

1. Evolution of the biodiversity effects over time

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package loading

```
library(here)
```

```
## here() starts at /Users/meganedeziel/Documents/GitHub/Lemna_BEf
```

```
library(tidyverse)
```

```
## -- Attaching core tidyverse packages ----- tidyverse 2.0.0 --
```

```
## v dplyr      1.1.2      v readr      2.1.4
```

```
## v forcats    1.0.0      v stringr   1.5.0
```

```
## v ggplot2    3.4.2      v tibble    3.2.1
```

```
## v lubridate  1.9.2      v tidyr     1.3.0
```

```
## v purrr      1.0.1
```

```
## -- Conflicts ----- tidyverse_conflicts() --
```

```
## x dplyr::filter() masks stats::filter()
```

```
## x dplyr::lag()     masks stats::lag()
```

```
## i Use the conflicted package (<http://conflicted.r-lib.org/>) to force all conflicts to become errors
```

```
library(nlme)
```

```
##
```

```
## Attaching package: 'nlme'
```

```
##
```

```
## The following object is masked from 'package:dplyr':
```

```
##
```

```
##      collapse
```

```
library(emmeans)
```

```
library(ggpubr)
```

```
library(grid)
```

```
library(car)
```

```
## Loading required package: carData
```

```
##
```

```
## Attaching package: 'car'
```

```
##
```

```
## The following object is masked from 'package:dplyr':
```

```
##
```

```
##      recode
```

```
##
```

```
## The following object is masked from 'package:purrr':
```

```
##
```

```
##      some
```

data importation

```
#Data on Net effect, CE, SE
NE_data<-read.csv(here("data", "NE_data.csv"), sep=";")
str(NE_data)

## 'data.frame': 132 obs. of 8 variables:
## $ Polyculture: chr "JOB1LmSp" "JOB1LmWc" "JOB1LmLt" "JOB1SpWc" ...
## $ Block : int 1 1 1 1 1 1 1 1 1 ...
## $ Composition: chr "LmSp" "LmWc" "LmLt" "SpWc" ...
## $ Days : int 0 0 0 0 0 0 0 0 0 ...
## $ RYT : num 1 1 1 1 1 ...
## $ NE : num -1.093 -0.226 -2.291 -0.327 -0.186 ...
## $ CE : num 0 0 0 0 0 ...
## $ SE : num -1.093 -0.226 -2.291 -0.327 -0.186 ...

#transform Composition, Block, Days as factor for further analyses
NE_data<-NE_data[, -1]
NE_data<-NE_data %>%
  mutate(across(c("Composition", "Block", "Days"),
    as.factor))
```

test homoscedasticity of variance

```
# Plot Raw data - NE against Days
PlotNE <- ggplot(NE_data, aes(x = Days, y = NE)) +
  geom_jitter(position=position_jitter(0.2)) +
  stat_summary(fun.data="mean_sdl", fun.args = list(mult=1),
    geom="pointrange", color = "red")+
  theme_classic() +
  labs(x="Days", y="NE (mg)") +
  scale_y_continuous(limits=c(-2000, 2000), breaks=seq(-2000, 2000, by=500))

leveneTest(NE ~ Days, data=NE_data) #the variance among the factor Days is NOT equal

## Levene's Test for Homogeneity of Variance (center = median)
##      Df F value    Pr(>F)
## group  3 40.109 < 2.2e-16 ***
##      128
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

PlotCE <- ggplot(NE_data, aes(x = Days, y = CE)) +
  geom_jitter(position=position_jitter(0.2)) +
  stat_summary(fun.data="mean_sdl", fun.args = list(mult=1),
    geom="pointrange", color = "red")+
  theme_classic() +
  labs(x="Days", y="CE (mg)") +
  scale_y_continuous(limits=c(-2000, 2000), breaks=seq(-2000, 2000, by=500))

leveneTest(CE ~ Days, data=NE_data) #variance among the factor Days is NOT equal

## Levene's Test for Homogeneity of Variance (center = median)
##      Df F value    Pr(>F)
## group  3 43.943 < 2.2e-16 ***
##      128
```

```
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

PlotSE <- ggplot(NE_data, aes(x = Days, y = SE)) +
  geom_jitter(position=position_jitter(0.2)) +
  stat_summary(fun.data="mean_sdl", fun.args = list(mult=1),
    geom="pointrange", color = "red")+
  theme_classic() +
  labs(x="Days", y="SE (mg)") +
  scale_y_continuous(limits=c(-2000, 2000), breaks=seq(-2000, 2000, by=500))

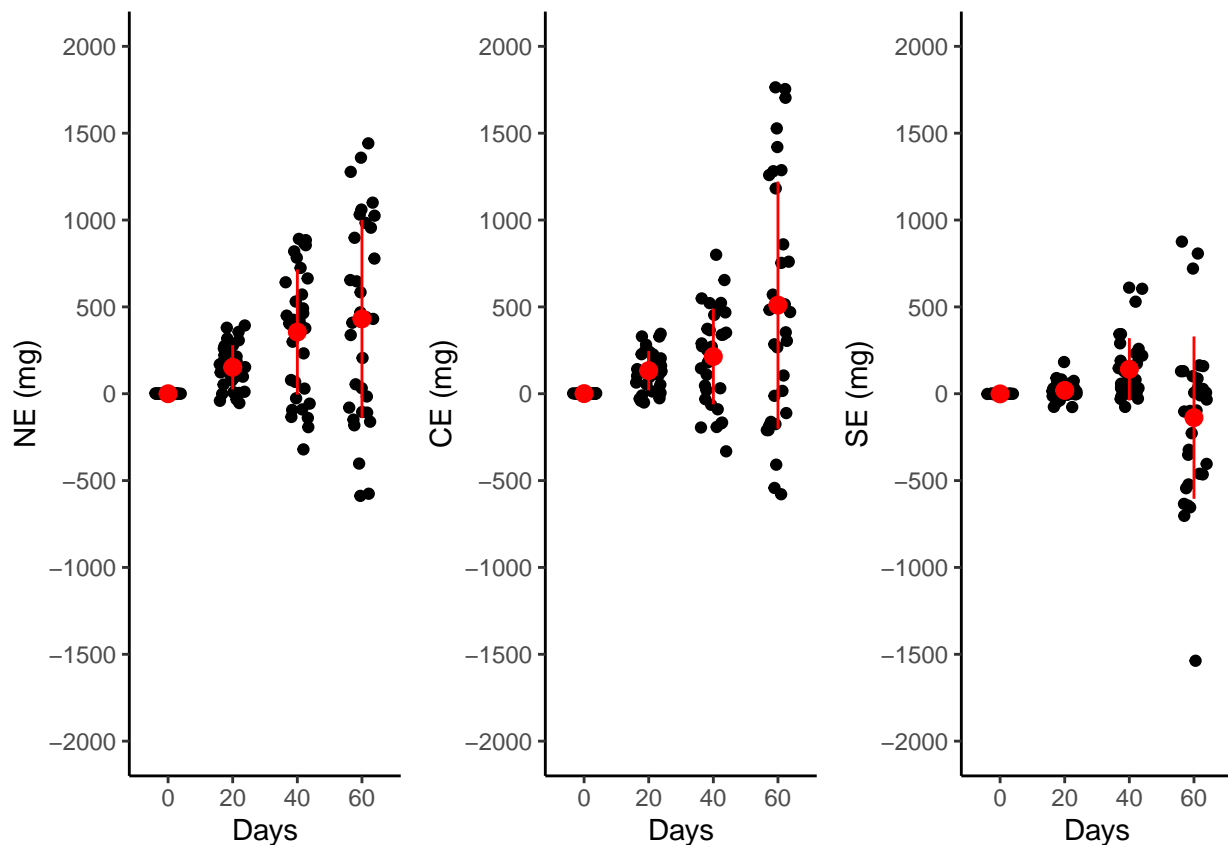
leveneTest(SE ~ Days, data=NE_data) #variance among the factor Days is NOT equal

## Levene's Test for Homogeneity of Variance (center = median)
##      Df F value    Pr(>F)
## group  3 21.849 1.724e-11 ***
##      128
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

#mean_sdl computes the mean plus or minus a constant times the standard deviation
 #(here the constant (mult) =1), so we see mean + standard deviation in Red
#We can clearly see that the variance of NE, CE and SE increases over time

ggarrange(PlotNE, PlotCE, PlotSE, nrow=1, ncol=3)

## Warning: Removed 1 rows containing non-finite values (`stat_summary()`).
## Warning: Removed 1 rows containing missing values (`geom_point()`).
```



why we use the nlme::lme function to compute the ANOVAs

Since the variance of NE, CE, SE increases over time (heteroscedasticity in the residuals), we decided to use the function nlme::lme to make our ANOVAs, because the function allows to weight the variance by the factor Days. Observations with higher variability will have lower weights, allowing the model to give them less influence. This method is particularly relevant with repeated measures data as we have here.

The class pdMat, which represent positive-definite matrices, is used to represent variance-covariance matrices of random effects. We have two blocks in the random effects variance-covariance matrix: one for Composition, and one for Block.

Sources: <https://stackoverflow.com/questions/36643713/how-to-specify-different-random-effects-in-nlme-vs-lme4> <https://biostatmatt.com/archives/2718>

The varIdent function allows different variances according to the levels of a classification factor.

ANOVA 1 : net effect through time

```
# Model testing the effect of Days on NE with Composition and Block as random factors and weighted by D

# Here we add a dummy factor just to include the whole dataset in a single block, to act as a grouping factor
NE_data$dummy<-factor(1)

#1.1-ASSESSING WHETHER THERE IS ANY STATISTICALLY SIGNIFICANT DIFFERENCE AMONG DAYS
#this first p-value aims to understand whether there is any statistically significant difference among days
#in the model, pdBlocked combines the covariance structures of pdIdent Composition and pdIdent Block (i.e. Block)
# -1 means that there is no default overall intercept being estimated for the factors Composition and Block
#pdIdent indicates that the random effects are assumed to have equal variances and no covariances.
#this whole syntax is actually a "trick" to specify non-nested crossed random effects in the nlme syntax
#an equivalent way to specify the random effects could be : random=list(dummy=pdBlocked(list(pdIdent(~Composition-1),
modNE <- lme(NC ~ Days,
             random=list(dummy=pdBlocked(list(pdIdent(~Composition-1),
                                             pdIdent(~Block-1))))),
             data=NE_data,
             weights=varIdent(form=~ 1 | Days), #residuals are allowed to have different variances for each day
             method="REML",
             control=list(msMaxIter=1000,
                          msMaxEval=1000))

anova(modNE) #there is at least one statistically significant difference among certain levels of factor Days

##              numDF denDF  F-value p-value
## (Intercept)      1   128  0.00909  0.9242
## Days              3   128 32.54603 <.0001

print(getVarCov(modNE))

## Random effects variance covariance matrix
##              CompositionLmLt CompositionLmSp CompositionLmSpLt
## CompositionLmLt          1.3912          0.0000          0.0000
## CompositionLmSp          0.0000          1.3912          0.0000
## CompositionLmSpLt        0.0000          0.0000          1.3912
## CompositionLmSpWc        0.0000          0.0000          0.0000
## CompositionLmSpWcLt      0.0000          0.0000          0.0000
## CompositionLmWc          0.0000          0.0000          0.0000
## CompositionLmWcLt        0.0000          0.0000          0.0000
```

## CompositionSpLt	0.0000	0.0000	0.0000	
## CompositionSpWc	0.0000	0.0000	0.0000	
## CompositionSpWcLt	0.0000	0.0000	0.0000	
## CompositionWcLt	0.0000	0.0000	0.0000	
## Block1	0.0000	0.0000	0.0000	
## Block2	0.0000	0.0000	0.0000	
## Block3	0.0000	0.0000	0.0000	
##	CompositionLmSpWc	CompositionLmSpWcLt	CompositionLmWc	
## CompositionLmLt	0.0000	0.0000	0.0000	
## CompositionLmSp	0.0000	0.0000	0.0000	
## CompositionLmSpLt	0.0000	0.0000	0.0000	
## CompositionLmSpWc	1.3912	0.0000	0.0000	
## CompositionLmSpWcLt	0.0000	1.3912	0.0000	
## CompositionLmWc	0.0000	0.0000	1.3912	
## CompositionLmWcLt	0.0000	0.0000	0.0000	
## CompositionSpLt	0.0000	0.0000	0.0000	
## CompositionSpWc	0.0000	0.0000	0.0000	
## CompositionSpWcLt	0.0000	0.0000	0.0000	
## CompositionWcLt	0.0000	0.0000	0.0000	
## Block1	0.0000	0.0000	0.0000	
## Block2	0.0000	0.0000	0.0000	
## Block3	0.0000	0.0000	0.0000	
##	CompositionLmWcLt	CompositionSpLt	CompositionSpWc	
## CompositionLmLt	0.0000	0.0000	0.0000	
## CompositionLmSp	0.0000	0.0000	0.0000	
## CompositionLmSpLt	0.0000	0.0000	0.0000	
## CompositionLmSpWc	0.0000	0.0000	0.0000	
## CompositionLmSpWcLt	0.0000	0.0000	0.0000	
## CompositionLmWc	0.0000	0.0000	0.0000	
## CompositionLmWcLt	1.3912	0.0000	0.0000	
## CompositionSpLt	0.0000	1.3912	0.0000	
## CompositionSpWc	0.0000	0.0000	1.3912	
## CompositionSpWcLt	0.0000	0.0000	0.0000	
## CompositionWcLt	0.0000	0.0000	0.0000	
## Block1	0.0000	0.0000	0.0000	
## Block2	0.0000	0.0000	0.0000	
## Block3	0.0000	0.0000	0.0000	
##	CompositionSpWcLt	CompositionWcLt	Block1	Block2
## CompositionLmLt	0.0000	0.0000	0.000000	0.000000
## CompositionLmSp	0.0000	0.0000	0.000000	0.000000
## CompositionLmSpLt	0.0000	0.0000	0.000000	0.000000
## CompositionLmSpWc	0.0000	0.0000	0.000000	0.000000
## CompositionLmSpWcLt	0.0000	0.0000	0.000000	0.000000
## CompositionLmWc	0.0000	0.0000	0.000000	0.000000
## CompositionLmWcLt	0.0000	0.0000	0.000000	0.000000
## CompositionSpLt	0.0000	0.0000	0.000000	0.000000
## CompositionSpWc	0.0000	0.0000	0.000000	0.000000
## CompositionSpWcLt	1.3912	0.0000	0.000000	0.000000
## CompositionWcLt	0.0000	1.3912	0.000000	0.000000
## Block1	0.0000	0.0000	0.039774	0.000000
## Block2	0.0000	0.0000	0.000000	0.039774
## Block3	0.0000	0.0000	0.000000	0.000000
##	Block3			
## CompositionLmLt	0.000000			

```
## CompositionLmSp      0.000000
## CompositionLmSpLt    0.000000
## CompositionLmSpWc    0.000000
## CompositionLmSpWcLt  0.000000
## CompositionLmWc      0.000000
## CompositionLmWcLt    0.000000
## CompositionSpLt      0.000000
## CompositionSpWc      0.000000
## CompositionSpWcLt    0.000000
## CompositionWcLt      0.000000
## Block1               0.000000
## Block2               0.000000
## Block3               0.039774
## Standard Deviations: 1.1795 1.1795 1.1795 1.1795 1.1795 1.1795 1.1795 1.1795 1.1795 1.1795 1.1795 1.1795
```

#1.2-ASSESSING OVERALL EFFECT OF DAYS : likelihood ratio test

#"when assessing the overall treatment effects using a likelihood ratio test, one should use maximum likelihood"

```
modNE.ml <- lme(Ne ~ Days,
  random=list(dummy=pdBlocked(list(pdIdent(~Composition-1),
                                   pdIdent(~Block-1)))),
  data=NE_data,
  weights=varIdent(form=~ 1 | Days),
  method="ML",
  control=list(msMaxIter=1000,
              msMaxEval=1000))
```

#null model

```
modNE0.ml <- lme(Ne ~ 1,
  random=list(dummy=pdBlocked(list(pdIdent(~Composition-1),
                                   pdIdent(~Block-1)))),
  data=NE_data,
  weights=varIdent(form=~ 1 | Days),
  method="ML",
  control=list(msMaxIter=1000,
              msMaxEval=1000))
```

#overall p-value modNE

anova(modNE0.ml, modNE.ml) #overall, the factor days has a statistically significant effect on net effe

```
##          Model df      AIC      BIC    logLik   Test  L.Ratio p-value
## modNE0.ml      1   7 1542.353 1562.532 -764.1764
## modNE.ml       2  10 1480.475 1509.303 -730.2372 1 vs 2 67.87832 <.0001
```

#1.3-COMPARISON BETWEEN EACH DAY AND DAY 0 (CONTROL LEVEL)

```
summary(modNE)
```

```
## Linear mixed-effects model fit by REML
```

```
## Data: NE_data
```

```
##      AIC      BIC    logLik
```

```
## 1451.485 1480.005 -715.7423
```

```
##
```

```
## Random effects:
```

```
## Composite Structure: Blocked
```

```
##
```

```
## Block 1: CompositionLmLt, CompositionLmSp, CompositionLmSpLt, CompositionLmSpWc, CompositionLmSpWcLt
```

```
## Formula: ~Composition - 1 | dummy
```

```
## Structure: Multiple of an Identity
##      CompositionLmLt CompositionLmSp CompositionLmSpLt CompositionLmSpWc
## StdDev:      1.179503      1.179503      1.179503      1.179503
##      CompositionLmSpWcLt CompositionLmWc CompositionLmWcLt CompositionSpLt
## StdDev:      1.179503      1.179503      1.179503      1.179503
##      CompositionSpWc CompositionSpWcLt CompositionWcLt
## StdDev:      1.179503      1.179503      1.179503
##
## Block 2: Block1, Block2, Block3
## Formula: ~Block - 1 | dummy
## Structure: Multiple of an Identity
##      Block1 Block2 Block3 Residual
## StdDev: 0.199435 0.199435 0.199435 0.2705557
##
## Variance function:
## Structure: Different standard deviations per stratum
## Formula: ~1 | Days
## Parameter estimates:
##      0      20      40      60
## 1.0000 471.8876 1334.1800 2108.5545
## Fixed effects: NE ~ Days
##      Value Std.Error DF t-value p-value
## (Intercept) 0.0349 0.37676 128 0.092725 0.9263
## Days20      152.2021 22.22487 128 6.848277 0.0000
## Days40      354.9528 62.83682 128 5.648802 0.0000
## Days60      430.9378 99.30807 128 4.339404 0.0000
## Correlation:
##      (Intr) Days20 Days40
## Days20 0
## Days40 0 0
## Days60 0 0 0
##
## Standardized Within-Group Residuals:
##      Min      Q1      Med      Q3      Max
## -1.87672122 -0.80706311 0.06059949 0.76522023 1.89863861
##
## Number of Observations: 132
## Number of Groups: 1
```

#here we can see that NE is significantly greater at day 20, at day 40 and at day 60 than at day 0

#WHICH TREATMENTS ARE DIFFERENT FROM EACH OTHER?

contrast(emmeans(modNE, specs="Days"), "pairwise") #the effect of days on NE isn't statistically differ

```
## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Composition' is absent, its contrast will be ignored

## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Block' is absent, its contrast will be ignored

## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Composition' is absent, its contrast will be ignored

## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Block' is absent, its contrast will be ignored

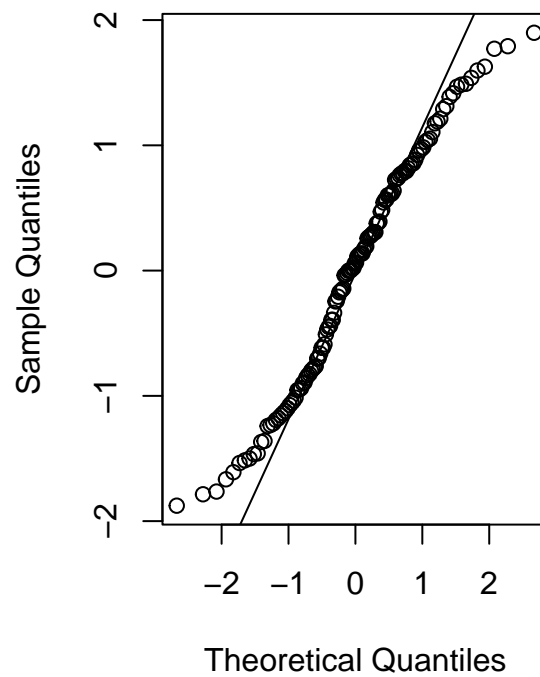
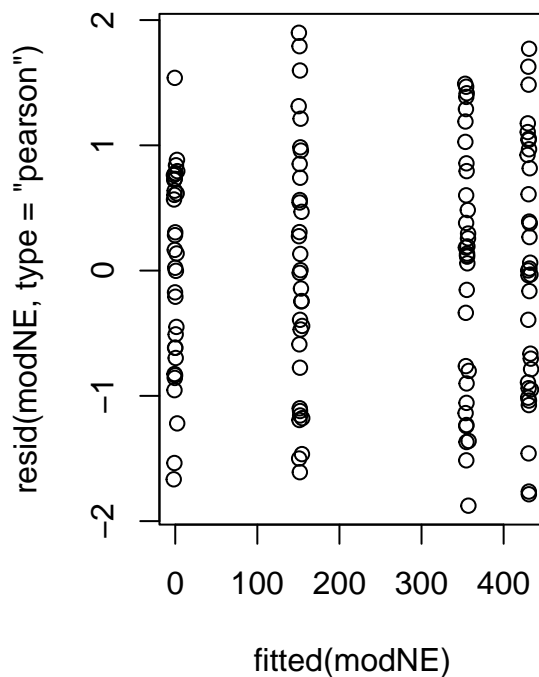
## contrast      estimate      SE df t.ratio p.value
```

```
## Days0 - Days20      -152  22.2 128  -6.848 <.0001
## Days0 - Days40      -355  62.8 128  -5.649 <.0001
## Days0 - Days60      -431  99.3 128  -4.339 0.0002
## Days20 - Days40     -203  66.7 128  -3.042 0.0149
## Days20 - Days60     -279 101.8 128  -2.739 0.0351
## Days40 - Days60      -76 117.5 128  -0.647 0.9166
##
## Degrees-of-freedom method: containment
## P value adjustment: tukey method for comparing a family of 4 estimates
```

#MODEL VALIDATION

```
par(mfrow=c(1,2))
plot(fitted(modNE), resid(modNE,type="pearson"))
qqnorm(resid(modNE,type="pearson"))
qqline(resid(modNE,type="pearson"))
```

Normal Q-Q Plot



##PLOT

```
hcl.colors(4, palette="Blue-Yellow")
```

```
## [1] "#2D3184" "#32AAB5" "#B3E7C5" "#F3F1E4"
```

#computing mean and standard deviation per day from raw data

```
mean<-as.vector(c(mean(NE_data[NE_data$Days==0, ]$NE), mean(NE_data[NE_data$Days==20, ]$NE), mean(NE_data[NE_data$Days==40, ]$NE), mean(NE_data[NE_data$Days==60, ]$NE)))
```

```
se<-as.vector(c(sd(NE_data[NE_data$Days==0, ]$NE)/sqrt(length(NE_data[NE_data$Days==0, ]$NE)), sd(NE_data[NE_data$Days==20, ]$NE)/sqrt(length(NE_data[NE_data$Days==20, ]$NE)), sd(NE_data[NE_data$Days==40, ]$NE)/sqrt(length(NE_data[NE_data$Days==40, ]$NE)), sd(NE_data[NE_data$Days==60, ]$NE)/sqrt(length(NE_data[NE_data$Days==60, ]$NE))))
```

```
NE_meanse<-data.frame(
  days=c("0", "20", "40", "60"),
  mean=mean,
  se=se,
  label=c("a", "b", "c", "c"))
```



```
NE_meanse$ci<-NE_meanse$se*1.96
```

```
NEp<-ggplot(NE_meanse) +
  geom_bar(aes(x=days, y=mean), colour="black", fill="#2D3184", stat="identity", alpha=0.7) +
  #geom_errorbar(aes(x=days, ymin=mean-se, ymax=mean+se), width=0.4, colour="red", alpha=0.9, size=1.3)
  geom_errorbar(aes(x=days, ymin=mean-ci, ymax=mean+ci), width=0.4, colour="black", alpha=0.9, size=1.3)
  geom_text(aes(x=days, y=mean+ci+100, label=label,), position=position_dodge(0.9), vjust=1, size=7) +
  theme_classic() +
  theme(text=element_text(size=15), plot.title=element_text(vjust=2), panel.grid.major.y = element_line
  labs(x="Days of growth", y="NE", title="Net biodiversity effect", pch=8) +
  scale_y_continuous(limits=c(0, 1000), breaks=seq(0, 1000, by=200))
```

```
## Warning: Using `size` aesthetic for lines was deprecated in ggplot2 3.4.0.
```

```
## i Please use `linewidth` instead.
```

```
## This warning is displayed once every 8 hours.
```

```
## Call `lifecycle::last_lifecycle_warnings()` to see where this warning was
```

```
## generated.
```

```
## Warning: The `size` argument of `element_line()` is deprecated as of ggplot2 3.4.0.
```

```
## i Please use the `linewidth` argument instead.
```

```
## This warning is displayed once every 8 hours.
```

```
## Call `lifecycle::last_lifecycle_warnings()` to see where this warning was
```

```
## generated.
```

ANOVA 2 : complementarity effect through time

```
modCE <- lme(CE ~ Days,
  random=list(dummy=pdBlocked(list(pdIdent(~Composition-1),
                                     pdIdent(~Block-1)))),
  data=NE_data,
  weights=varIdent(form=~ 1 | Days),
  method="REML",
  control=list(msMaxIter=1000,
               msMaxEval=1000))
anova(modCE) #there is at least one statistically significant difference among certain levels of factor
```

```
##          numDF denDF    F-value p-value
```

```
## (Intercept)      1   128  5.127877  0.0252
```

```
## Days              3   128 27.354690 <.0001
```

```
modCE.ml <- lme(CE ~ Days,
  random=list(dummy=pdBlocked(list(pdIdent(~Composition-1),
                                     pdIdent(~Block-1)))),
  data=NE_data,
  weights=varIdent(form=~ 1 | Days),
  method="ML",
  control=list(msMaxIter=1000,
               msMaxEval=1000))
```

```
#null model
```

```
modCE0.ml <- lme(CE ~ 1,
  random=list(dummy=pdBlocked(list(pdIdent(~Composition-1),
                                     pdIdent(~Block-1)))),
  data=NE_data,
  weights=varIdent(form=~ 1 | Days),
  method="ML",
```

```

control=list(msMaxIter=1000,
             msMaxEval=1000))

anova(modCEO.ml, modCE.ml) #overall, the factor days has a statistically significant effect on compleme

##           Model df          AIC          BIC    logLik    Test  L.Ratio p-value
## modCEO.ml      1   7 1447.319 1467.499 -716.6597
## modCE.ml       2  10 1394.119 1422.947 -687.0593 1 vs 2 59.20085 <.0001

summary(modCE)

## Linear mixed-effects model fit by REML
## Data: NE_data
##           AIC          BIC    logLik
## 1365.367 1393.888 -672.6836
##
## Random effects:
## Composite Structure: Blocked
##
## Block 1: CompositionLmLt, CompositionLmSp, CompositionLmSpLt, CompositionLmSpWc, CompositionLmSpWcLt
## Formula: ~Composition - 1 | dummy
## Structure: Multiple of an Identity
##           CompositionLmLt CompositionLmSp CompositionLmSpLt CompositionLmSpWc
## StdDev:      1.203187      1.203187      1.203187      1.203187
##           CompositionLmSpWcLt CompositionLmWc CompositionLmWcLt CompositionSpLt
## StdDev:      1.203187      1.203187      1.203187      1.203187
##           CompositionSpWc CompositionSpWcLt CompositionWcLt
## StdDev:      1.203187      1.203187      1.203187
##
## Block 2: Block1, Block2, Block3
## Formula: ~Block - 1 | dummy
## Structure: Multiple of an Identity
##           Block1      Block2      Block3      Residual
## StdDev: 0.02276269 0.02276269 0.02276269 0.04379333
##
## Variance function:
## Structure: Different standard deviations per stratum
## Formula: ~1 | Days
## Parameter estimates:
##           0          20          40          60
## 1.000 2592.952 6290.353 17723.190
## Fixed effects: CE ~ Days
##           Value Std.Error DF t-value p-value
## (Intercept) 0.8222 0.36309 128 2.264408 0.0252
## Days20      132.0337 19.76722 128 6.679426 0.0000
## Days40      213.3245 47.95413 128 4.448511 0.0000
## Days60      567.7921 135.11167 128 4.202391 0.0000
## Correlation:
## (Intr) Days20 Days40
## Days20 0
## Days40 0 0
## Days60 0 0 0
##
## Standardized Within-Group Residuals:

```

```
##           Min           Q1           Med           Q3           Max
## -1.98913850 -0.66697556  0.04653119  0.61160543  2.40300123
##
## Number of Observations: 132
## Number of Groups: 1
```

#here we can see that CE is significantly greater at day 20, at day 40 and at day 60 than at day 0

```
contrast(emmeans(modCE, specs="Days"), "pairwise") #the effect of days on CE isn't statistically differ
```

```
## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Composition' is absent, its contrast will be ignored
```

```
## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Block' is absent, its contrast will be ignored
```

```
## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Composition' is absent, its contrast will be ignored
```

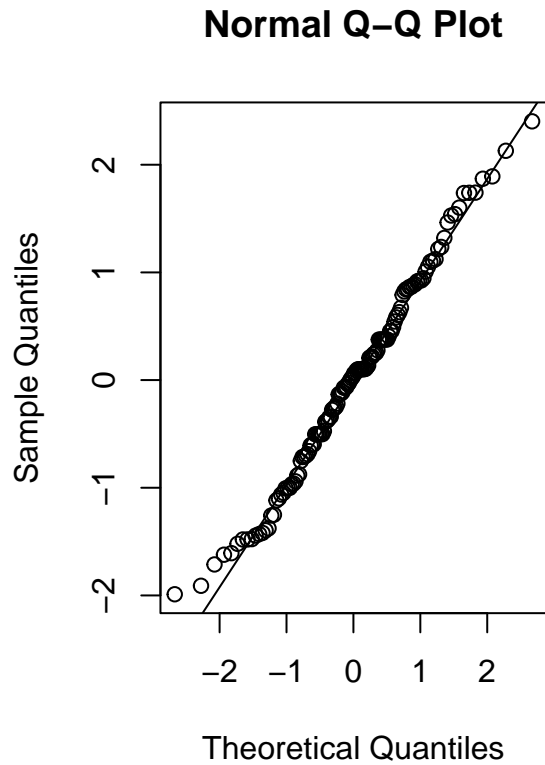
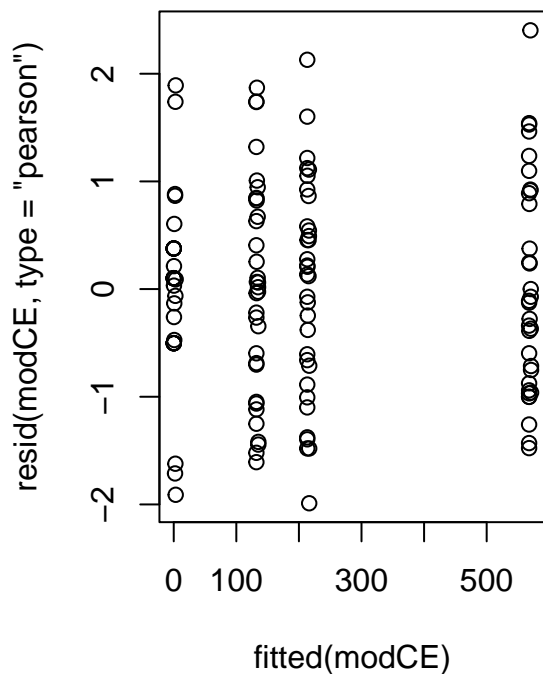
```
## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Block' is absent, its contrast will be ignored
```

```
## contrast      estimate    SE  df t.ratio p.value
## Days0 - Days20   -132.0   19.8 128  -6.679  <.0001
## Days0 - Days40   -213.3   48.0 128  -4.449  0.0001
## Days0 - Days60   -567.8  135.1 128  -4.202  0.0003
## Days20 - Days40   -81.3   51.9 128  -1.567  0.4010
## Days20 - Days60  -435.8  136.6 128  -3.191  0.0095
## Days40 - Days60  -354.5  143.4 128  -2.472  0.0692
##
```

```
## Degrees-of-freedom method: containment
```

```
## P value adjustment: tukey method for comparing a family of 4 estimates
```

```
par(mfrow=c(1,2))
plot(fitted(modCE), resid(modCE,type="pearson"))
qqnorm(resid(modCE,type="pearson"))
qqline(resid(modCE,type="pearson"))
```



```
meanCE<-as.vector(c(mean(NE_data[NE_data$Days==0, ]$CE), mean(NE_data[NE_data$Days==20, ]$CE), mean(NE_data[NE_data$Days==40, ]$CE), mean(NE_data[NE_data$Days==60, ]$CE)))
seCE<-as.vector(c(sd(NE_data[NE_data$Days==0, ]$CE)/sqrt(length(NE_data[NE_data$Days==0, ]$CE)), sd(NE_data[NE_data$Days==20, ]$CE)/sqrt(length(NE_data[NE_data$Days==20, ]$CE)), sd(NE_data[NE_data$Days==40, ]$CE)/sqrt(length(NE_data[NE_data$Days==40, ]$CE)), sd(NE_data[NE_data$Days==60, ]$CE)/sqrt(length(NE_data[NE_data$Days==60, ]$CE))))

CE_meanse<-data.frame(
  days=c("0", "20", "40", "60"),
  mean=meanCE,
  se=seCE,
  label=c("a", "b", "bc", "c"))
CE_meanse$ci<-CE_meanse$se*1.96

CEp<-ggplot(CE_meanse) +
  geom_bar(aes(x=days, y=mean), colour="black", fill="#2D3184", stat="identity", alpha=0.7) +
  #geom_errorbar(aes(x=days, ymin=mean-se, ymax=mean+se), width=0.4, colour="red", alpha=0.9, size=1.3)
  geom_errorbar(aes(x=days, ymin=mean-ci, ymax=mean+ci), width=0.4, colour="black", alpha=0.9, size=1.3)
  geom_text(aes(x=days, y=mean+ci+100, label=label), position=position_dodge(0.9), vjust=1, size=7) +
  theme_classic() +
  theme(text=element_text(size=15), plot.title=element_text(vjust=2), panel.grid.major.y = element_line(linetype="none"),
  labs(x="Days of growth", y="CE", title="Complementarity effect", pch=8) +
  scale_y_continuous(limits=c(0, 1000), breaks=seq(0, 1000, by=200))
```

ANOVA 3 : Selection effect through time

```
modSE <- lme(SE ~ Days,
  random=list(dummy=pdBlocked(list(pdIdent(~Composition-1),
    pdIdent(~Block-1)))),
  data=NE_data,
  weights=varIdent(form=~ 1 | Days),
  method="REML",
```

```

        control=list(msMaxIter=1000,
                     msMaxEval=1000))
anova(modSE) #there is at least one statistically significant difference among certain levels of factor

##           numDF denDF   F-value p-value
## (Intercept)      1   128 18.915587 <.0001
## Days              3   128  9.535597 <.0001

modSE.ml <- lme(SE ~ Days,
               random=list(dummy=pdBlocked(list(pdIdent(~Composition-1),
                                                  pdIdent(~Block-1)))),
               data=NE_data,
               weights=varIdent(form=~ 1 | Days),
               method="ML",
               control=list(msMaxIter=1000,
                           msMaxEval=1000))

#null model
modSE0.ml <- lme(SE ~ 1,
               random=list(dummy=pdBlocked(list(pdIdent(~Composition-1),
                                                  pdIdent(~Block-1)))),
               data=NE_data,
               weights=varIdent(form=~ 1 | Days),
               method="ML",
               control=list(msMaxIter=1000,
                           msMaxEval=1000))

anova(modSE0.ml, modSE.ml) #overall, the factor days has a statistically significant effect on compleme

##           Model df      AIC      BIC   logLik   Test  L.Ratio p-value
## modSE0.ml      1   7 1357.574 1377.754 -671.7871
## modSE.ml       2  10 1339.454 1368.282 -659.7269 1 vs 2 24.12035 <.0001

summary(modSE)

## Linear mixed-effects model fit by REML
##   Data: NE_data
##           AIC      BIC   logLik
##   1315.568 1344.089 -647.7842
##
## Random effects:
## Composite Structure: Blocked
##
## Block 1: CompositionLmLt, CompositionLmSp, CompositionLmSpLt, CompositionLmSpWc, CompositionLmSpWcL
## Formula: ~Composition - 1 | dummy
## Structure: Multiple of an Identity
##           CompositionLmLt CompositionLmSp CompositionLmSpLt CompositionLmSpWc
## StdDev:           0.4732538           0.4732538           0.4732538           0.4732538
##           CompositionLmSpWcLt CompositionLmWc CompositionLmWcLt CompositionSpLt
## StdDev:           0.4732538           0.4732538           0.4732538           0.4732538
##           CompositionSpWc CompositionSpWcLt CompositionWcLt
## StdDev:           0.4732538           0.4732538           0.4732538
##
## Block 2: Block1, Block2, Block3
## Formula: ~Block - 1 | dummy
## Structure: Multiple of an Identity

```

```
##           Block1    Block2    Block3 Residual
## StdDev: 0.1760824 0.1760824 0.1760824 0.2573057
##
## Variance function:
## Structure: Different standard deviations per stratum
## Formula: ~1 | Days
## Parameter estimates:
##           0         20         40         60
##    1.0000 199.7767 695.0623 1815.6250
## Fixed effects: SE ~ Days
##           Value Std.Error  DF    t-value p-value
## (Intercept)  -0.78725   0.18084 128 -4.353388  0.0000
## Days20        20.16838   8.94835 128  2.253867  0.0259
## Days40       141.62831  31.13268 128  4.549184  0.0000
## Days60       -136.85423  81.32397 128 -1.682828  0.0948
## Correlation:
##      (Intr) Days20 Days40
## Days20 -0.001
## Days40  0.000  0.000
## Days60  0.000  0.000  0.000
##
## Standardized Within-Group Residuals:
##           Min           Q1           Med           Q3           Max
## -2.99493534 -0.61529622 -0.04568812  0.56281949  3.16007848
##
## Number of Observations: 132
## Number of Groups: 1
```

#here we can see that CE is significantly greater at day 20, at day 40 than at day 0, but CE isn't sign

contrast(emmeans(modSE, specs="Days"), "pairwise") #the effect of days on SE is only different at Day 4

```
## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Composition' is absent, its contrast will be ignored
```

```
## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Block' is absent, its contrast will be ignored
```

```
## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Composition' is absent, its contrast will be ignored
```

```
## Warning in model.matrix.default(trms, m, contrasts.arg = contrasts): variable
## 'Block' is absent, its contrast will be ignored
```

```
## contrast      estimate    SE  df t.ratio p.value
## Days0 - Days20    -20.2   8.95 128  -2.254  0.1146
## Days0 - Days40   -141.6  31.13 128  -4.549  0.0001
## Days0 - Days60    136.9  81.32 128   1.683  0.3370
## Days20 - Days40  -121.5  32.39 128  -3.750  0.0015
## Days20 - Days60   157.0  81.81 128   1.919  0.2251
## Days40 - Days60   278.5  87.08 128   3.198  0.0093
##
```

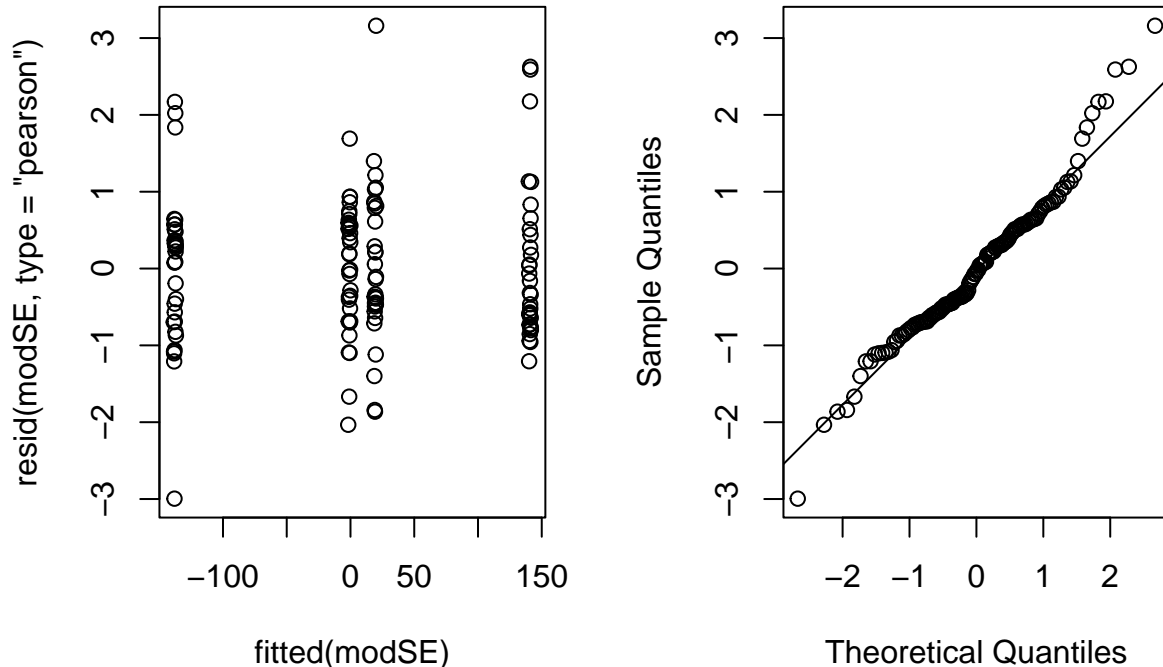
```
## Degrees-of-freedom method: containment
```

```
## P value adjustment: tukey method for comparing a family of 4 estimates
```

```
par(mfrow=c(1,2))
plot(fitted(modSE), resid(modSE,type="pearson"))
```

```
qqnorm(resid(modSE,type="pearson"))
qqline(resid(modSE,type="pearson"))
```

Normal Q-Q Plot



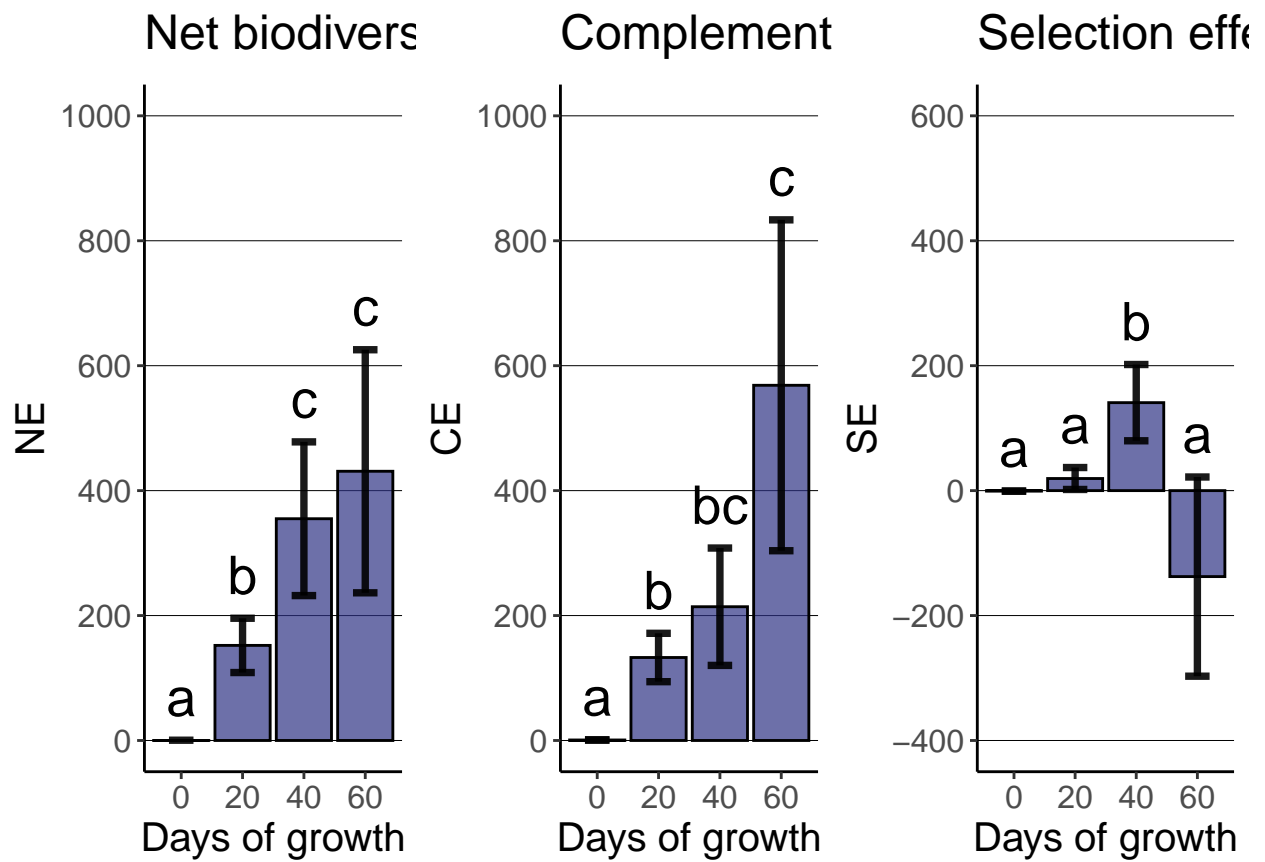
```
meanSE<-as.vector(c(mean(NE_data[NE_data$Days==0, ]$SE), mean(NE_data[NE_data$Days==20, ]$SE), mean(NE_data[NE_data$Days==40, ]$SE), mean(NE_data[NE_data$Days==60, ]$SE)))
seSE<-as.vector(c(sd(NE_data[NE_data$Days==0, ]$SE)/sqrt(length(NE_data[NE_data$Days==0, ]$SE)), sd(NE_data[NE_data$Days==20, ]$SE)/sqrt(length(NE_data[NE_data$Days==20, ]$SE)), sd(NE_data[NE_data$Days==40, ]$SE)/sqrt(length(NE_data[NE_data$Days==40, ]$SE)), sd(NE_data[NE_data$Days==60, ]$SE)/sqrt(length(NE_data[NE_data$Days==60, ]$SE))))

SE_meanse<-data.frame(
  days=c("0", "20", "40", "60"),
  mean=meanSE,
  se=seSE,
  label=c("a", "a", "b", "a"))
SE_meanse$ci<-SE_meanse$se*1.96

SEp<-ggplot(SE_meanse) +
  geom_bar(aes(x=days, y=mean), colour="black", fill="#2D3184", stat="identity", alpha=0.7) +
  #geom_errorbar(aes(x=days, ymin=mean-se, ymax=mean+se), width=0.4, colour="red", alpha=0.9, size=1.3)
  geom_errorbar(aes(x=days, ymin=mean-ci, ymax=mean+ci), width=0.4, colour="black", alpha=0.9, size=1.3)
  geom_text(aes(x=days, y=mean+ci+100, label=label,), position=position_dodge(0.9), vjust=1, size=7) +
  theme_classic() +
  theme(text=element_text(size=15), plot.title=element_text(vjust=2), panel.grid.major.y = element_line())
  labs(x="Days of growth", y="SE", title="Selection effect", pch=8) +
  scale_y_continuous(limits=c(-400, 600), breaks=seq(-400, 600, by=200))
```

###Final plots BEF through time

```
BEFplots<-ggarrange(NEp, CEp, SEp, nrow=1, ncol=3, common.legend = TRUE)
BEFplots
```



```
#annotate_figure(BEFplots, bottom = textGrob("Days of growth", rot = 90, vjust = 1, gp = gpar(cex = 1.3
```