

# Herbivory Script

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2023-01-27

#Load Library

```
library(here)
library(tidyverse)
library(lubridate)
library(performance)
library(ggpubr)
```

## Load In Data

```
#Reading in experimental treatment metadata
experiment.metadata <- read_csv(here::here("Data", "Snail_Metadata", "Experiment_Treatment_Metadata.csv"))

#Reading in organism morphometric data
snail.data <- read_csv(here::here("Data", "Snail_Metadata", "Tegula_funebris_Morphometric_Data.csv"))
  select(Snail_ID:Experiment_End_Date)

#joining experimental treatment and snail data
experiment.data <- left_join(experiment.metadata, snail.data, by = join_by(Snail_ID))
```

#HERBIVORY SCRIPT

#Linear Regression

```
algae.drying.rates <- read_csv(here::here("Data", "Herbivory_Data", "Algae_Drying_Weights.csv"))
```

```
## Rows: 30 Columns: 13
## -- Column specification -----
## Delimiter: ","
## chr (1): Treatment
## dbl (12): Algae_ID, Initial_Algae_Weight, 0:00, 0:20, 0:40, 1:00, 1:20, 1:40...
##
## i Use 'spec()' to retrieve the full column specification for this data.
## i Specify the column types or set 'show_col_types = FALSE' to quiet this message.
```

```
#algae.drying.rates %>% view()
```

```
algae.rates <- algae.drying.rates %>%
```

```

pivot_longer("0:00":"3:00", names_to="Time", values_to="Algae_Weight")

drying.rates <- algae.rates %>%
  mutate(Algae_Weight_Change=Initial_Algae_Weight-Algae_Weight,
         Percent_change = Algae_Weight_Change/Initial_Algae_Weight*100,
         Time=as.numeric(hm(Time))/60,
         Weight_Change_Rate=Algae_Weight_Change/Time,
         Size=case_when(Initial_Algae_Weight >= 1.2 ~ "large", #mean weight = 1.2
                        Initial_Algae_Weight <= 1.2 ~ "small")) %>%
  dplyr::select(Time, Treatment, Initial_Algae_Weight,Percent_change, Algae_Weight, Algae_ID, Size) %>%
  mutate_all(~replace(., is.nan(.), 0))

### Linear regression model (Y = Algae_Weight, X = Time)
model_1 <- lm(Percent_change ~ Time*Treatment, data = drying.rates)

### non-linear regression model (Y = Algae_Weight, X = Time)
model_2 <- lm(Percent_change ~ poly(Time, 2), data = drying.rates) #non-linear

#checking the models
summary(model_1)

```

```

##
## Call:
## lm(formula = Percent_change ~ Time * Treatment, data = drying.rates)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -19.3785  -3.4092   0.1742   3.2634  10.6715
##
## Coefficients:
##              Estimate Std. Error t value Pr(>|t|)
## (Intercept)      4.580283   0.729527   6.278 1.22e-09 ***
## Time              0.280116   0.006833  40.997 < 2e-16 ***
## TreatmentNon-Blotted -1.171060   1.031706  -1.135   0.257
## Time:TreatmentNon-Blotted 0.012259   0.009663   1.269   0.206
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 4.807 on 296 degrees of freedom
## Multiple R-squared:  0.9223, Adjusted R-squared:  0.9215
## F-statistic: 1171 on 3 and 296 DF, p-value: < 2.2e-16

```

```
summary(model_2)
```

```

##
## Call:
## lm(formula = Percent_change ~ poly(Time, 2), data = drying.rates)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -16.3726  -2.4530  -0.1072   2.5889  11.2950
##

```

```
## Coefficients:
##           Estimate Std. Error t value Pr(>|t|)
## (Intercept)   29.7568    0.2562 116.126 < 2e-16 ***
## poly(Time, 2)1 284.8106    4.4383  64.171 < 2e-16 ***
## poly(Time, 2)2 -32.0514    4.4383  -7.222 4.34e-12 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 4.438 on 297 degrees of freedom
## Multiple R-squared:  0.9335, Adjusted R-squared:  0.9331
## F-statistic: 2085 on 2 and 297 DF, p-value: < 2.2e-16
```

```
AIC(model_1)
```

```
## [1] 1799.404
```

```
AIC(model_2)
```

```
## [1] 1750.515
```

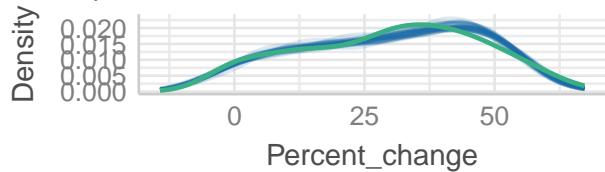
```
#selecting the model
model=model_2

# Plot the four diagnostic plots
check_model(model, panel = TRUE)
```

```
## Not enough model terms in the conditional part of the model to check for
## multicollinearity.
```

### Posterior Predictive Check

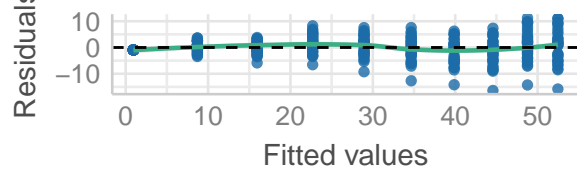
Model-predicted lines should resemble observed data



— Observed data — Model-predicted data

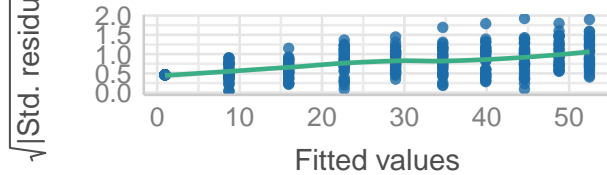
### Linearity

Reference line should be flat and horizontal



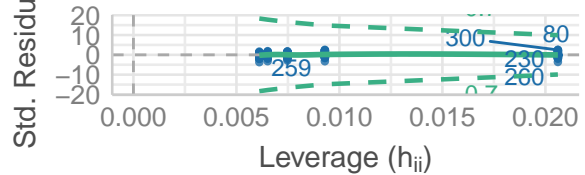
### Homogeneity of Variance

Reference line should be flat and horizontal



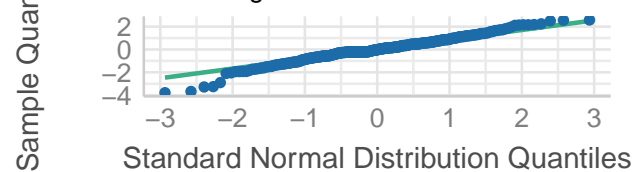
### Influential Observations

Points should be inside the contour lines



### Normality of Residuals

Dots should fall along the line



```
#model$coefficients[1]
```

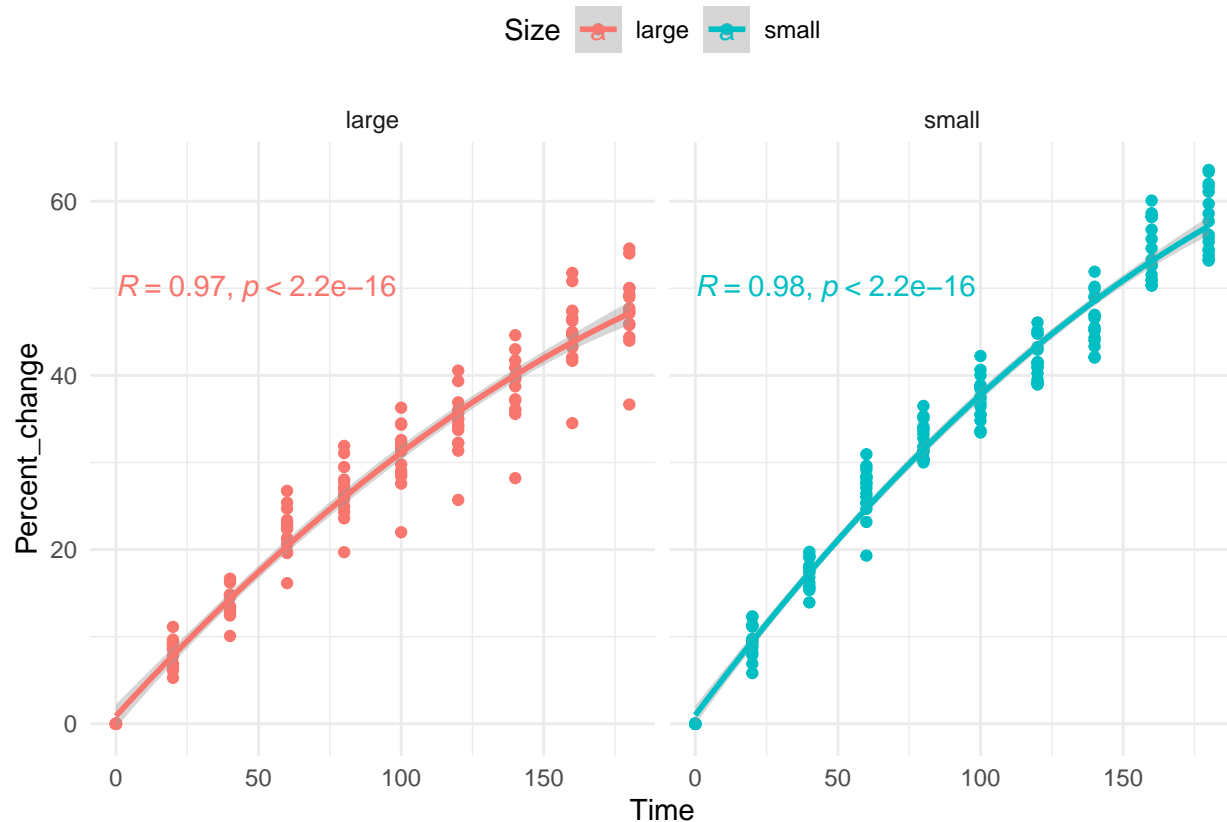
*#Only using size categories for linear regression since there is not a difference between blotted and n*

```
small_model <- lm(Percent_change ~ poly(Time, 2), data = drying.rates %>% filter(Size=="small"))
```

```
large_model <- lm(Percent_change ~ poly(Time, 2), data = drying.rates %>% filter(Size=="large"))
```

*#plotting regressions*

```
ggplot(drying.rates, aes(Time, Percent_change, color = Size)) +
  geom_point(aes(color = Size))+
  geom_smooth(method = "lm", formula = "y~ poly(x, 2)", se = TRUE)+
  stat_cor(label.y = 50)+
  facet_wrap(~Size) + theme_minimal() +
  theme(legend.position = "top")
```



```
##### HERBIVORY DATA #####
#reading in data
herbivory.data <- read_csv(here::here("Data", "Herbivory_Data", "Algae_Spreadsheet.csv"))
#algae.data %>% view()

#Joining datasets
herbivory.treatment <- left_join(herbivory.data, experiment.data)

#Cleaning data
herbivory.treatment <- herbivory.treatment %>%
  mutate(Size=case_when(Algae_Weight_Before >= 1.2 ~ "large",
                        Algae_Weight_Before < 1.2 ~ "small")) %>%
  mutate(Trial_Duration = as.numeric(mdy_hm(Collection_Time_After)-mdy_hm(Placement_Time_Before))*24) %>%
  dplyr::select(Temp_Treatment, pH_Treatment, Algae_ID, Final_Blotted_Wet_Mass_g,
                Drying_Duration_Before, Algae_Weight_Before, Drying_Duration_After, Algae_Weight_After,
                Time_Point, Trial_Duration, Identity, Size)

#Normalizing data from polynomial regression if data is either small or large for experimental algal wei.
normalized.data <- herbivory.treatment %>%
  mutate(percent_change_before = if_else(Size == "small",
    predict(small_model, terms = "Time", newdata = data.frame(Time = Drying_Duration_Before)),
    predict(large_model, terms = "Time", newdata = data.frame(Time = Drying_Duration_Before))),
    percent_change_after = if_else(Size == "small",
    predict(small_model, terms = "Time", newdata = data.frame(Time = Drying_Duration_After)),
    predict(large_model, terms = "Time", newdata = data.frame(Time = Drying_Duration_After))))
```

```

#Calculating percent change of algae
herbivory.data <- normalized.data %>%
  mutate(weight_change_before=(percent_change_before*0.01)*Algae_Weight_Before,
         weight_change_after=(percent_change_after*0.01)*Algae_Weight_After) %>%
  mutate(normalized_algae_weight_before=Algae_Weight_Before + weight_change_before,
         normalized_algae_weight_after=Algae_Weight_After + weight_change_after) %>%
  #mutate(algae_weight_change=Algae_Weight_Before-Algae_Weight_After) %>%
  mutate(normalized_algae_weight_change=normalized_algae_weight_before-normalized_algae_weight_after,
         algae_weight_change=Algae_Weight_Before-Algae_Weight_After)

treatment.data<- herbivory.data %>% filter(Identity=='Treatment')

autogenic.control.data<- herbivory.data %>% filter(Identity=='Control') %>%
  group_by(Temp_Treatment, pH_Treatment, Time_Point) %>%
  mutate(control_weight_change=algae_weight_change,
         control_normalized_weight_change=normalized_algae_weight_change) %>%
  dplyr::select(Temp_Treatment, pH_Treatment, Time_Point, control_weight_change,
               control_normalized_weight_change)

controlled.data <- left_join(treatment.data, autogenic.control.data)
controlled.data <- left_join(controlled.data, experiment.data)

#average rate of algae consumption per hour per gram of snail (controlled)
herbivory.data.final <- controlled.data %>%
  mutate(normalized_algae_weight_change_per_snail_hr=(normalized_algae_weight_change+
               control_normalized_weight_change)/Initial_Blotted_Wet_Mass_g/Trial_Duration,
         algae_weight_change_per_snail_hr=(algae_weight_change+
               control_weight_change)/Initial_Blotted_Wet_Mass_g/Trial_Duration) %>%
  group_by(Temp_Treatment, pH_Treatment, Algae_ID) %>%
  summarise(normalized_algae_weight_change_per_snail_hr= mean(normalized_algae_weight_change_per_snail_hr),
            algae_weight_change_per_snail_hr = mean(algae_weight_change_per_snail_hr))

# Collect, check, and present data in long format.
as_tibble(herbivory.data.final)

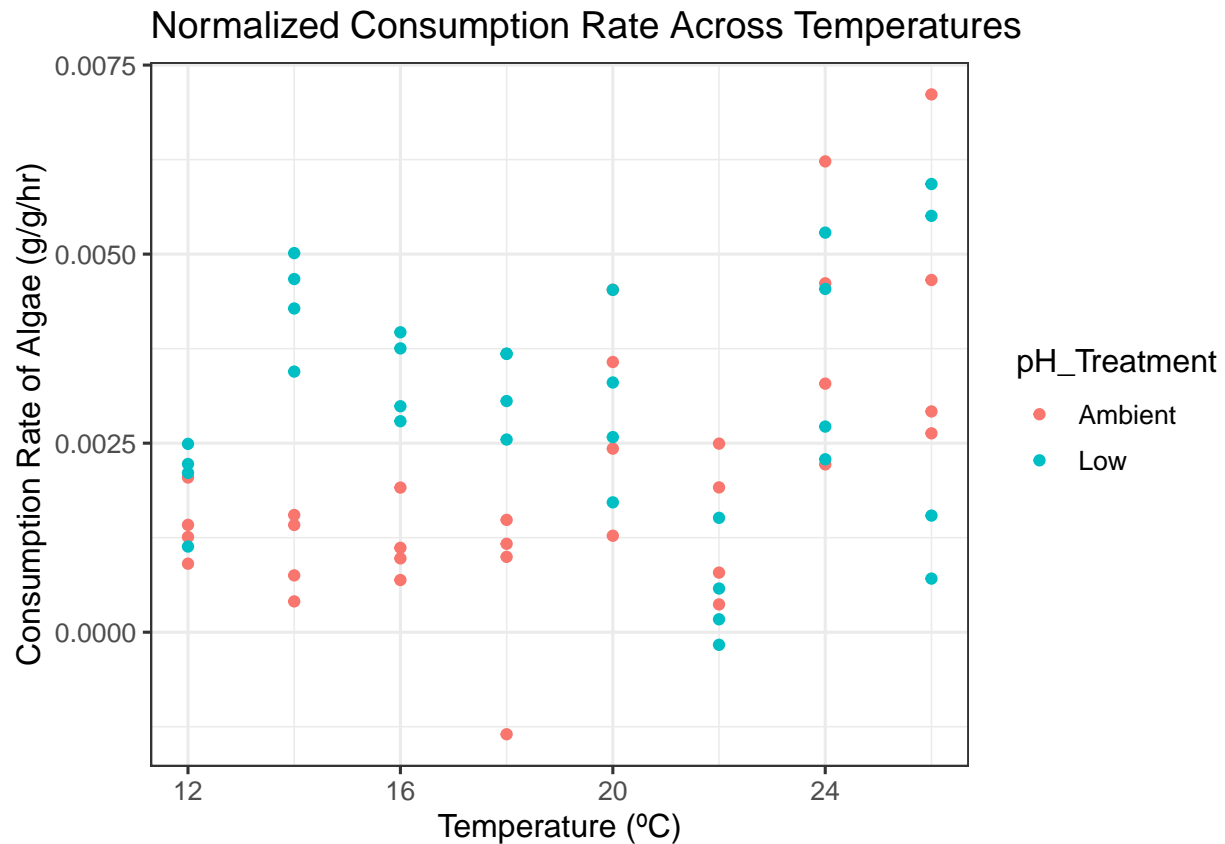
```

```

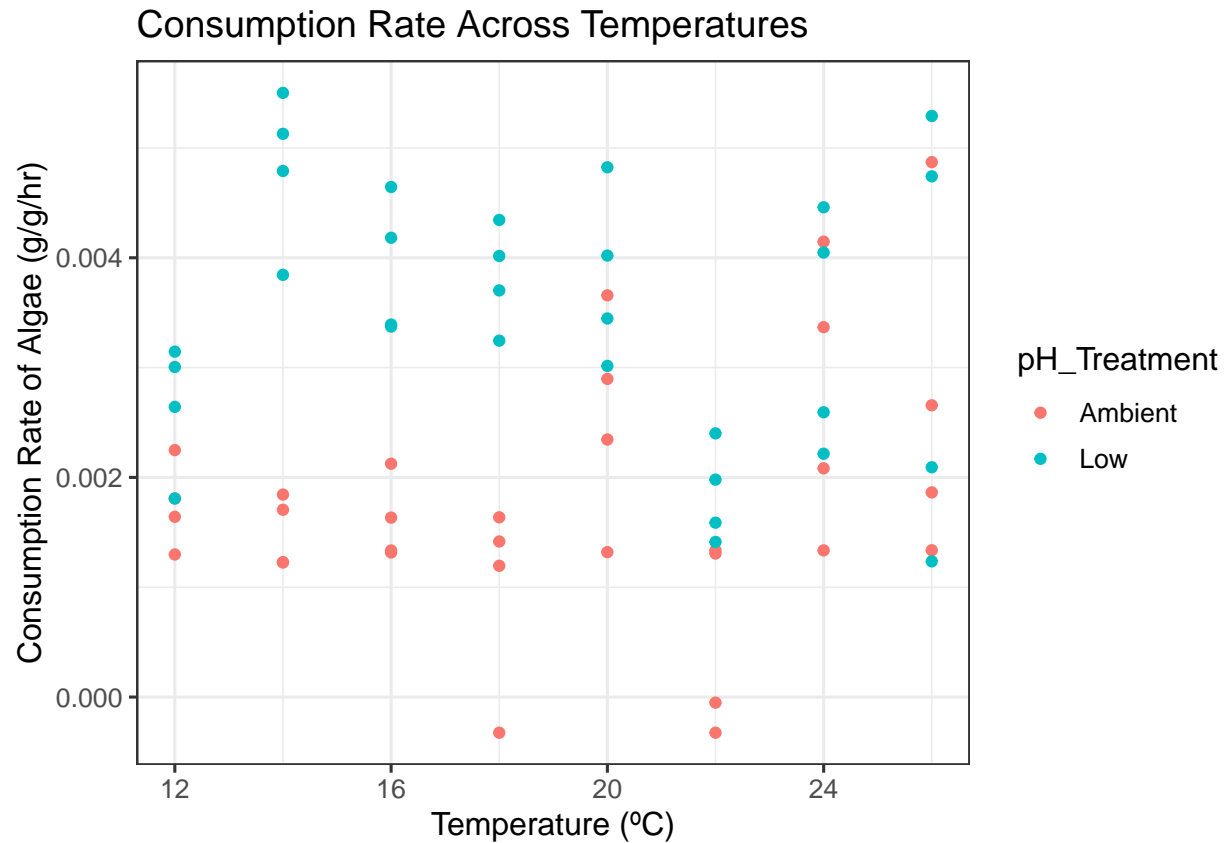
## # A tibble: 64 x 5
##   Temp_Treatment pH_Treatment Algae_ID normalized_algae_weight_change_per_sna~1
##   <dbl> <chr> <chr> <dbl>
## 1      12 Ambient      41      0.00205
## 2      12 Ambient      42      0.000906
## 3      12 Ambient      43      0.00142
## 4      12 Ambient      44      0.00126
## 5      12 Low         1      0.00222
## 6      12 Low         2      0.00211
## 7      12 Low         3      0.00249
## 8      12 Low         4      0.00113
## 9      14 Ambient     46      0.000752
## 10     14 Ambient     47      0.00155
## # i 54 more rows
## # i abbreviated name: 1: normalized_algae_weight_change_per_snail_hr
## # i 1 more variable: algae_weight_change_per_snail_hr <dbl>

```

```
#Plotting the data using ggplot
ggplot(herbivory.data.final, aes(Temp_Treatment, normalized_algae_weight_change_per_snail_hr, color=pH_
geom_point() +
theme_bw(base_size = 12) +
labs(x = 'Temperature (°C)',
y = 'Consumption Rate of Algae (g/g/hr)',
title = 'Normalized Consumption Rate Across Temperatures')
```



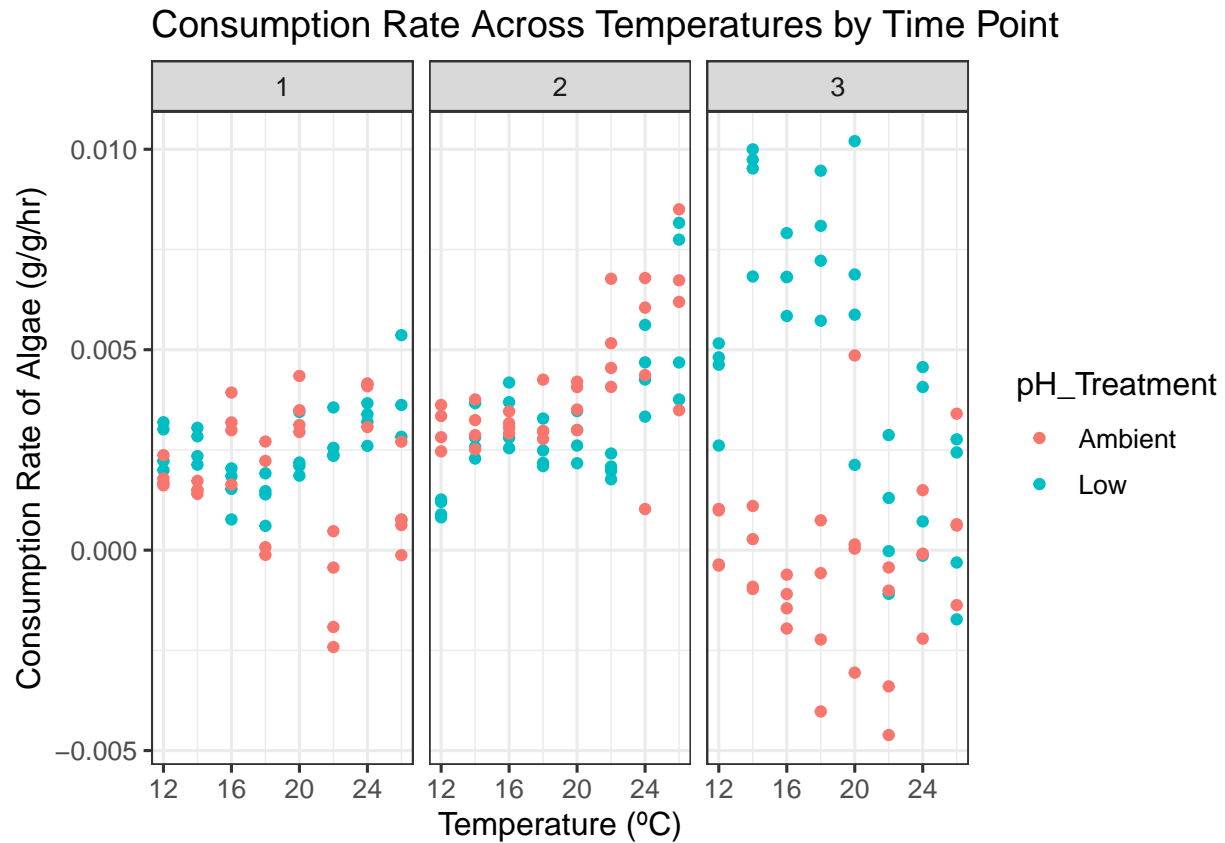
```
#Plotting the data using ggplot
ggplot(herbivory.data.final, aes(Temp_Treatment, algae_weight_change_per_snail_hr, color=pH_Treatment))
geom_point() +
theme_bw(base_size = 12) +
labs(x = 'Temperature (°C)',
y = 'Consumption Rate of Algae (g/g/hr)',
title = 'Consumption Rate Across Temperatures')
```



```
time.point.data <- controlled.data %>%
  mutate(normalized_algae_weight_change_per_snail_hr=(normalized_algae_weight_change+
    control_normalized_weight_change)/Initial_Blotting_Wet_Mass_g/Trial_Duration,
    algae_weight_change_per_snail_hr=(algae_weight_change+
    control_weight_change)/Initial_Blotting_Wet_Mass_g/Trial_Duration) %>%
  select(Time_Point, Temp_Treatment, pH_Treatment, normalized_algae_weight_change_per_snail_hr, algae_w

ggplot(time.point.data, aes(Temp_Treatment, algae_weight_change_per_snail_hr, color=pH_Treatment)) +
  geom_point() +
  theme_bw(base_size = 12) +
  labs(x = 'Temperature (°C)',
    y = 'Consumption Rate of Algae (g/g/hr)',
    title = 'Consumption Rate Across Temperatures by Time Point') +
  facet_wrap(~Time_Point)
```





```
ggplot(time.point.data, aes(Temp_Treatment, normalized_algae_weight_change_per_snail_hr, color=pH_Treatment)) +
  geom_point() +
  theme_bw(base_size = 12) +
  labs(x = 'Temperature (°C)',
       y = 'Consumption Rate of Algae (g/g/hr)',
       title = 'Normalized Consumption Rate Across Temperatures by Time Point') +
  facet_wrap(~Time_Point)
```

Normalized Consumption Rate Across Temperatures by Time Poin

