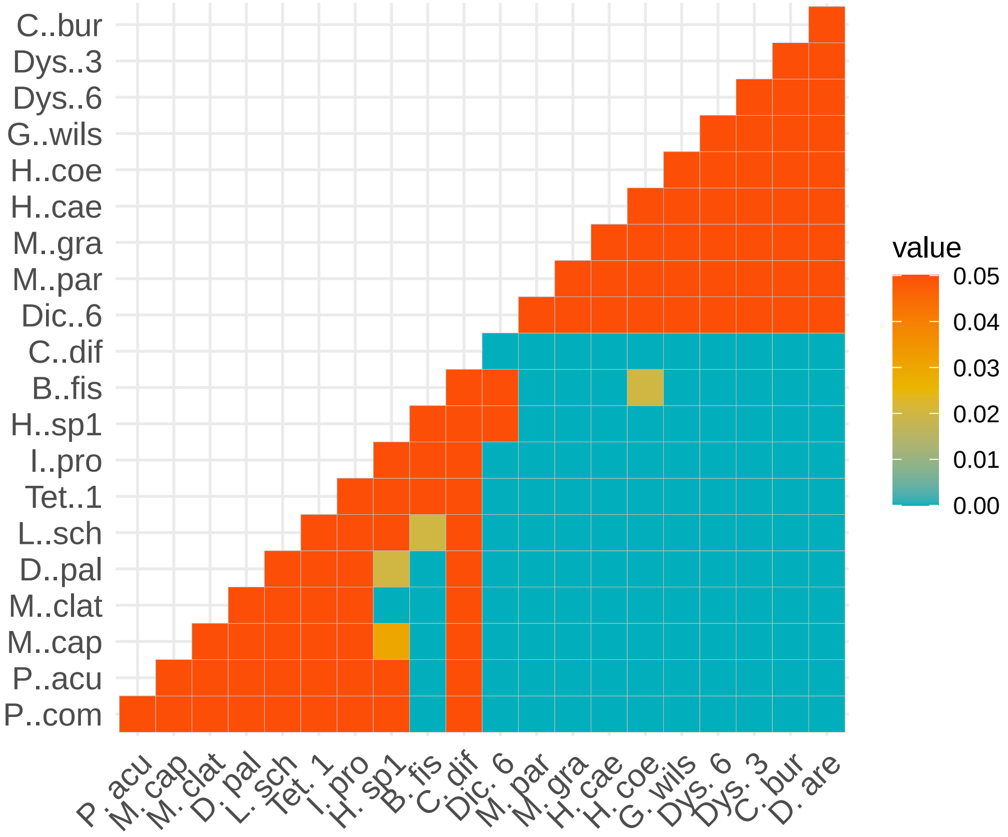
```
#heatmap to look at significant Tukey results and generate Fig. 2b. We reorganized
#significant p values from pairwise comparisons in Table S5 into a correlation matrix
#(TableS10).
#TableS10 is not provided.
hp<-read_excel("TableS10.xls")
```

```
## New names:
## * `` -> ...1
```

```
hp_df<-data.frame(hp, row.names=1)
hp_df[hp_df > .05] <- 0.05 #converting all values >0.05 to 0.05
hp_df.mat<-as.matrix(hp_df)
view(hp_df)
Fig.2b<-ggcorrplot(hp_df.mat, type="lower", hc.order=TRUE, method = "square") +
  scale_fill_gradient2(limit = c(0,0.05), low = "#00AFBB", high = "#FC4E07" ,
                       mid = "#E7B800", midpoint = 0.025)
```

```
## Scale for 'fill' is already present. Adding another scale for 'fill', which
## will replace the existing scale.
```

```
Fig.2b
```



SAC data analysis

```
# references
#https://ncss-wpengine.netdna-ssl.com/wp-content/themes/ncss/pdf/
#Procedures/NCSS/Mixed_Models-Repeated_Measures.pdf;
#https://rcompanion.org/handbook/I_09.html; https://ademos.people.uic.edu/Chapter18.html
##citation:Mangiafico, S.S. 2016. Summary and Analysis of Extension Program
##Evaluation in R, version 1.18.1. rcompanion.org/handbook/.
#(Pdf version rcompanion.org/documents/RHandbookProgramEvaluation.pdf.)

SAC<-read_excel("Table S2.xls")# Importing and formatting data xcel sheets to R
#statistical analysis
SAC_df<-as.data.frame(SAC)
SAC_df$correction<-as.numeric(SAC$correction)
SAC_df$hours <- factor(SAC$hours)#making hours a factor
```

SAC: fitting a linear model with fixed effects

```
#For the corAR1 structure, a value for the first order
#correlation can be specified. In this case,
#the value of 0.429 is found using the ACF function
#determining autocorrelation in residuals

#Fit linear model using generalized least squares
SAC.model.gls = gls(correction ~ species + hours + species*hours,
             data=SAC_df)

SAC.model.gls.ACF<-ACF(SAC.model.gls)
view(SAC.model.gls.ACF)
```

SAC:determining autocorrelation in residuals

SAC:linear mixed effects base model

```
SAC.model.lme = lme(correction ~ species + hours + species*hours,
            random = ~1|trial,
            data=SAC_df)
SAC.model.lme.ACF<-ACF(SAC.model.lme)
SAC.model.lme.ACF
```

```
##    lag           ACF
## 1    0  1.0000000000
## 2    1  0.5549906734
## 3    2  0.2866825429
## 4    3  0.1197080976
## 5    4 -0.0974173112
## 6    5 -0.1530004105
## 7    6 -0.2003259008
## 8    7 -0.1748970127
## 9    8 -0.1465801341
## 10   9 -0.0762675327
## 11  10 -0.0227106137
## 12  11  0.0217988360
## 13  12  0.0495616115
## 14  13  0.0003156655
## 15  14 -0.0397561815
## 16  15 -0.1433857252
## 17  16 -0.0827107547
## 18  17 -0.0441271181
```

```
# Value 0.5549906734 is the value for the first order correlation of model SAC.model.lme
```

SAC:fixed effects

```
#fixed effects model
SAC.model.fixed = gls(correction ~ species + hours + species*hours,
                  data=SAC_df,
                  method="REML")
```

SAC:linear mixed effects base mode with regressive lag structure

```
#linear mixed effects base model with autoregressive lag 1 structure
SAC.model.coAR1.rand = lme(correction ~ species + hours + species*hours,
            random = ~1|trial,
            correlation = corAR1(form = ~ 1|trial, value = 0.5550),
            data=SAC_df,
            method="REML")
```

SAC:comparing models

```
#test random effects
testmodel<-anova(SAC.model.fixed, SAC.model.coAR1.rand, SAC.model.lme)
testmodel
```

```
##                        Model df      AIC      BIC    logLik   Test  L.Ratio
## SAC.model.fixed            1 55 178.6604 348.4782 -34.33018
## SAC.model.coAR1.rand       2 57 121.9944 297.9874  -3.99721 1 vs 2 60.66594
## SAC.model.lme              3 56 179.6868 352.5922 -33.84341 2 vs 3 59.69239
##                        p-value
## SAC.model.fixed
## SAC.model.coAR1.rand  <.0001
## SAC.model.lme         <.0001
```

```
#random effects seems to be better fit  (AIC=121.99)
```

SAC:ANOVA analysis of the model

```
SAC.model.coAR1.rand_anova<-anova(SAC.model.coAR1.rand, type = "marginal")
SAC.model.coAR1.rand_anova
```

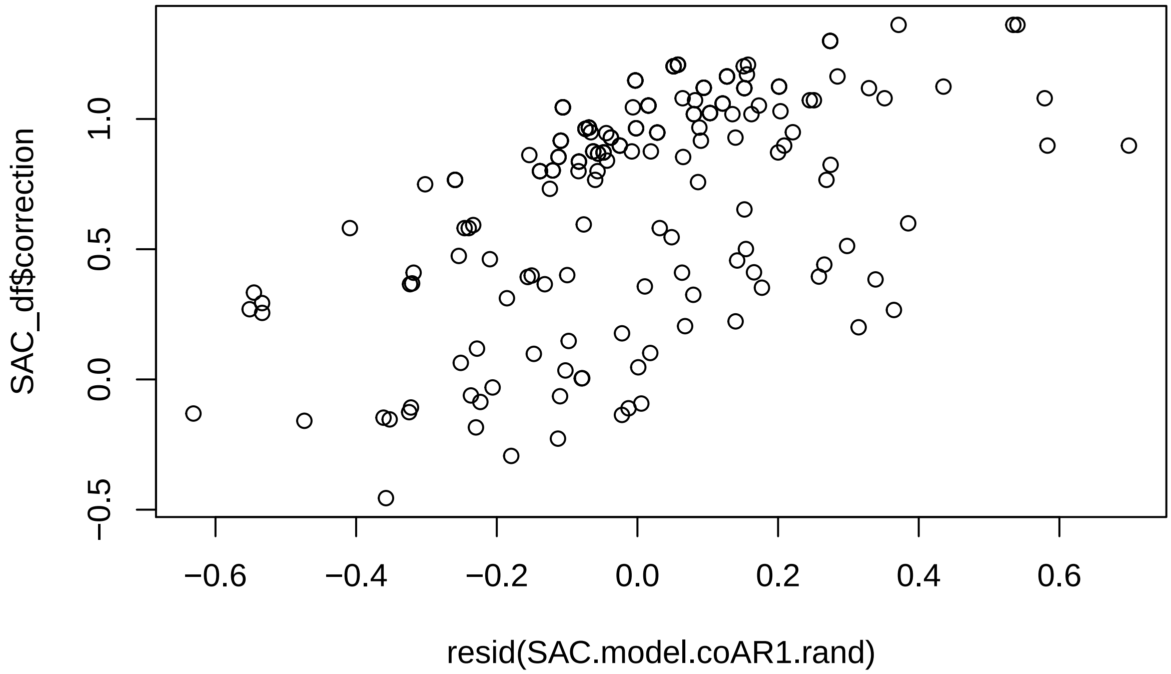
```
##                numDF denDF   F-value p-value
## (Intercept)        1   159  0.497054  0.4818
## species            8   159 11.596270  <.0001
## hours              5   159  9.499972  <.0001
## species:hours     40   159  2.563586  <.0001
```

SAC:Tukey's post hoc analysis to determine pairwise differences

```
SAC.model.coAR1.rand.tukey <- emmeans(SAC.model.coAR1.rand,
                                      list(pairwise ~ species*hours),
                                      adjust = "tukey", cld = FALSE)
SAC.model.coAR1.rand.tukey_df <-
  as.data.frame(SAC.model.coAR1.rand.tukey$`pairwise differences of species`)
write_csv(SAC.model.coAR1.rand.tukey_df, "SAC.model.coAR1.rand.tukey_df.csv")
#used to Table S2 and determine letter display which show significant
#differences between species Tukey's pairwise comparisons in Fig. 3.
```

SAC:Testing Linearity assumption

```
SAC.model.rand.lin<-plot(resid(SAC.model.coAR1.rand), SAC_df$correction)
```



```
#Seems to be randomnly distributed points
```

SAC:TEsting homogeneity of variace

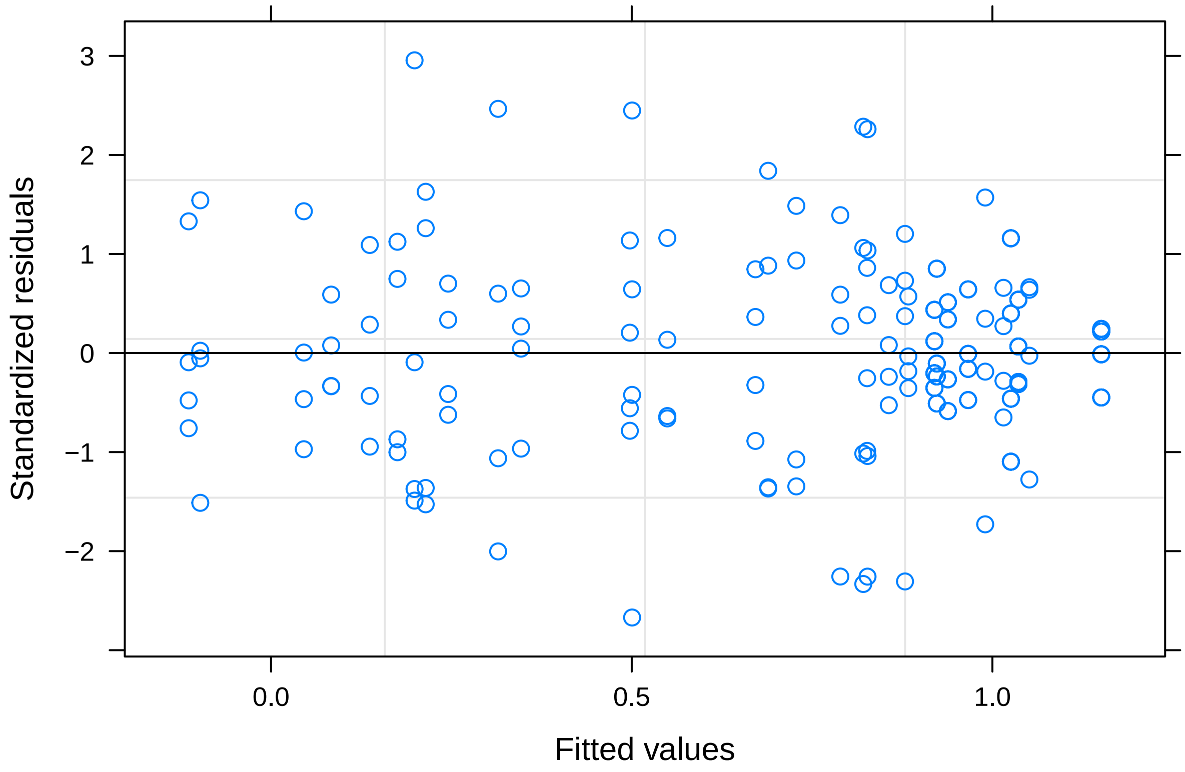
```
#extracts the residuals and places them in a new column in our original data table
SAC_df$mod_resid<- residuals(SAC.model.coAR1.rand)
#creates a new column with the absolute value of the residuals
SAC_df$Abs_mod_resid <-abs(SAC_df$mod_resid)
#squares the absolute values of the residuals to provide the more robust estimate
SAC_df$mod_resid2 <- SAC_df$mod_resid^2
#ANOVA of the squared residuals
Levene.SAC.model.rand <- lm(mod_resid2 ~ trial, data=SAC_df)
anova(Levene.SAC.model.rand)
```

```
## Analysis of Variance Table
##
## Response: mod_resid2
##            Df  Sum Sq   Mean Sq F value Pr(>F)
## trial       1 0.00001 0.0000130  0.0023 0.9621
## Residuals 214 1.23382 0.0057655
```

```
# variance of the residuals is equal because p value > 0.05.
```

SAC:Visual solution

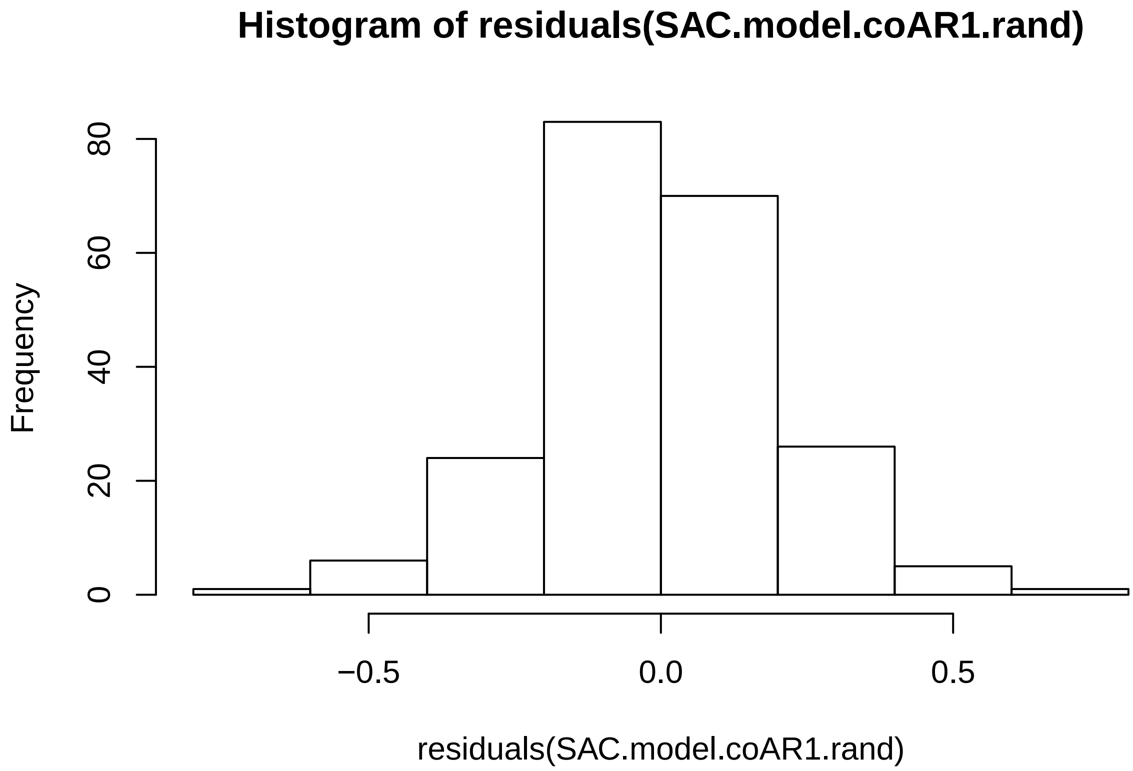
```
plot(SAC.model.coAR1.rand)
```



```
#Seems to show even spread around the centered line
```

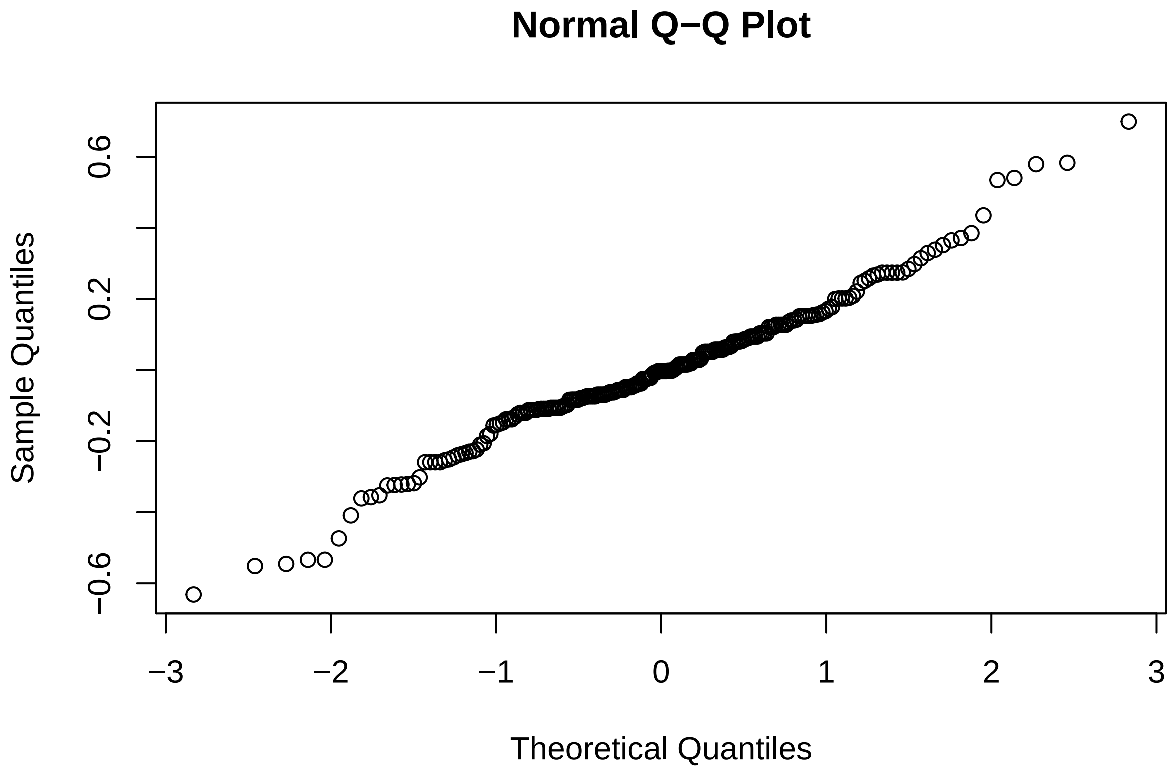
SAC:Histogram of model

```
hist(residuals(SAC.model.coAR1.rand))
```



##SAC:Assumption 3: Residuals of the model are normally distributed

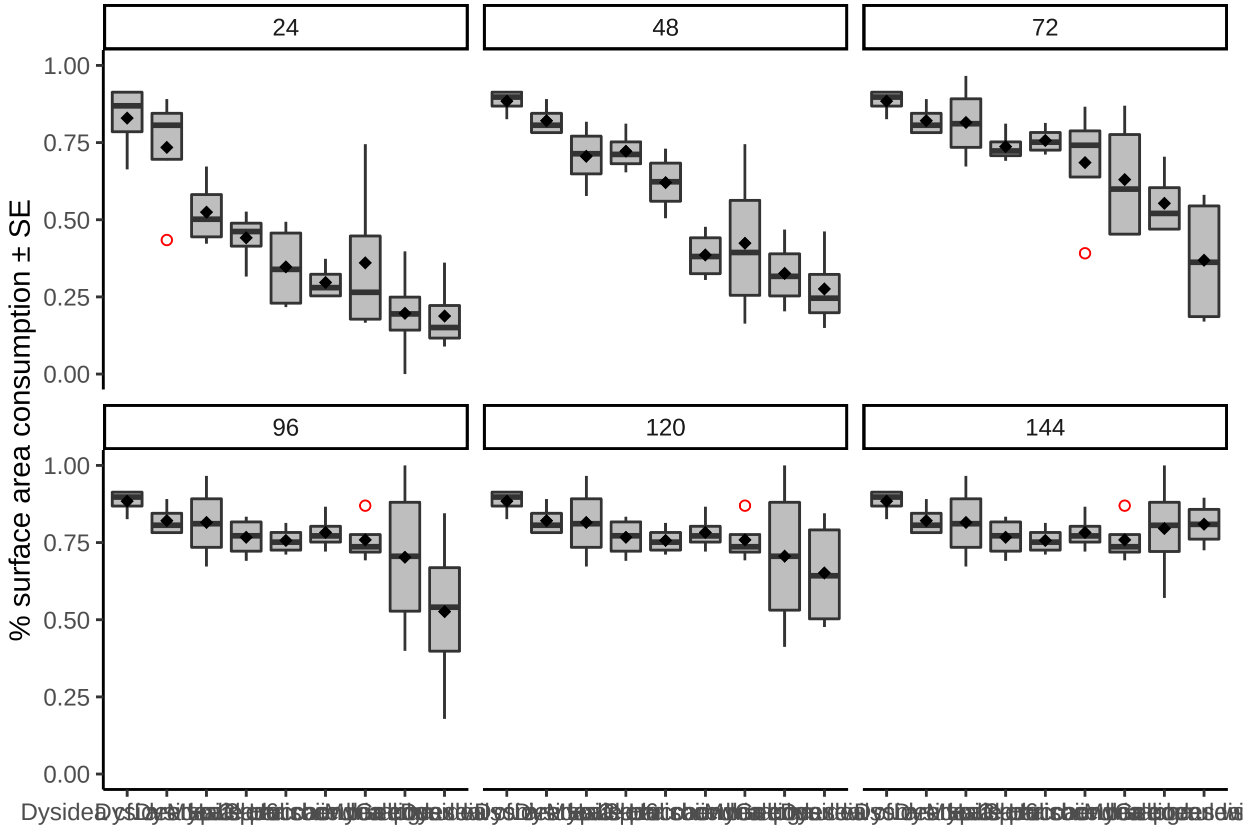
```
qqnorm(resid(SAC.model.coAR1.rand, Main="qqplot"))
```



```
#identifies outliers that may be exerting undue influence on the model.
#In this case there don't seem to be any outliers.
```

SAC:Subsetting the data to only include the first three time points for graphing purposes

```
Fig.3a<-ggplot(SAC_df, aes(x=reorder(species, -normalized), y=normalized)) +
  theme_classic() + theme(legend.title = element_blank(),axis.text.x =
                          element_text(angle = 45, hjust = 1, size = 6))+
  geom_boxplot(outlier.colour = "red", outlier.shape = 1, fill="grey")+
  stat_summary(fun=mean, geom="point", shape=18, size=2, color="black", fill="black")+
  theme_classic() +
  facet_wrap(~hours) +
  labs(y = "% surface area consumption ± SE", x = element_blank())
Fig.3a
```



SAC:Heatmasp for Fig. 3

SAC:heatmaps at 24 hour Fig. 3

```
#heatmap to look at significant Tukey results and generate Fig. 2b.
#We reorganized significant p values from pairwise comparisons in Table 7
#into a correlation matrix (hm24,hm48,hm72,hm96).These tables are not provided
hp24<-read_excel("hm24.xls")
```

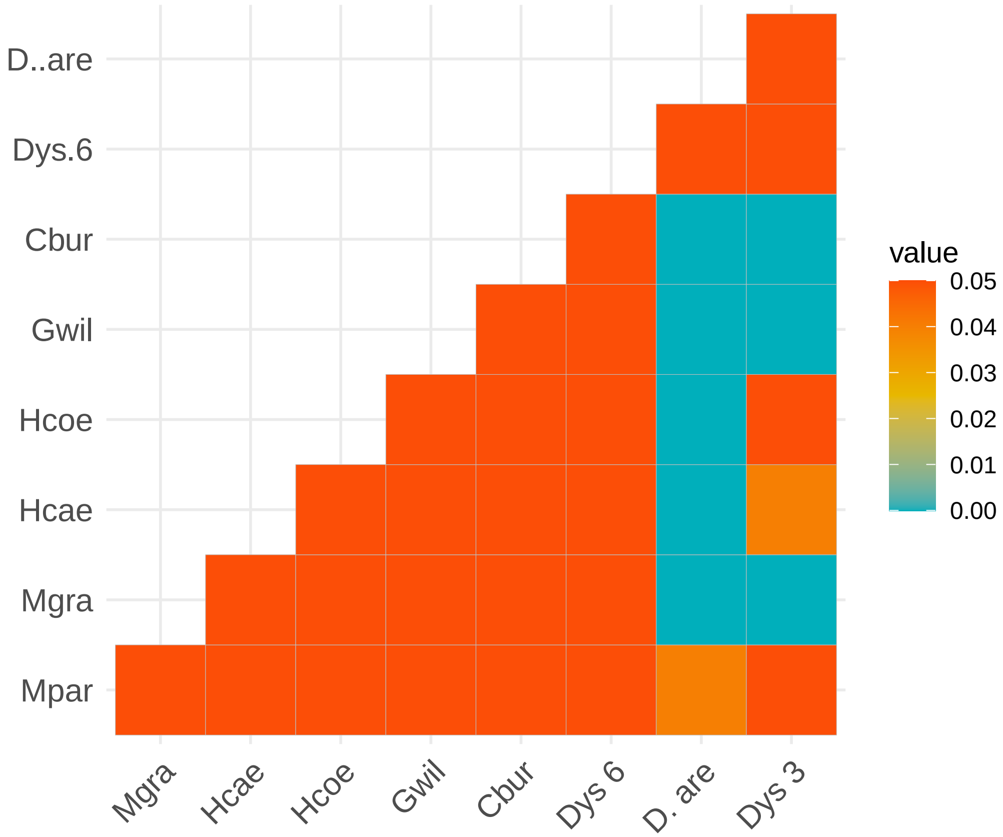
```
## New names:
## * `` -> ...1
```

```
hp_df24<-data.frame(hp24, row.names=1)
hp_df24[hp_df24 > .05] <- 0.05 #converting all values >0.05 to 0.05
hp_df24.mat<-as.matrix(hp_df24)
view(hp_df24)

Fig.3hm_24<-ggcorrplot(hp_df24.mat, type="lower", hc.order=TRUE, method = "square") +
  scale_fill_gradient2(limit = c(0,0.05), low = "#00AFBB", high = "#FC4E07" ,
                      mid = "#E7B800", midpoint = 0.025)
```

```
## Scale for 'fill' is already present. Adding another scale for 'fill', which
## will replace the existing scale.
```

```
Fig.3hm_24
```

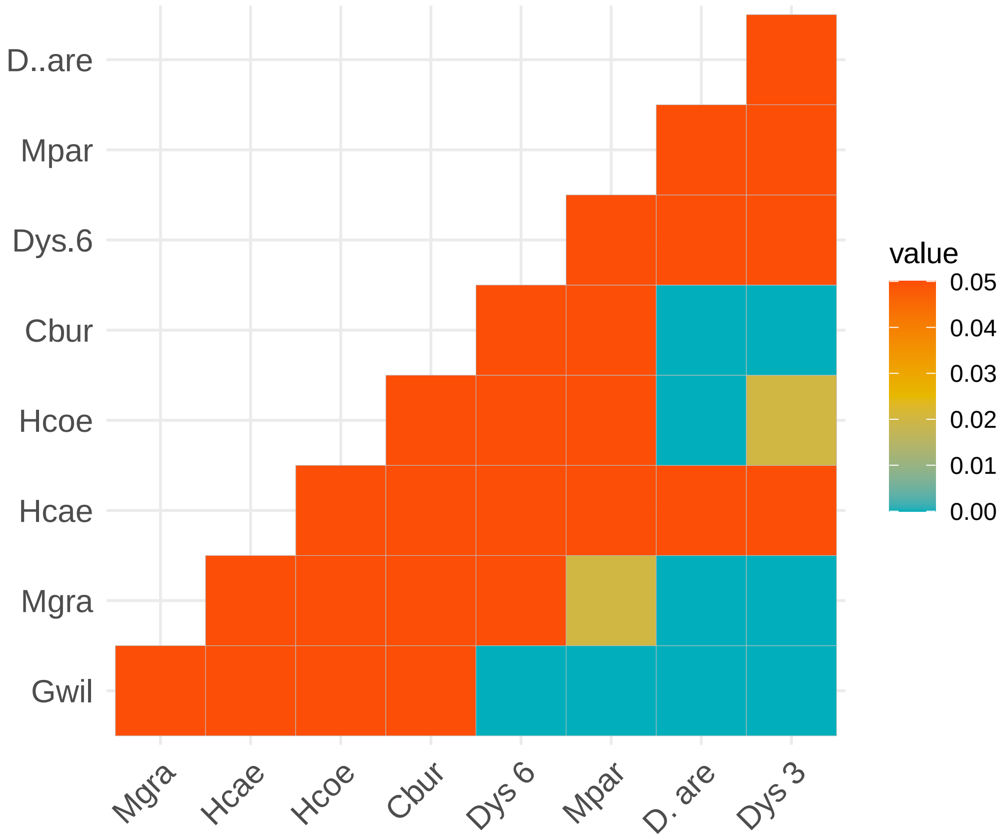


##SAC:heatmaps at 48 hour Fig. 3

```
hp48<-read_excel("hm48.xls")
hp_df48<-data.frame(hp48, row.names=1)
hp_df48[hp_df48 > .05] <- 0.05 #converting all values >0.05 to 0.05
hp_df48.mat<-as.matrix(hp_df48)
Fig.3hm_48<-ggcorrplot(hp_df48.mat, type="lower",
                       hc.order=TRUE, method = "square") +
  scale_fill_gradient2(limit = c(0,0.05), low = "#00AFBB",
                       high = "#FC4E07" , mid = "#E7B800", midpoint = 0.025)
```

```
## Scale for 'fill' is already present. Adding another scale for 'fill', which
## will replace the existing scale.
```

```
Fig.3hm_48
```

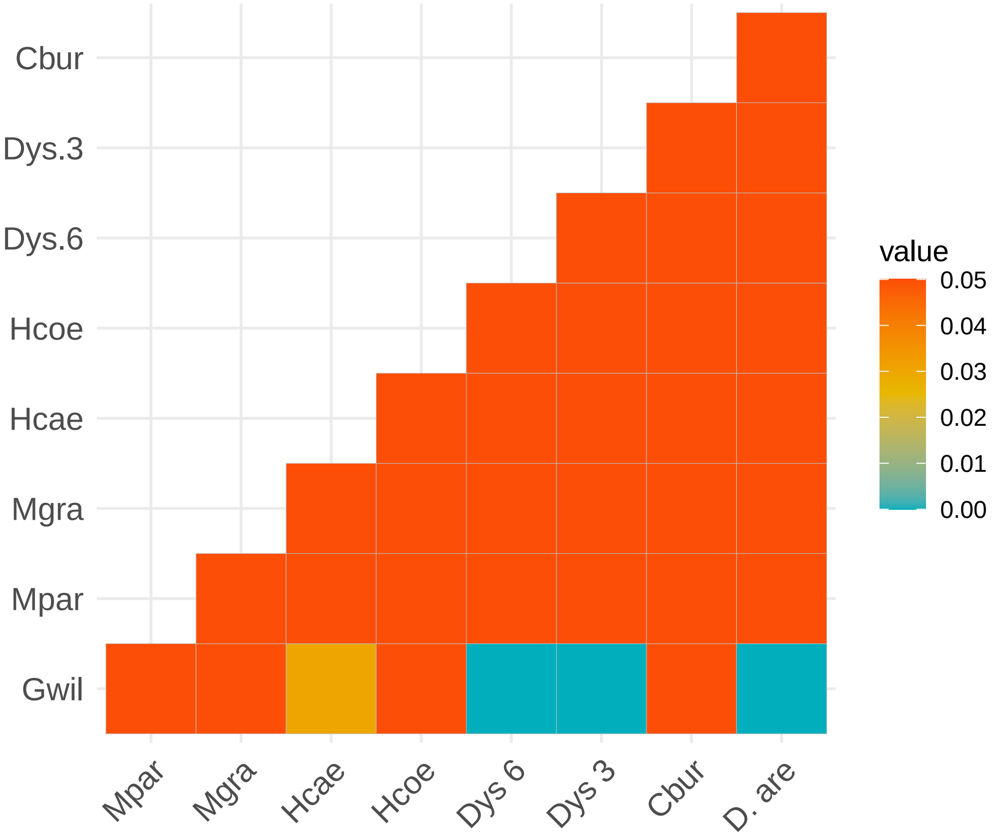


##SAC:heatmaps at 72 hour Fig. 3

```
hp72<-read_excel("hm72.xls")
hp_df72<-data.frame(hp72, row.names=1)
hp_df72[hp_df72 > .05] <- 0.05 #converting all values >0.05 to 0.05
hp_df72.mat<-as.matrix(hp_df72)
Fig.3hm_72<-ggcorrplot(hp_df72.mat, type="lower", hc.order=TRUE,
                       method = "square") +
  scale_fill_gradient2(limit = c(0,0.05),
                       low = "#00AFBB", high = "#FC4E07" , mid = "#E7B800",
                       midpoint = 0.025)
```

```
## Scale for 'fill' is already present. Adding another scale for 'fill', which
## will replace the existing scale.
```

```
Fig.3hm_72
```

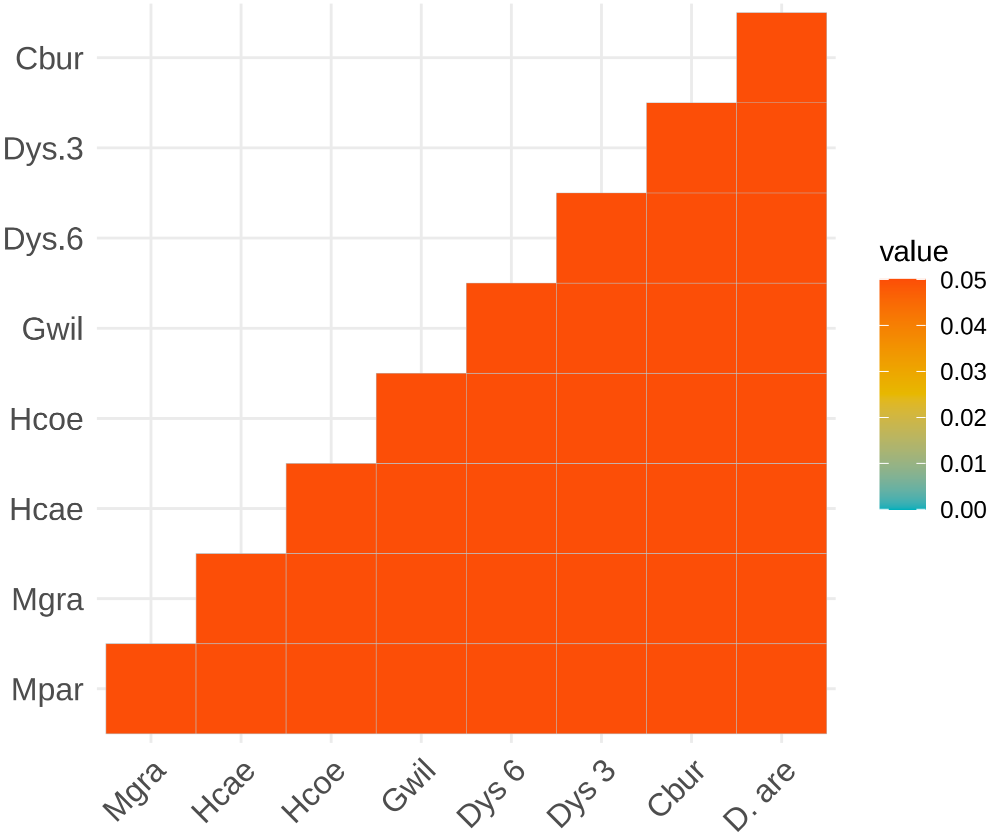


##SAC:heatmaps at 96 hour Fig. 3

```
hp96<-read_excel("hm96.xls")
hp_df96<-data.frame(hp96, row.names=1)
hp_df96[hp_df96 > .05] <- 0.05 #converting all values >0.05 to 0.05
hp_df96.mat<-as.matrix(hp_df96)
Fig.3hm_96<-ggcorrplot(hp_df96.mat, type="lower", hc.order=TRUE,
                       method = "square") + scale_fill_gradient2(limit = c(0,0.05),
                                                                  low = "#00AFBB",
                                                                  high = "#FC4E07" ,
                                                                  mid = "#E7B800",
                                                                  midpoint = 0.025)
```

```
## Scale for 'fill' is already present. Adding another scale for 'fill', which
## will replace the existing scale.
```

```
Fig.3hm_96
```

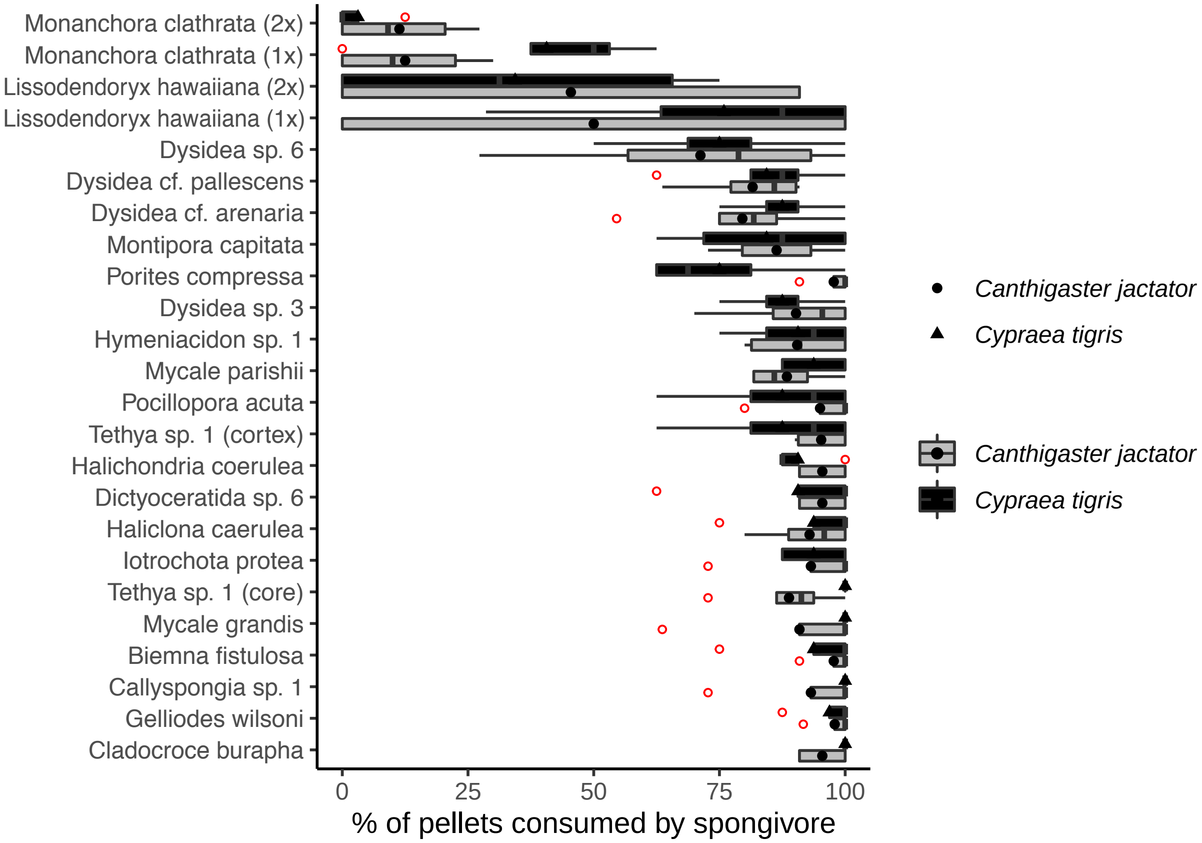


Note that the `echo = FALSE` parameter was added to the code chunk to prevent printing of the R code that generated the plot.

palatability assay of crude extracts from prey species (Fig.4)

```
###Chemical palatability script for Fig. 4####
chem <-read_excel("Table S9.xls")

Fig.4<-ggplot(chem, aes(x=reorder(Species, -PercentAccept), y=PercentAccept,
                        fill=Spongivore)) +
  geom_boxplot(outlier.colour = "red", outlier.shape = 1, outlier.size = 1) +
  scale_fill_manual("legend", values=c("Canthigaster jactator" = "grey",
                                       "Cypraea tigris" ="black")) + theme_classic() +
  theme(legend.title = element_blank(),
                         legend.text = element_text(face = "italic"))+
  labs(y = "% of pellets consumed by spongivore", x = element_blank())+
  stat_summary(fun=mean, geom = "point",
                                         size=1.5, aes(shape = Spongivore),
                                         position = position_dodge(width = .75))+
  rotate()
Fig.4
```


Monanchora clathrata (2x)
Monanchora clathrata (1x)
Lissodendoryx hawaiiana (2x)
Lissodendoryx hawaiiana (1x)
Dysidea sp. 6
Dysidea cf. pallescens
Dysidea cf. arenaria
Montipora capitata
Porites compressa
Dysidea sp. 3
Hymeniacidon sp. 1
Mycale parishii
Pocillopora acuta
Tethya sp. 1 (cortex)
Halichondria coerulea
Dictyoceratida sp. 6
Haliclona caerulea
Iotrochota protea
Tethya sp. 1 (core)
Mycale grandis
Biemna fistulosa
Callyspongia sp. 1
Gelliodes wilsoni
Cladocroce burapha
0
25
50
75
100
% of pellets consumed by spongivore
● Canthigaster jactator
▲ Cypraea tigris
Canthigaster jactator
Cypraea tigris

```
##END##
```