

Final_plant_analyses

Caroline Edwards

5/7/2021

```
rm(list=ls()) #clear your working directory
setwd("~/quant_bio/GitHub/QB2021_Edwards/2.Worksheets/13.DiversityProject/")

package.list<-c('vegan','ade4','viridis','gplots','BiodiversityR','indicspecies','mobsim')
for (package in package.list){
  if (!require(package, character.only = TRUE, quietly =TRUE)){
    install.packages(package)
    library(package, character.only = TRUE)
  }
}

## This is vegan 2.5-6

## Warning: package 'ade4' was built under R version 3.6.2

## Warning: package 'gplots' was built under R version 3.6.2

##
## Attaching package: 'gplots'

## The following object is masked from 'package:stats':
##
##     lowess

## Warning: package 'BiodiversityR' was built under R version 3.6.2

## Registered S3 methods overwritten by 'lme4':
##   method                                  from
##   cooks.distance.influence.merMod         car
##   influence.merMod                        car
##   dfbeta.influence.merMod                 car
##   dfbetas.influence.merMod                car

## BiodiversityR 2.12-3: Use command BiodiversityRGUI() to launch the Graphical User Interface;
## to see changes use BiodiversityRGUI(changeLog=TRUE, backward.compatibility.messages=TRUE)

## Warning: package 'mobsim' was built under R version 3.6.2
```

```

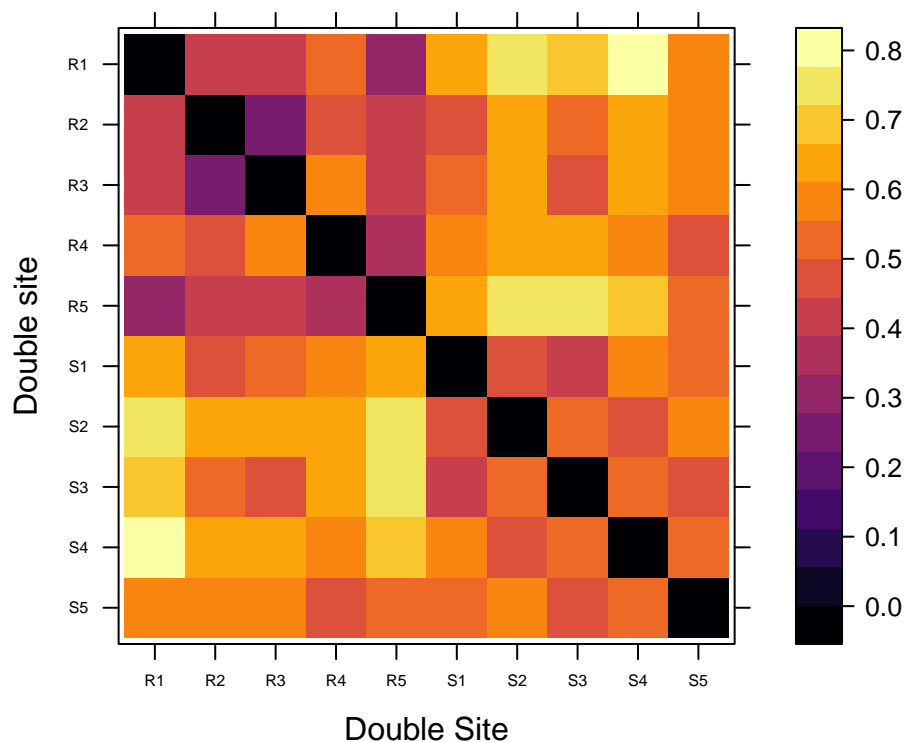
#Clustering analyses
#Make site by species matrix for plant species
veg_sxs<-read.csv("veg_data.csv")
rownames(veg_sxs)<-(veg_sxs[,1])
veg_sxs<-veg_sxs[1:47,-1]
veg_sxs<-as.data.frame(t(veg_sxs))

#Make a heatmap between all sites using species relative abundances using bray-curtis distance
veg.db<-vegdist(veg_sxs, method = "bray")

order<-rev(attr(veg.db,"Labels"))
levelplot(as.matrix(veg.db)[, order], aspect = "iso", col.regions = inferno, xlab = "Double Site",
          ylab = "Double site", scales = list(cex = 0.5), main = "Bray-Curtis Distance")

```

Bray-Curtis Distance

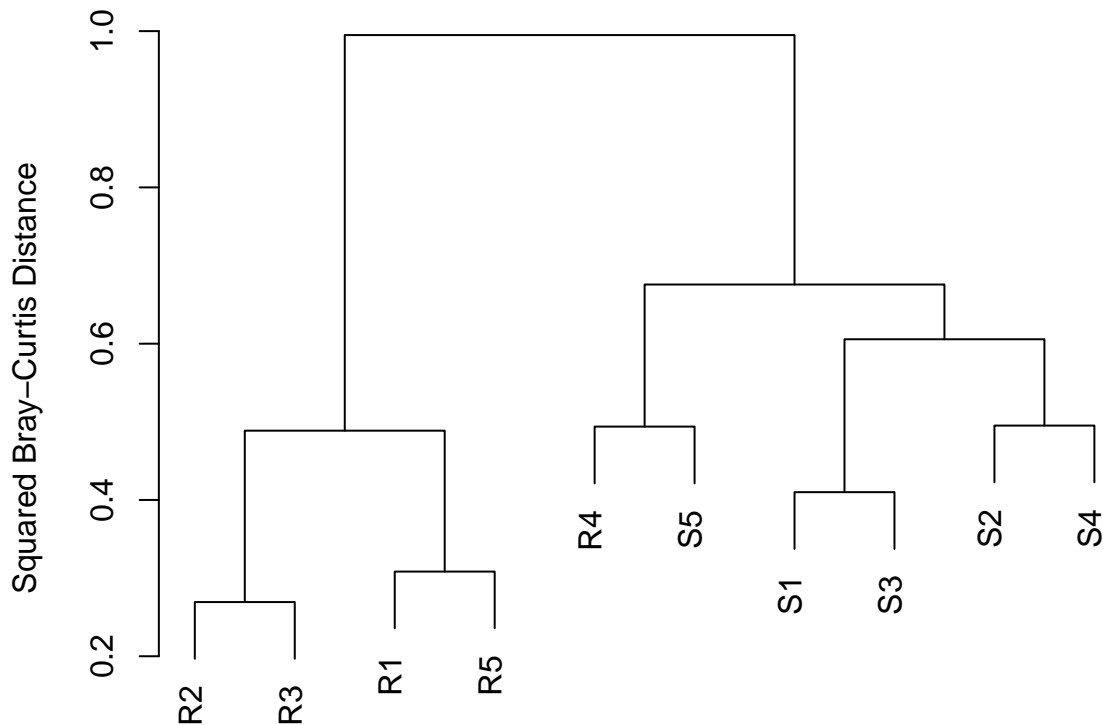


```

#Make a cluster analysis for all sites using species relative abundances using bray-curtis distance
veg.ward<-hclust(veg.db, method="ward.D2")
par(mar = c(1,5,2,2)+0.1)
plot(veg.ward, main = "Plants on ridge-to-snowbed gradient: Ward's clustering", ylab = "Squared Bray-Cu

```

Plants on ridge-to-snowbed gradient: Ward's clustering



```
#Mantel tests for correlation between plant community and fungal community, environmental dissimilarity
#Read in data and wrangle
veg<-read.csv("veg_data.csv")
row.names(veg)<-veg[,1]
veg<-veg[, -1]

otu_sxs<-read.csv("OTU_table.csv")
otu_sxs<-t(otu_sxs)
env_var<-read.delim("variables.txt")
env_var$site<- c(rep("S1", 5), rep("S2", 5), rep("S3", 5), rep("S4", 5), rep("S5", 5), rep("R1", 5), rep("R2", 5), rep("R3", 5), rep("R4", 5), rep("R5", 5))

otu_names<-read.csv("modified_OTU_table.csv")
OTU_ID<-otu_names$X.OTU.ID
colnames(otu_sxs)<-OTU_ID

env_var<-read.delim("variables.txt")
env_var$site<- c(rep("S1", 5), rep("S2", 5), rep("S3", 5), rep("S4", 5), rep("S5", 5), rep("R1", 5), rep("R2", 5), rep("R3", 5), rep("R4", 5), rep("R5", 5))

#Take mean of all samples of environmental data at each site, so there is only one value per site
env_P<-tapply(env_var$P, env_var$site, mean)
env_N<-tapply(env_var$N, env_var$site, mean)
env_C<-tapply(env_var$C, env_var$site, mean)
env_V<-c(rep("R", 5), rep("S", 5))
env<-cbind(env_P, env_N, env_C, env_V)
colnames(env)<-c("P", "N", "C", "Site_type")
env<-as.data.frame(env)
```

```
#Rarefaction of otu samples
site_species.r <- rrarefy(otu_sxs, 1000)
```

```
## Warning in rrarefy(otu_sxs, 1000): some row sums < 'sample' and are not rarefied
```

```
richness <- rowSums((site_species.r > 0) * 1)
minimum.r <- min(rowSums(site_species.r))
rarefy <- rarefy(x = site_species.r, sample = minimum.r, se = TRUE)

#Remove samples containing less than 1000 reads (R1.14, R1.55.2, R2.25, S2.78.2)
df.site_species.r <- as.data.frame(site_species.r)
rarefied_site_species <- data.frame()
for (i in 1:nrow(df.site_species.r)){
  if (rowSums(df.site_species.r[i,]) >= 1000){
    rarefied_site_species <- rbind(rarefied_site_species, df.site_species.r[i,])
  }
}
otu<-as.data.frame(rarefied_site_species)
otu_site<-c(rep("S1", 5), rep("S2", 5), rep("S3", 5), rep("S4", 5), rep("S5", 5), rep("R1", 4), rep("R2", 4))

#Add up all samples of fungi data at each site, so there is one total value per site
otu_total<-matrix(nrow=10,ncol=807)
colnames(otu_total)<-OTU_ID
for (i in 1:(ncol(otu))){
  otu_total[,i]<-tapply(otu[,i], otu_site, sum)
}

#Mantel test of the correlation between plant and fungal community dissimilarity and also plant and env
env$P<-as.numeric(env$P)
env$C<-as.numeric(env$C)
env$N<-as.numeric(env$N)

otu.dist<- vegdist(otu_total, method="bray")
env.dist<- vegdist(scale(env[, -4]), method="euclid")
veg.dist<- vegdist(veg_sxs, method = "bray")

mantel(veg.dist, otu.dist)
```

```
##
## Mantel statistic based on Pearson's product-moment correlation
##
## Call:
## mantel(xdis = veg.dist, ydis = otu.dist)
##
## Mantel statistic r: 0.4095
##      Significance: 0.014
##
## Upper quantiles of permutations (null model):
##  90%  95% 97.5%  99%
## 0.207 0.287 0.338 0.452
## Permutation: free
## Number of permutations: 999
```

```
mantel(veg.dist, env.dist)
```

```
##
## Mantel statistic based on Pearson's product-moment correlation
##
## Call:
## mantel(xdis = veg.dist, ydis = env.dist)
##
## Mantel statistic r: 0.2052
##      Significance: 0.091
##
## Upper quantiles of permutations (null model):
##   90%   95% 97.5%   99%
## 0.190 0.268 0.320 0.361
## Permutation: free
## Number of permutations: 999
```

```
#Indicator species analyses
```

```
#Look at indicator species for the two site types in plants and fungi
```

```
indval_veg<-multipatt(veg_sxs, cluster=c(rep("R", 5), rep("S", 5)), func = "r.g", control=how(nperm=999))
summary(indval_veg)
```

```
##
## Multilevel pattern analysis
## -----
##
## Association function: r.g
## Significance level (alpha): 0.05
##
## Total number of species: 47
## Selected number of species: 6
## Number of species associated to 1 group: 6
##
## List of species associated to each combination:
##
## Group R  #sps.  4
##              stat p.value
## Cladonia.arbuscula 0.946  0.010 **
## Trientalis.europaea 0.768  0.048 *
## Empetrum.nigrum    0.691  0.029 *
## Cared.bigelowii    0.669  0.039 *
##
## Group S  #sps.  2
##              stat p.value
## Salix.herbaceae 0.908  0.010 **
## Polytrichum.sp. 0.770  0.028 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
indval_otu<-multipatt(otu_total, cluster=c(rep("R", 5), rep("S", 5)), func = "r.g", control=how(nperm=999))
summary(indval_otu)
```

```
##
## Multilevel pattern analysis
## -----
##
## Association function: r.g
## Significance level (alpha): 0.05
##
## Total number of species: 807
## Selected number of species: 15
## Number of species associated to 1 group: 15
##
## List of species associated to each combination:
##
## Group R #sps. 9
##      stat p.value
## 793 0.874 0.011 *
## 444 0.838 0.011 *
## 1125 0.826 0.027 *
## 672 0.754 0.027 *
## 667 0.737 0.050 *
## 681 0.731 0.011 *
## 106 0.680 0.027 *
## 1119 0.654 0.011 *
## 1091 0.421 0.044 *
##
## Group S #sps. 6
##      stat p.value
## 410 0.743 0.049 *
## 451 0.737 0.044 *
## 848 0.697 0.011 *
## 334 0.626 0.020 *
## 376 0.593 0.030 *
## 799 0.515 0.011 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
site_type<-c(rep("R", 5), rep("S", 5))
adonis(veg_sxs ~ site_type, method = "bray", permutations = 999)
```

```
##
## Call:
## adonis(formula = veg_sxs ~ site_type, permutations = 999, method = "bray")
##
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##           Df SumsOfSqs MeanSqs F.Model      R2 Pr(>F)
## site_type  1   0.54964 0.54964  4.9473 0.38211 0.007 **
## Residuals  8   0.88880 0.11110           0.61789
## Total      9   1.43844           1.00000
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```

#PCoA of plant communities at each site with plant species mapped on
plant.pcoa<- cmdscale(veg.db, eig=TRUE, k=3)
explainvar1<-round(plant.pcoa$eig[1]/sum(plant.pcoa$eig),3)*100
explainvar2<-round(plant.pcoa$eig[2]/sum(plant.pcoa$eig),3)*100
explainvar3<-round(plant.pcoa$eig[3]/sum(plant.pcoa$eig),3)*100
sum.eig<-sum(explainvar1, explainvar2, explainvar3)

#png("pcoa_plants.png", units="in", width=7, height=5, res = 600)

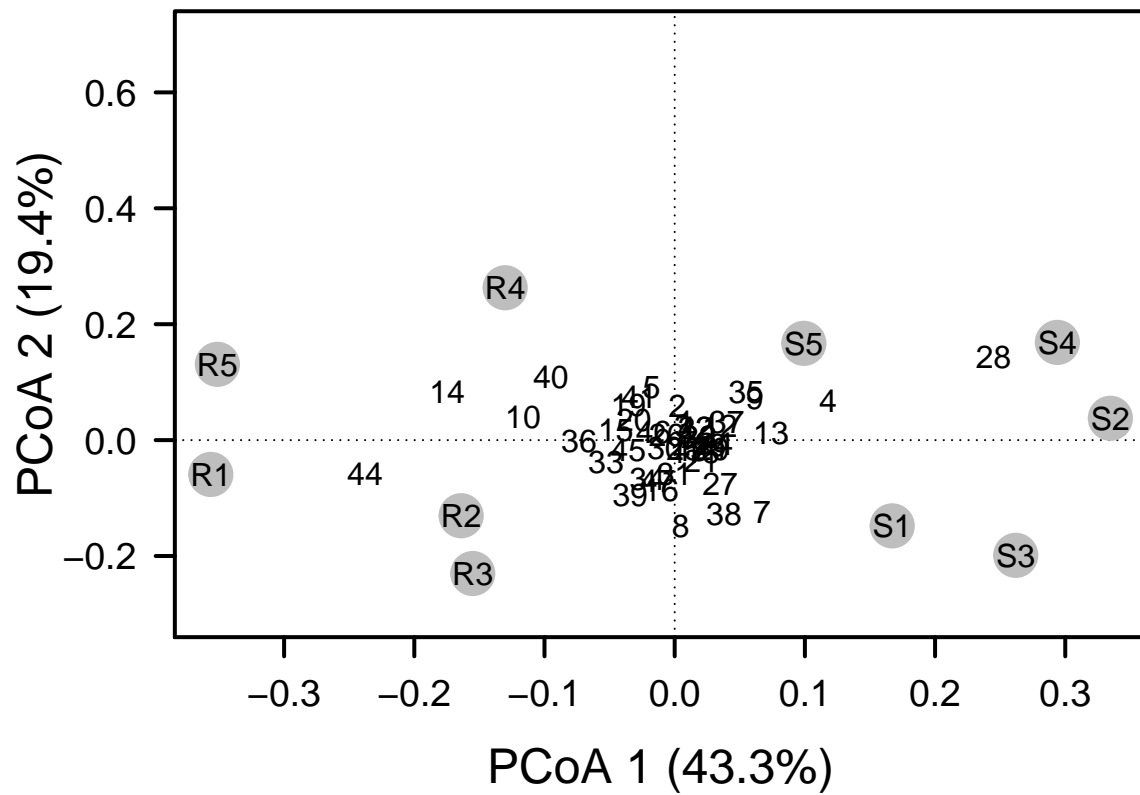
par(mar = c(5,5,1,2)+0.1)
plot(plant.pcoa$point[,1], plant.pcoa$points[,2], ylim = c(-0.3,0.7),
     xlab=paste("PCoA 1 (", explainvar1, "%)", sep = ""),
     ylab=paste("PCoA 2 (", explainvar2, "%)", sep = ""),
     pch=16, cex=2.0, type="n", cex.lab=1.5, cex.axis=1.2, axes=FALSE)
axis(side=1, labels=T, lwd.ticks = 2, cex.axis=1.2, las=1)
axis(side=2, labels=T, lwd.ticks = 2, cex.axis=1.2, las=1)
abline(h=0, v=0, lty=3)
box(lwd=2)

points(plant.pcoa$points[,1], plant.pcoa$points[,2],
       pch=19, cex=3, bg="gray", col="gray")
text(plant.pcoa$points[,1], plant.pcoa$points[,2],
     labels = row.names(plant.pcoa$points))

plantREL<- veg_sxs
for(i in 1:nrow(veg_sxs)){
  plantREL[i,]=veg_sxs[i,]/sum(veg_sxs[i,])
}

plant.pcoa<-add.spec.scores(plant.pcoa, plantREL, method="pcoa.scores")
text(plant.pcoa$cproj[,1], plant.pcoa$cproj[,2],
     labels=c(1:47), col="black")

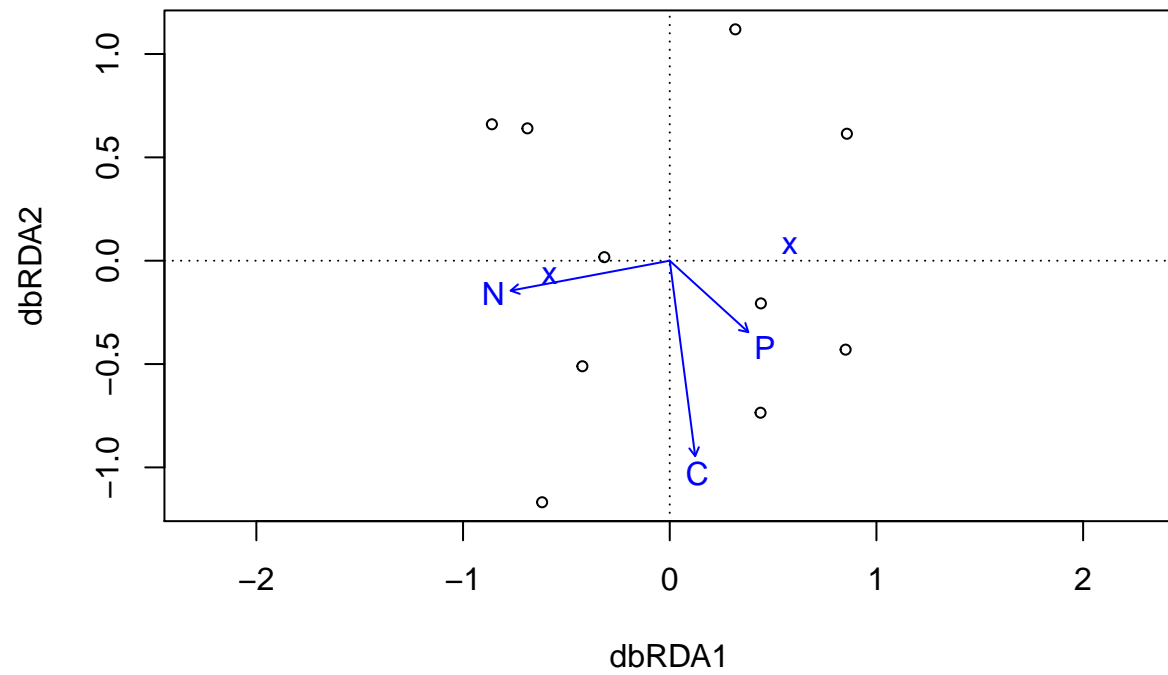
```



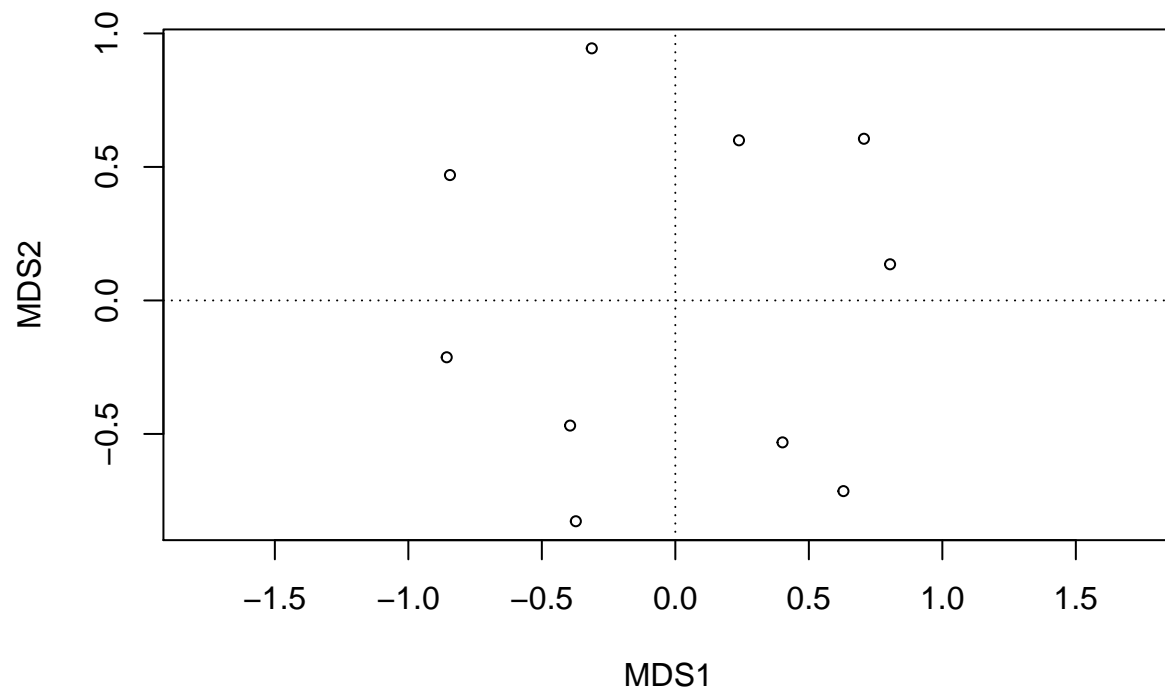
```
#dev.off()
```

```
#PCoA with all the soil environmental variables and site type
```

```
veg.db<-vegdist(veg_sxs, method = "bray", diag = TRUE)
doubts.dbrda<-dbrda(veg.db ~ ., as.data.frame(env))
ordiplot(doubts.dbrda)
```

```
doubs.dbrda.mod0<-dbrda(veg.db~1, as.data.frame(env))
ordiplot(doubs.dbrda.mod0)
```



```
doubs.dbrda.mod1<-dbrda(veg.db~., as.data.frame(env))
doubs.dbrda<-ordiR2step(doubs.dbrda.mod0, doubs.dbrda.mod1, perm.max=200)
```

```
## Step: R2.adj= 0
## Call: veg.db ~ 1
##
##               R2.adjusted
## <All variables> 0.363522123
## + Site_type    0.304872511
## + N            0.192894584
## + C            0.015557983
## + P            0.001422119
## <none>         0.000000000
##
##           Df    AIC      F Pr(>F)
## + Site_type 1 1.7676 4.9473 0.014 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.3048725
## Call: veg.db ~ Site_type
##
##               R2.adjusted
## + C            0.3636883
## <All variables> 0.3635221
```

```
## <none>          0.3048725
## + N             0.3022031
## + P             0.2976075
```

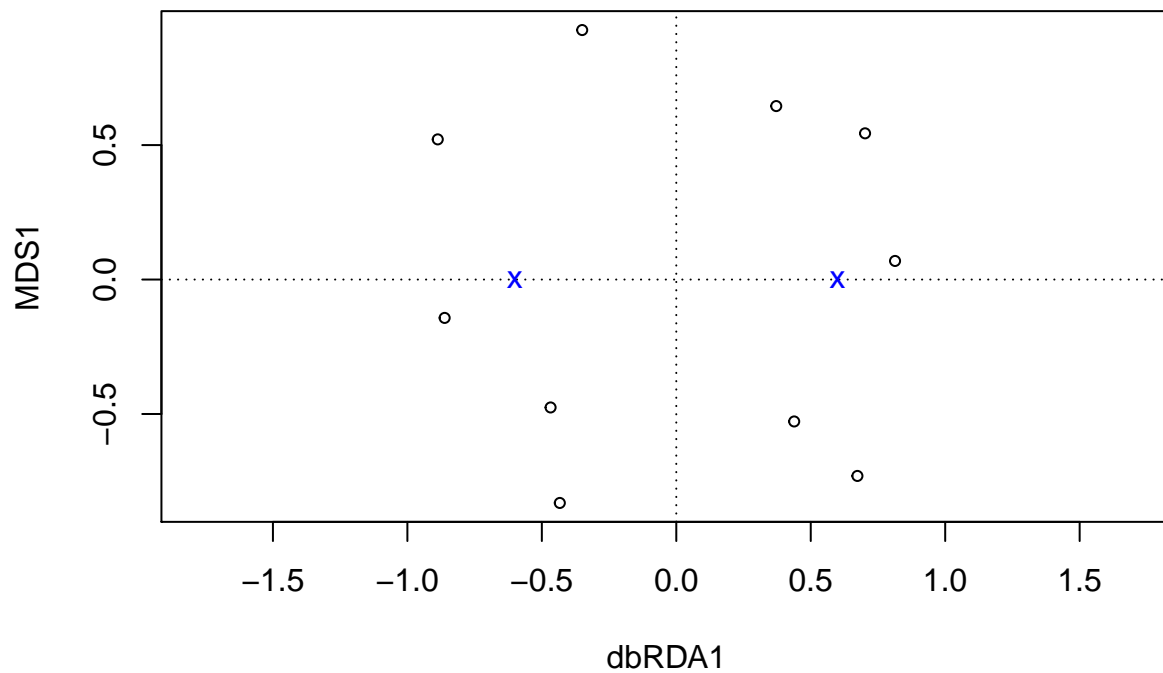
```
doubs.dbrda$call
```

```
## dbrda(formula = veg.db ~ Site_type, data = as.data.frame(env))
```

```
doubs.dbrda$anova
```

```
##           R2.adj Df    AIC      F Pr(>F)
## + Site_type  0.30487  1 1.7676 4.9473  0.014 *
## <All variables> 0.36352
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
ordiplot(doubs.dbrda)
```



```
permutest(doubs.dbrda, permutations = 999)
```

```
##
## Permutation test for dbrda under reduced model
##
```

```
## Permutation: free
## Number of permutations: 999
##
## Model: dbrda(formula = veg.db ~ Site_type, data = as.data.frame(env))
## Permutation test for all constrained eigenvalues
##           Df Inertia      F Pr(>F)
## Model      1 0.54964 4.9473 0.011 *
## Residual   8 0.88880
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
envfit(doubs.dbrda, env, perm=999)
```

```
##
## ***VECTORS
##
##      dbrDA1      MDS1      r2 Pr(>r)
## P -0.64388 -0.76513 0.3629 0.207
## N  0.99360  0.11292 0.6315 0.035 *
## C -0.16129 -0.98691 0.4454 0.136
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## Permutation: free
## Number of permutations: 999
##
## ***FACTORS:
##
## Centroids:
##           dbrDA1      MDS1
## Site_typeR -0.5998 0.0000
## Site_typeS  0.5998 0.0000
##
## Goodness of fit:
##           r2 Pr(>r)
## Site_type 0.4738 0.008 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## Permutation: free
## Number of permutations: 999
```

```
dbrda.explainvar1<-round(doubs.dbrda$CCA$eig[1]/
                        sum(c(doubs.dbrda$CCA$eig, doubs.dbrda$CA$eig)),3)*100
dbrda.explainvar2<-round(doubs.dbrda$CCA$eig[2]/
                        sum(c(doubs.dbrda$CCA$eig, doubs.dbrda$CA$eig)),3)*100

#png("dbRDA_plants_4.png", units="in", width=5, height=5, res = 600)

par(mar=c(5,5,4,4)+0.1)

plot(scores(doubs.dbrda, display="wa"),xlim=c(-1.3,1.1), ylim=c(-1.1,2.7),
      xlab=paste("dbRDA 1 (", dbrda.explainvar1, "%)", sep=""),
      ylab=paste("dbRDA 2 (", dbrda.explainvar2, "%)", sep=""),
      pch=16, cex=2.0, type="n", cex.lab=1.5, cex.axis=1.2, axes=FALSE)
```

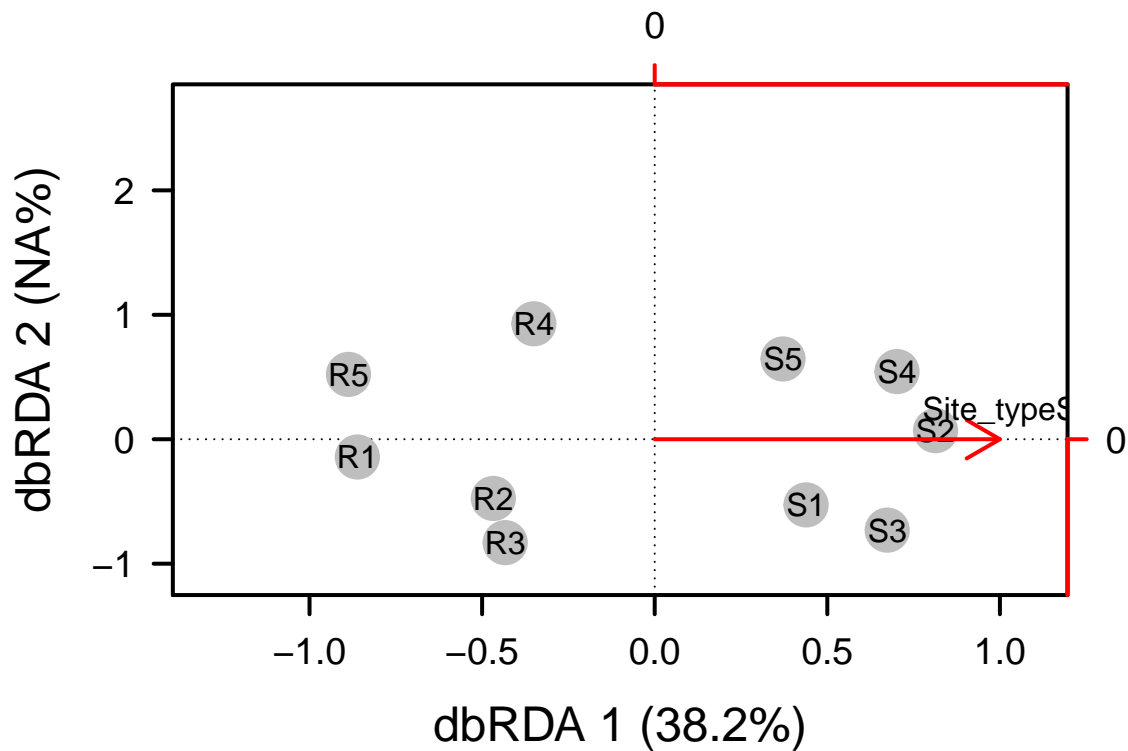
```

axis(side=1, labels=T, lwd.ticks=2, cex.axis=1.2, las=1)
axis(side=2, labels=T, lwd.ticks=2, cex.axis=1.2, las=1)
abline(h=0,v=0,lty=3)
box(lwd=2)

points(scores(doubs.dbrda, display="wa"),
       pch=19, cex=3, bg="gray", col="gray")
text(scores(doubs.dbrda, display="wa"),
     labels=row.names(scores(doubs.dbrda, display="wa"))))

vectors<-scores(doubs.dbrda, display = "bp")
arrows(0,0,vectors[,1], vectors[,2],
      lwd=2, lty=1, length=0.2, col="red")
text(vectors[,1], vectors[,2], pos=3,
     labels=row.names(vectors))
axis(side=3, lwd.ticks = 2, cex.axis=1.2, las=1, col="red", lwd=2.2,
     at=pretty(range(vectors[,1]))*2, labels = pretty(range(vectors[,1])))
axis(side=4, lwd.ticks = 2, cex.axis=1.2, las=1, col="red", lwd=2.2,
     at=pretty(range(vectors[,2]))*2, labels = pretty(range(vectors[,2])))

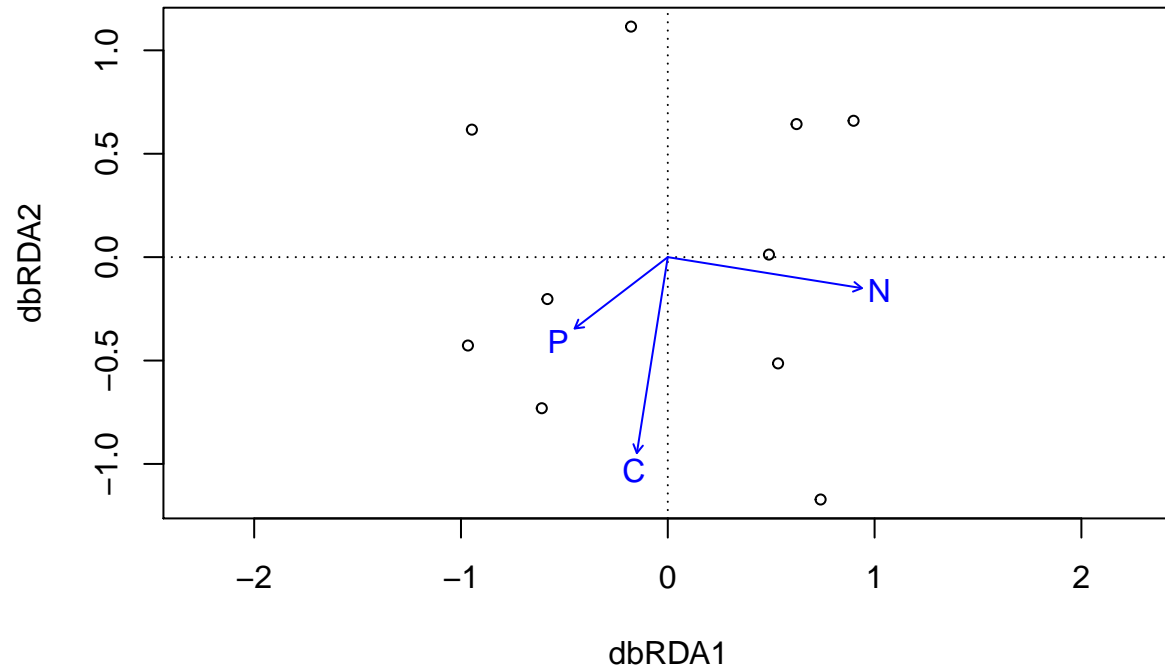
```



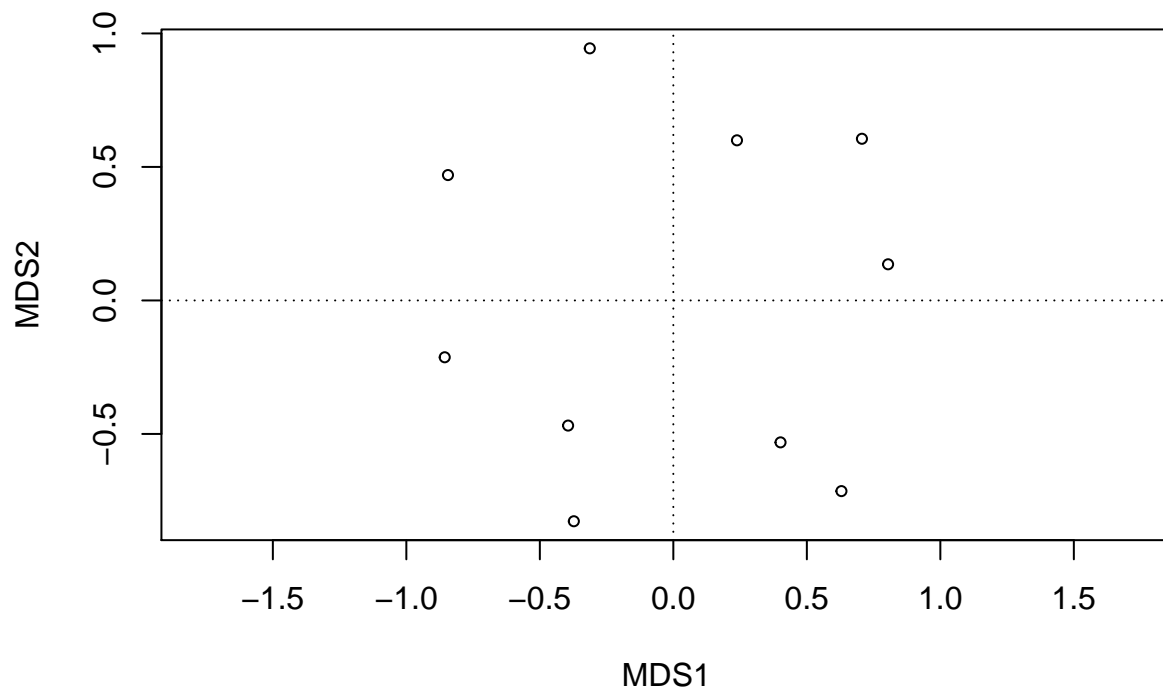
```
#dev.off()
```

```
#PCoA with just the soil environmental variables and not site type
```

```
veg.db<-vegdist(veg_sxs, method = "bray", diag = TRUE)
env_subset<-env[, -4]
doubs.dbrda<-dbrda(veg.db ~ ., as.data.frame(env_subset))
ordiplot(doubs.dbrda)
```



```
doubs.dbrda.mod0<-dbrda(veg.db~1, as.data.frame(env_subset))
ordiplot(doubs.dbrda.mod0)
```



```
doubs.dbrda.mod1<-dbrda(veg.db~., as.data.frame(env_subset))
doubs.dbrda<-ordiR2step(doubs.dbrda.mod0, doubs.dbrda.mod1, perm.max=200)
```

```
## Step: R2.adj= 0
## Call: veg.db ~ 1
##
##               R2.adjusted
## <All variables> 0.199096257
## + N             0.192894584
## + C             0.015557983
## + P             0.001422119
## <none>          0.000000000
##
##      Df    AIC      F Pr(>F)
## + N   1 3.2612 3.151 0.018 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.1928946
## Call: veg.db ~ N
##
##               R2.adjusted
## + C             0.2371528
## <All variables> 0.1990963
## <none>          0.1928946
## + P             0.1688228
```

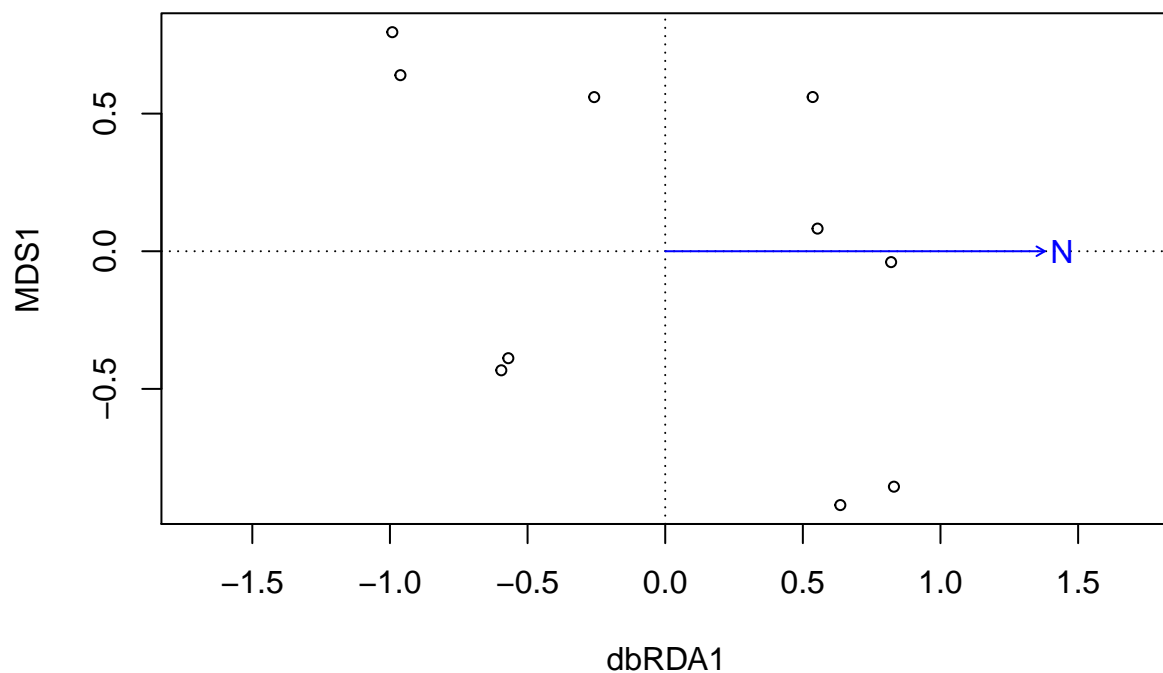
```
doubs.dbrda$call
```

```
## dbrda(formula = veg.db ~ N, data = as.data.frame(env_subset))
```

```
doubs.dbrda$anova
```

```
##           R2.adj Df    AIC    F Pr(>F)
## + N           0.1929  1 3.2612 3.151 0.018 *
## <All variables> 0.1991
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
ordiplot(doubs.dbrda)
```



```
permutest(doubs.dbrda, permutations = 999)
```

```
##
## Permutation test for dbrda under reduced model
##
## Permutation: free
## Number of permutations: 999
##
## Model: dbrda(formula = veg.db ~ N, data = as.data.frame(env_subset))
```



```
## Permutation test for all constrained eigenvalues
##           Df Inertia      F Pr(>F)
## Model      1 0.40647 3.151 0.022 *
## Residual   8 1.03198
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
envfit(doubs.dbrda, env_subset, perm=999)
```

```
##
## ***VECTORS
##
##      dbrDA1      MDS1      r2 Pr(>r)
## P -0.81953 -0.57303 0.2455 0.375
## N 0.86848 0.49573 0.9361 0.001 ***
## C -0.60531 0.79599 0.0090 0.963
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## Permutation: free
## Number of permutations: 999
```

```
dbrda.explainvar1<-round(doubs.dbrda$CCA$eig[1]/
                        sum(c(doubs.dbrda$CCA$eig, doubs.dbrda$CA$eig)),3)*100
dbrda.explainvar2<-round(doubs.dbrda$CCA$eig[2]/
                        sum(c(doubs.dbrda$CCA$eig, doubs.dbrda$CA$eig)),3)*100

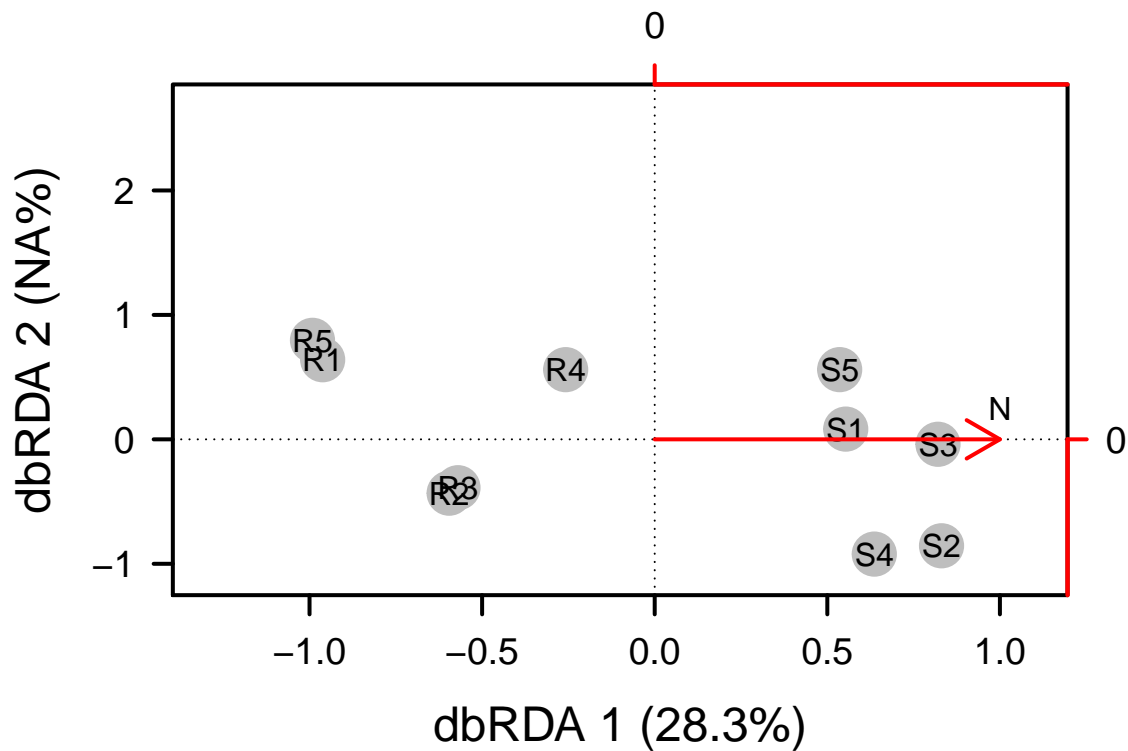
#png("dbRDA_plants_3.png", units="in", width=5, height=5, res = 600)
par(mar=c(5,5,4,4)+0.1)

plot(scores(doubs.dbrda, display="wa"),xlim=c(-1.3,1.1), ylim=c(-1.1,2.7),
      xlab=paste("dbRDA 1 (", dbrda.explainvar1, "%)", sep=""),
      ylab=paste("dbRDA 2 (", dbrda.explainvar2, "%)", sep=""),
      pch=16, cex=2.0, type="n", cex.lab=1.5, cex.axis=1.2, axes=FALSE)

axis(side=1, labels=T, lwd.ticks=2, cex.axis=1.2, las=1)
axis(side=2, labels=T, lwd.ticks=2, cex.axis=1.2, las=1)
abline(h=0,v=0,lty=3)
box(lwd=2)

points(scores(doubs.dbrda, display="wa"),
       pch=19, cex=3, bg="gray", col="gray")
text(scores(doubs.dbrda, display="wa"),
     labels=row.names(scores(doubs.dbrda, display="wa"))))

vectors<-scores(doubs.dbrda, display = "bp")
arrows(0,0,vectors[,1], vectors[,2],
      lwd=2, lty=1, length=0.2, col="red")
text(vectors[,1], vectors[,2], pos=3,
     labels=row.names(vectors))
axis(side=3, lwd.ticks = 2, cex.axis=1.2, las=1, col="red", lwd=2.2,
     at=pretty(range(vectors[,1]))*2, labels = pretty(range(vectors[,1])))
axis(side=4, lwd.ticks = 2, cex.axis=1.2, las=1, col="red", lwd=2.2,
     at=pretty(range(vectors[,2]))*2, labels = pretty(range(vectors[,2])))
```



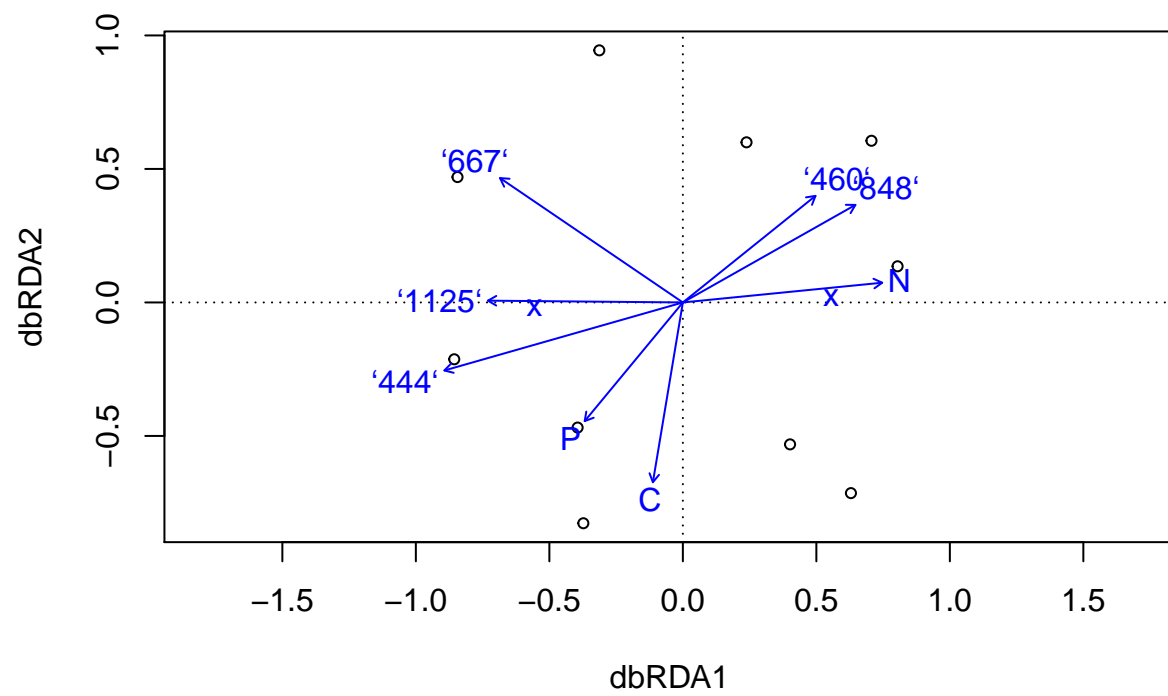
```
#dev.off()
```

```
#Add OTU and soil environmental variables together as total plant environment matrix
#Only use OTUs that were highly significant (**) and >0.7 association
otu_total<-as.data.frame(otu_total)
```

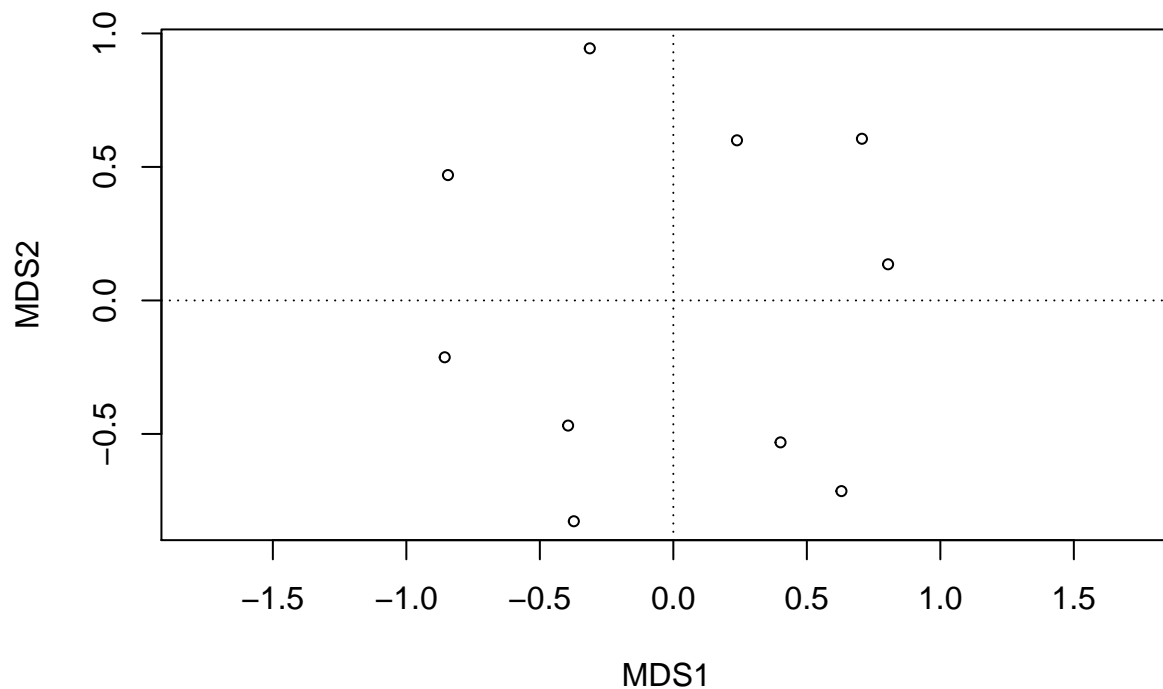
```
#For the ridge sites
env$'667'<-otu_total$'667'
env$'444'<-otu_total$'444'
env$'1125'<-otu_total$'1125'
```

```
#For the snowbed sites
env$'848'<-otu_total$'848'
env$'460'<-otu_total$'460'
```

```
#Do PCoA analysis again with new environmental matrix including fungi
veg.db<-vegdist(veg_sxs, method = "bray", diag = TRUE)
doubts.dbrda<-dbrda(veg.db ~ ., as.data.frame(env))
ordiplot(doubts.dbrda)
```



```
doubs.dbrda.mod0<-dbrda(veg.db~1, as.data.frame(env))
ordiplot(doubs.dbrda.mod0)
```



*#This function is no longer working - the error says it could be due to "fitting too many terms".
 #Before the presentation, it was working fine, so I have included the plot "dbRDA_with_fungi.png"
 #which is the output from this part that isn't working anymore*

```
# douds.dbrda.mod1<-dbrda(veg.db~., as.data.frame(env))
# douds.dbrda<-ordiR2step(douds.dbrda.mod0, douds.dbrda.mod1, perm.max=200)
#
# douds.dbrda$call
# douds.dbrda$anova
# ordiplot(douds.dbrda)
#
# permutest(douds.dbrda, permutations = 999)
# envfit(douds.dbrda, env, perm=999)
#
# dbrda.explainvar1<-round(douds.dbrda$CCA$eig[1]/
#                           sum(c(douds.dbrda$CCA$eig, douds.dbrda$CA$eig)),3)*100
# dbrda.explainvar2<-round(douds.dbrda$CCA$eig[2]/
#                           sum(c(douds.dbrda$CCA$eig, douds.dbrda$CA$eig)),3)*100
#
# #png("dbRDA_with_fungi.png", units="in", width=7, height=5, res = 600)
#
# par(mar=c(5,5,4,4)+0.1)
#
# plot(scores(douds.dbrda, display="wa"),xlim=c(-1.3,1.1), ylim=c(-1.1,2.7),
#       xlab=paste("dbRDA 1 (", dbrda.explainvar1, "%)", sep=""),
#       ylab=paste("dbRDA 2 (", dbrda.explainvar2, "%)", sep=""),
```

```

#      pch=16, cex=2.0, type="n", cex.lab=1.5, cex.axis=1.2, axes=FALSE)
#
# axis(side=1, labels=T, lwd.ticks=2, cex.axis=1.2, las=1)
# axis(side=2, labels=T, lwd.ticks=2, cex.axis=1.2, las=1)
# abline(h=0,v=0,lty=3)
# box(lwd=2)
#
# points(scores(doubs.dbrda, display="wa"),
#        pch=19, cex=3, bg="gray", col="gray")
# text(scores(doubs.dbrda, display="wa"),
#       labels=row.names(scores(doubs.dbrda, display="wa"))))
#
# vectors<-scores(doubs.dbrda, display = "bp")
# arrows(0,0,vectors[,1], vectors[,2],
#        lwd=2, lty=1, length=0.2, col="red")
# text(vectors[,1], vectors[,2], pos=3,
#       labels=row.names(vectors))
# axis(side=3, lwd.ticks = 2, cex.axis=1.2, las=1, col="red", lwd=2.2,
#       at=pretty(range(vectors[,1]))*2, labels = pretty(range(vectors[,1])))
# axis(side=4, lwd.ticks = 2, cex.axis=1.2, las=1, col="red", lwd=2.2,
#       at=pretty(range(vectors[,2]))*2, labels = pretty(range(vectors[,2])))
#
#dev.off()

```