

Effect of Water Stress Treatment on Growth Parameters

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Title: Comparative study of effect of water stress in different growth parameters of cultivated crops and wild weed species

Introduction

Water stress affects plant growth and development, including plant height, leaf length, width, dry matter, and chlorophyll content (Picón-Toro et al., 2012). Leaves are essential indicators of growth and yield since they play a significant role in physiological processes (Zhang et al., 2015). The plant's main response to water stress is to reduce its plant height, leaf area with increasing leaf numbers and thus improve water use efficiency (Ramezanifar et al., 2022). Photosynthesis is increased by higher chlorophyll levels and chlorophyll levels are higher in plants that are more resilient to water stress (Jangpromma et al., 2010). The chlorophyll content is a proxy for overall photosynthetic efficiency in plants and therefore is a good indicator of water stress tolerance in crop species (Hasanuzzaman et al., 2022).

Objectives

To know the response of different cultivated and wild species to the water stress To know the effect of water stress on different growth parameters of cultivated and wild species

Learning Objectives

The main objective of this project is to get familiar with the R coding and use the R for the analysis of biological data.

Hypothesis

1. Ho = There will be no significant difference in the growth parameter of plants due to water stress
2. H1 = There will be significant difference in the growth parameter of plants due to water stress

Materials and Methods

2.1 Greenhouse Pot Experiment

Solanum lycopersicum, Spinacea oleracea, Hordeum Vulgare, Beta vulgaris and Raphanus sativus among the cultivated crop and Amaranthus retroflexus, Lolium perenne, Sonchus oleraceus, Portulaca oleracea among the weeds were selected for the pot experiment in the greenhouse. At first the Samplings were prepared in the normal condition. The plants were transplanted after two weeks with a single plant in the pot. Two hundred ten plants were transplanted. The 10X2X3X7 factorial design experiment was organized in a completely randomized design (CRD) in the controlled condition. After transplanting, the plants were watered up to field condition and left to grow. The plants in the control received weekly irrigation that reached up to the field's capacity, the plants in the intermediate up to half the field's capacity, and the stressed plants received half of the field's capacity every two weeks.

2.2 Measured Parameters

Parameters like soil humidity, electrical conductivity, plant height, leaf number, leaf length, leaf width, leaf area, and chlorophyll content were measured in every two-week interval. Other parameters, aerial fresh weight, aerial dry weight, root length, roots fresh weight, and roots dry weight were measured at the end of the experiment on the 6th week of experiment setup.

WET Sensor was used to monitor electrical conductivity and soil humidity. Electrical conductivity was measured in milli Siemens per centimeters (mS/cm) and soil humidity was recorded in percentages. Using a SPAD meter, the amount of chlorophyll was calculated in micromoles per square meter (mmolem⁻²). Using a digimizer, we measured the length of the roots and the area of the leaves. Fresh weight was calculated using a weighing machine and the dry weight was calculated after the contents had been dried in the oven for more than 24 hours.

2.3 Statistical Analysis

Google Sheet was used to enter the data. All statistical analysis was conducted in R Studio version 4.2.1. Parametric data that meet the assumption of the analysis of variance were subjected to factorial analysis of variance (ANOVA) using the linear mixed model. Model residuals were checked for normality using the shapiro Test. Post hoc means separation was achieved using Tukey Honestly significant difference (TukeyHSD) with the aid of the agricolae package in R. Missing data were drop or imputed using column means prior to analysis.

```
#install.packages("pacman")
# Pacman package to load all the other packages
pacman::p_load(dplyr, ggplot2, readxl, tidyverse, agricolae,
               googlesheets4, car, rstatix, FactoMineR, writexl, missMDA,
               factoextra, lmerTest, ggpubr, tidyverse, agricolae,
               kableExtra)
```

Import and tidy the data

```
##fromweb <-read_sheet
('https://docs.google.com/spreadsheets/
#d/1WM6hDgwCFxbLpT37HLzU-iYZAEGbDHP6CpU8BDwWmZc')
```

```
## [1] "https://docs.google.com/spreadsheets/\n#d/1WM6hDgwCFxbLpT37HLzU-iYZAEGbDHP6CpU8BDwWmZc"
```

```
ws0 <- read_excel("data/wstress.xlsx")
ws1 <- ws0
```

```
#####transform to factor
for(i in 1:7){
  ws0[[i]] <- as.factor(ws0[[i]])
}
###check for success
levels(ws0$Week)
```

```
## [1] "W1" "W2" "W3" "W4" "W5" "W6"
```

```
##### rename group 2 amaranthus for distinction
ws0$Species <- as.character(ws0$Species)
ws0$Species <- ifelse(
  ws0$Group == "G2" & ws0$Species == "Amaranthus retroflexus",
  "G2retroflexus", ws0$Species)
ws0$Species <- as.factor(ws0$Species)

var.numeric <- c(
  "Soil_humidity", "Electrical_conductivity", "Plant_height",
  "Leaf_number", "Leaf_length", "Leaf_width", "Leaf_area",
  "Chlorophyll_content",
  "Aerial_fresh_weight", "Aerial_dry_weight",
  "Root_length", "Roots_fresh_weight", "Roots_dry_weight")

for (i in var.numeric) {
  ws0[[i]] <- as.numeric(ws0[[i]])
}

levels(ws0$Treatment) <- c("Control", "Intermediate", "WaterStress")
#class(ws0$Species) #to verify if conversion to factor was succesful
```

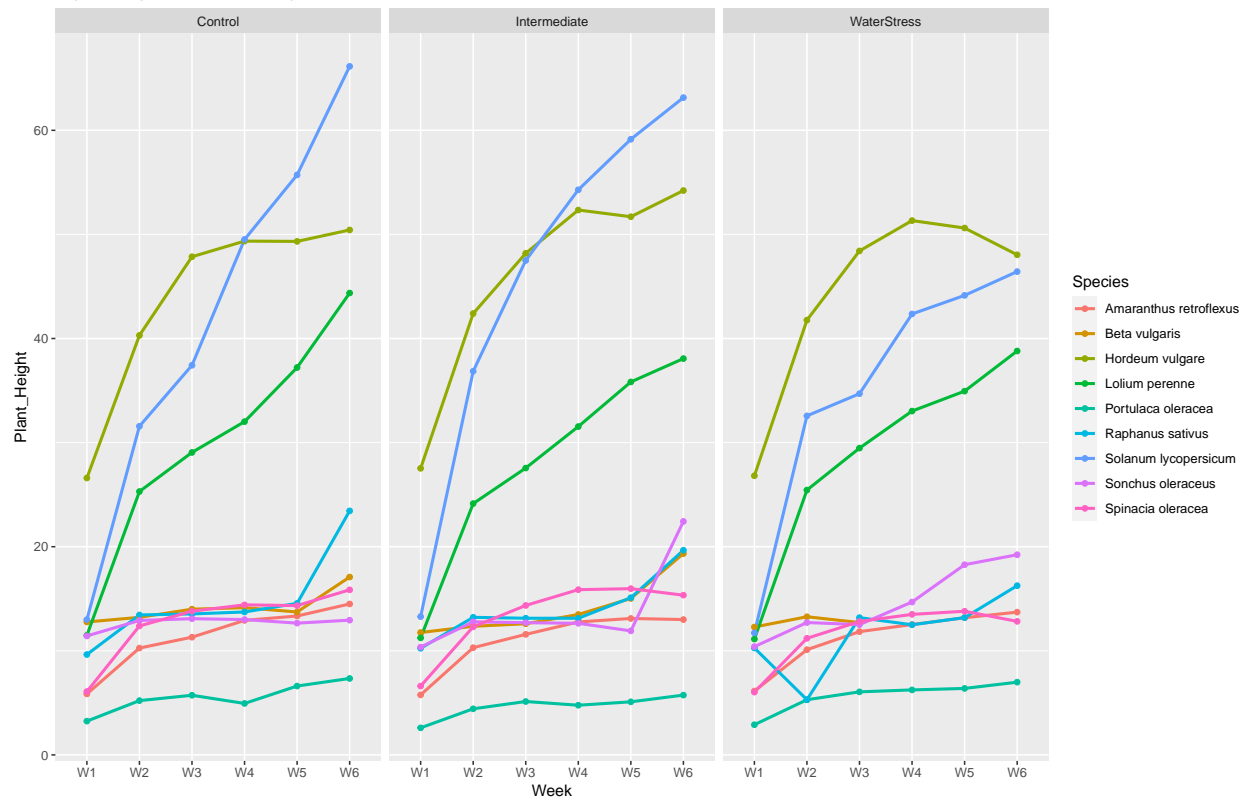
Variation of some variables accross weeks

```
## create a summmary table for the some variable, to be used in plot
sumtable <- ws0%>% group_by(Species, Treatment, Week) %>%
  summarize(Plant_Height=mean(Plant_height),
            Leaf_Length=mean(Leaf_length),
            Leaf_Width=mean(Leaf_width),
            chlorophyll=mean(Chlorophyll_content))

#####line graph to show over weeks effect of treatment on selected va
#riable
pheight <- ggplot(sumtable, aes(x=Week, y=Plant_Height, group=Species,
  col=Species))+
  geom_line(size=1)+
  geom_point()+
  facet_wrap(~Treatment)+ggtitle("Fig.1 line graph of Plant Height")

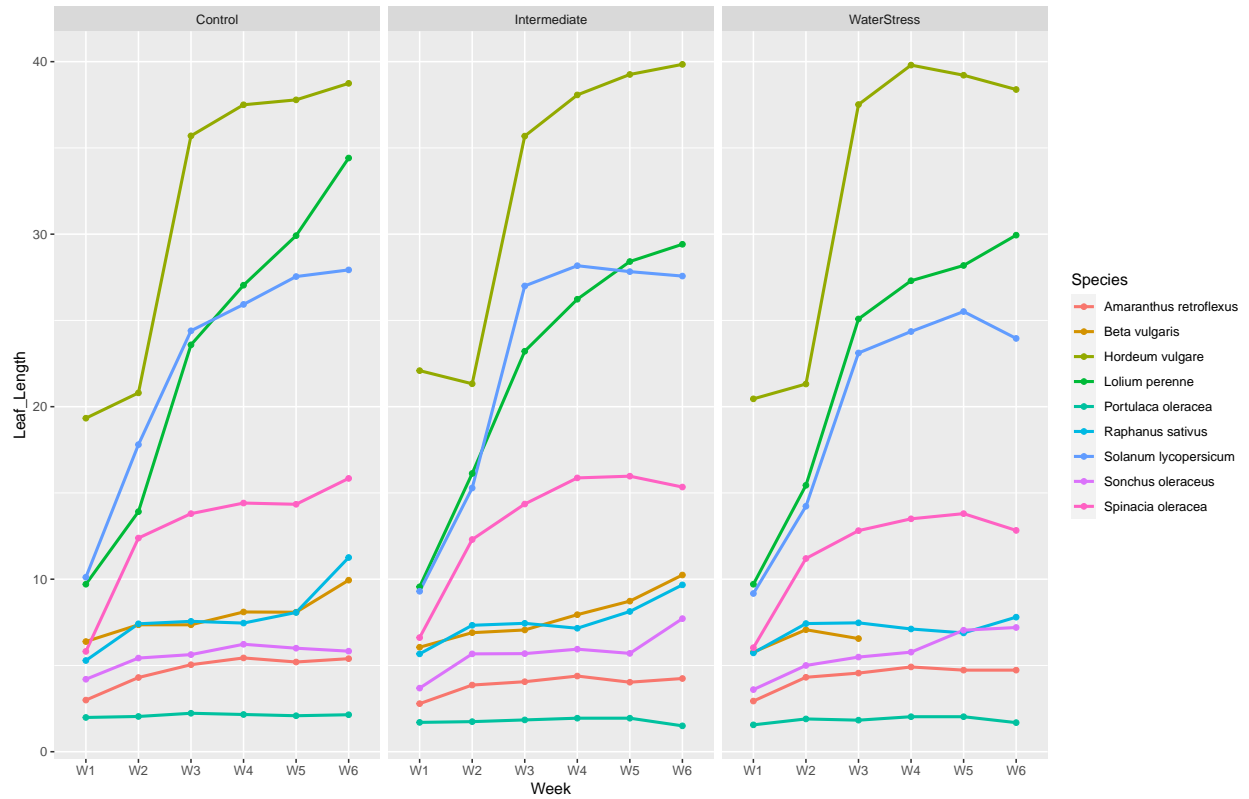
pheight
```

Fig.1 line graph of Plant Height



```
l1length <- ggplot(sumtable, aes(x=Week, y=Leaf_Length, group=Species,
col=Species))+
  geom_line(size=1)+
  geom_point()+
  facet_wrap(~Treatment)+ggtitle("Fig.2 line graph of Leave Length")
l1length
```

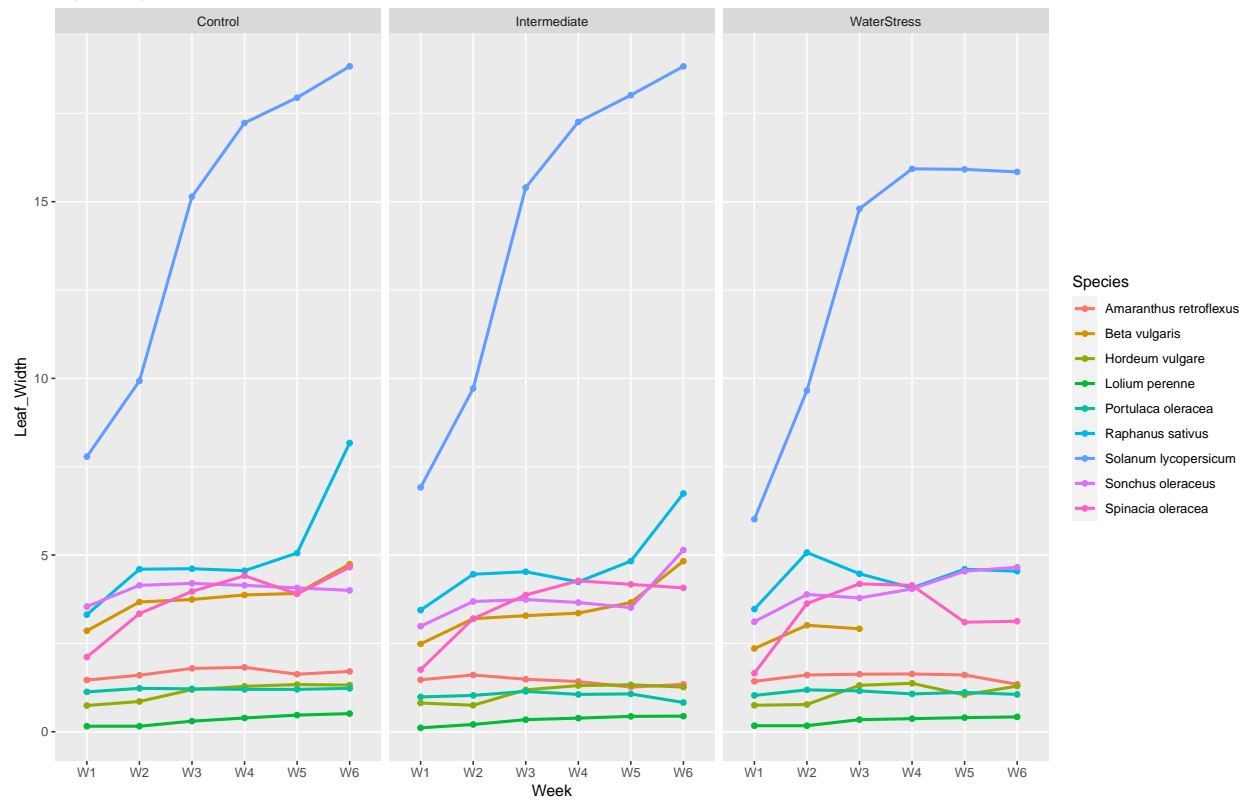
Fig.2 line graph of Leaf Length



```
lwidth <- ggplot(sumtable, aes(x=Week, y= Leaf_Width, group=Species,
col=Species))+
  geom_line(size=1)+
  geom_point()+
  facet_wrap(~Treatment)+ggtitle("Fig.3 line graph of Leave Width")
```

lwidth

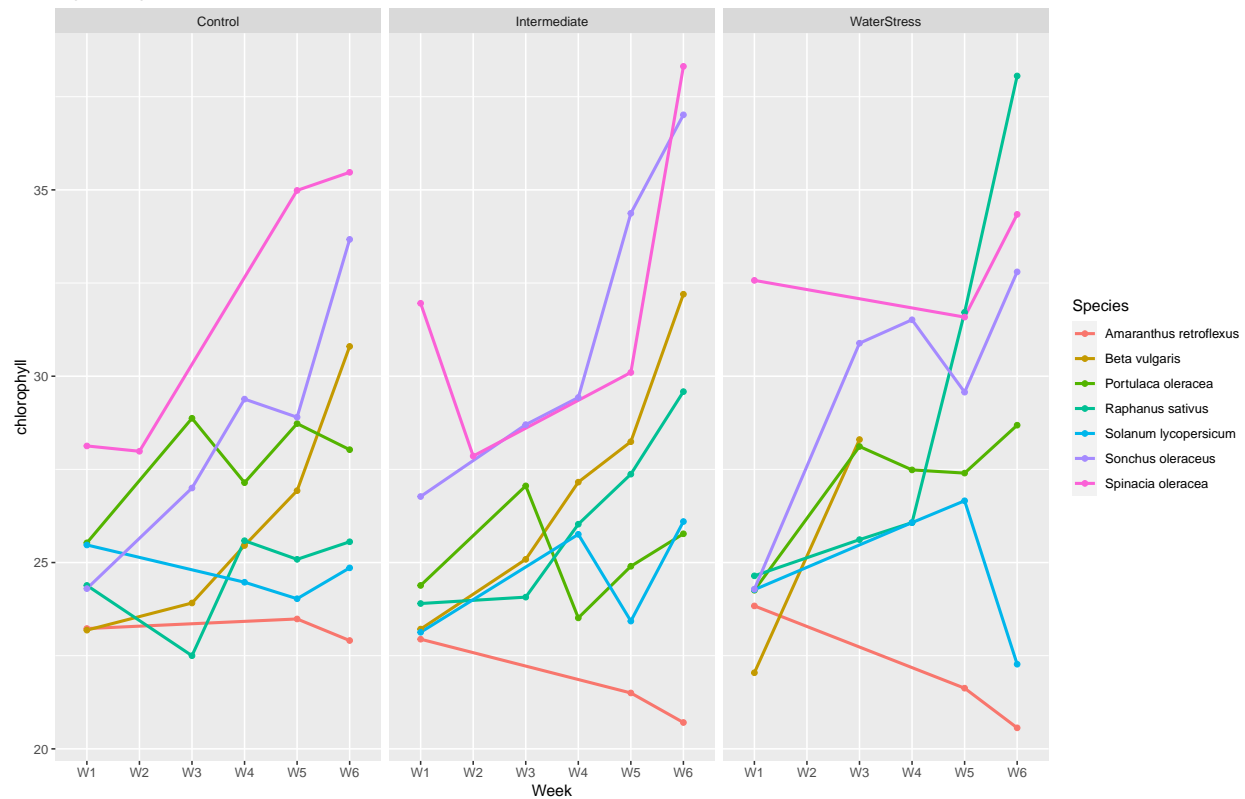
Fig.3 line graph of Leaf Width



```
sumtable <- na.omit(sumtable) ###to exclude NA values from chlorophyll variable
chlorophyll <- ggplot(sumtable, aes(x=Week, y=chlorophyll, group=Species,
  col=Species))+
  geom_line(size=1)+
  geom_point()+
  facet_wrap(~Treatment)+ggtitle("Fig.4 line graph of Chlorophyll Content")

chlorophyll
```

Fig.4 line graph of Chlorophyll Content



#####3

Principal component analysis

```
data <- ws0 %>% filter(Week=="W6")

#import the dataset to be used for the principal component analysis
a <- read.csv("data/myframe.csv", header=TRUE, row.names = "PlantId")
##PlantId has been used as the rowname in order to display individual point in

newV <- names(a) ##Create a new vector with the names of the variables
####conduct the PCA Analysis
res.pcac <- PCA(a, ncp = 2, graph = FALSE)

##-----
```

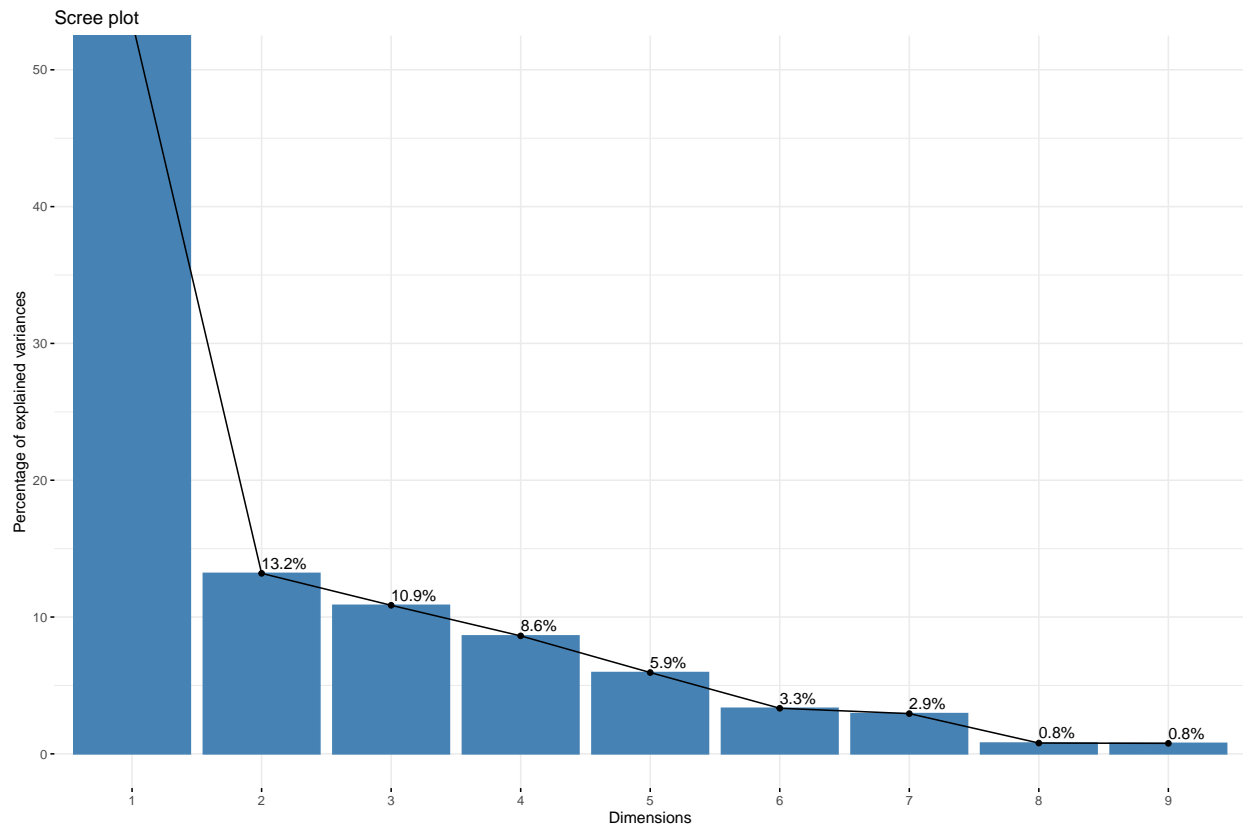
Scree Plot

The Eigenvalues are shown visually in the scree plot. The graph was created to show how many significant elements there were in the datasets. In this instance, the first two principal components together account for more than 60%. As a result, just the first “major component” was chosen to be taken into consideration when looking at the factors that best explain the water stress that the plants experienced.


```
eig.val <- get_eigenvalue(res.pcac)
eig.val
```

```
##      eigenvalue variance.percent cumulative.variance.percent
## Dim.1 4.81913983      53.5459981          53.54600
## Dim.2 1.18738796      13.1931996          66.73920
## Dim.3 0.97732119      10.8591244          77.59832
## Dim.4 0.77615643       8.6239604          86.22228
## Dim.5 0.53496641       5.9440712          92.16635
## Dim.6 0.29996500       3.3329444          95.49930
## Dim.7 0.26478232       2.9420258          98.44132
## Dim.8 0.07104183       0.7893537          99.23068
## Dim.9 0.06923903       0.7693225         100.00000
```

```
## Scree plot show the amount of variable explained by each component
scree <- fviz_eig(res.pcac, addlabels = TRUE, ylim = c(0, 50))
scree
```



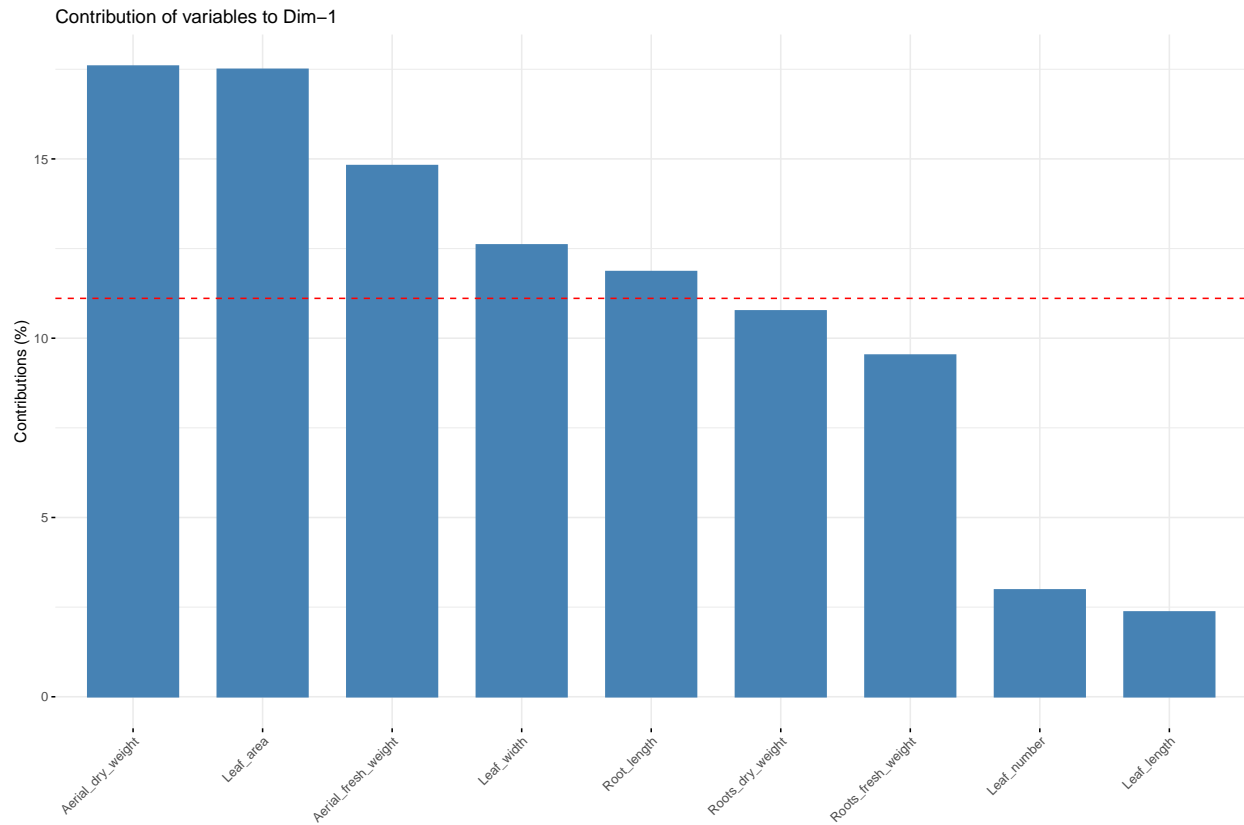
```
##-----
```

Contribution

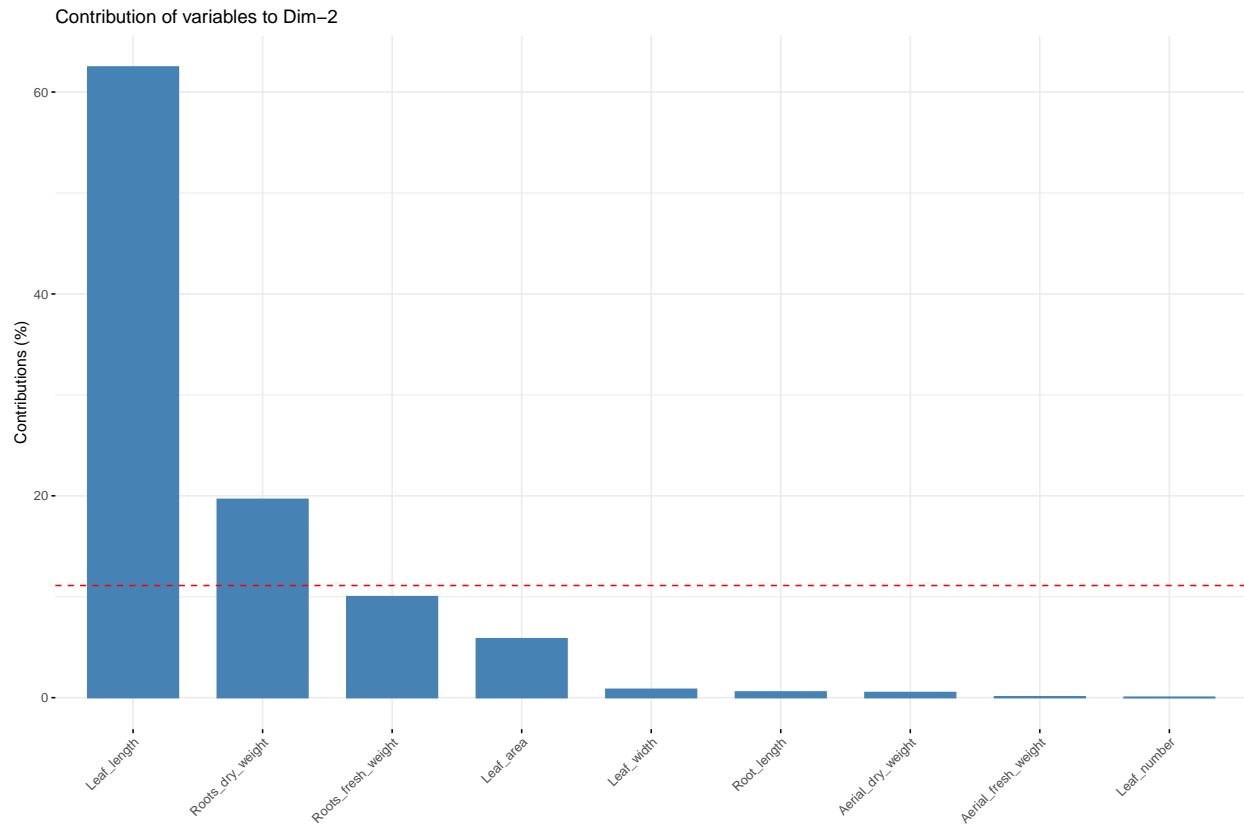
Variables are displayed together with their percentage contributions to each principle component in the principal component analysis bar plots. Aerial dry weight, leaf area, fresh aerial weight, leaf breadth, and root lengths contribute more in the first dimension, and in the second, leaf length and root dry weight are

more important. As a result, we can infer that these characteristics may be crucial for describing plant stress.

```
# Contributions of variables to PC1  
c1 <- fviz_contrib(res.pcac, choice = "var", axes = 1, top = 10)  
c1
```



```
# Contributions of variables to PC2  
c2 <- fviz_contrib(res.pcac, choice = "var", axes = 2, top = 10)  
c2
```

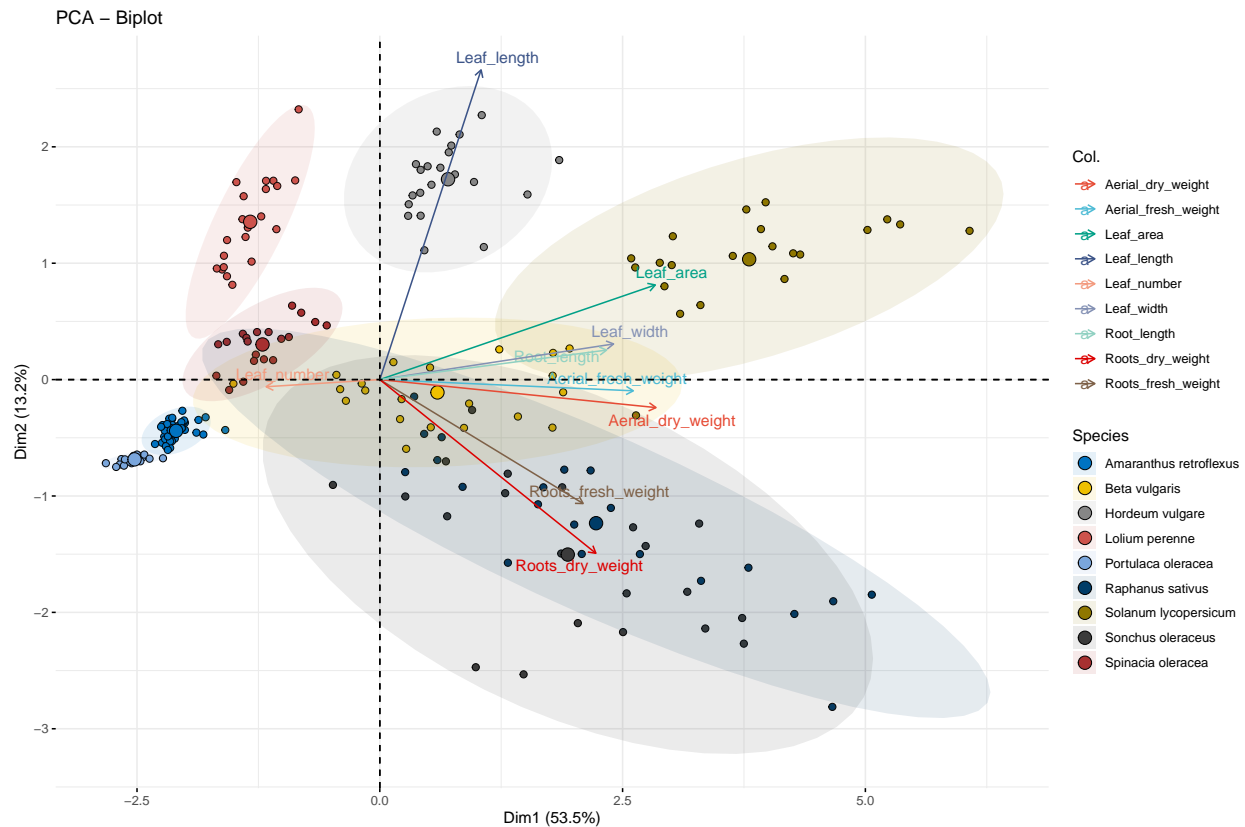


PCA Biplot

To find the key factors that best explain the changes in the data, a Principal Component Analysis biplot was created. These are the groups of observations that share a common trait. The many species are represented by the points. The lines with greater lengths are the ones that are primarily responsible for the species' variation. The biplot demonstrates how the species varies as a result of several parameters. Different species exhibit their responses to water stress in different ways. For instance, *Hordeum vulgare* differs primarily as a result of the leaf length.

```
#### Plot the biplot
print1 <- fviz_pca_biplot(res.pcac,
# Fill individuals by groups
geom.ind = "point",
pointshape = 21,
pointsize = 2.0,
addEllipses = TRUE,
fill.ind = data$Species,
col.ind = "black",
# Color variable by groups
col.var = newV,
legend.title = list(fill = "Species"),
repel = TRUE # Avoid label overplotting
)+
ggpubr::fill_palette("jco")+ # Individual fill color
ggpubr::color_palette("npg") # Variable colors
```

```
print1
```



Cluster Analysis

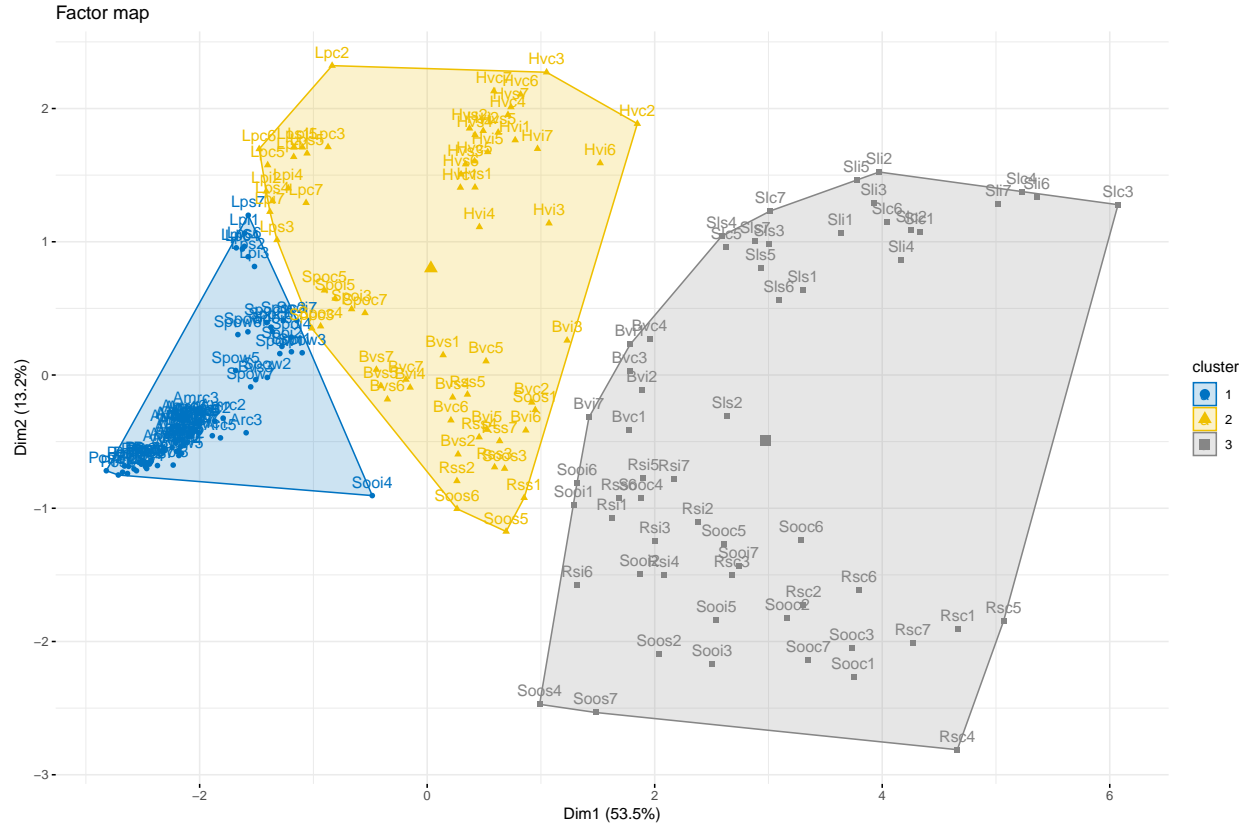
clusters are visible here, and for each cluster, a different set of variables determines the variance of the clusters. For cluster 1, for instance, the relevant variables are the number of leaves, their width, the weight of the roots and the weight of the aerial parts. Beta vulgaris is the dominant species found in this cluster. This indicates that the Beta vulgaris' response to water stress depended significantly on the number of leaves, the size of the leaves, the fresh weight of the roots, and the fresh weight of the aerial parts.

```
#- Conduct the Cluster analysis using the principal component results
```

```
res.hcpc <- HCPC (res.pcac, graph = FALSE)

cluster <- fviz_cluster(res.hcpc,
  repel = FALSE, # Avoid label overlapping
  show.clust.cent = TRUE, # Show cluster centers
  palette = "jco",
  ggtheme = theme_minimal(),
  main = "Factor map"
)

cluster
```



The most important variable contributing to cluster

```
mycl <- res.hcpc$desc.var$quanti
mycl
```

```
## $'1'
##
##          v.test Mean in category Overall mean sd in category
## Leaf_number      5.347200    13.49878298    11.1018768    6.45828599
## Roots_fresh_weight -6.305440     0.05781609     1.4114286    0.06883973
## Leaf_width        -6.484251     1.63400472     4.3145639    0.98675817
## Leaf_length       -7.357696     7.42883296    14.9371832    6.94554959
## Roots_dry_weight  -8.126645     0.01145977     0.1903238    0.03283389
## Aerial_fresh_weight -8.679526     1.51701149     6.4214762    1.43612034
## Aerial_dry_weight  -9.625577     0.10172414     0.5798571    0.10870404
## Leaf_area        -10.334543     4.45536369    29.8694602    4.44202016
## Root_length      -10.904383     5.91568966    16.0425333    3.80004127
##
## Overall sd      p.value
## Leaf_number      5.4501044 8.932526e-08
## Roots_fresh_weight 2.6101120 2.873757e-10
## Leaf_width        5.0262699 8.917384e-11
## Leaf_length      12.4074598 1.871111e-13
## Roots_dry_weight  0.2676036 4.413367e-16
## Aerial_fresh_weight 6.8703018 3.974214e-18
## Aerial_dry_weight  0.6039517 6.235609e-22
## Leaf_area        29.8994779 4.917495e-25
## Root_length      11.2915415 1.098363e-27
##
## $'2'
```

```
##          v.test Mean in category Overall mean sd in category
## Leaf_length      7.600593      24.6800000    14.937183    13.8505451
## Root_length      3.734479      20.3990308    16.042533     9.3050744
## Roots_fresh_weight -2.117945      0.8403077     1.411429     0.7489233
## Leaf_number      -2.476060      9.7076923    11.101877     4.3135674
## Leaf_width       -3.224792      2.6400000     4.314564     1.9856640
##          Overall sd      p.value
## Leaf_length      12.407460 2.947767e-14
## Root_length      11.291542 1.881040e-04
## Roots_fresh_weight 2.610112 3.417971e-02
## Leaf_number      5.450104 1.328411e-02
## Leaf_width       5.026270 1.260642e-03
##
## $'3'
##          v.test Mean in category Overall mean sd in category
## Aerial_dry_weight 11.532974      1.3598276     0.5798571     0.5295264
## Aerial_fresh_weight 10.676482      14.6351724     6.4214762     6.4604121
## Leaf_width       10.478228      10.2120690     4.3145639     6.0737444
## Roots_dry_weight 10.472391      0.5041379     0.1903238     0.2919969
## Leaf_area        10.129276      63.7832759    29.8694602    28.0812135
## Roots_fresh_weight 9.136789      4.0818966     1.4114286     3.7128943
## Root_length      8.152407      26.3505172    16.0425333     7.8476518
## Leaf_number      -3.331033      9.0689655    11.1018768     3.0335030
##          Overall sd      p.value
## Aerial_dry_weight 0.6039517 8.998054e-31
## Aerial_fresh_weight 6.8703018 1.311481e-26
## Leaf_width       5.0262699 1.087620e-25
## Roots_dry_weight 0.2676036 1.156834e-25
## Leaf_area        29.8994779 4.096722e-24
## Roots_fresh_weight 2.6101120 6.433435e-20
## Root_length      11.2915415 3.567520e-16
## Leaf_number      5.4501044 8.652433e-04
```

```
### store a dataframe containing the original variable and assigned cluster
b <- res.hcpc$data.clust
```

```
#NEXT we
```

```
###combine with the species and the treatment column with the dataframe
#to see the cluster where all the Species in the control treatment are?
```

```
comb <- b %>% mutate(Species=data$Species, Treatment=data$Treatment) %>%
  arrange(Species)
```

```
filt <- comb[, -c(1:9)]
```

```
#c1 <-filt %>% filter(clust=="3")
```

```
#summary(c1$Species)
```

```
## inference from the cluster analysis result in filt dataframe
```

```
# Most of the species clustered together without separation of individuals under
# different treatment
# Species with similar characteristics clustered together
```

Anova analysis and Result

According to the anova table, leaf length is significant for five species, leaf area for six species, aerial fresh weight for eight species, aerial dry weight for seven species, roots fresh weight for four species, and roots dry weight for three species are all significant. In this instance as well, the difference between cultivated species and weeds in the treatment's impact on some growth parameters is quite substantial. *Portulaca oleracea*, *Raphanus sativus*, and *Solanum lycopersicum* shows the significance in same parameters.

```
### create a vector with names of variables for which anova analysis would be
### conducted
variables <- c("Soil_humidity", "Plant_height",
              "Leaf_width", "Leaf_number", "Leaf_length",
              "Leaf_area", "Aerial_fresh_weight", "Aerial_dry_weight",
              "Root_length", "Roots_fresh_weight", "Roots_dry_weight")
class(variables)
```

```
## [1] "character"
```

```
##### We create an empty dataframe where the results of the Anova would be
##### written
```

```
Tr <- levels(as.factor(ws0$Treatment))

mean.st <- paste("mean", Tr) ##this will be used to store the mean

class(Tr)
```

```
## [1] "character"
```

```
column.names<-c("Species", "variables", mean.st)
sp <- levels(as.factor(ws0$Species))

rows <- length(sp) * length(variables)

results <- as.data.frame(matrix(nrow=rows, ncol=length(column.names)) )
names(results) <- column.names

results[, "pvalue"] = NA ## create additional column to store the p value
                        ## with initial value NA
results[, "significance"] = NA #create additional column to store the sig
                        ## with initial value NA

ss0 <- ws0%>%filter(Week=="W6") ###subset the week 6 data from the dataframe
## sst <- ws0[ws0$Week=="W6", ]

i <- sp[1]
j <- variables[1]
k <- 1
```

```

for(i in sp ){
  for(j in variables){

    ss1 <- ss0[ss0$Species==i, ]
    ss1 <- ss1 %>%drop_na_(j) ## remove NA from the new subset with only one sp
    lm.1 <- lm(ss1[[j]]~Treatment, data=ss1 ) # fit the lm
    sumoa <- anova(lm.1) #create anova model
    pvalue <- sumoa$`Pr(>F)`[1] #extract the pvalue
    hsd <- HSD.test(lm.1, "Treatment") #conduct poshoc test
    saf <- hsd$means ##store the mean and standard error
    saf1 <- saf[, 1] ##extract only the mean
    safs <- signif(saf1, digits = 3) ####reduce the mean to 3 decimal p
    saf2 <- saf[, 2] ##extract the sd
    safs2 <- signif(saf2, digits = 3) ## reduce sd to 3 dcp
    letter <- hsd$groups$groups[order(rownames(hsd$groups))]
    results$Species[k] <- i
    results$variables[k] <- j
    ## write into result the mean, letter from posthoc and sd
    results[k, mean.st] <- paste(safs, "+", safs2, letter)
    results[k, "pvalue"] <- pvalue #### write the pvalue
    k <- k+1

  }
}

###check the pvalue column, then print significance in sig column if p<0.05
results$significance <- ifelse(results$pvalue >= 0.05, "N.S", "***")

##reduce the p value column to 23 decimal places
results$pvalue <-format(results$pvalue, scientific = TRUE, digit=3)

#### Combine pvalue and significance in a single column
results$pvalue <- paste(results$pvalue, results$significance)

### subset the significant variables
grp1 <- results %>% filter(significance=="***")

##create a new frame with only the species, variable and pvalue
finalframe <- grp1[, -7]

```

Boxplot showing the results of the Anova

```

###
###using slightly modified version of the previous code
### anova is created to get the pvalu, max and a plot is created
k <- 1
newv <- c("Leaf_area", "Leaf_number", "Roots_fresh_weight", "", "Aerial_fresh_weight","Aerial_dry_weight")

```



```

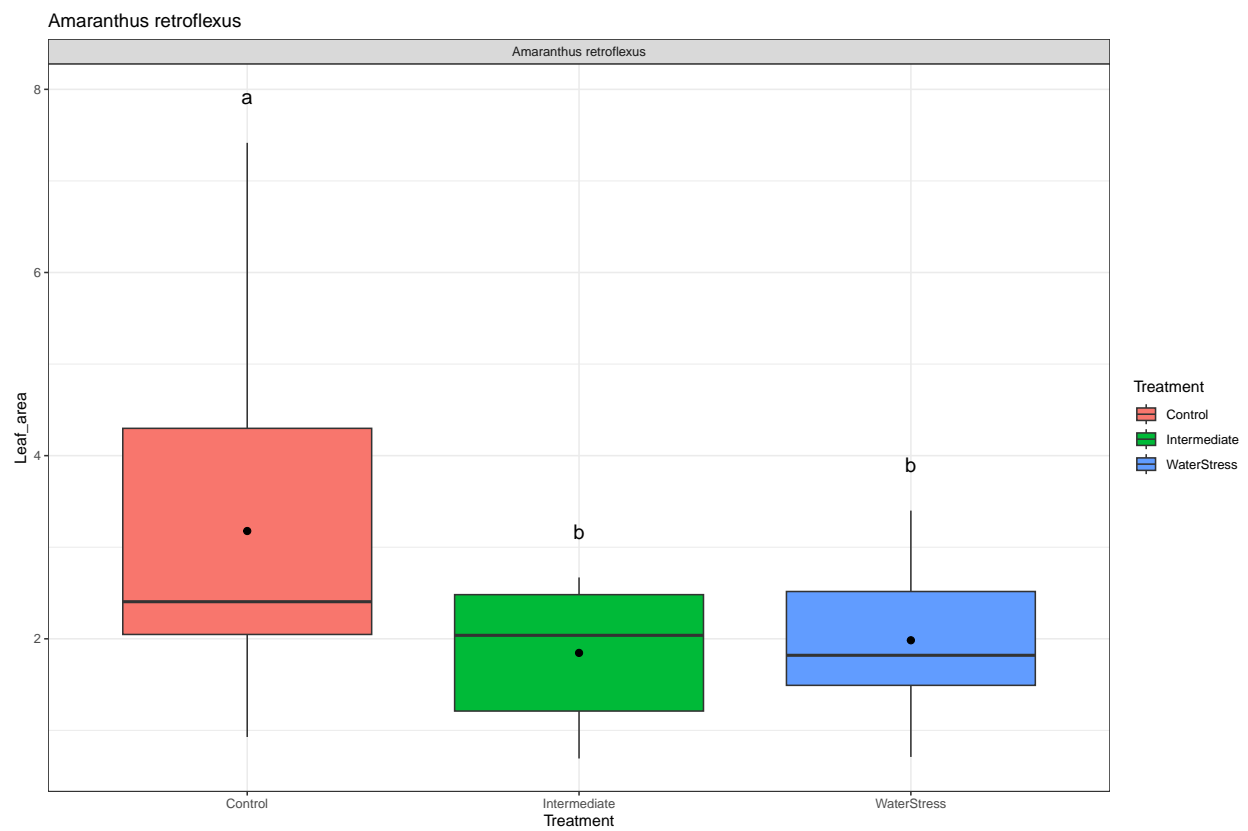
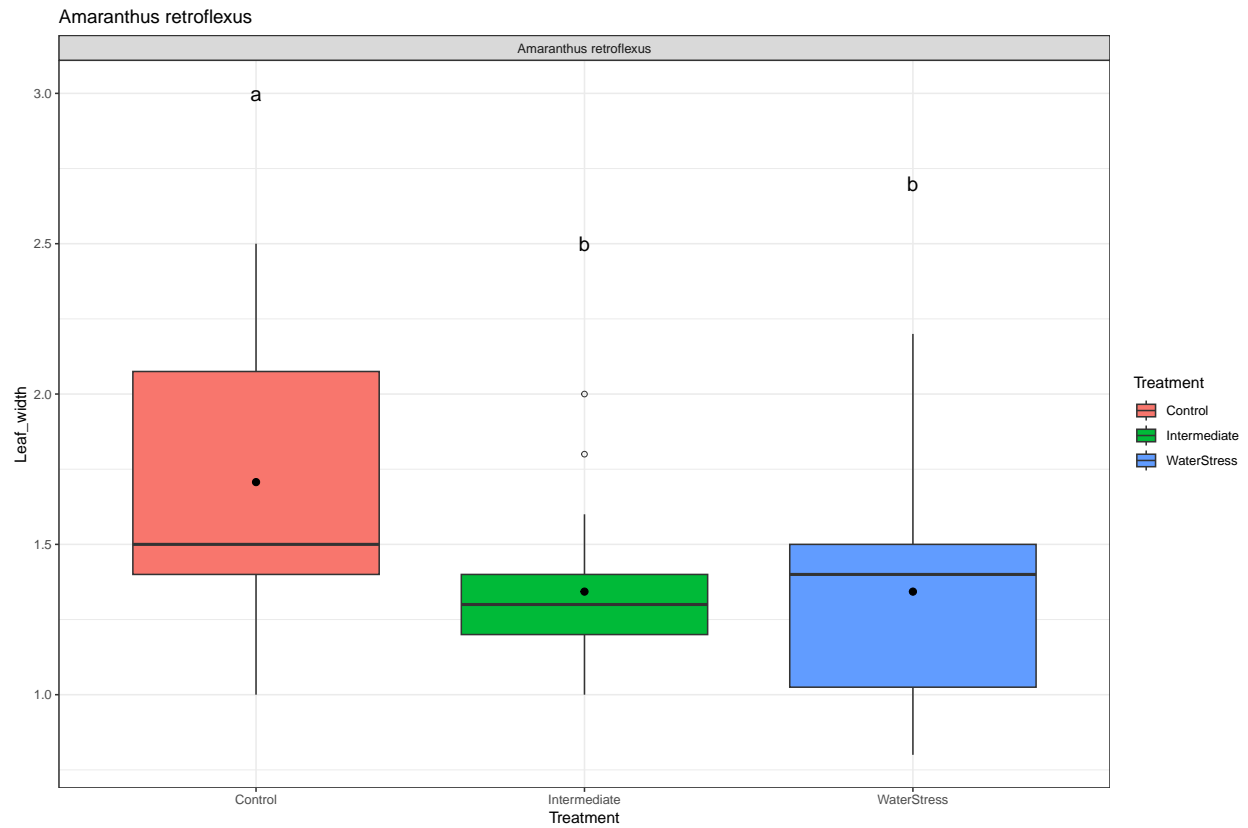
vec <- as.vector(newV)
for(i in sp ){
  for(v in vec){

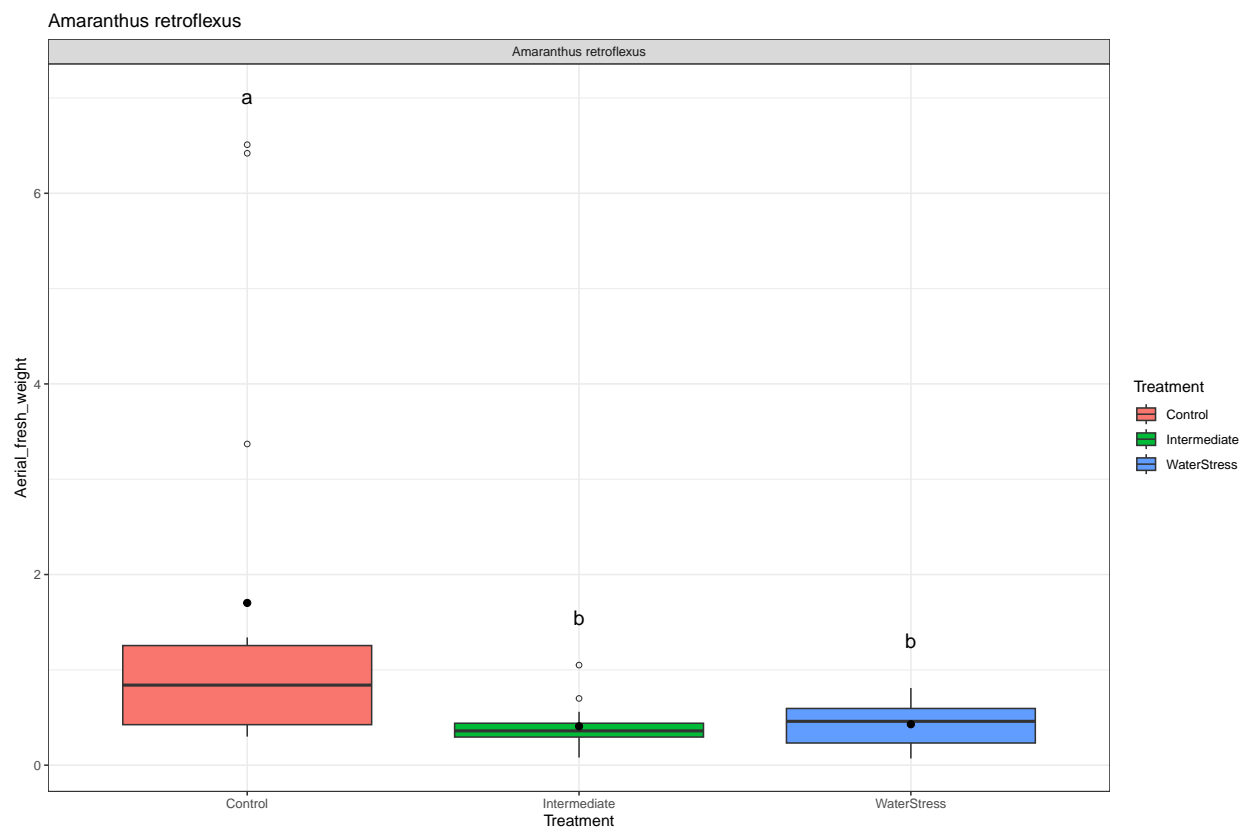
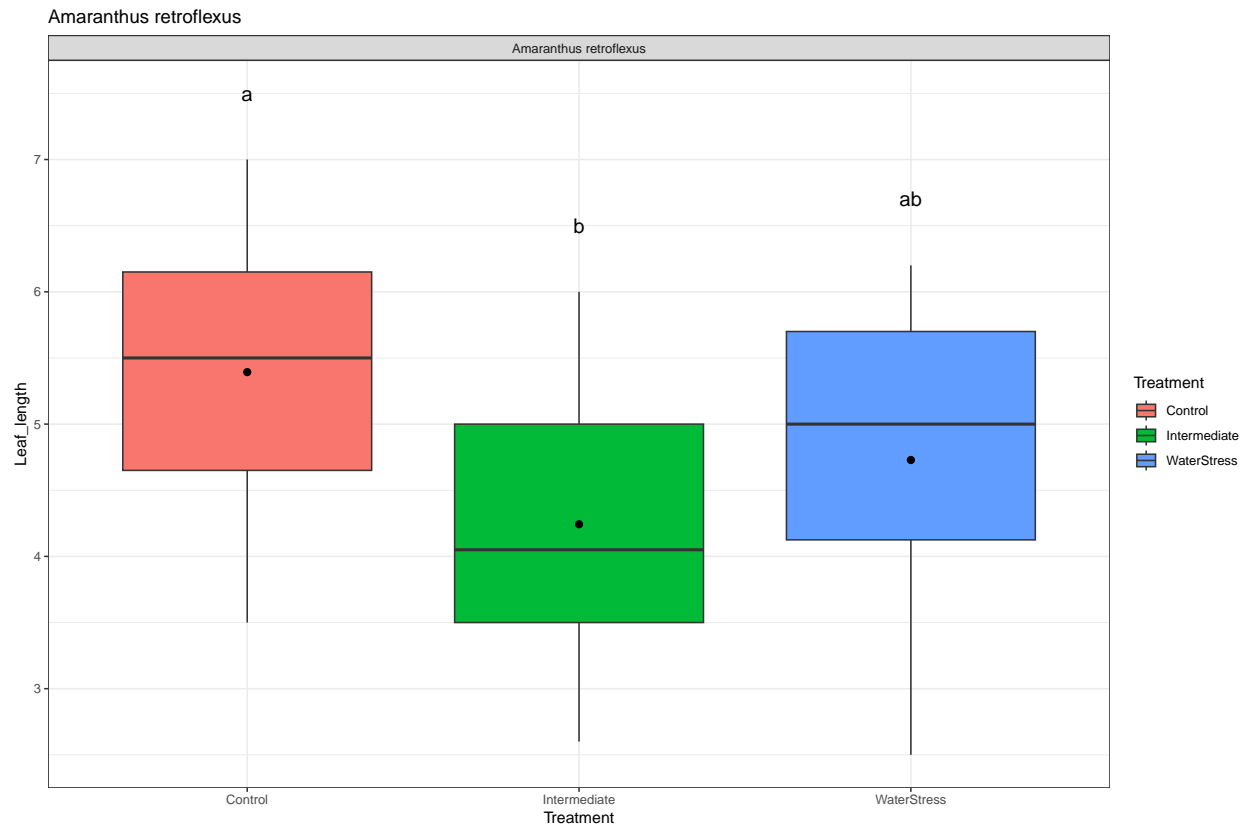
    ss1 <- ss0[ss0$Species==i, ]
    ss1 <- ss1 %>%drop_na_(v)
    levels(ss1$Treatment) <- c("Control", "Intermediate", "WaterStress")
    lm.1 <- aov(ss1[[v]]~Treatment, data=ss1 )
    sumoa <- anova(lm.1) #create anova model
    pvalue <- sumoa$`Pr(>F)`[1]
    hsd <- HSD.test(lm.1, "Treatment")
    maxi <- hsd$means$Max
    ylabv <- names(ss1[v])
    letter <- hsd$groups$groups[order(rownames(hsd$groups))]
    p <- ggplot(ss1, aes(x=Treatment, y=ss1[[ylabv]])) +
    geom_boxplot(aes(x=Treatment, y=ss1[[ylabv]], fill=Treatment), outlier.shape=1)+
    ylab(ylabv)+stat_summary(fun=mean, geom="point", size=2)+
    annotate("text", x = 1:3, y =0.5+maxi, label = letter, size=5)+
    theme_bw()+ggtitle(i)
    p <- p+facet_wrap(~Species)
    nameofp <- paste(i, ylabv)
    ggsave(path="figs", plot =p, file = paste(i, ylabv , ".jpg"), dpi=400)

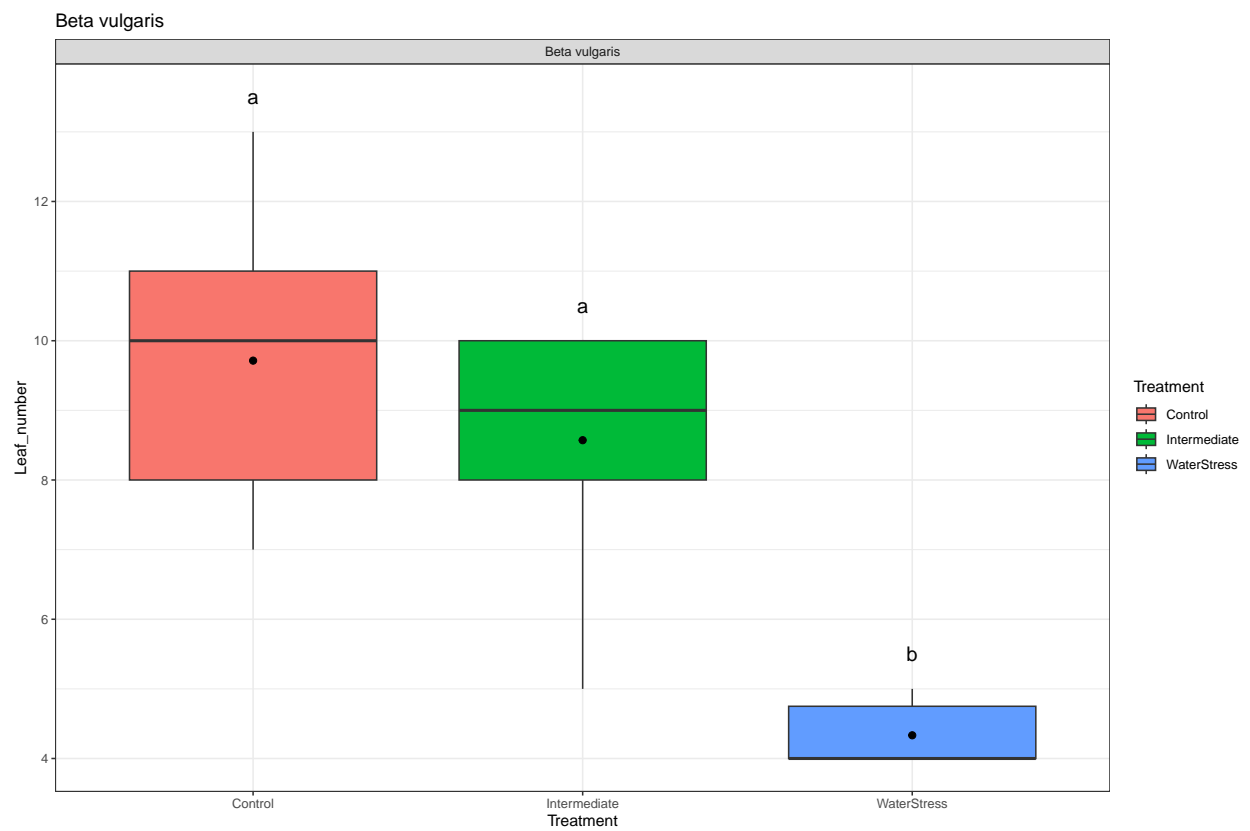
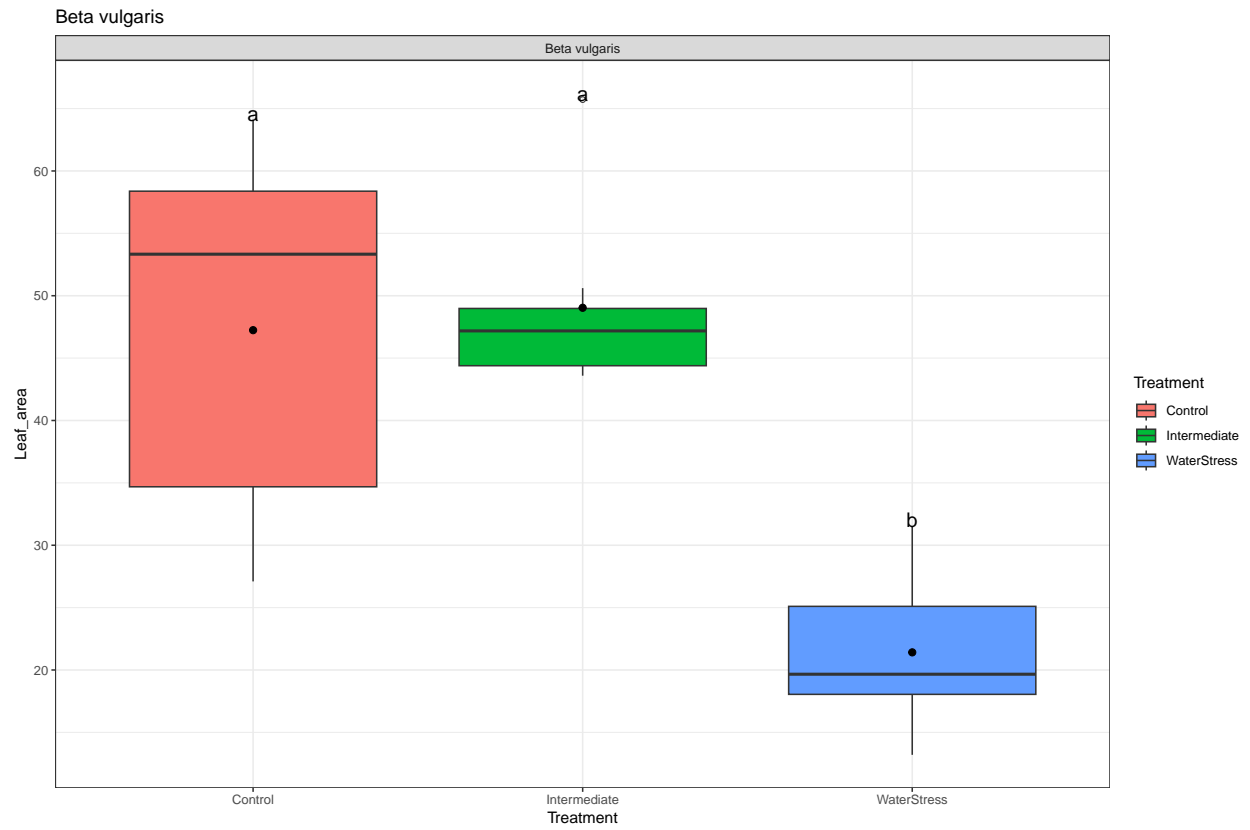
    if(pvalue<0.05) {
      print(p)
    }

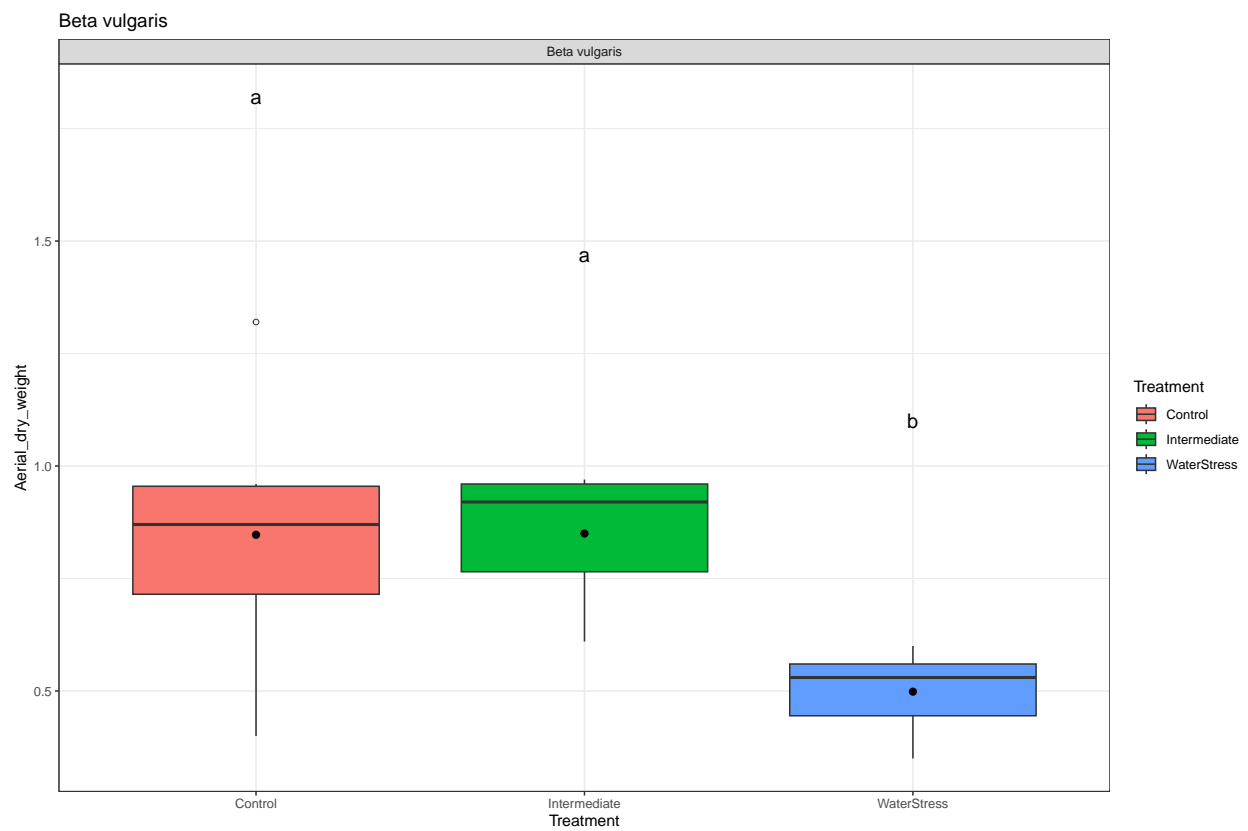
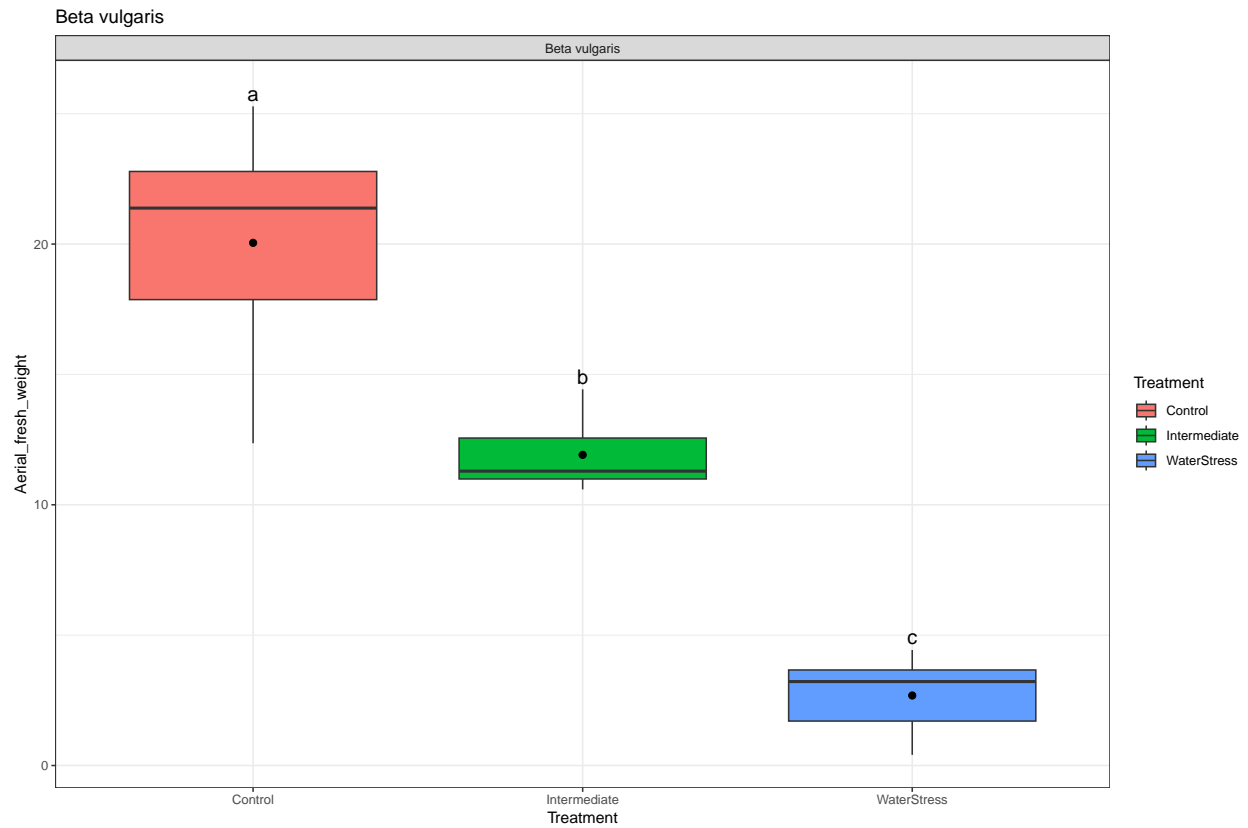
    k <- k+1
  }
}

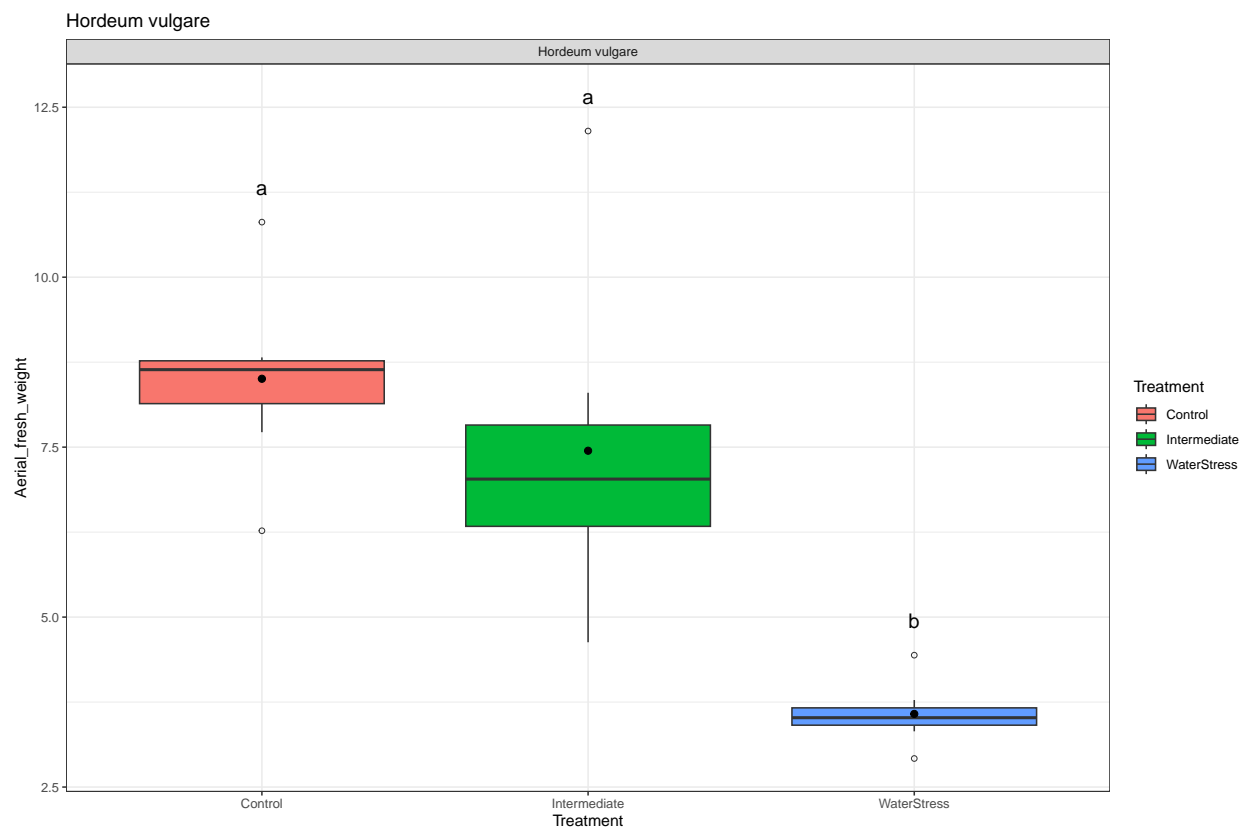
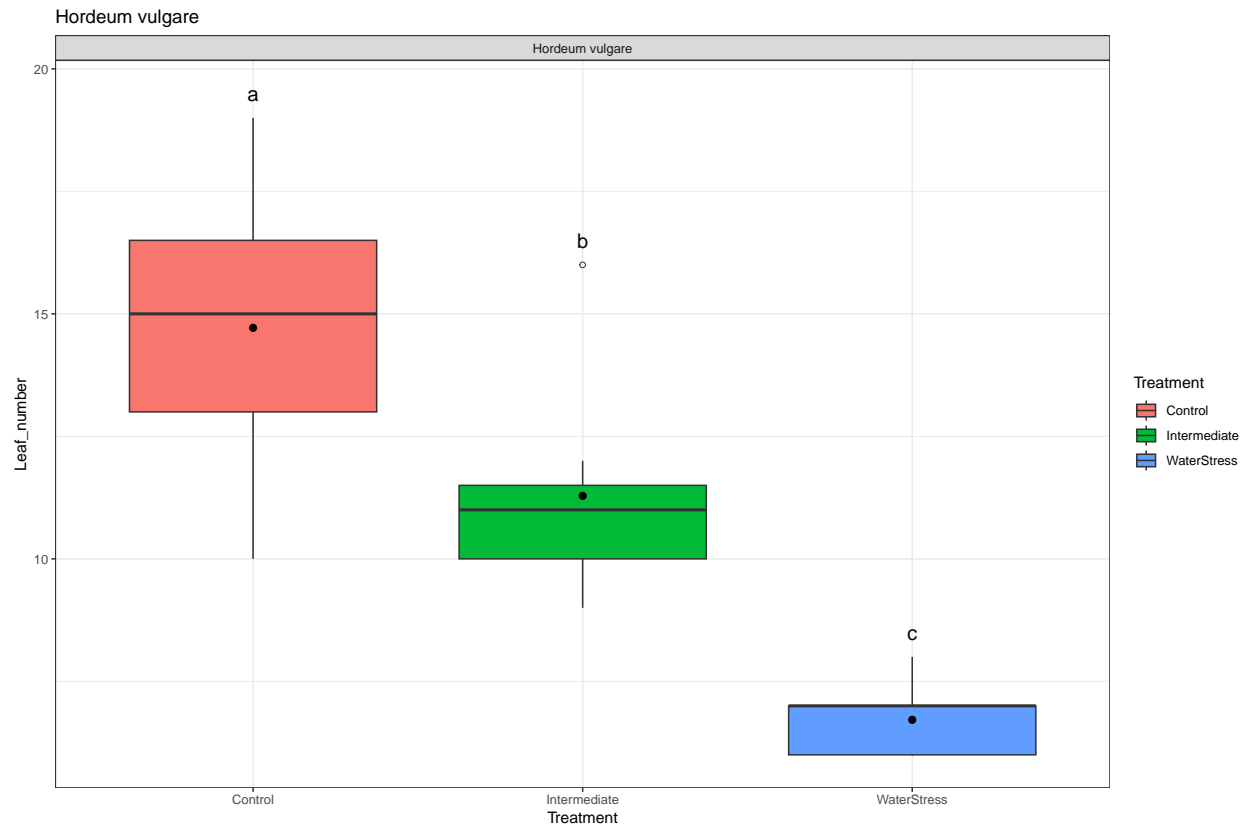
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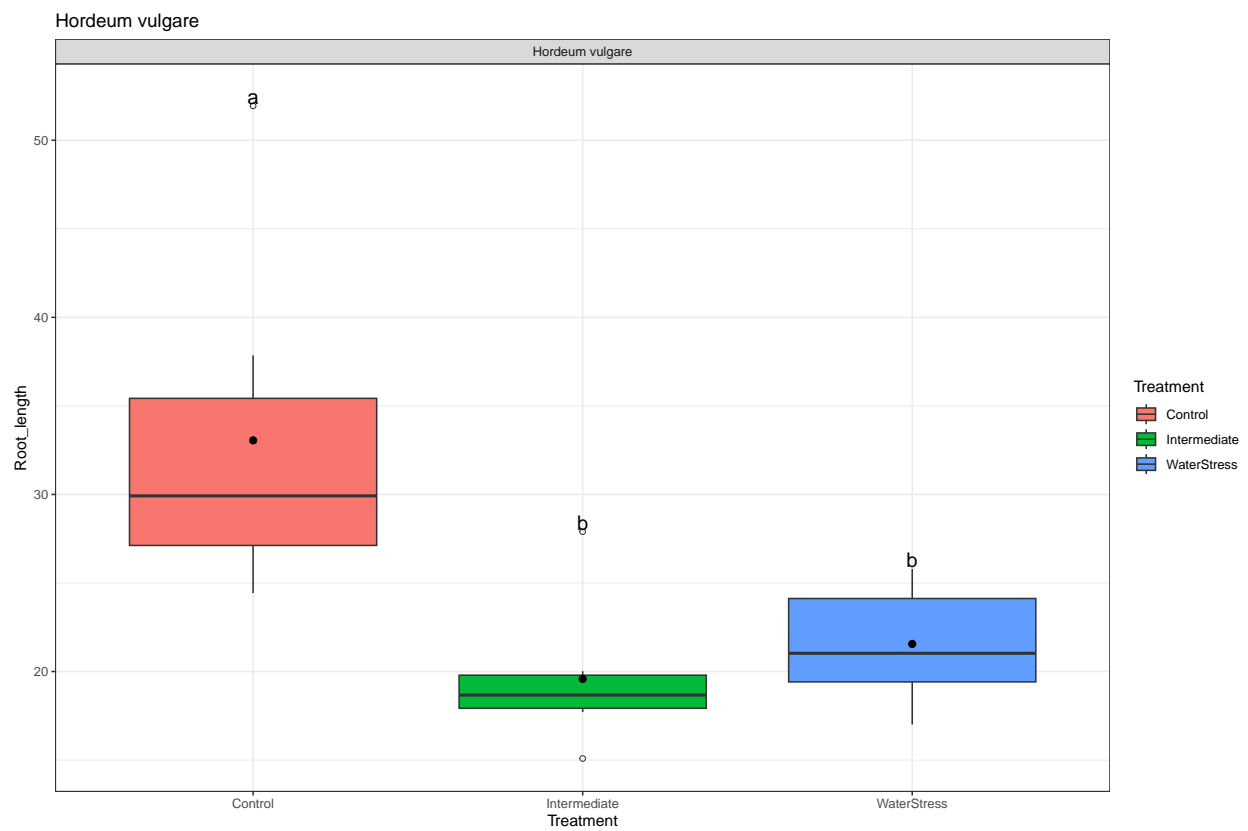
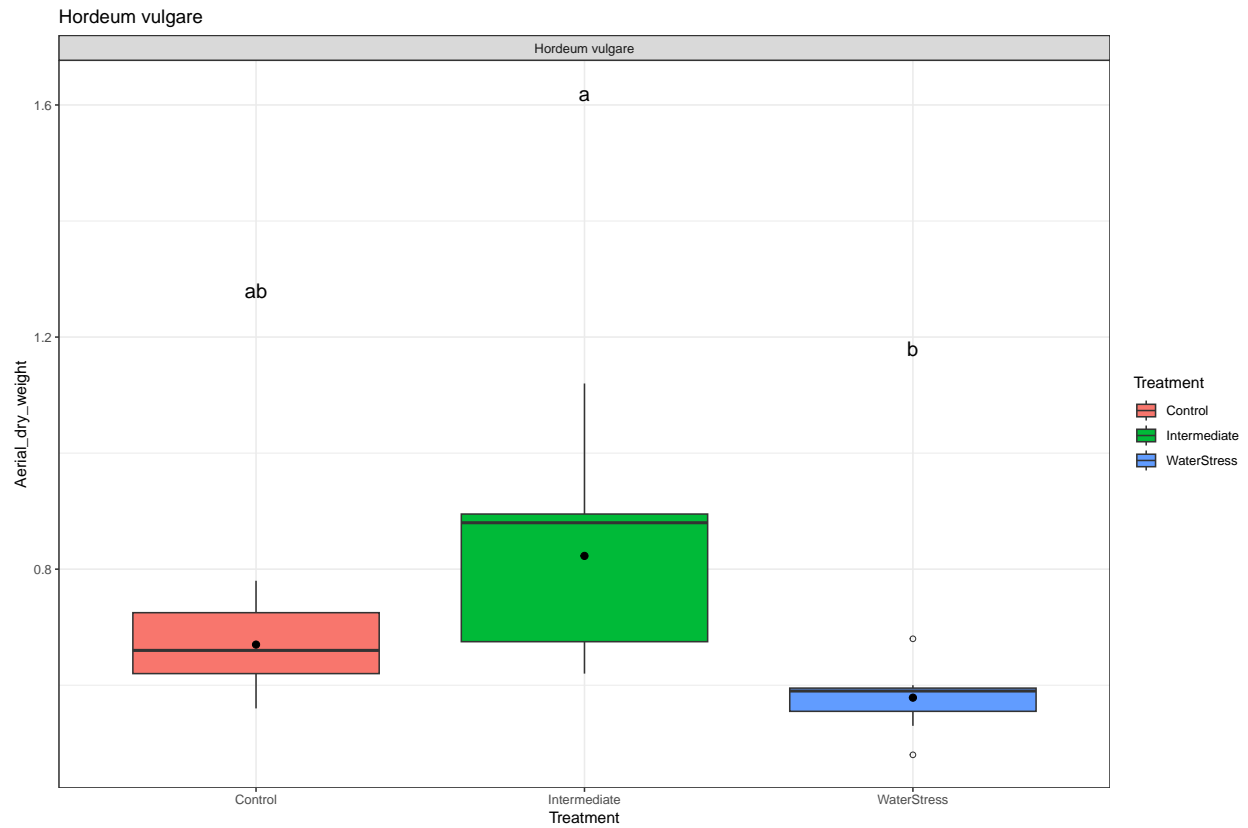


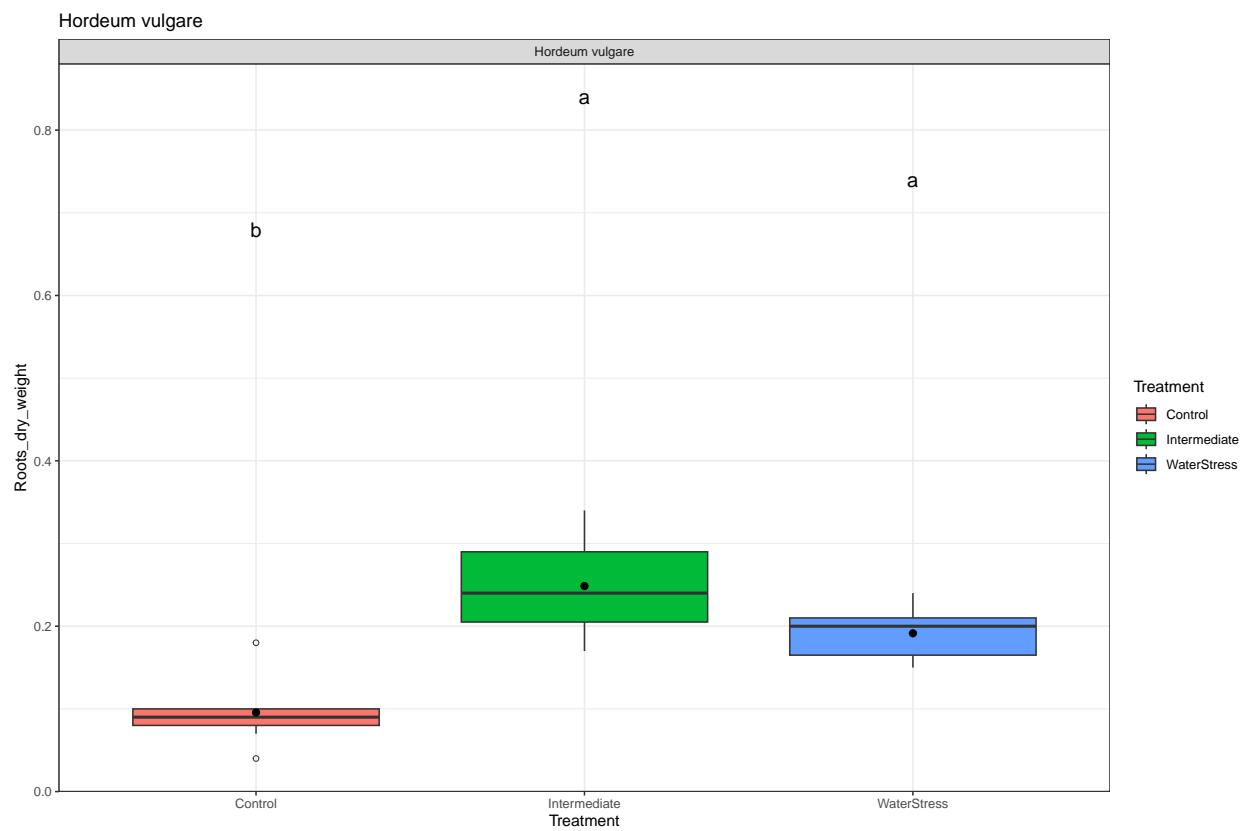
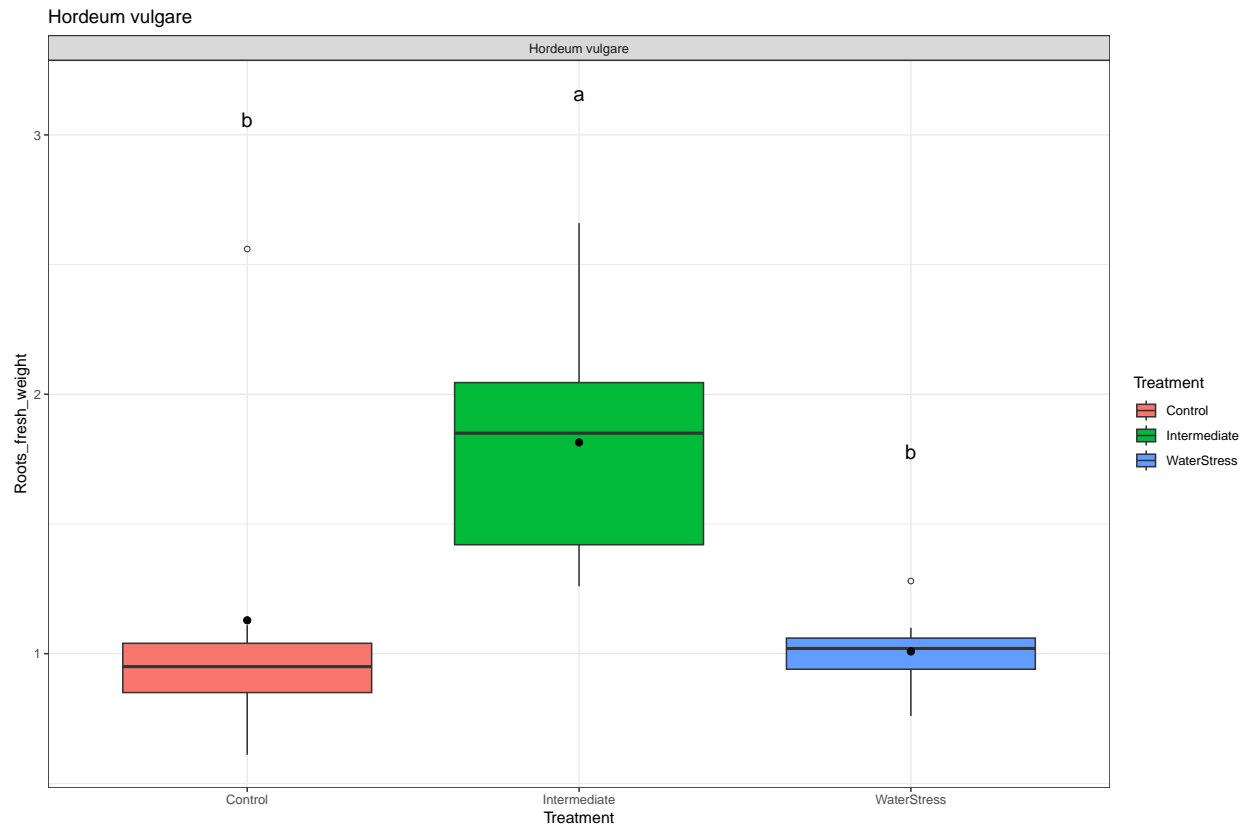


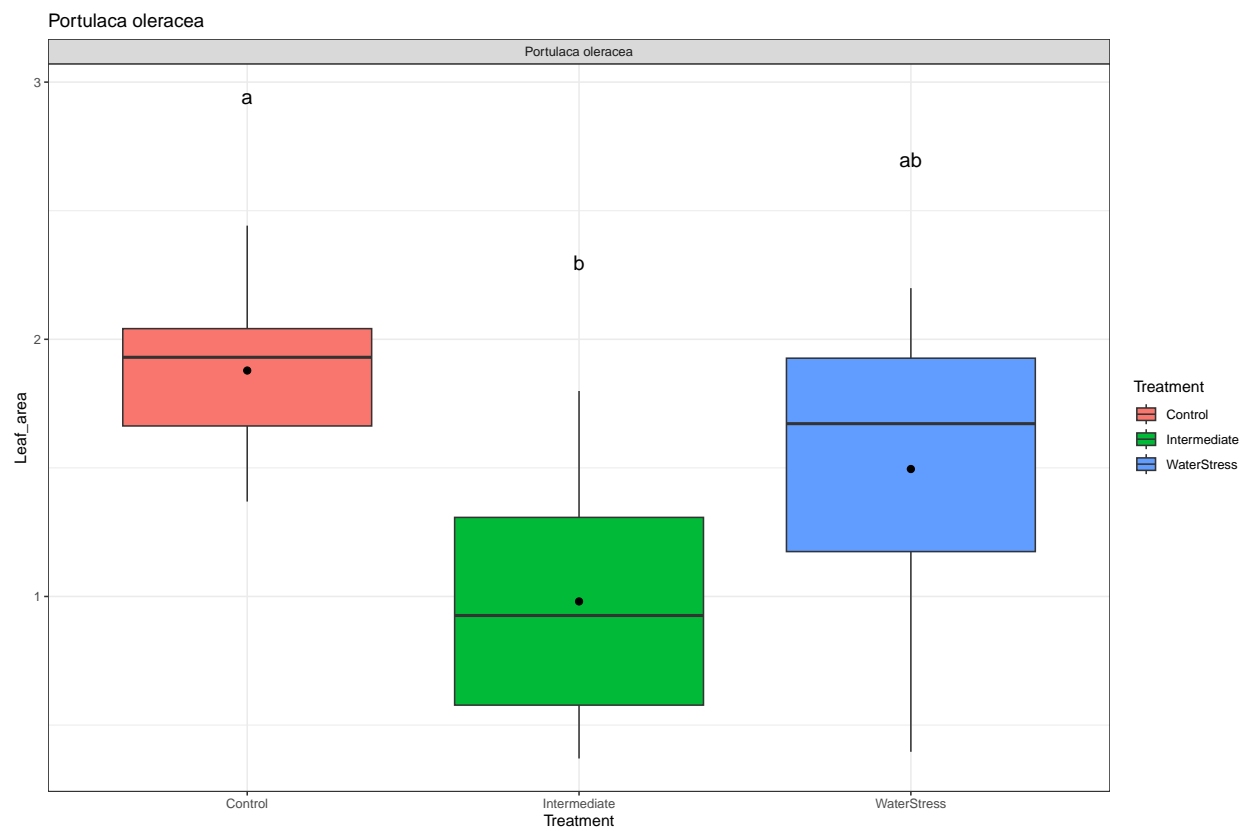
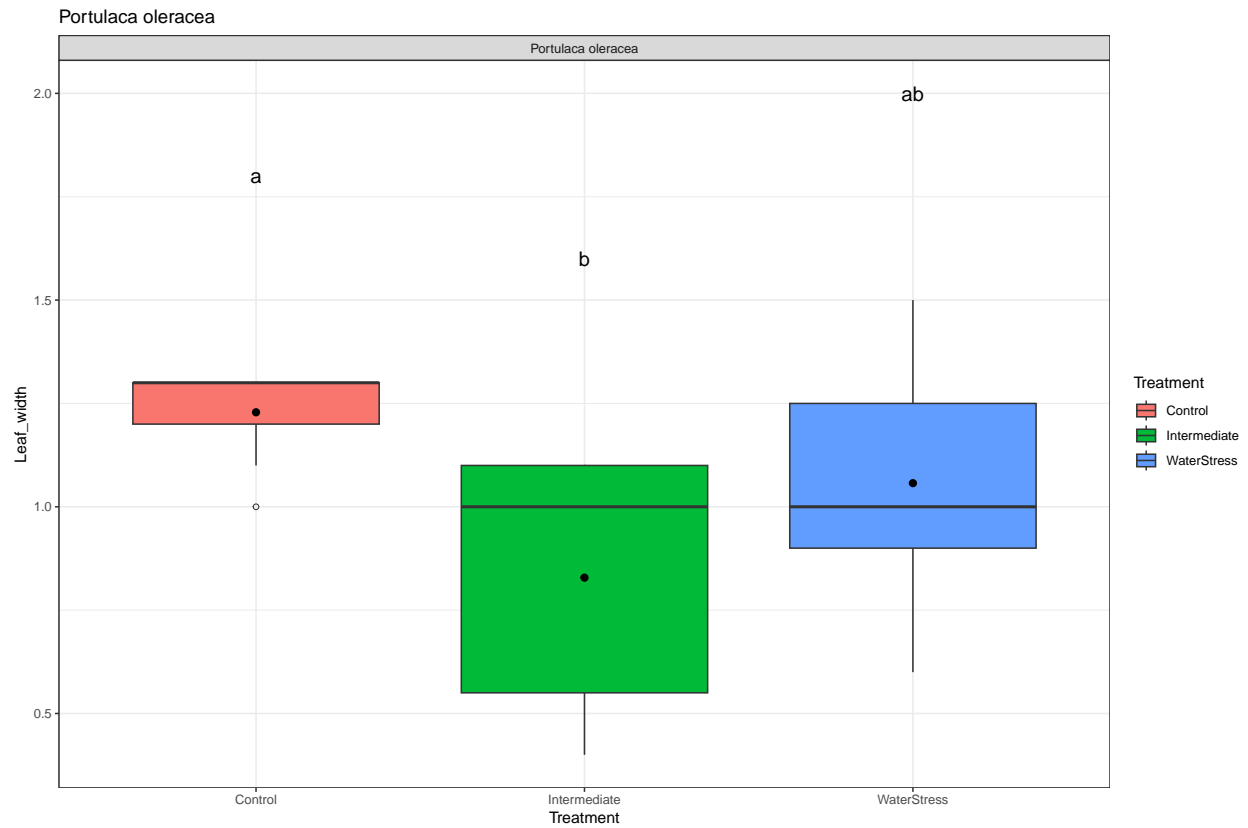


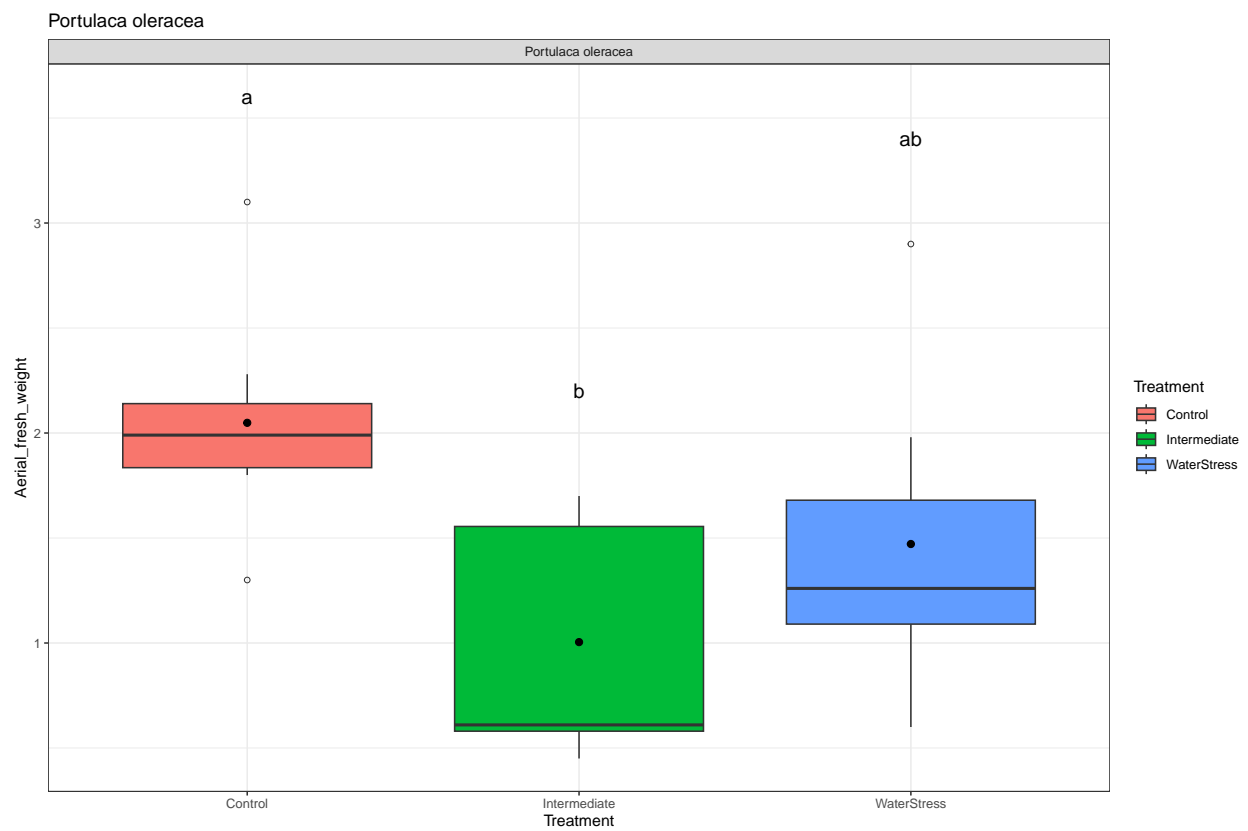
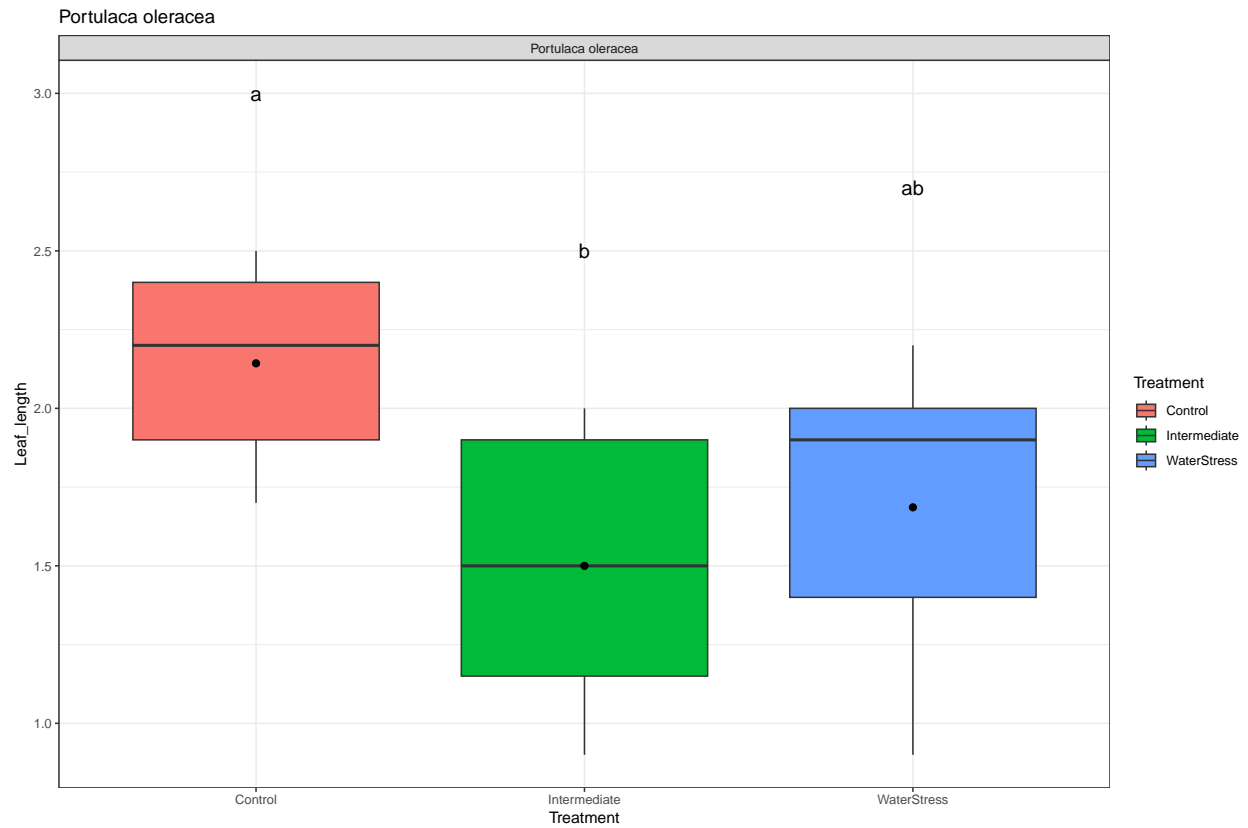


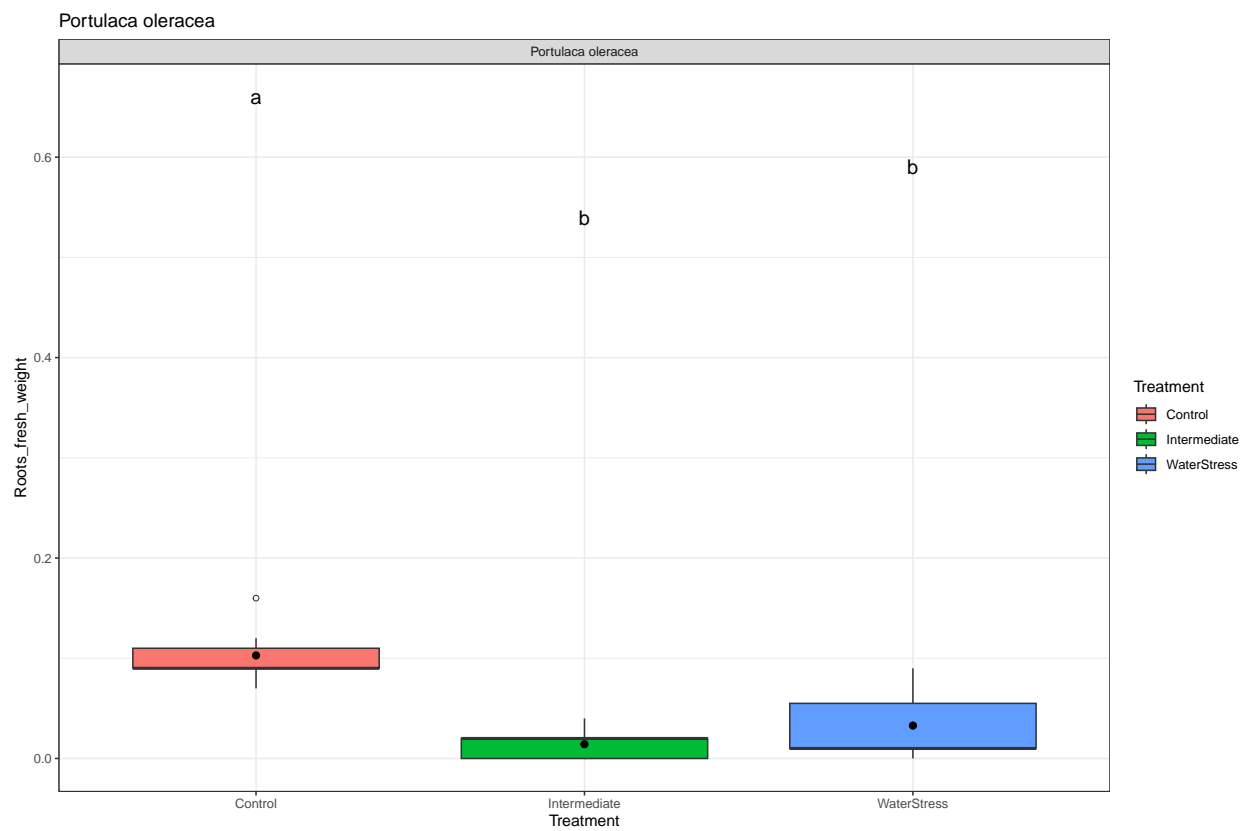
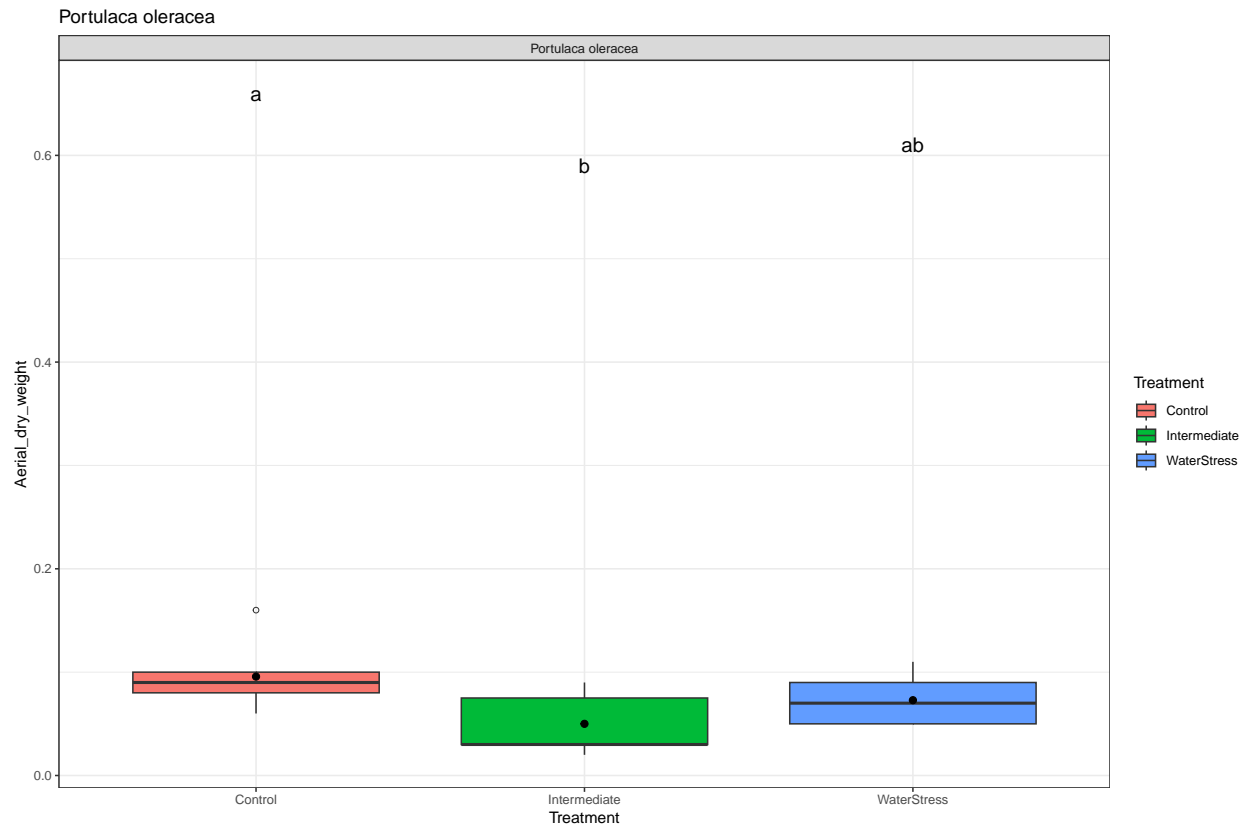


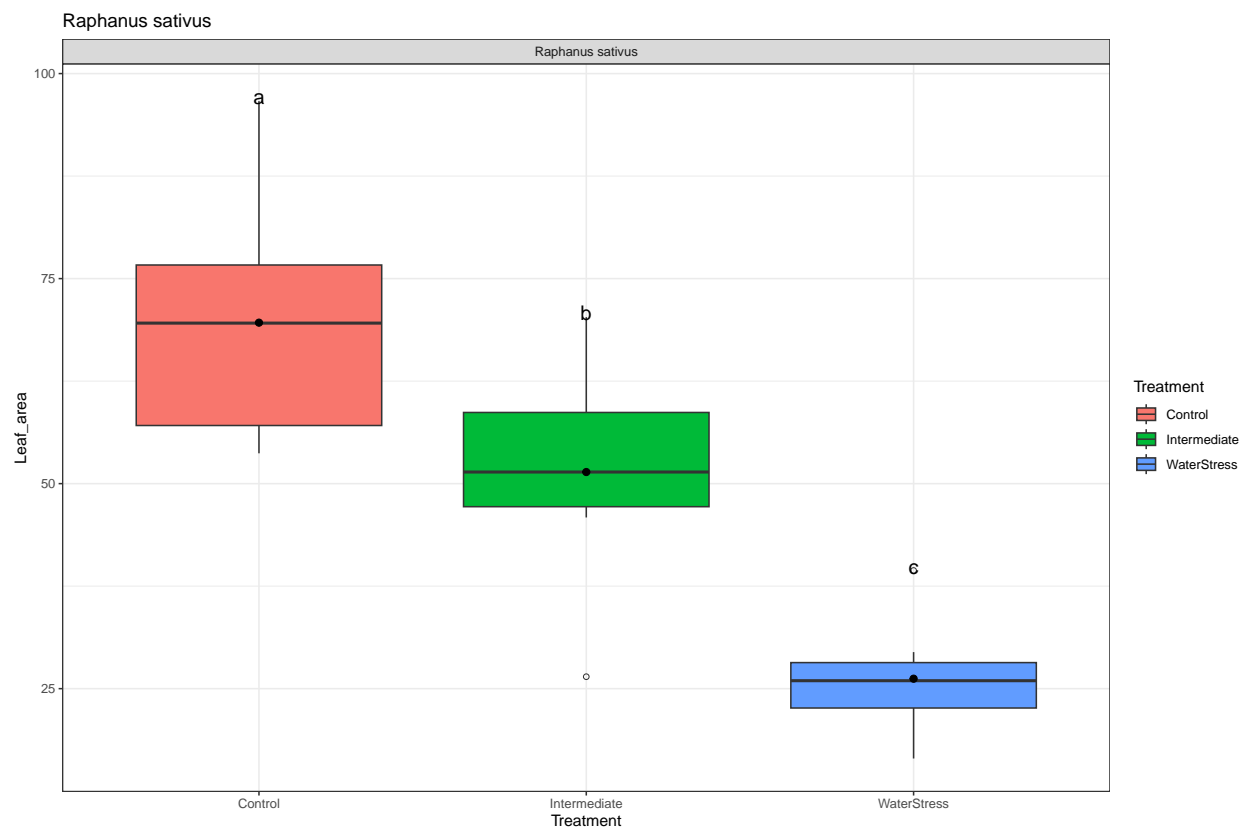
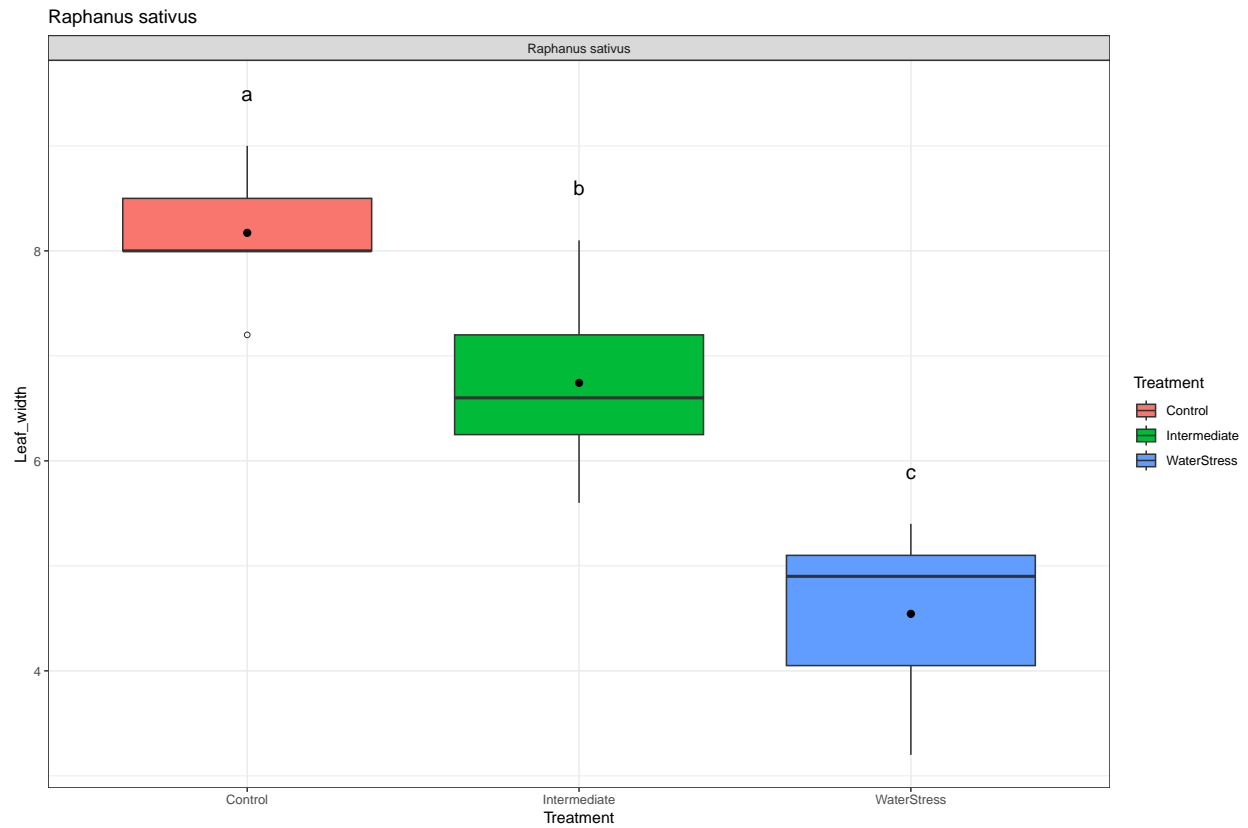


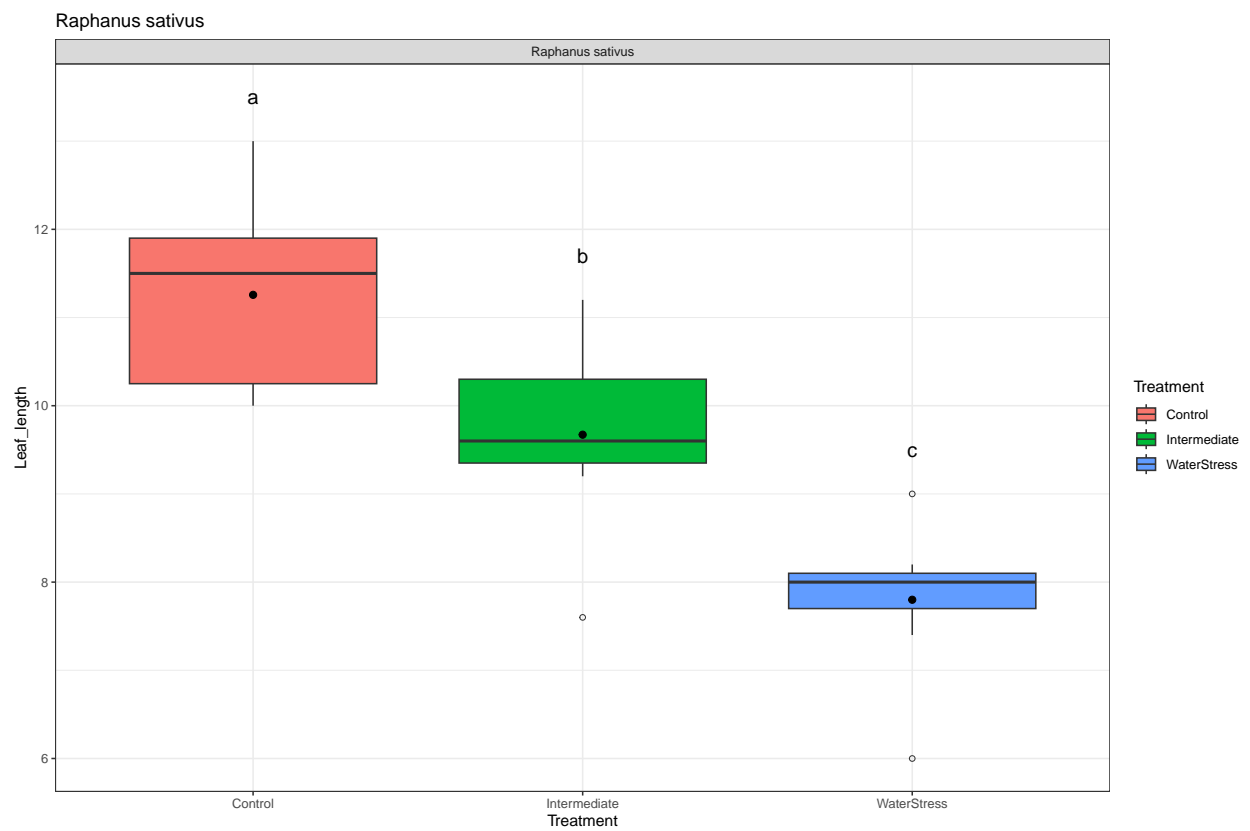
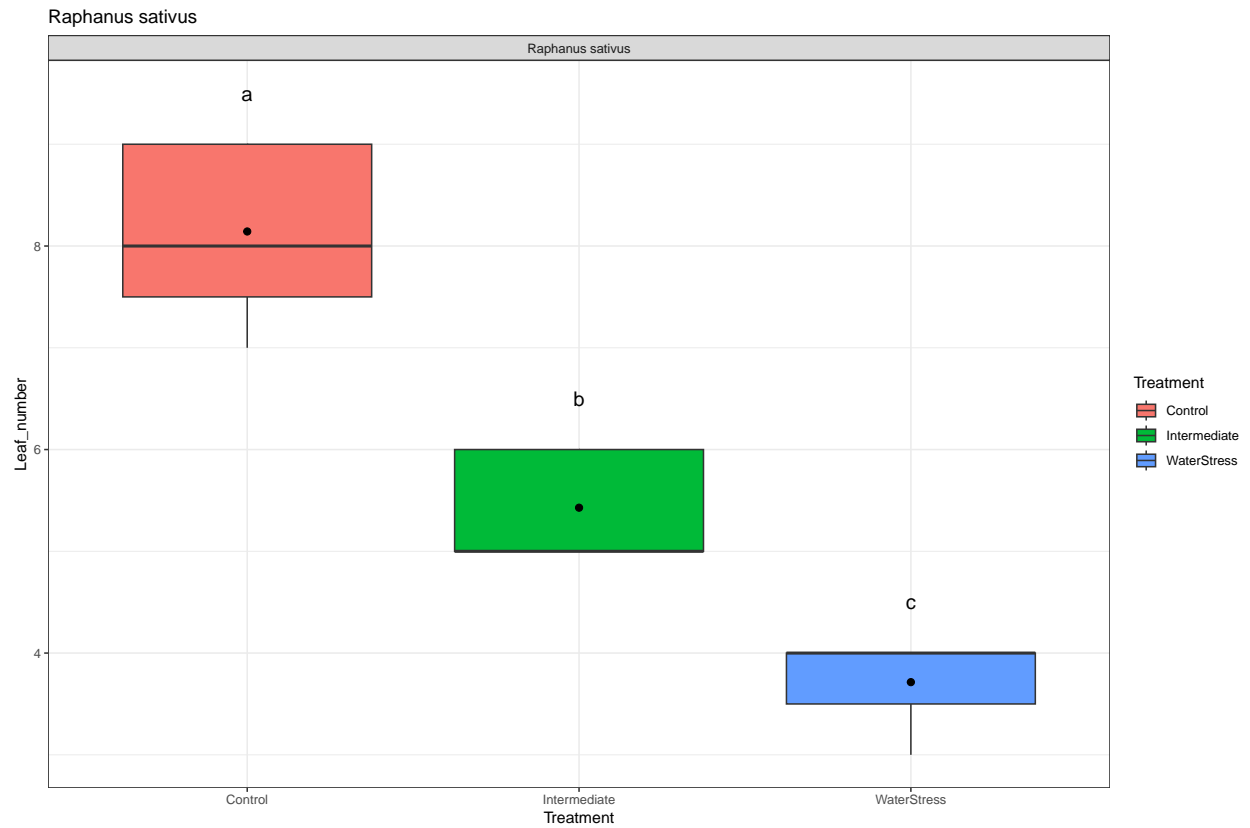


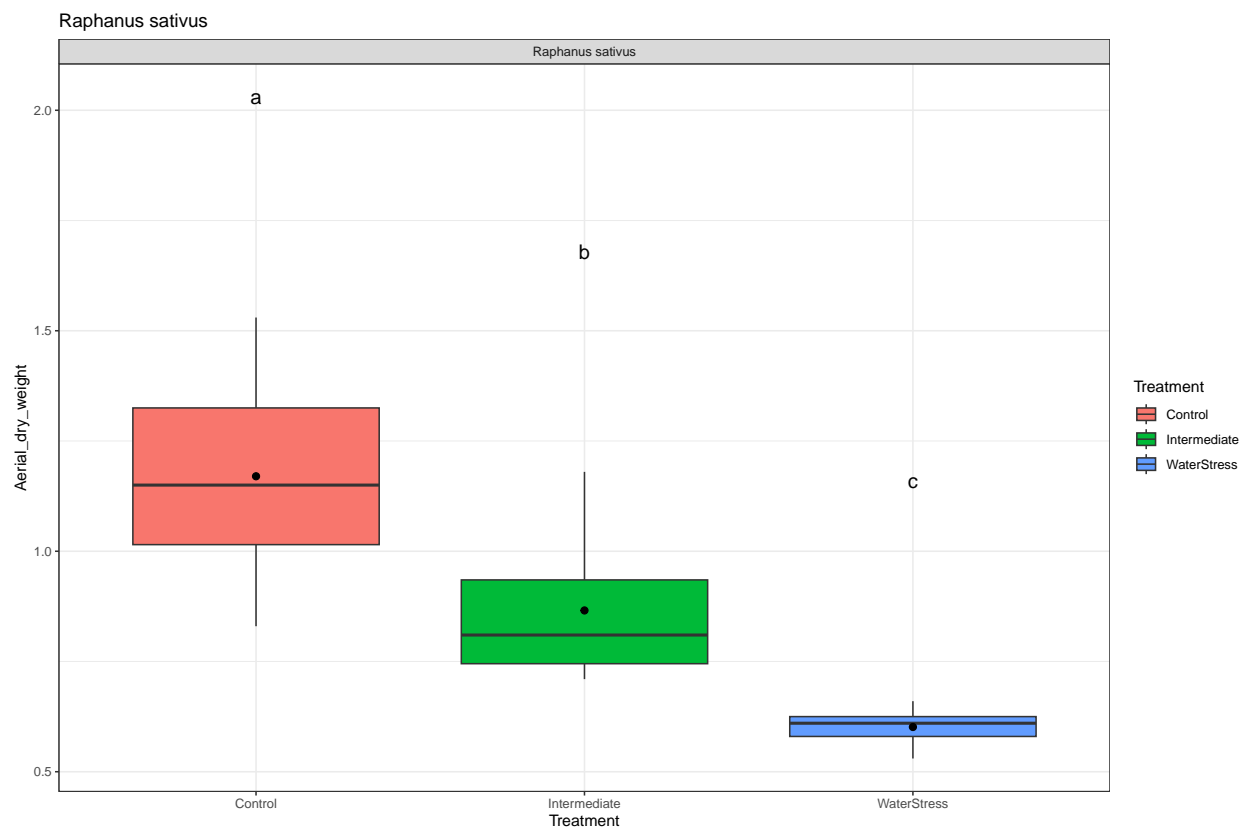
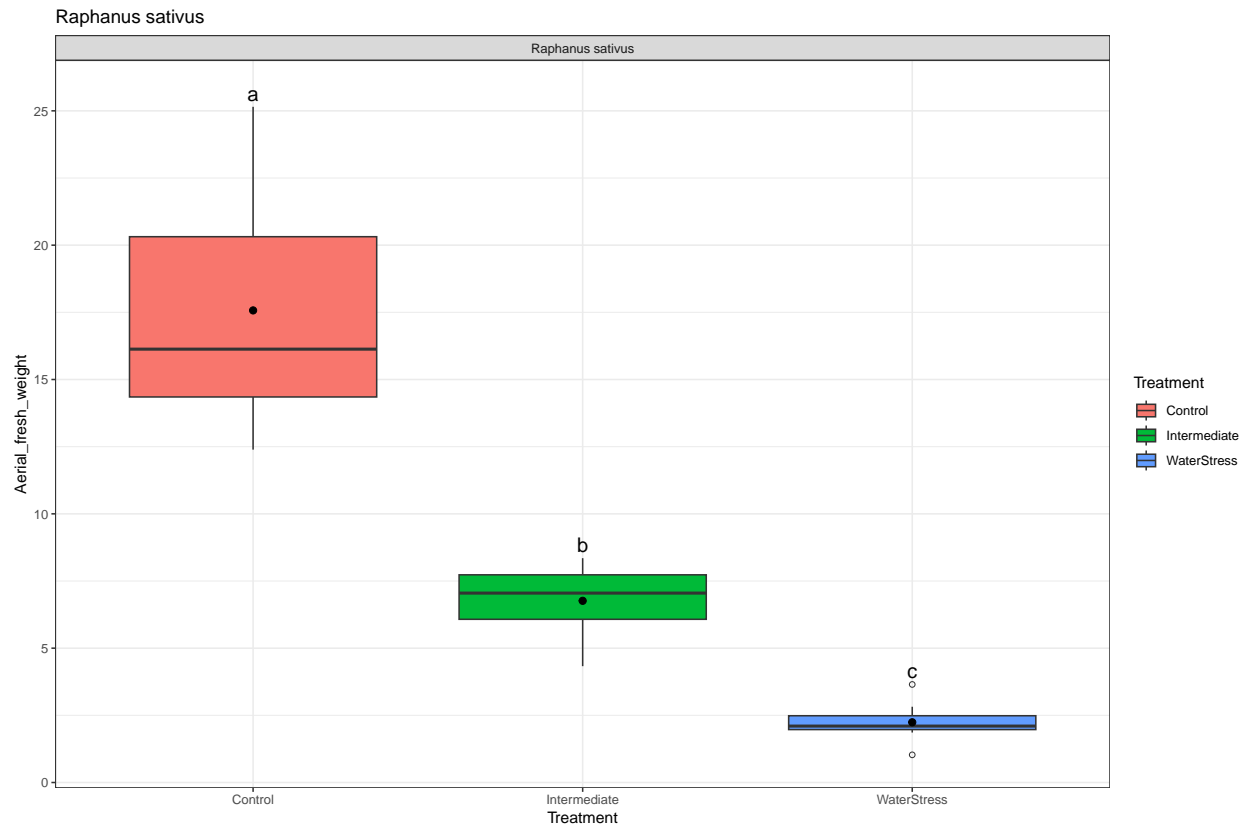


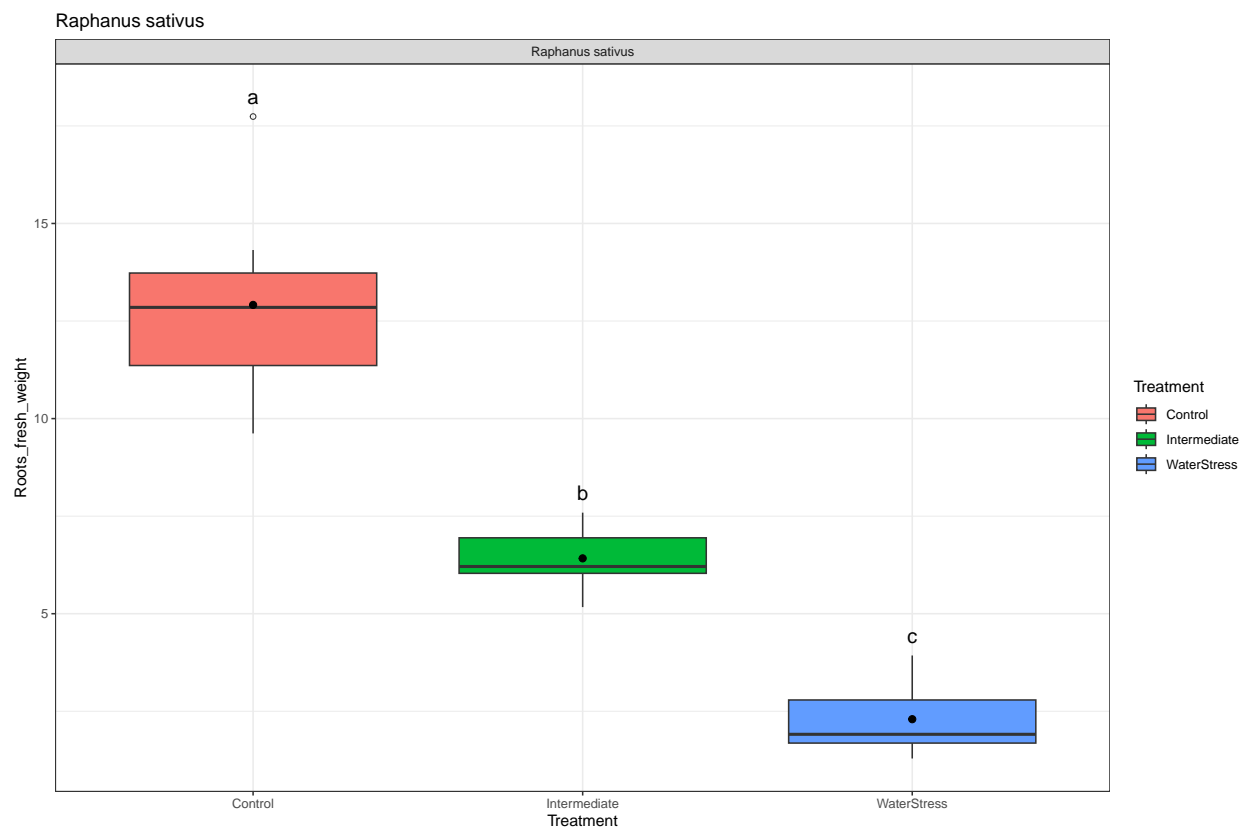
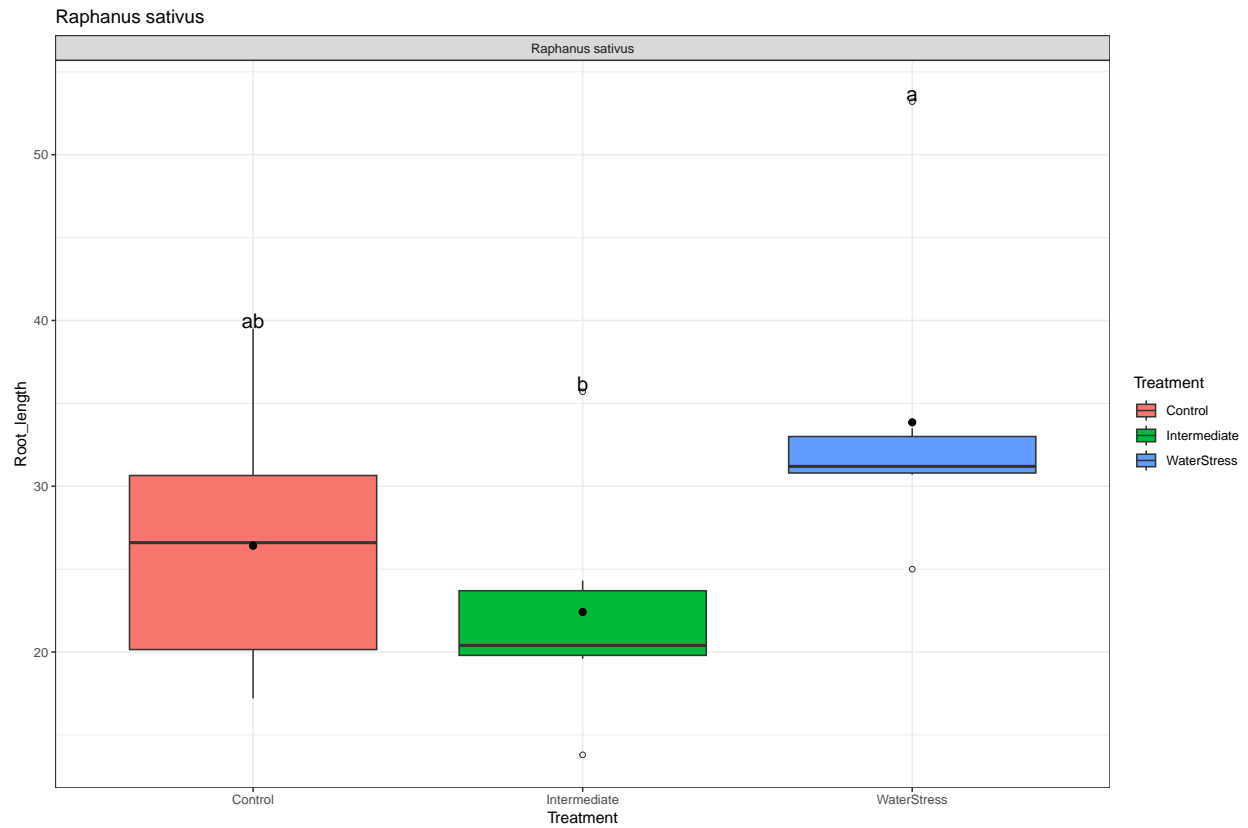


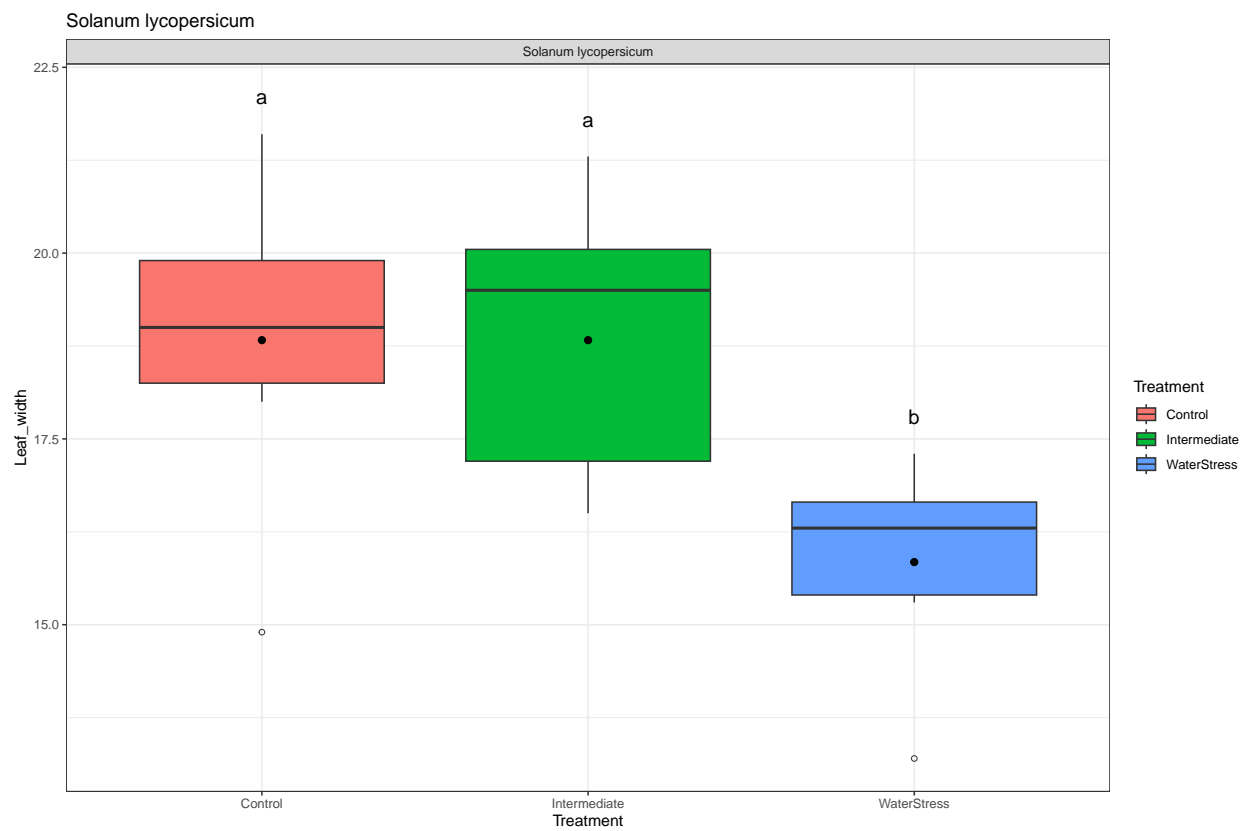
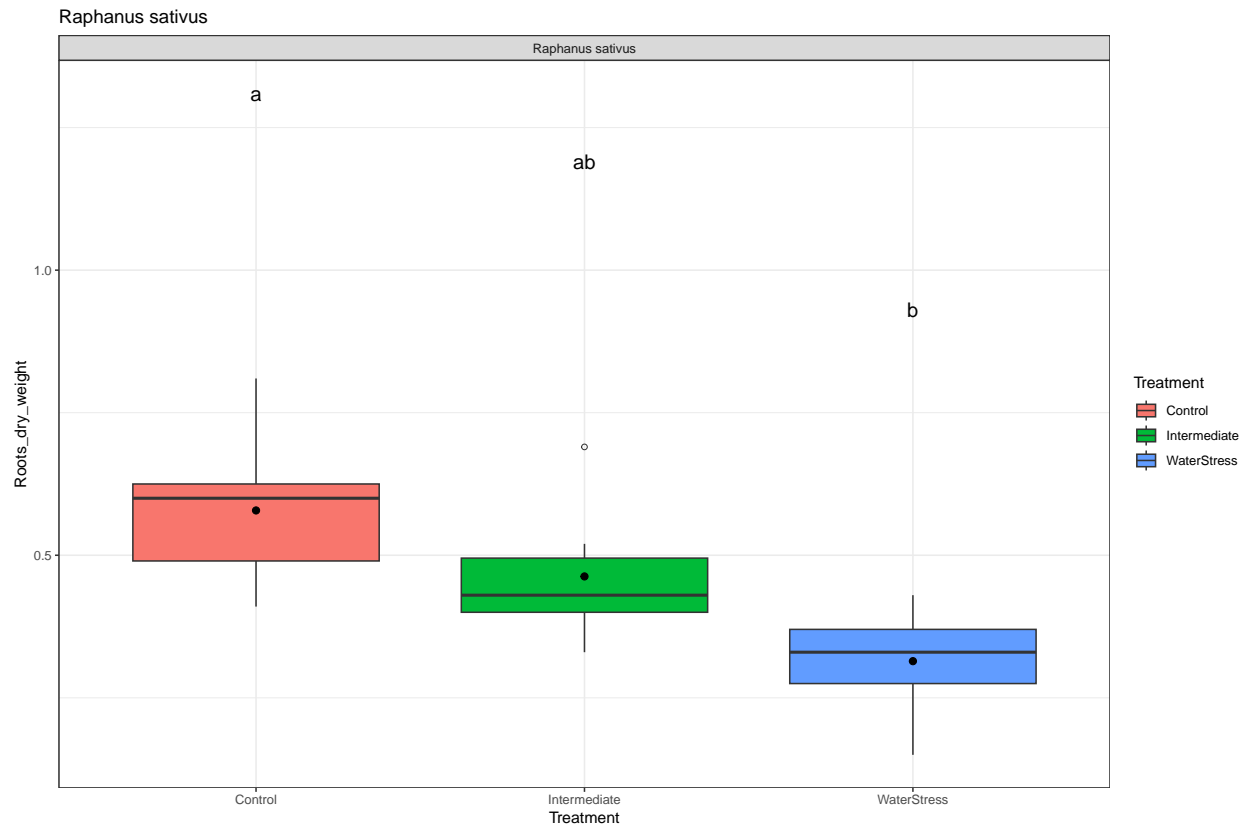


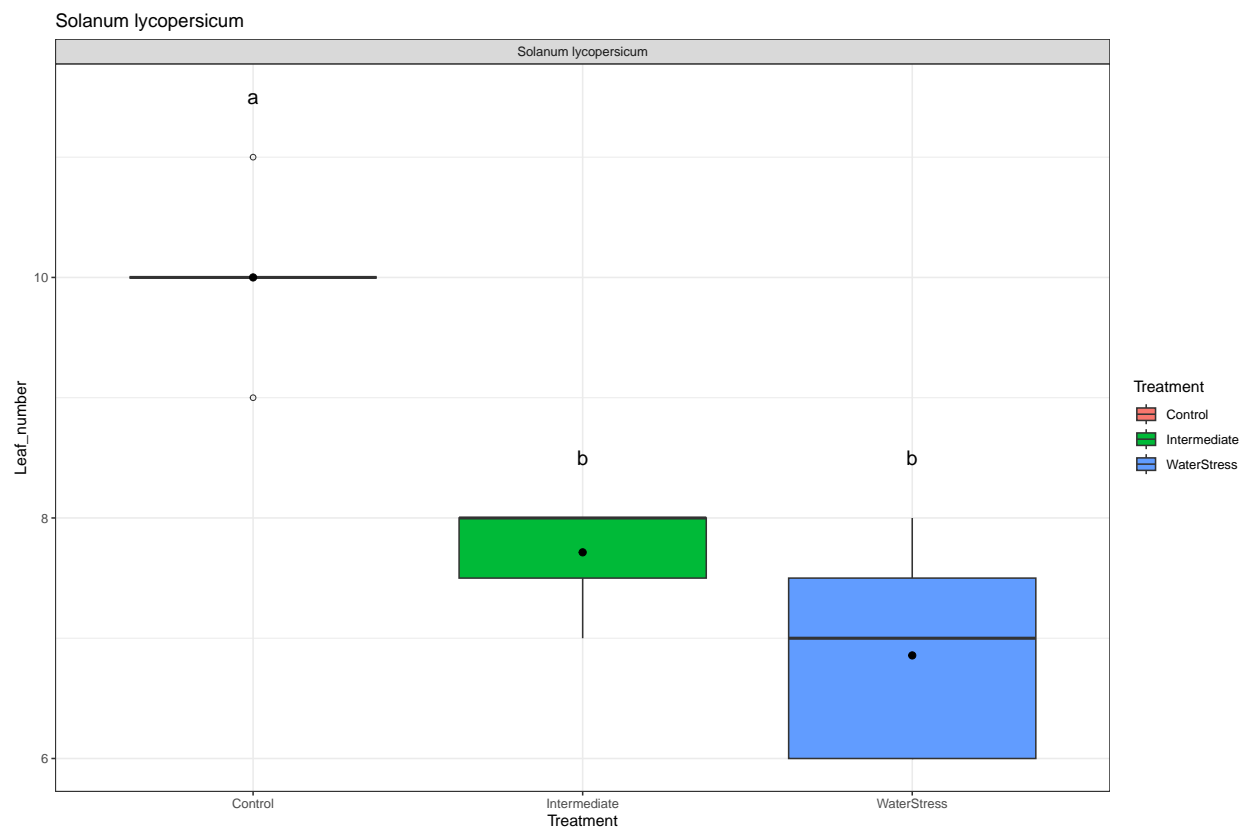
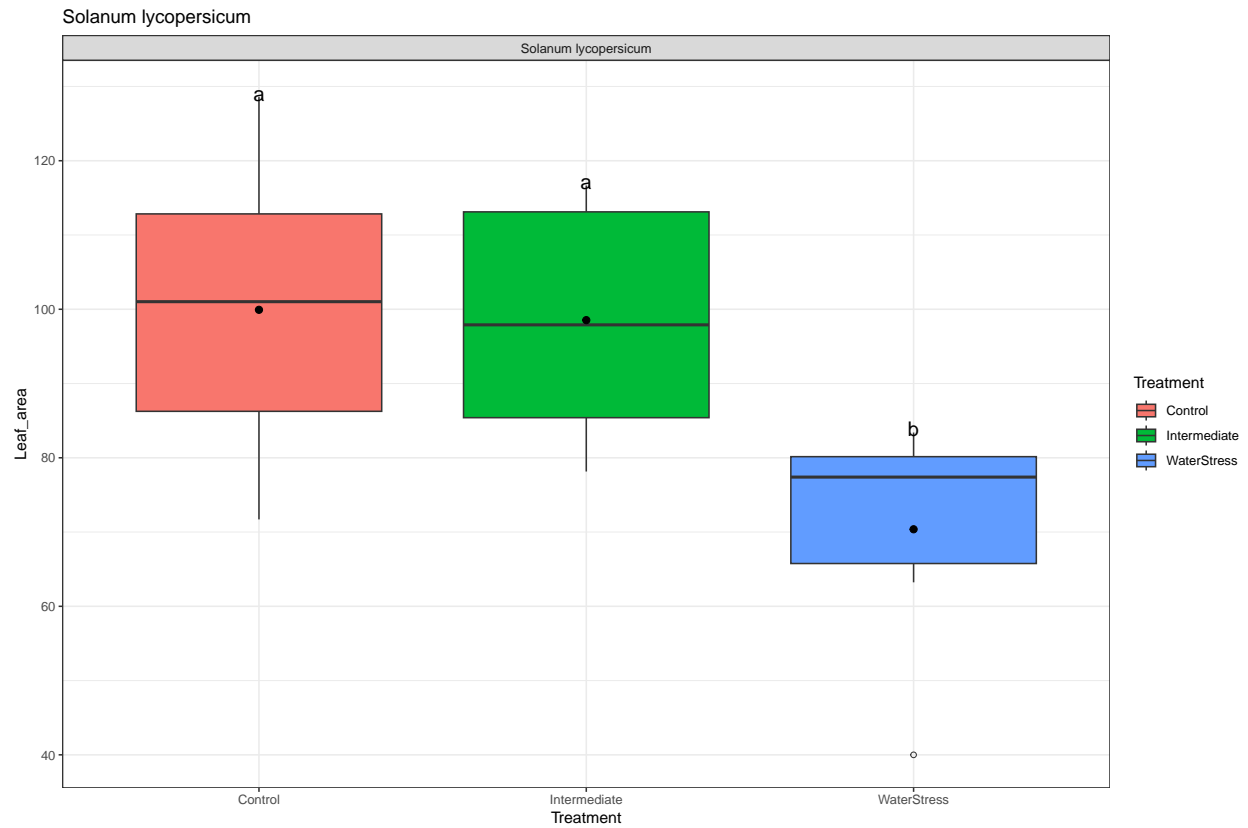


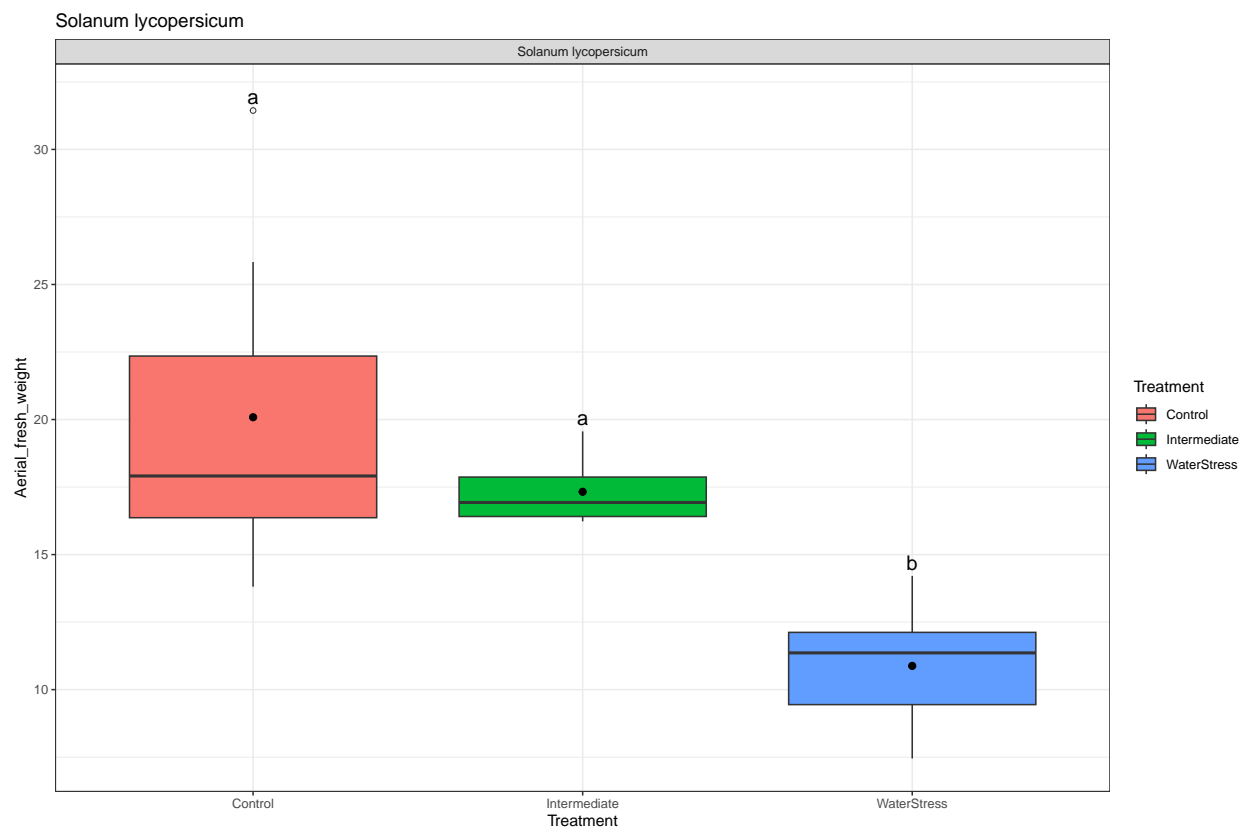
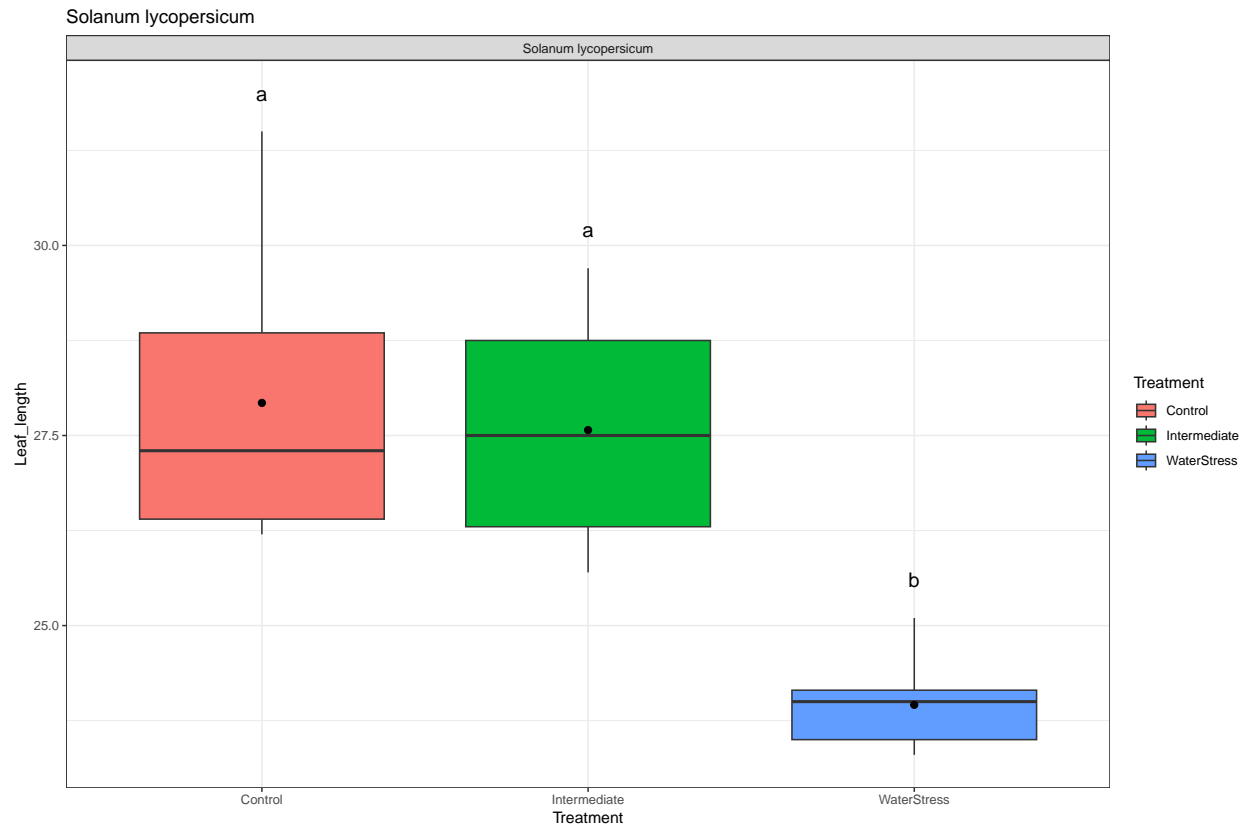


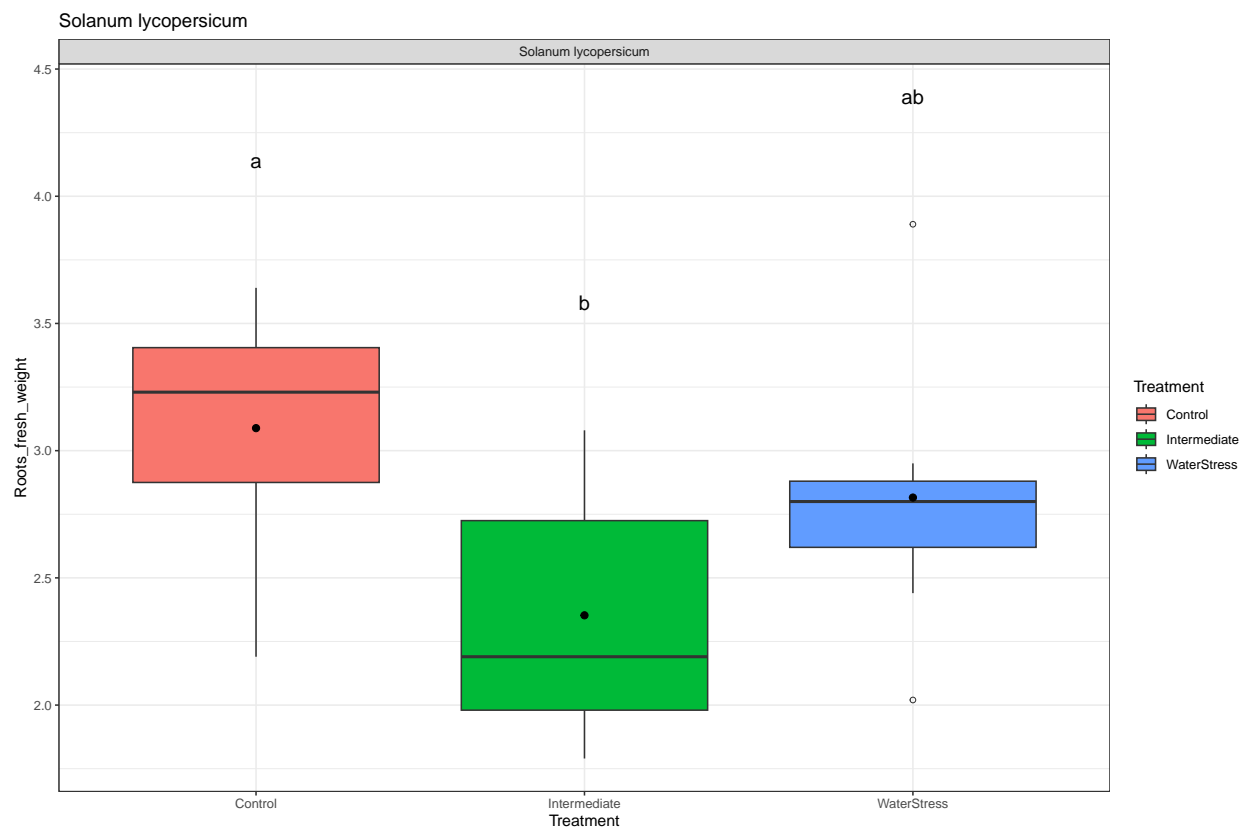
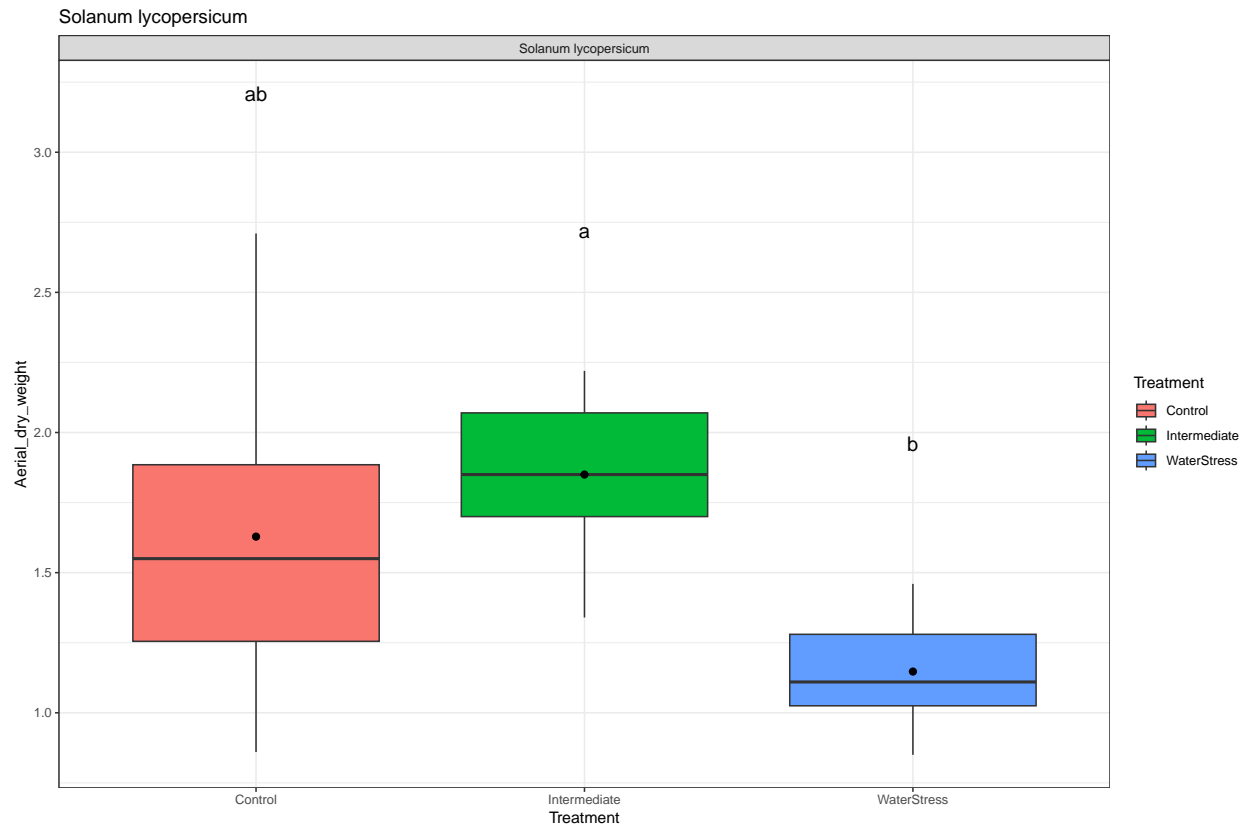


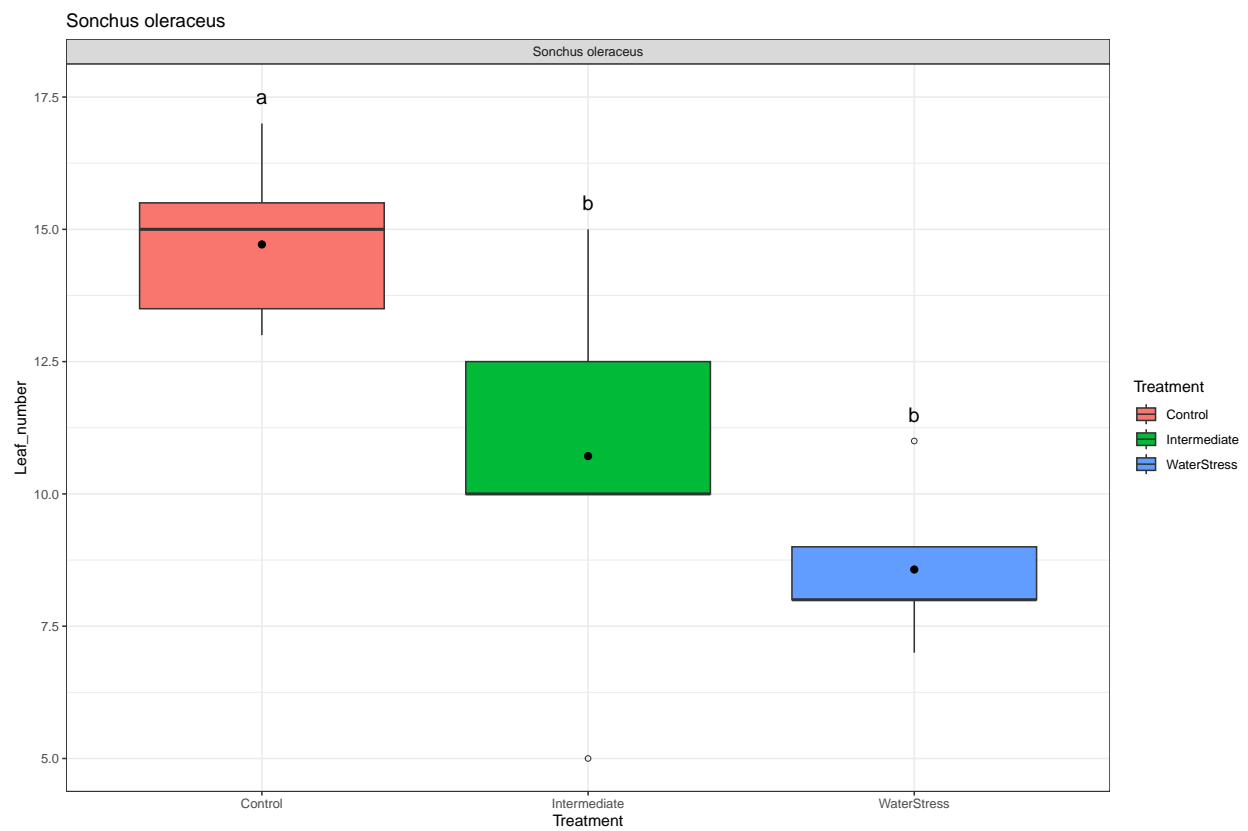
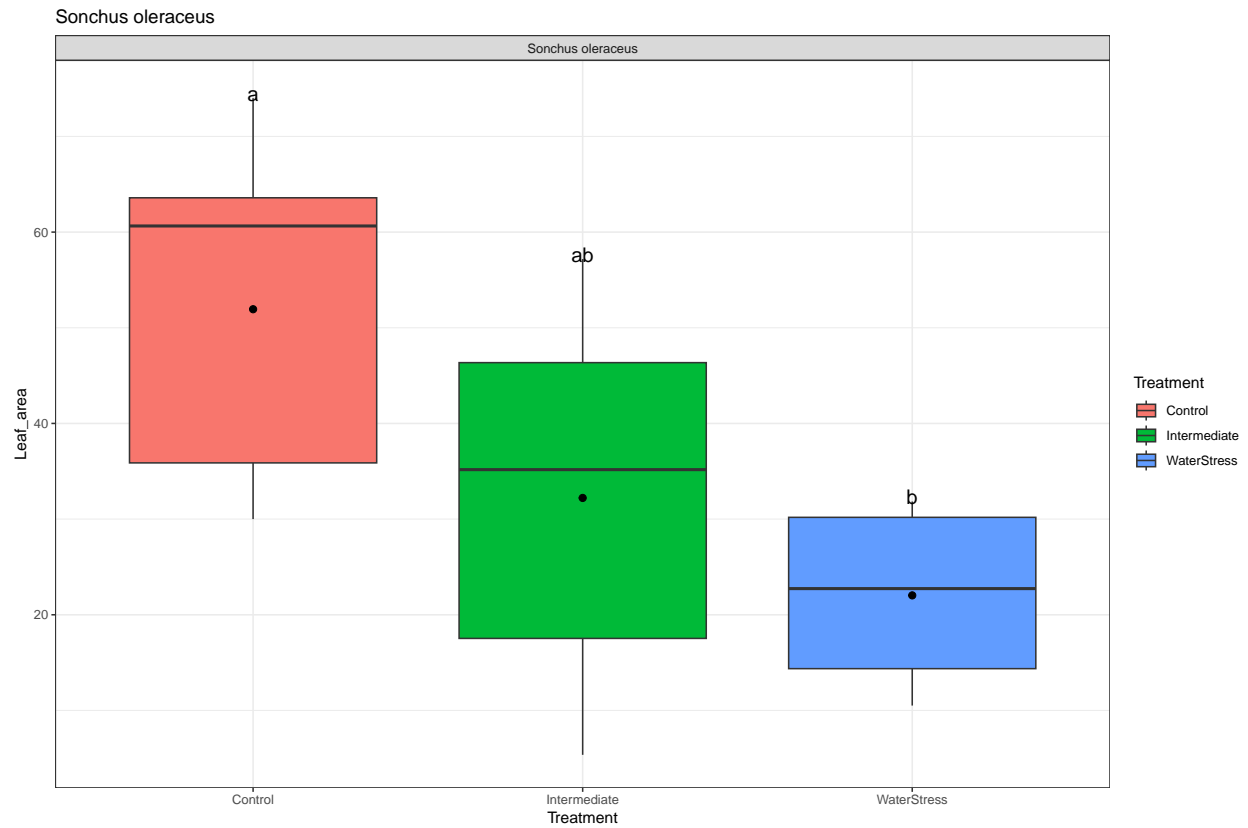


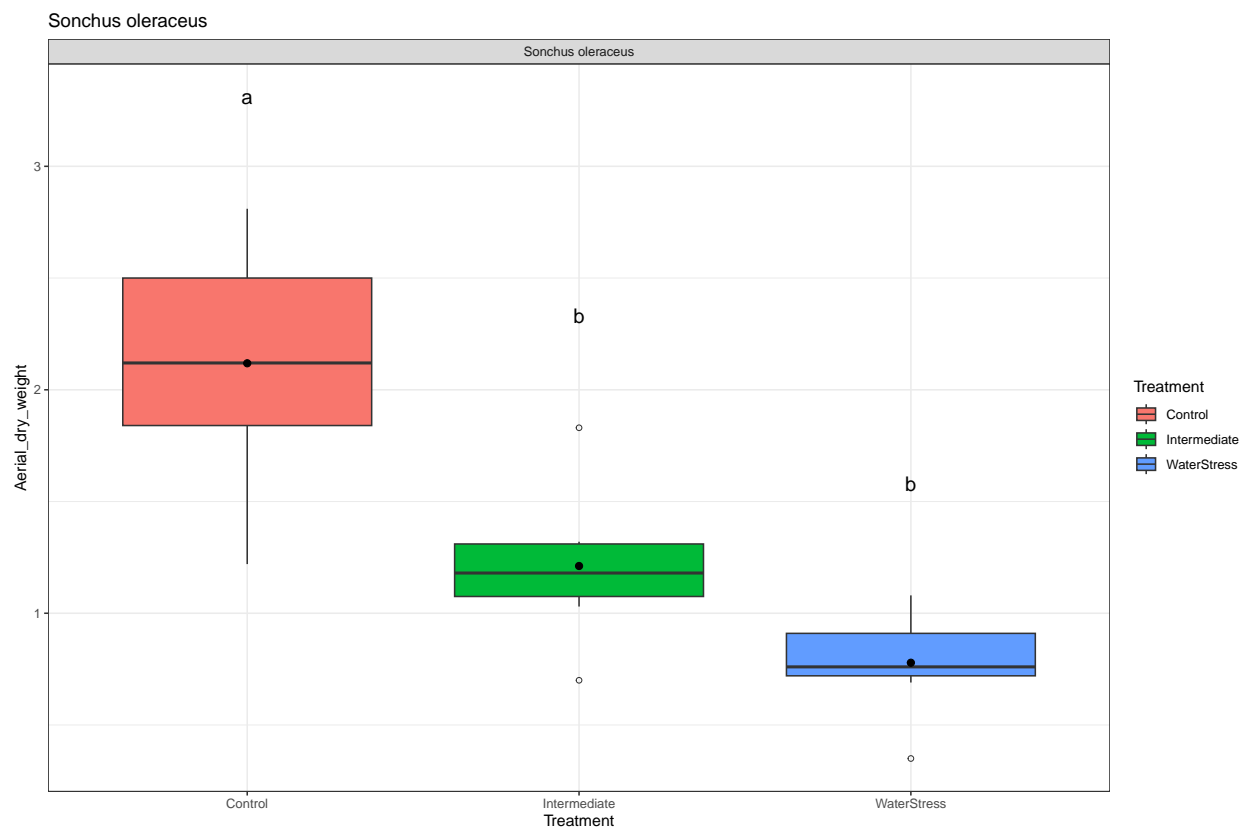
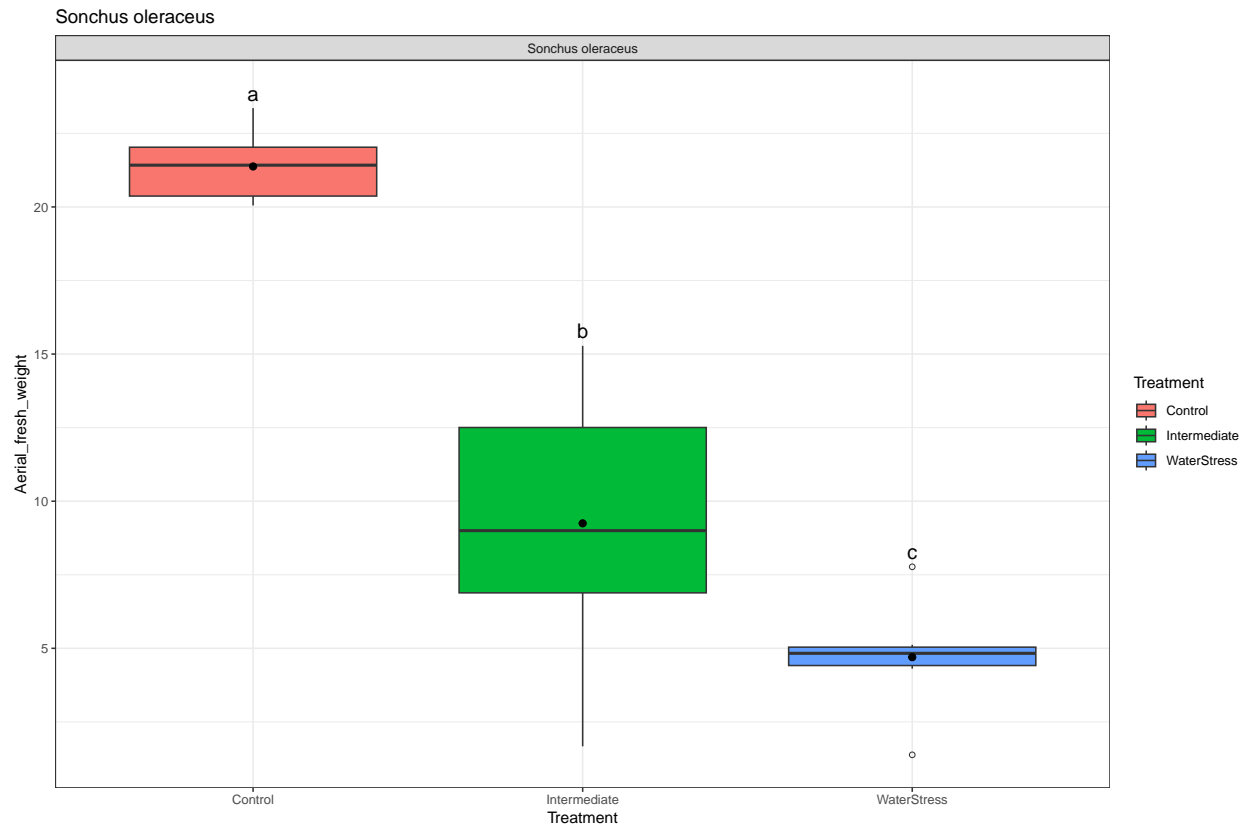


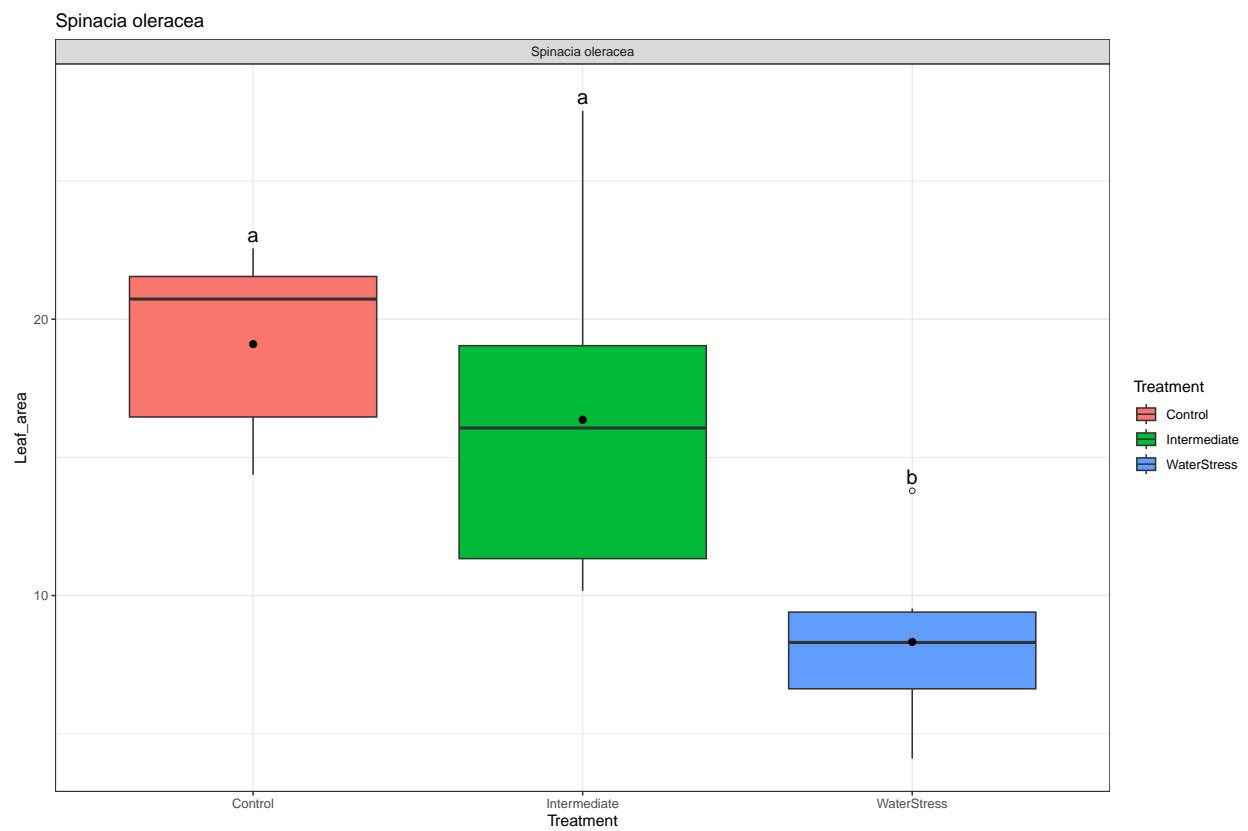
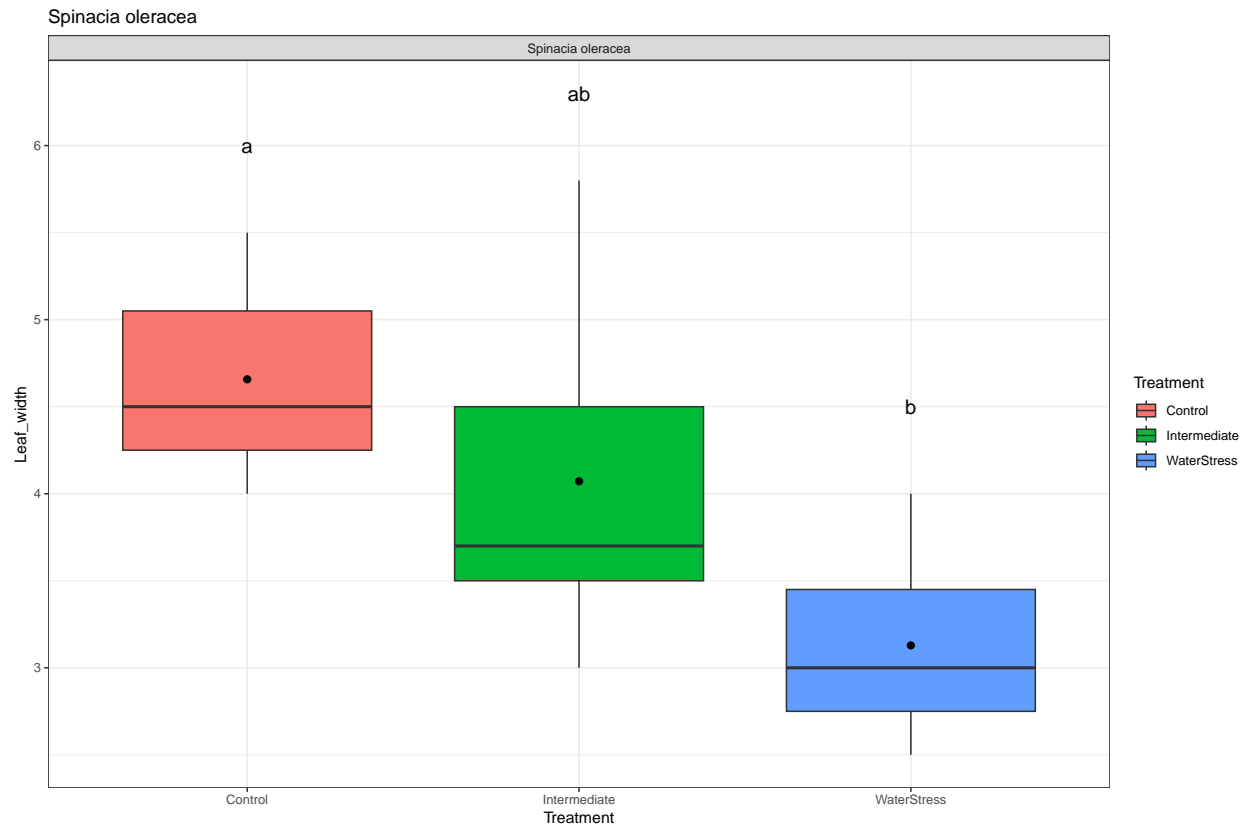


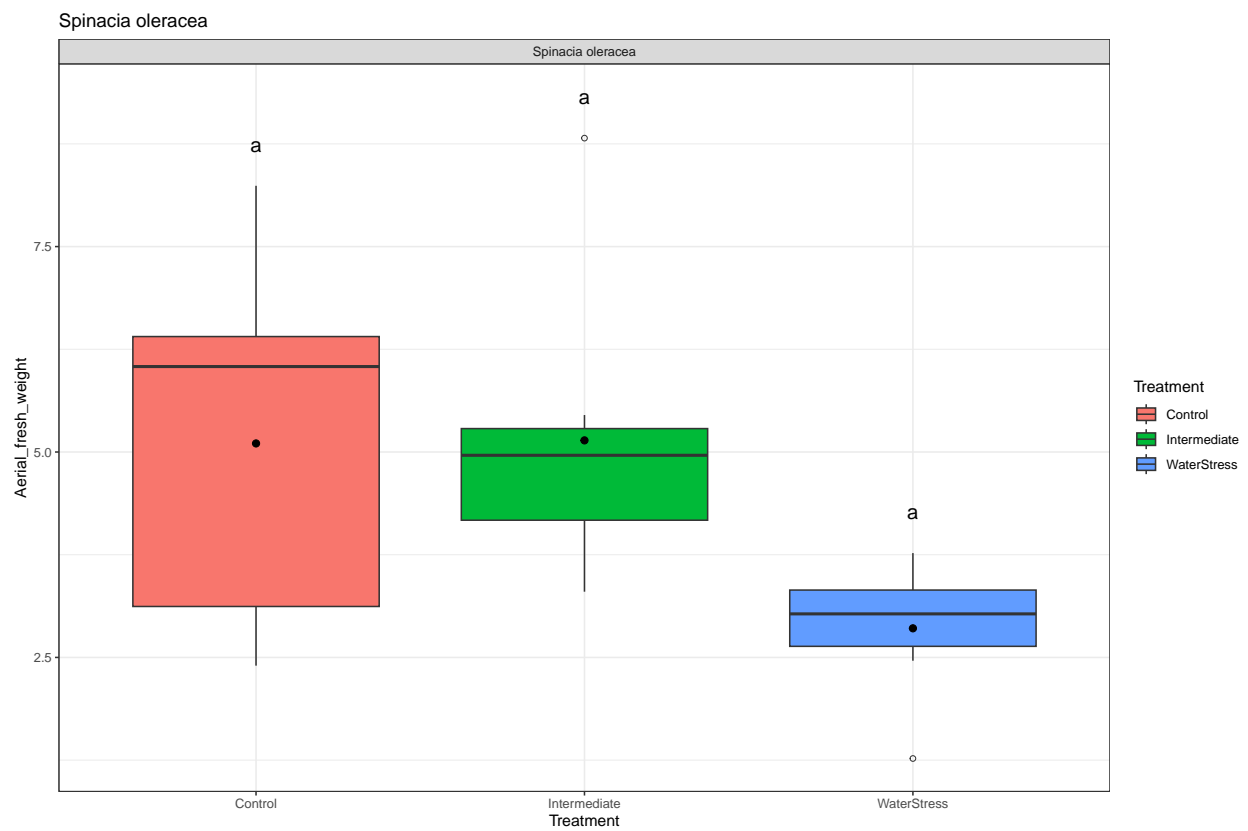
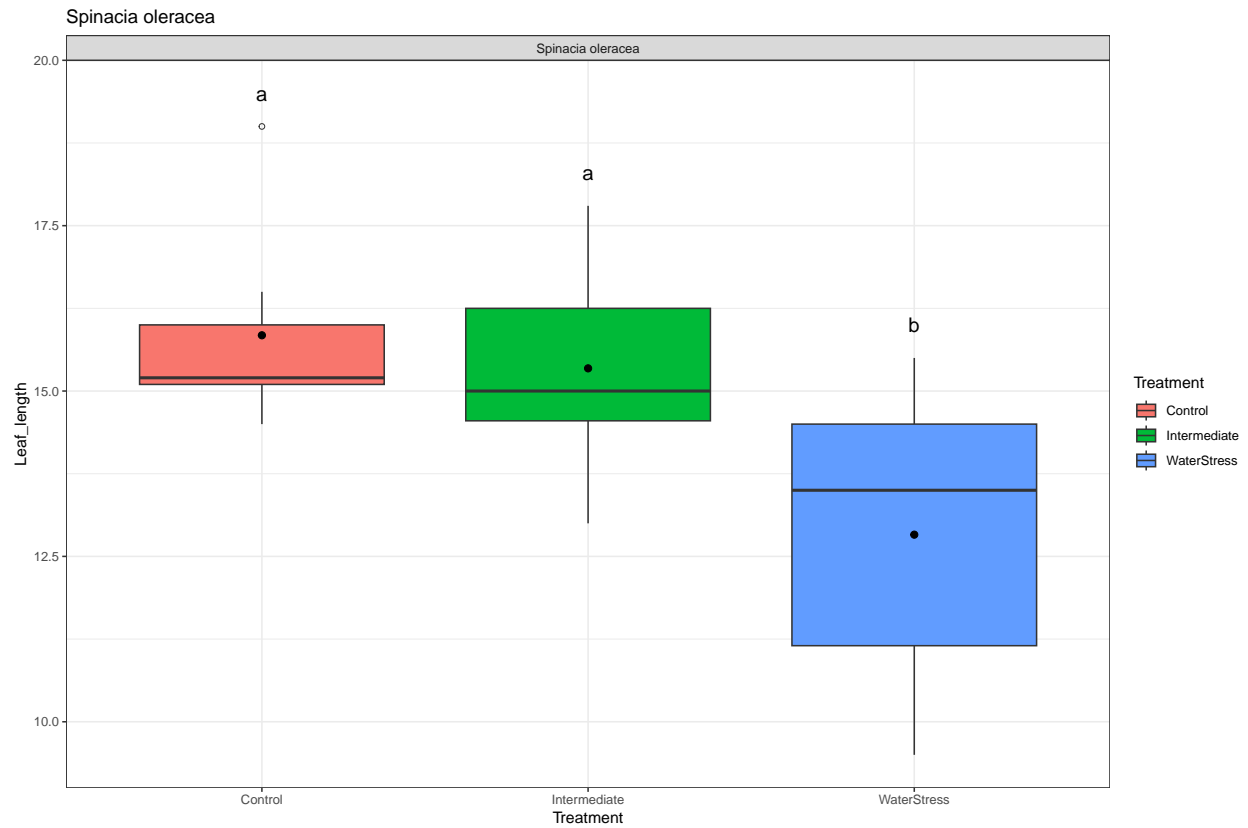


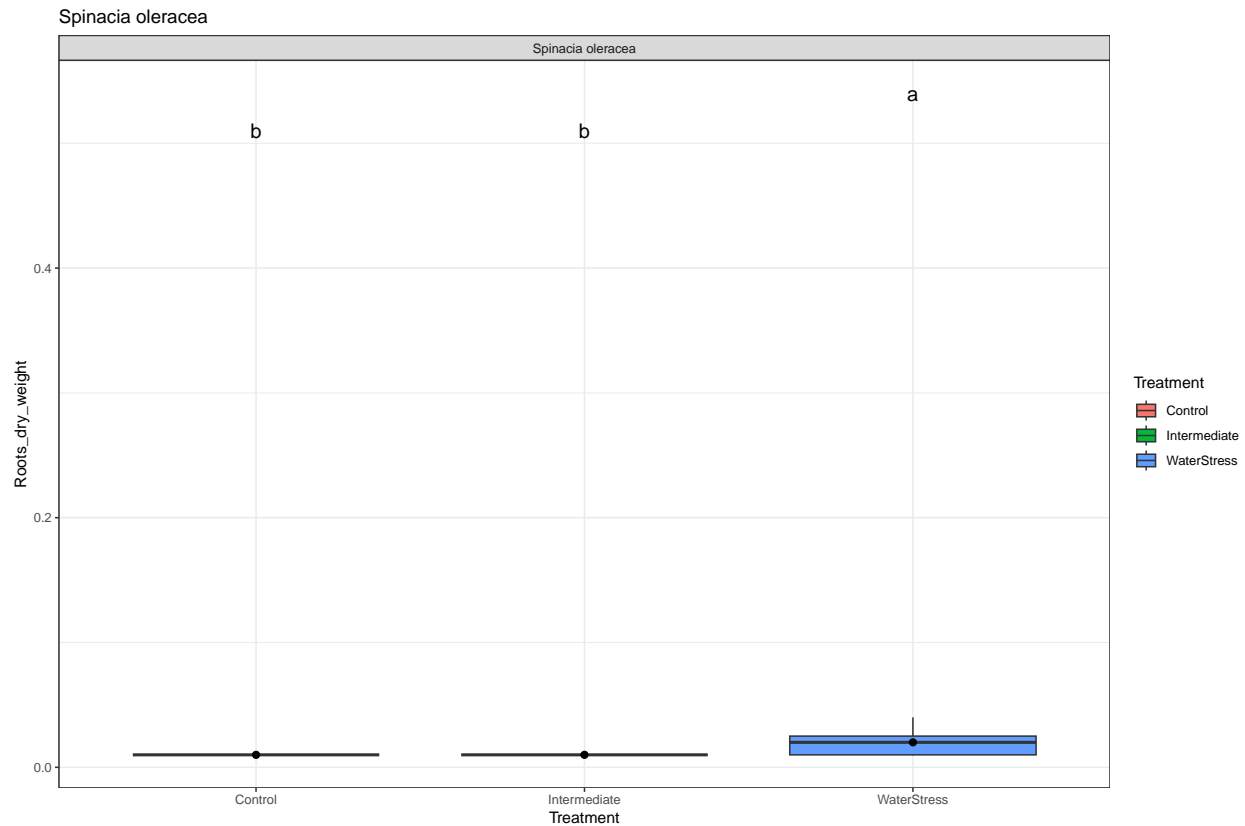












Only boxplot where pvalue<0.05 is shown. However, all boxplots are in
figs folder

The ANOVA Table

```
kable(finalframe, format = "latex", booktabs = TRUE) %>%
  kable_styling(latex_options = "scale_down")
```

```
#write_xlsx(finalframe, "final.xlsx")
```

Correlation Between Soil humidity and Electrical conductivity

The scatterplot in the image below depicts the correlation between electrical conductivity and soil humidity. Because the data points are clustered around the line in the figure, there is a higher correlation between soil humidity and electrical conductivity. The value of R also demonstrates how well the data match the model.

```
d2 <- data[, c("Electrical_conductivity", "Soil_humidity")]
dzclean <- na.omit(d2) ### some groups have missing data
dcpr <- dzclean[, c("Electrical_conductivity", "Soil_humidity")]

sus <- ggscatter(dzclean, x = "Electrical_conductivity", # correlation plot
```

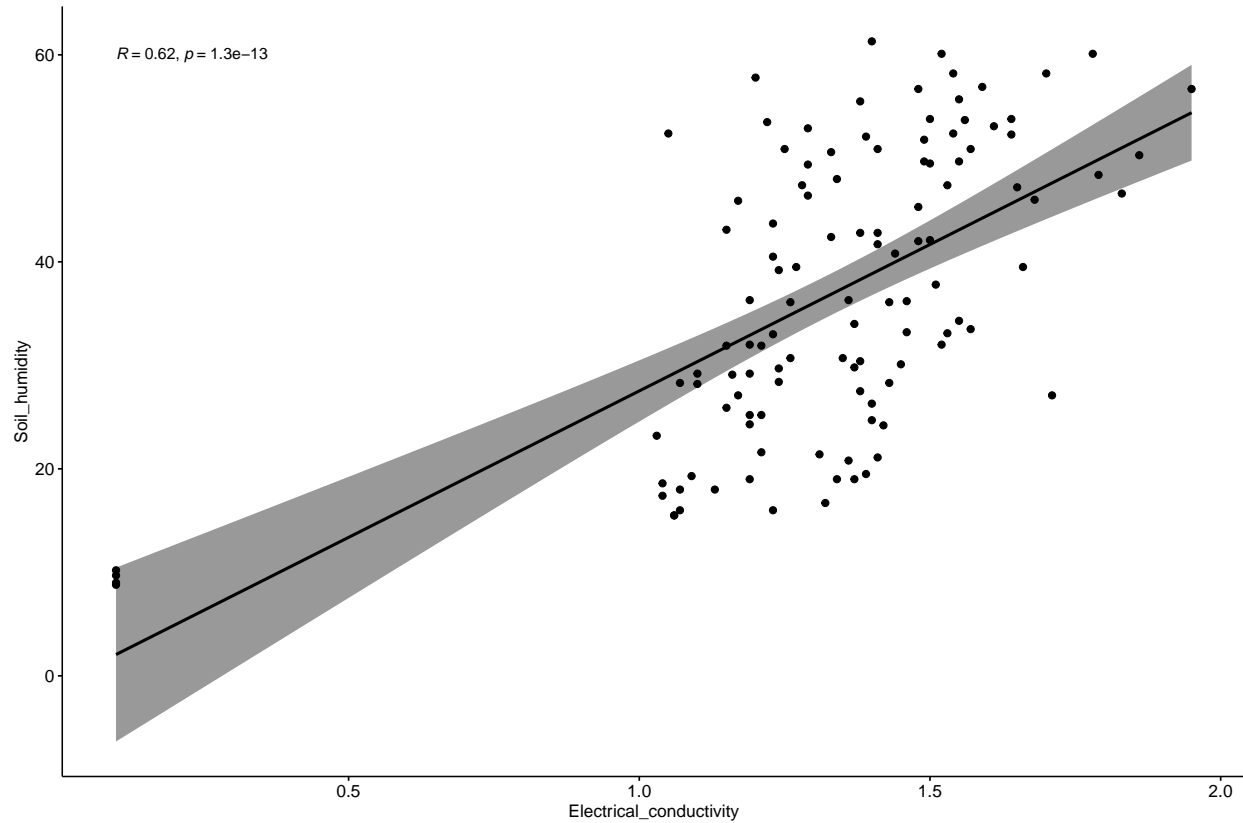

Species	variables	mean Control	mean Intermediate	mean WaterStress	pvalue
Amaranthus retroflexus	Soil_humidity	50.6 + 4.28 a	37.1 + 8.52 b	22.7 + 9.01 c	3.37e-11 ***
Amaranthus retroflexus	Leaf_width	1.71 + 0.441 a	1.34 + 0.293 b	1.34 + 0.403 b	2.24e-02 ***
Amaranthus retroflexus	Leaf_length	5.39 + 1.16 a	4.24 + 0.997 b	4.73 + 1.21 ab	3.47e-02 ***
Amaranthus retroflexus	Leaf_area	3.18 + 1.91 a	1.85 + 0.71 b	1.98 + 0.822 b	1.52e-02 ***
Amaranthus retroflexus	Aerial_fresh_weight	1.7 + 2.16 a	0.408 + 0.239 b	0.429 + 0.239 b	1.34e-02 ***
Beta vulgaris	Soil_humidity	14.9 + 5.21 a	7.74 + 4.68 b	2.61 + 2.63 b	1.88e-04 ***
Beta vulgaris	Leaf_number	9.71 + 2.29 a	8.57 + 1.9 a	4.33 + 0.516 b	1.29e-04 ***
Beta vulgaris	Leaf_area	47.2 + 15.3 a	49 + 7.75 a	21.4 + 6.6 b	3.85e-04 ***
Beta vulgaris	Aerial_fresh_weight	20 + 4.38 a	11.9 + 1.4 b	2.69 + 1.44 c	3.91e-09 ***
Beta vulgaris	Aerial_dry_weight	0.847 + 0.288 a	0.85 + 0.139 a	0.499 + 0.0901 b	3.67e-03 ***
Hordeum vulgare	Soil_humidity	55.1 + 3.83 a	7.01 + 2.26 b	3.77 + 1.08 b	1.91e-18 ***
Hordeum vulgare	Plant_height	50.4 + 4.48 ab	54.2 + 3.22 a	48 + 2.91 b	1.61e-02 ***
Hordeum vulgare	Leaf_number	14.7 + 2.98 a	11.3 + 2.29 b	6.71 + 0.756 c	1.10e-05 ***
Hordeum vulgare	Aerial_fresh_weight	8.51 + 1.36 a	7.45 + 2.38 a	3.58 + 0.464 b	4.62e-05 ***
Hordeum vulgare	Aerial_dry_weight	0.67 + 0.0794 ab	0.823 + 0.178 a	0.579 + 0.062 b	4.00e-03 ***
Hordeum vulgare	Root_length	33.1 + 9.44 a	19.6 + 4 b	21.6 + 3.21 b	1.44e-03 ***
Hordeum vulgare	Roots_fresh_weight	1.13 + 0.652 b	1.81 + 0.499 a	1.01 + 0.162 b	1.23e-02 ***
Hordeum vulgare	Roots_dry_weight	0.0957 + 0.0428 b	0.249 + 0.0634 a	0.191 + 0.0324 a	4.79e-05 ***
Lolium perenne	Soil_humidity	56.1 + 3.76 a	25 + 3.81 b	8.34 + 2.9 c	6.09e-15 ***
Portulaca oleracea	Soil_humidity	37.4 + 4.88 a	32 + 3.48 b	15.6 + 2.34 c	5.46e-09 ***
Portulaca oleracea	Leaf_width	1.23 + 0.125 a	0.829 + 0.315 b	1.06 + 0.305 ab	3.47e-02 ***
Portulaca oleracea	Leaf_length	2.14 + 0.331 a	1.5 + 0.447 b	1.69 + 0.467 ab	2.86e-02 ***
Portulaca oleracea	Leaf_area	1.88 + 0.356 a	0.981 + 0.552 b	1.5 + 0.625 ab	1.66e-02 ***
Portulaca oleracea	Aerial_fresh_weight	2.05 + 0.551 a	1 + 0.565 b	1.47 + 0.753 ab	2.09e-02 ***
Portulaca oleracea	Aerial_dry_weight	0.0957 + 0.0315 a	0.05 + 0.0289 b	0.0729 + 0.0269 ab	2.98e-02 ***
Portulaca oleracea	Roots_fresh_weight	0.103 + 0.0293 a	0.0143 + 0.0151 b	0.0329 + 0.0368 b	3.96e-05 ***
Raphanus sativus	Soil_humidity	22.5 + 4.07 a	4.59 + 1.07 b	2.83 + 0.629 b	1.18e-11 ***
Raphanus sativus	Plant_height	23.4 + 2.05 a	19.7 + 1.35 b	16.3 + 1.71 c	1.75e-06 ***
Raphanus sativus	Leaf_width	8.17 + 0.568 a	6.74 + 0.832 b	4.54 + 0.836 c	2.01e-07 ***
Raphanus sativus	Leaf_number	8.14 + 0.9 a	5.43 + 0.535 b	3.71 + 0.488 c	1.28e-09 ***
Raphanus sativus	Leaf_length	11.3 + 1.13 a	9.67 + 1.14 b	7.8 + 0.924 c	4.64e-05 ***
Raphanus sativus	Leaf_area	69.6 + 15.3 a	51.4 + 14.1 b	26.2 + 7.36 c	2.28e-05 ***
Raphanus sativus	Aerial_fresh_weight	17.6 + 4.8 a	6.76 + 1.42 b	2.24 + 0.815 c	4.01e-08 ***
Raphanus sativus	Aerial_dry_weight	1.17 + 0.251 a	0.866 + 0.166 b	0.601 + 0.0422 c	4.51e-05 ***
Raphanus sativus	Root_length	26.4 + 7.87 ab	22.4 + 6.74 b	33.9 + 8.95 a	4.27e-02 ***
Raphanus sativus	Roots_fresh_weight	12.9 + 2.63 a	6.42 + 0.824 b	2.3 + 0.996 c	3.20e-09 ***
Raphanus sativus	Roots_dry_weight	0.579 + 0.131 a	0.463 + 0.117 ab	0.314 + 0.0941 b	1.67e-03 ***
Solanum lycopersicum	Soil_humidity	46.6 + 4.85 a	7.07 + 1.7 b	4.99 + 1.44 b	1.08e-15 ***
Solanum lycopersicum	Plant_height	66.1 + 9.87 a	63.1 + 2.85 a	46.4 + 3.69 b	2.78e-05 ***
Solanum lycopersicum	Leaf_width	18.8 + 2.11 a	18.8 + 1.85 a	15.8 + 1.37 b	8.01e-03 ***
Solanum lycopersicum	Leaf_number	10 + 0.577 a	7.71 + 0.488 b	6.86 + 0.9 b	2.32e-07 ***
Solanum lycopersicum	Leaf_length	27.9 + 2.01 a	27.6 + 1.54 a	24 + 0.613 b	1.53e-04 ***
Solanum lycopersicum	Leaf_area	99.9 + 19.9 a	98.5 + 16.5 a	70.4 + 15.3 b	7.66e-03 ***
Solanum lycopersicum	Aerial_fresh_weight	20.1 + 6.27 a	17.3 + 1.28 a	10.9 + 2.37 b	1.17e-03 ***
Solanum lycopersicum	Aerial_dry_weight	1.63 + 0.608 ab	1.85 + 0.3 a	1.15 + 0.208 b	1.47e-02 ***
Solanum lycopersicum	Roots_fresh_weight	3.09 + 0.493 a	2.35 + 0.488 b	2.82 + 0.57 ab	4.82e-02 ***
Sonchus oleraceus	Soil_humidity	5.67 + 1.35 a	3.79 + 2.01 ab	3.5 + 1.13 b	3.43e-02 ***
Sonchus oleraceus	Leaf_number	14.7 + 1.5 a	10.7 + 3.15 b	8.57 + 1.27 b	1.56e-04 ***
Sonchus oleraceus	Leaf_area	51.9 + 17.5 a	32.2 + 19.3 ab	22 + 9.11 b	8.06e-03 ***
Sonchus oleraceus	Aerial_fresh_weight	21.4 + 1.24 a	9.25 + 4.76 b	4.7 + 1.87 c	1.78e-08 ***
Sonchus oleraceus	Aerial_dry_weight	2.12 + 0.565 a	1.21 + 0.343 b	0.779 + 0.246 b	2.91e-05 ***
Spinacia oleracea	Soil_humidity	49.9 + 4.07 a	34.3 + 4.27 b	15.5 + 8.4 c	1.24e-08 ***
Spinacia oleracea	Plant_height	15.9 + 1.53 a	15.3 + 1.56 a	12.8 + 2.25 b	1.29e-02 ***
Spinacia oleracea	Leaf_width	4.66 + 0.624 a	4.07 + 0.983 ab	3.13 + 0.605 b	4.88e-03 ***
Spinacia oleracea	Leaf_length	15.8 + 1.52 a	15.3 + 1.56 a	12.8 + 2.25 b	1.30e-02 ***
Spinacia oleracea	Leaf_area	19.1 + 3.25 a	16.4 + 6.34 a	8.32 + 3.06 b	7.72e-04 ***
Spinacia oleracea	Aerial_fresh_weight	5.1 + 2.22 a	5.14 + 1.83 a	2.85 + 0.818 a	3.57e-02 ***
Spinacia oleracea	Roots_dry_weight	0.01 + 0 b	0.01 + 0 b	0.02 + 0.0115 a	1.60e-02 ***

```

y = "Soil_humidity",
add = "reg.line", conf.int = TRUE,
cor.coef = TRUE, cor.method = "pearson",
xlab = "Electrical_conductivity",
ylab = "Soil_humidity")

```

sus



Conclusion

For all the same conditions, not all species react to water stress in the same way. Consequently, various parameters are required for the evaluation to explain their response. From the cluster analysis, the parameters measured from the root and the leaf like root length, leaf length, and root weight are the most important variable defining clusters for most of the species, and may therefore can be used in future studies of plant response to water stress. In most species, leaf area, aerial fresh weight, and aerial dry weight play significant roles in predicting how they will react to the treatment. The treatments are more significant in crop species compared to weed species, indicating that cultivated plants are more susceptible to the water stress than weeds.

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