Ulva and Hypnea Photosynthesis and GROWTH Run5 - Sept-Nov 2021

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run5.6 GROWTH RATE Analysis, Script Chunks, and Plots

This is the analysis of the final run of the Ulva and Hypnea salinity and nutrient experiments conducted on the lanai in St. John 616. These experiments incorporated three temperature levels. Data gaps for both species filled by end of April 2022. This output reflects all data totally five treatments for each species.

Packages loaded:

```
library(lme4)
library(lmerTest)
library(effects)
library(car)
library(MuMIn)
library (dplyr)
library(emmeans)
library(DHARMa)
library(performance)
library(patchwork)
```

Load and prepare the dataset

Open growth/weight dataset

```
run5.6_growth <- read.csv("/Users/Angela/src/work/limu/algal_growth_photosynthesis/data/run5-6_growth_a
```

Make a new column for weight change (difference final from initial)

```
run5.6_growth$growth_rate_percent <-
  (run5.6_growth$final.weight - run5.6_growth$Initial.weight) / run5.6_growth$Initial.weight * 100
```

Also make a new column for daily growth rate from 8 day study (steady growth rate assumed rather than exponential), which may or may not be used

```
run5.6_growth$steady_growth_daily <- run5.6_growth$growth_rate_percent / 8
```

Make a new column that keeps only the numerical values for temperature (removes C)

```
run5.6_growth$temp_clean <- as.factor(substr(run5.6_growth$temperature, 1, 2))
```

Change levels to factors

```
run5.6_growth$temperature <- as.factor(run5.6_growth$temp_clean)
run5.6_growth$run <- as.factor(run5.6_growth$run)
run5.6_growth$treatment <- as.factor(as.character(run5.6_growth$treatment))
```

Create subset of the data to isolate the species

```
hypnea <- subset(run5.6_growth, Species == "Hm")
ulva <- subset(run5.6_growth, Species == "Ul")
```

Run the model

ULVA

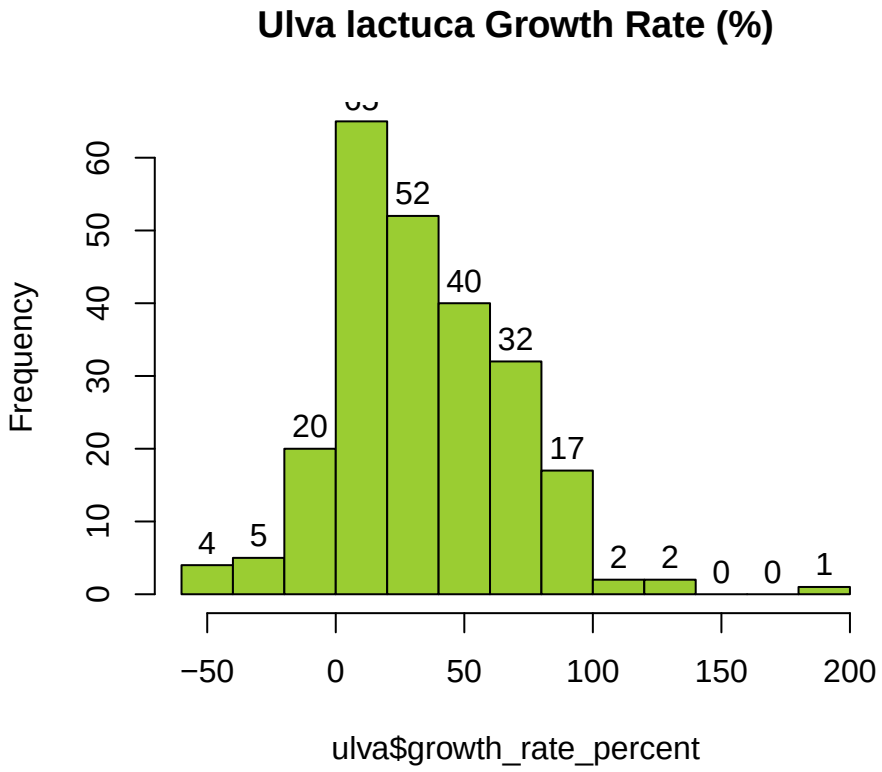
run model without interaction since interaction caused collinearity issues

```
run5.6_growth_model_noint <- lmer(formula = growth_rate_percent ~ treatment +
                                    temperature + (1 | run) + (1 | plant.ID) +
                                    (1 | RLC.order), data = ulva)
```

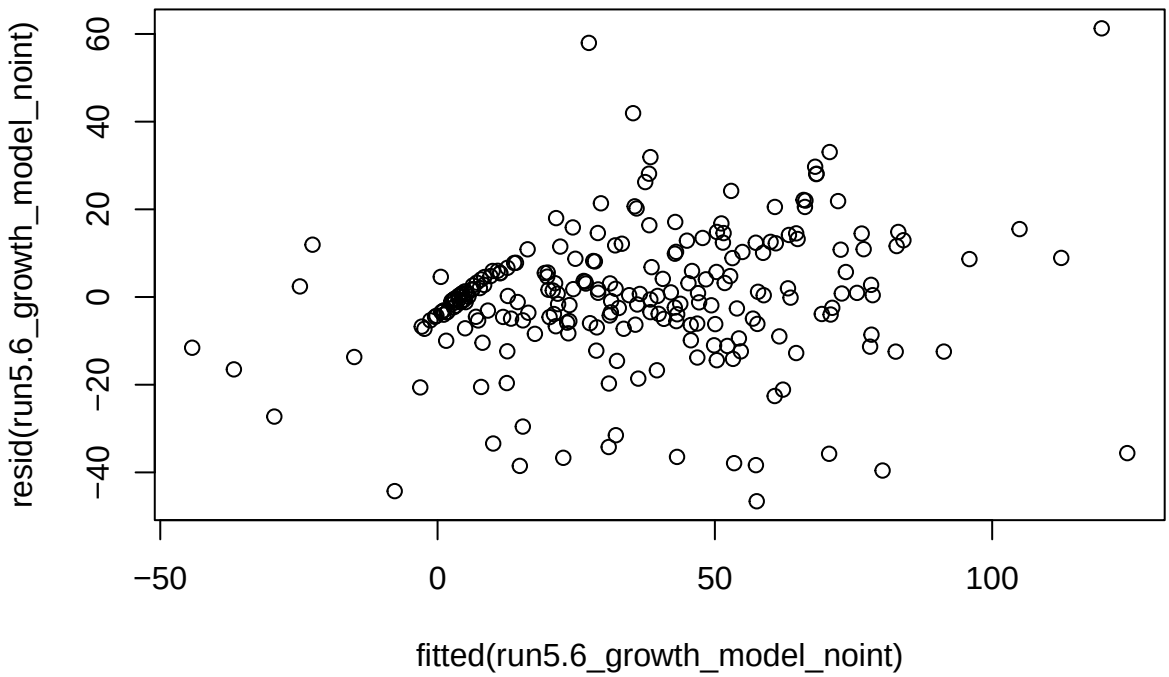
```
## boundary (singular) fit: see ?isSingular
```

#make a histogram of the data for Ulva

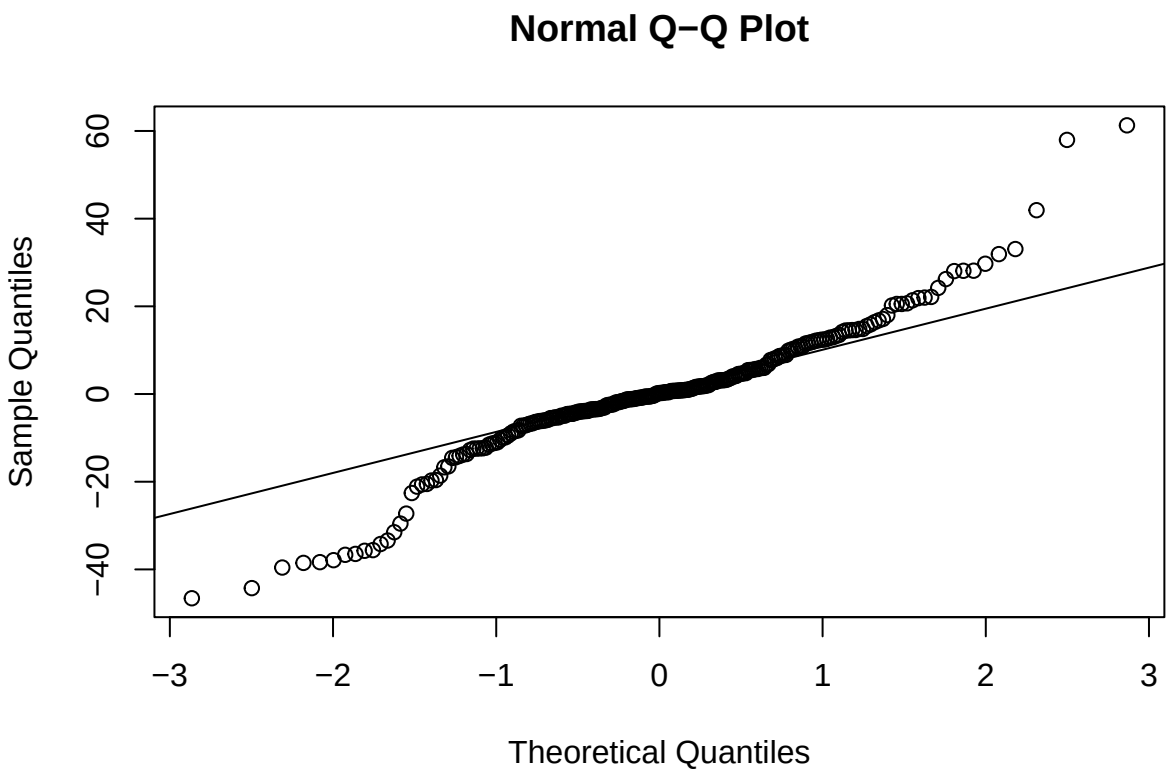
```
hist(ulva$growth_rate_percent, main = paste("Ulva lactuca Growth Rate (%)"),
     col = "olivedrab3", labels = TRUE)
```




```
plot(resid(run5.6_growth_model_noint) ~ fitted(run5.6_growth_model_noint))
```

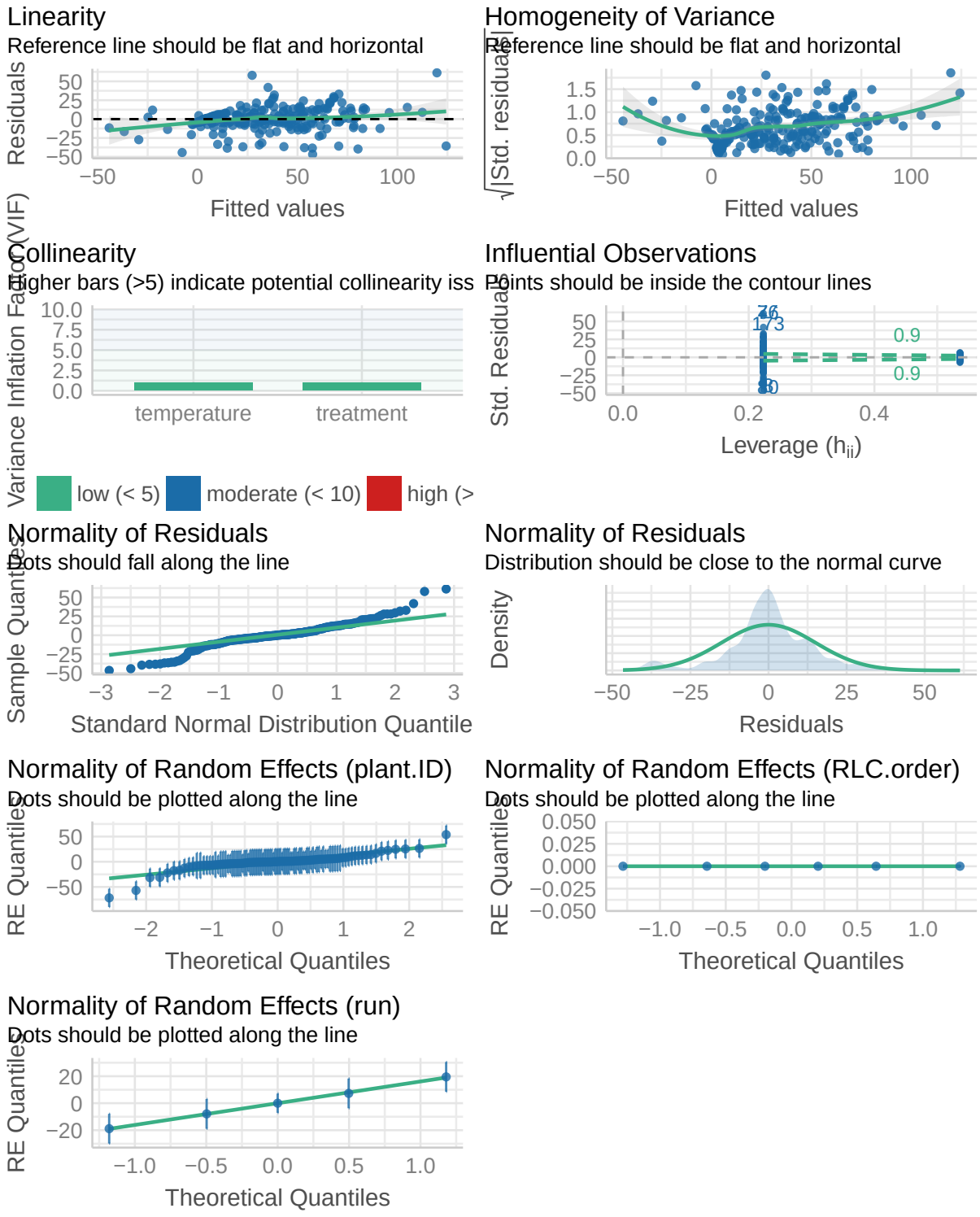



```
qqnorm(resid(run5.6_growth_model_noint))
qqline(resid(run5.6_growth_model_noint))
```



Check the performance of the model for Ulva

```
performance::check_model(run5.6_growth_model_noint)
```



These outputs show the model is acceptable

##Run ANOVA and Tukey's comparison

ANOVA shows that there is no significant difference between temperatures but the salinity/nutrient treatments are very close to significant. Tukey's is run on treatments to see pairwise comparisons

```
anova(run5.6_growth_model_noint, type = c("III"), ddf = "Satterthwaite")
```

```
## Type III Analysis of Variance Table with Satterthwaite's method
##              Sum Sq Mean Sq NumDF   DenDF F value  Pr(>F)
## treatment   11448.5 2862.13     4   4.823  9.0470 0.01793 *
## temperature    35.5   17.76     2 106.302  0.0562 0.94542
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
ulva_growth_model_aov <- aov(growth_rate_percent ~ treatment + temperature, data = ulva)
TukeyHSD(ulva_growth_model_aov, "treatment", ordered = FALSE)
```

```
##   Tukey multiple comparisons of means
##     95% family-wise confidence level
##
## Fit: aov(formula = growth_rate_percent ~ treatment + temperature, data = ulva)
##
## $treatment
##          diff        lwr      upr     p adj
## 1-0 26.122972   8.891291 43.35465 0.0004144
## 2-0 33.643252  16.411571 50.87493 0.0000019
## 3-0 40.933674  23.701993 58.16536 0.0000000
## 4-0 45.547350  28.315668 62.77903 0.0000000
## 2-1  7.520280  -9.711401 24.75196 0.7514921
## 3-1 14.810702  -2.420979 32.04238 0.1293111
## 4-1 19.424377   2.192696 36.65606 0.0183743
## 3-2  7.290422  -9.941259 24.52210 0.7723133
## 4-2 11.904098  -5.327584 29.13578 0.3206085
## 4-3  4.613675 -12.618006 21.84536 0.9478415
```

```
r.squaredGLMM(run5.6_growth_model_noint)
```

```
## Warning: 'r.squaredGLMM' now calculates a revised statistic. See the help page.
```

```
##            R2m       R2c
## [1,] 0.2104576 0.7429601
```

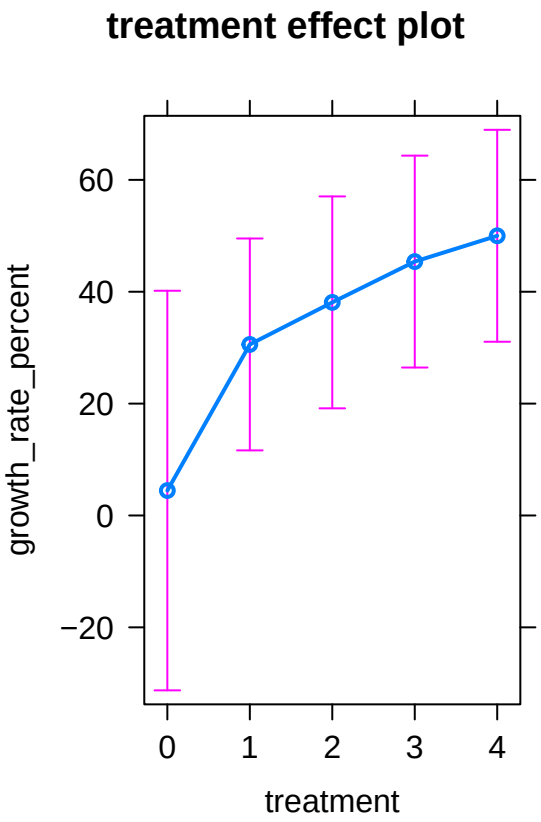
```
summary(run5.6_growth_model_noint)
```

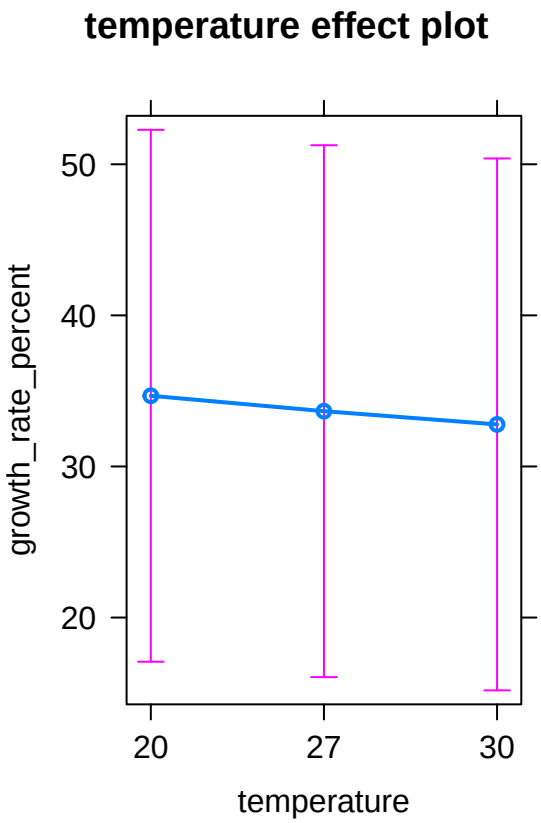
```
## Linear mixed model fit by REML. t-tests use Satterthwaite's method [
## lmerModLmerTest]
## Formula: growth_rate_percent ~ treatment + temperature + (1 | run) + (1 |
##     plant.ID) + (1 | RLC.order)
##    Data: ulva
##
## REML criterion at convergence: 2146.5
##
## Scaled residuals:
##     Min      1Q  Median      3Q     Max
## -2.6188 -0.3140  0.0118  0.3964  3.4453
```

```
##
## Random effects:
##  Groups    Name        Variance Std.Dev.
##  plant.ID  (Intercept) 340.4    18.45
##  RLC.order (Intercept)   0.0     0.00
##  run       (Intercept) 315.0    17.75
##  Residual              316.4    17.79
## Number of obs: 240, groups:  plant.ID, 96; RLC.order, 6; run, 5
##
## Fixed effects:
##                Estimate Std. Error      df t value Pr(>|t|)
## (Intercept)       5.429     18.421   2.821   0.295    0.789
## treatment1       26.123     20.521   2.780   1.273    0.299
## treatment2       33.643     20.521   2.780   1.639    0.207
## treatment3       40.934     20.521   2.780   1.995    0.147
## treatment4       45.547     20.521   2.780   2.220    0.120
## temperature27    -1.019      5.657 106.302  -0.180    0.857
## temperature30    -1.894      5.657 106.302  -0.335    0.738
##
## Correlation of Fixed Effects:
##             (Intr) trtmn1 trtmn2 trtmn3 trtmn4 tmpr27
## treatment1  -0.869
## treatment2  -0.869  0.984
## treatment3  -0.869  0.984  0.984
## treatment4  -0.869  0.984  0.984  0.984
## temperatr27 -0.154  0.000  0.000  0.000  0.000
## temperatr30 -0.154  0.000  0.000  0.000  0.000  0.500
## optimizer (nloptwrap) convergence code: 0 (OK)
## boundary (singular) fit: see ?isSingular
```

Effects Plots

```
plot(allEffects(run5.6_growth_model_noint))
```



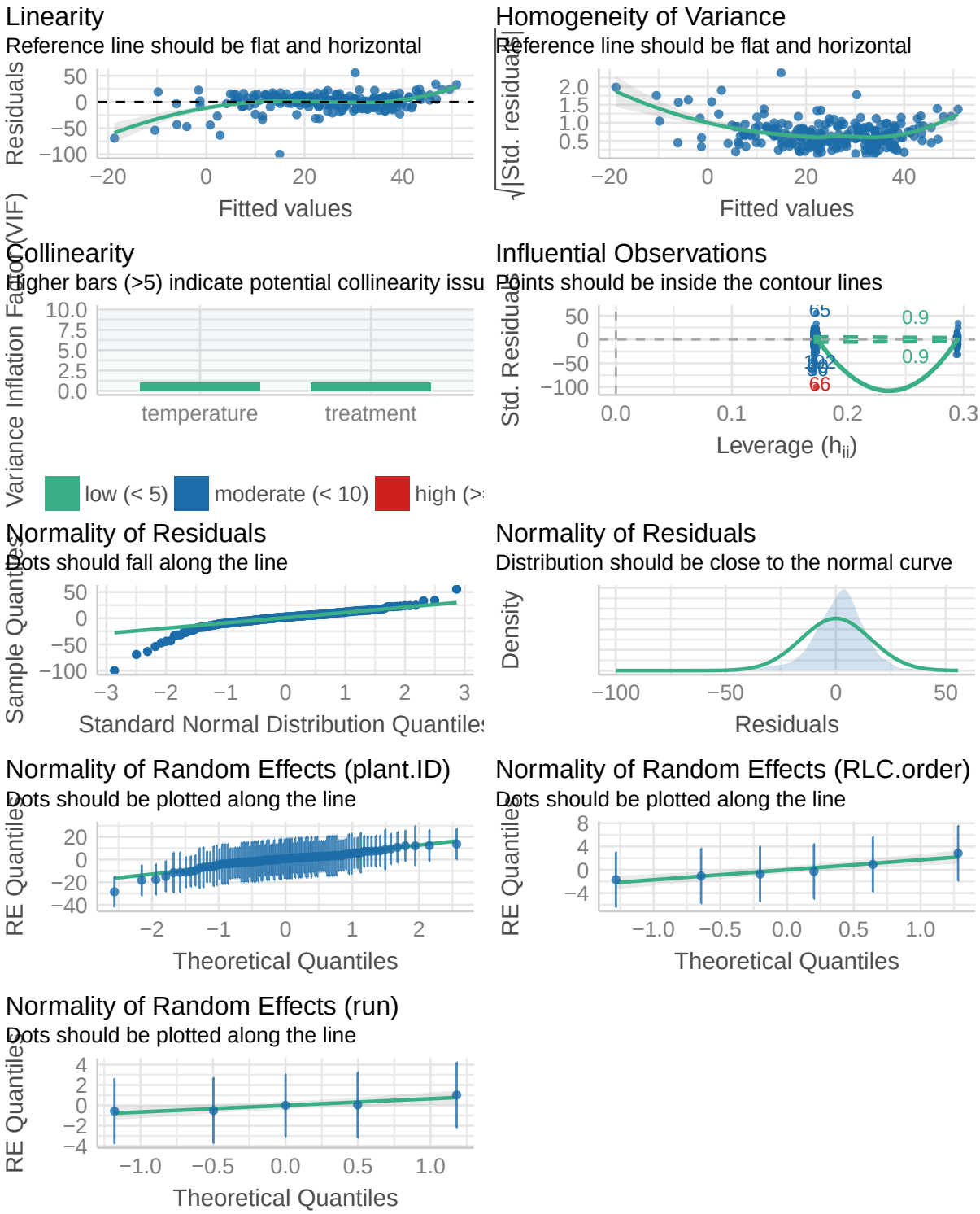


HYPNEA

```
run5.6_growth_model_noint <- lmer(formula = growth_rate_percent ~ treatment +
                                    temperature + (1 | run) + (1 | plant.ID) +
                                    (1 | RLC.order), data = hypnea)
```

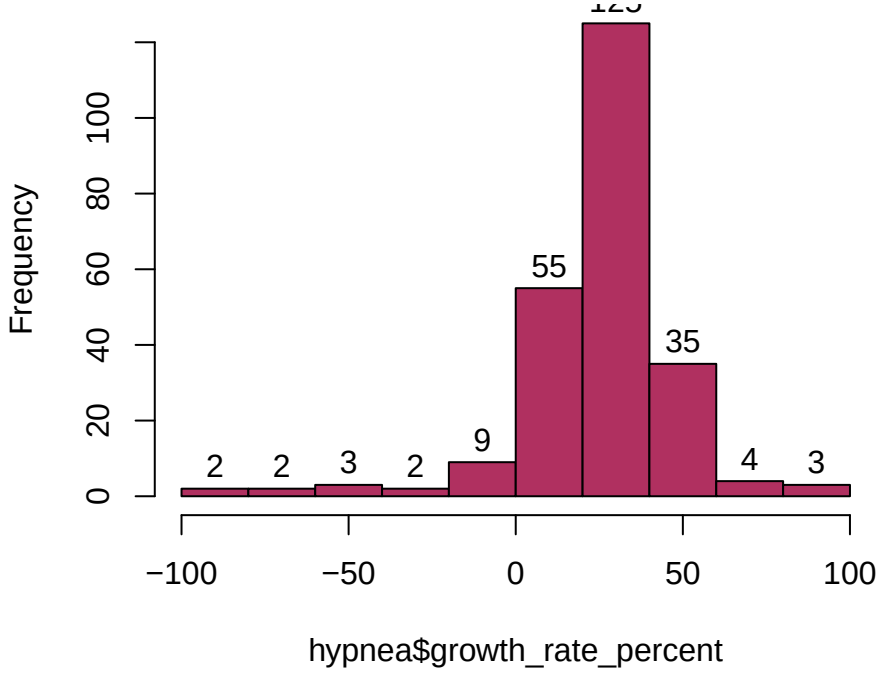
Check the performance of the model for Hypnea

```
performance::check_model(run5.6_growth_model_noint)
```

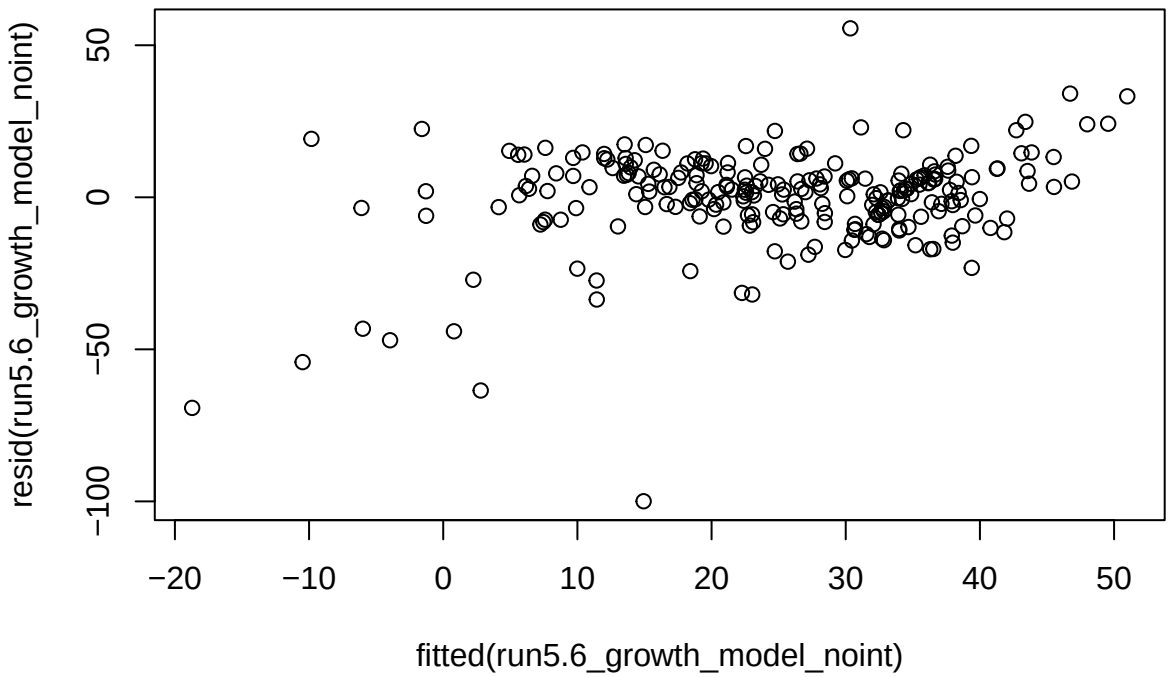


These outputs show the model is acceptable for the data

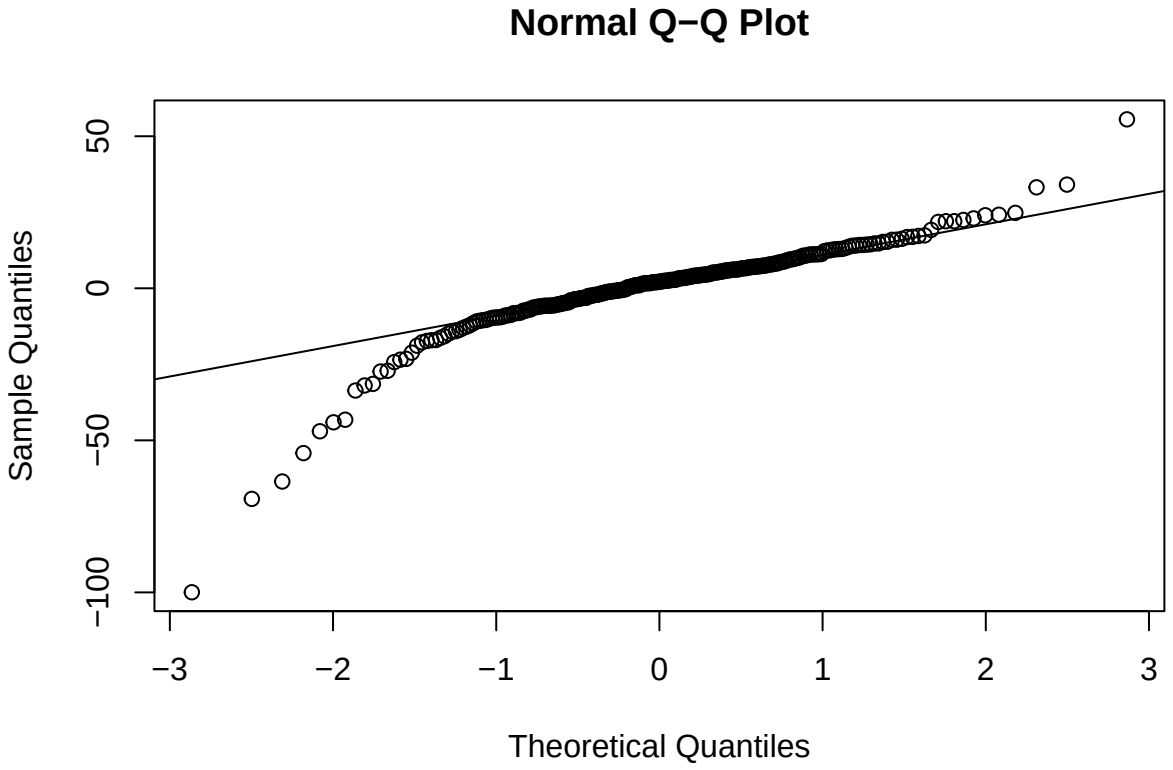
```
hist(hypnea$growth_rate_percent, main = paste("Hypnea musciformis Growth Rate (%)"),
     col = "maroon", labels = TRUE)
```




```
plot(resid(run5.6_growth_model_noint) ~ fitted(run5.6_growth_model_noint))
```




```
qqnorm(resid(run5.6_growth_model_noint))
qqline(resid(run5.6_growth_model_noint))
```




```
anova(run5.6_growth_model_noint, type = c("III"), ddf = "Satterthwaite")
```

```
## Type III Analysis of Variance Table with Satterthwaite's method
##              Sum Sq Mean Sq NumDF   DenDF F value  Pr(>F)
## treatment   17326.0  4331.5     4  3.0249 13.9859 0.02726 *
## temperature    56.8    28.4     2 28.7741  0.0917 0.91269
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
hypnea_growth_model_aov <- aov(growth_rate_percent ~ treatment + temperature, data = hypnea)
TukeyHSD(hypnea_growth_model_aov, "treatment", ordered = FALSE)
```

```
##   Tukey multiple comparisons of means
##     95% family-wise confidence level
##
## Fit: aov(formula = growth_rate_percent ~ treatment + temperature, data = hypnea)
##
## $treatment
##              diff         lwr          upr     p adj
## 2-1   -15.4100923 -27.0807398  -3.73944481 0.0031807
## 3-1   -11.5769131 -23.2475606   0.09373443 0.0530291
## 3.5-1  -0.7223387 -12.3929862  10.94830879 0.9998110
## 4-1   -24.2978542 -35.9685017 -12.62720672 0.0000003
## 3-2     3.8331792  -7.8374683  15.50382674 0.8955467
## 3.5-2  14.6877536   3.0171061  26.35840110 0.0057452
## 4-2    -8.8877619 -20.5584094   2.78288559 0.2262656
## 3.5-3  10.8545744  -0.8160731  22.52522186 0.0819446
## 4-3   -12.7209412 -24.3915887  -1.05029365 0.0249597
## 4-3.5 -23.5755155 -35.2461630 -11.90486801 0.0000008
```

```
r.squaredGLMM(run5.6_growth_model_noint)
```

```
##            R2m       R2c
## [1,] 0.1644597 0.4022791
```

```
summary(run5.6_growth_model_noint)
```

```
## Linear mixed model fit by REML. t-tests use Satterthwaite's method [
## lmerModLmerTest]
## Formula: growth_rate_percent ~ treatment + temperature + (1 | run) + (1 |
##     plant.ID) + (1 | RLC.order)
##    Data: hypnea
##
## REML criterion at convergence: 2081.8
##
## Scaled residuals:
##     Min      1Q  Median      3Q     Max
## -5.6796 -0.3253  0.1225  0.4421  3.1569
##
## Random effects:
##  Groups    Name        Variance Std.Dev.
##  plant.ID  (Intercept) 111.159  10.543
##  RLC.order (Intercept)   8.876   2.979
##  run       (Intercept)   3.190   1.786
##  Residual              309.705  17.598
## Number of obs: 240, groups:  plant.ID, 96; RLC.order, 6; run, 5
##
## Fixed effects:
##               Estimate Std. Error       df t value Pr(>|t|)
## (Intercept)    35.7815     4.1631  17.1258   8.595 1.28e-07 ***
## treatment2    -15.4101     3.5923 161.5491  -4.290 3.07e-05 ***
## treatment3    -11.5769     3.5923 161.5491  -3.223  0.00154 **
## treatment3.5   -0.7223     4.6393   2.4459  -0.156  0.88820
## treatment4    -24.2979     3.5923 161.5491  -6.764 2.34e-10 ***
## temperature27   0.1798     4.4743  19.3094   0.040  0.96836
## temperature30  -1.5010     4.2516  53.3627  -0.353  0.72546
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Correlation of Fixed Effects:
##             (Intr) trtmn2 trtmn3 trt3.5 trtmn4 tmpr27
## treatment2  -0.431
## treatment3  -0.431  0.500
## treatmnt3.5 -0.495  0.387  0.387
## treatment4  -0.431  0.500  0.500  0.387
## temperatr27 -0.524  0.000  0.000  0.000  0.000
## temperatr30 -0.514  0.000  0.000  0.000  0.000  0.486
```

Effects Plots

```
plot(allEffects(run5.6_growth_model_noint))
```