

6. Worksheet: Among Site (Beta) Diversity – Part 2

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OVERVIEW

In this worksheet, we continue to explore concepts, statistics, and visualizations related to β -diversity. Now that you know how to formally quantify β -diversity, we will learn how to test hypotheses about β -diversity using multivariate statistics.

Directions:

1. In the Markdown version of this document in your cloned repo, change “Student Name” on line 3 (above) with your name.
2. Complete as much of the worksheet as possible during class.
3. Use the handout as a guide; it contains a more complete description of data sets along with examples of proper scripting needed to carry out the exercises.
4. Answer questions in the worksheet. Space for your answers is provided in this document and is indicated by the “>” character. If you need a second paragraph be sure to start the first line with “>”. You should notice that the answer is highlighted in green by RStudio (color may vary if you changed the editor theme).
5. Before you leave the classroom today, you should **push** this file to your GitHub repo, at whatever stage you are. This will enable you to pull your work onto your own computer.
6. When you have completed the worksheet, **Knit** the text and code into a single PDF file by pressing the **Knit** button in the RStudio scripting panel. This will save the PDF output in your Posit.cloud workspace: `/cloud/project/QB-2025/Week4-Beta/`
7. After Knitting, please submit the worksheet by making a **push** to your GitHub repo and then create a **pull request** via GitHub. Your pull request should include this file (**6.BetaDiversity_2_Worksheet.Rmd**) with all code blocks filled out and questions answered) and the PDF output of **Knitr** (**6.BetaDiversity_2_Worksheet.pdf**).

The completed exercise is due on **Wednesday, February 12th, 2025 before 12:00 PM (noon)**.

1) R SETUP

Typically, the first thing you will do in either an R script or an RMarkdown file is setup your environment. This includes things such as setting the working directory and loading any packages that you will need.

In the R code chunk below, provide the code to:

1. clear your R environment,
2. print your current working directory,
3. set your working directory to your **Week4-Beta/** folder.
4. load the **vegan** R package (be sure to install if needed).

```
#Code to clear R environment
rm(list = ls())
getwd()
```

```
## [1] "/cloud/project/QB2025_Guevara/Week4-Beta"
setwd("/cloud/project/QB2025_Guevara/Week4-Beta")
getwd()
```

```
## [1] "/cloud/project/QB2025_Guevara/Week4-Beta"
library(vegan)
```

```
## Loading required package: permute
## Loading required package: lattice
## This is vegan 2.6-8
```

2) LOADING DATA

Load dataset

In the R code chunk below, load the `doubs` dataset from the `ade4` package

```
# note, please do not print the dataset when submitting
data(doubs)
```

```
## Warning in data(doubs): data set 'doubs' not found
```

3) HYPOTHESIS TESTING

A. Multivariate Procedures for Categorical Designs

Earlier work done in the Doubs River suggested that the river has four distinct regions of habitat quality: the first region (sites 1-14) of “high quality”; the second (sites 15 - 19) and fourth (sites 26 - 30) of “moderate quality”; and the third (sites 20 - 25) of “low quality”.

In the code chunk below, test the hypothesis that fish community composition varies with river quality.

1. create a factor vector that categorizes habitat quality in the Doubs River,
2. use the multivariate analyses for categorical predictors to describe how fish community structure relates to habitat quality.

```
#Create "Factors" vector
quality <- c(rep("HQ", 13), rep("MQ", 5), rep("LQ", 6), rep("MQ", 5))
```

```
#Calling fish dataframe from doubs as fish
```

```
library(ade4)
data(doubs)
fish <- doubs$fish
```

```
fish <- fish[-8, ] #Removes site 8 from data
```

```
#Run PERMANOVA with adonis function
```

```
adonis2(fish ~ quality, method = "bray", permutations = 999)
```

```
## Permutation test for adonis under reduced model
## Permutation: free
## Number of permutations: 999
##
## adonis2(formula = fish ~ quality, permutations = 999, method = "bray")
##           Df SumOfSqs      R2      F Pr(>F)
## Model      2   3.0947 0.45765 10.97  0.001 ***
```

```

## Residual 26    3.6674 0.54235
## Total      28    6.7621 1.00000
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

#load indicpecies because multipatt is function from this package
library(indicpecies)
indval <- multipatt(fish, cluster = quality, func = "IndVal.g",
                    control = how(nperm = 999))
summary(indval)

##
## Multilevel pattern analysis
## -----
##
## Association function: IndVal.g
## Significance level (alpha): 0.05
##
## Total number of species: 27
## Selected number of species: 23
## Number of species associated to 1 group: 1
## Number of species associated to 2 groups: 22
##
## List of species associated to each combination:
##
## Group MQ #sps. 1
##      stat p.value
## Teso 0.686    0.02 *
##
## Group HQ+MQ #sps. 2
##      stat p.value
## Satr 0.860    0.005 **
## Phph 0.859    0.013 *
##
## Group LQ+MQ #sps. 20
##      stat p.value
## Alal 0.935    0.001 ***
## Gogo 0.933    0.001 ***
## Ruru 0.916    0.001 ***
## Legi 0.901    0.001 ***
## Baba 0.895    0.001 ***
## Chna 0.866    0.001 ***
## Spbi 0.866    0.002 **
## Cyca 0.866    0.001 ***
## Acce 0.866    0.002 **
## Lele 0.863    0.008 **
## Titi 0.853    0.003 **
## Chto 0.829    0.005 **
## Rham 0.829    0.001 ***
## Anan 0.829    0.001 ***
## Eslu 0.827    0.024 *
## Pefl 0.806    0.014 *
## Blbj 0.791    0.003 **
## Scer 0.766    0.006 **
## Abbr 0.750    0.008 **

```

```

## Icme 0.661    0.018 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

fish.rel <- decostand(fish, method = "total")
phi <- multipatt(fish.rel, cluster = quality, func = "r.g", control = how(nperm = 999))
summary(phi)

##
## Multilevel pattern analysis
## -----
##
## Association function: r.g
## Significance level (alpha): 0.05
##
## Total number of species: 27
## Selected number of species: 18
## Number of species associated to 1 group: 9
## Number of species associated to 2 groups: 9
##
## List of species associated to each combination:
##
## Group HQ #sps. 3
##      stat p.value
## Phph 0.802    0.001 ***
## Neba 0.734    0.001 ***
## Satr 0.650    0.001 ***
##
## Group LQ #sps. 2
##      stat p.value
## Alal 0.693    0.001 ***
## Ruru 0.473    0.029 *
##
## Group MQ #sps. 4
##      stat p.value
## Anan 0.571    0.006 **
## Spbi 0.557    0.007 **
## Chto 0.542    0.007 **
## Icme 0.475    0.030 *
##
## Group LQ+MQ #sps. 9
##      stat p.value
## Legi 0.658    0.002 **
## Baba 0.645    0.002 **
## Rham 0.600    0.004 **
## Acce 0.594    0.003 **
## Cyca 0.586    0.004 **
## Chna 0.571    0.010 **
## Blbj 0.571    0.005 **
## Gogo 0.523    0.010 **
## Abbr 0.499    0.025 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

```

Question 1: Based on the PERMANOVA, IndVal, and phi coefficient analyses, what did you learn about the

relationship between habitat quality and the fish species composition? Are the different analyses consistent with one another and do they agree with the visualizations (heat maps, cluster dendograms, ordinations) that you created?

Answer 1: It seems that Satr and Phph are fish species that are found in waters of higher quality. Generally the high or low quality have the lowest number of species at their locations. The different analyses are super consistent with each other, however, they are similar enough that we see some of the same species in the same water quality using any of the analyses. This is likely due to the Indval method measuring species specificity and fidelity without standardizing while phi measures species correlation and does standardize.

B. Multivariate Procedures for Continuous Designs

i. Mantel Test

In the R code chunk below, do the following:

1. create distance matrices for both fish communities and environmental factors, and
2. use a Mantel test to test the hypothesis that fish assemblages are correlated with stream environmental variables.

```
#Define matrices
fish.dist <- vegdist(doubs$fish[-8,], method = "bray")
env.dist <- vegdist(scale(doubs$env[-8,]), method = "euclid")

#Mantel test
mantel(fish.dist,env.dist)

##
## Mantel statistic based on Pearson's product-moment correlation
##
## Call:
## mantel(xdis = fish.dist, ydis = env.dist)
##
## Mantel statistic r: 0.604
##      Significance: 0.001
##
## Upper quantiles of permutations (null model):
##   90%   95%  97.5%   99%
## 0.109 0.138 0.174 0.200
## Permutation: free
## Number of permutations: 999
```

Question 2: What do the results from our Mantel test suggest about fish diversity and stream environmental conditions? How does this relate to your hypothesis about stream quality influencing fish communities?

Answer 2:

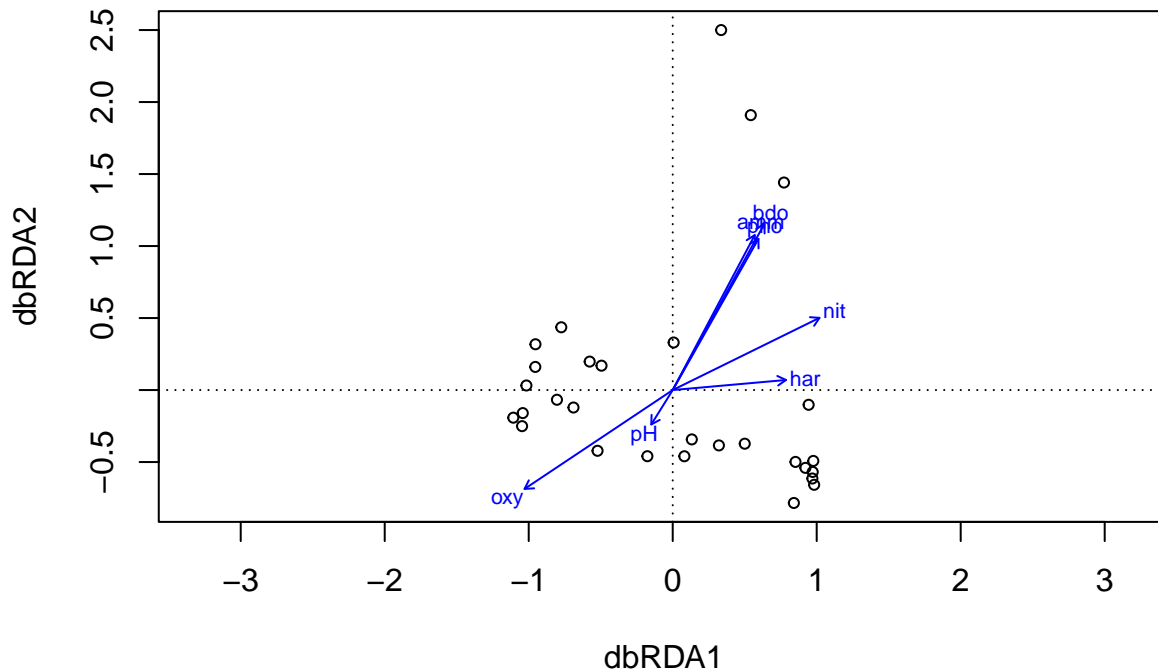
There is a pretty strong and statistically significant correlation between stream env. conditions and fish community composition as $r = 0.604$ basically meaning that fish diversity increases along with better/improved env. conditions. I would say that this fits, although, my previous hypothesis was discussing which fish would be potential indicators of river quality. This finding does indeed seem to make sense, the greater the water conditions, the less stress the fish have to contend with in order to establish a population there to boost the local species richness. ### ii. Constrained Ordination In the R code chunk below, do the following:

1. create an environmental matrix of the water chemistry data included in the `doubs` dataset using forward and reverse selection of variables,
2. conduct a redundancy analysis on the fish assemblages of the Doubs River,

3. use a permutation test to determine the significance of the constrained analysis,
4. use a permutation test to determine the correlation of each environmental factor on the constrained axes,
5. calculate the explained variation on the first and second constrained axes,
6. plot the constrained ordination results including labeled points for each site, and
7. add vectors that demonstrate the influence of each environmental factor the constrained ordination.

```
#Define environmental matrix
env.chem <- as.matrix(doubs$en[-8,5:11])

#Perform dbRDA
fish.db <- vegdist(fish, method = "bray")
doubs.dbrda <- dbrda(fish.db ~ ., as.data.frame(env.chem))
ordiplot(doubs.dbrda)
```

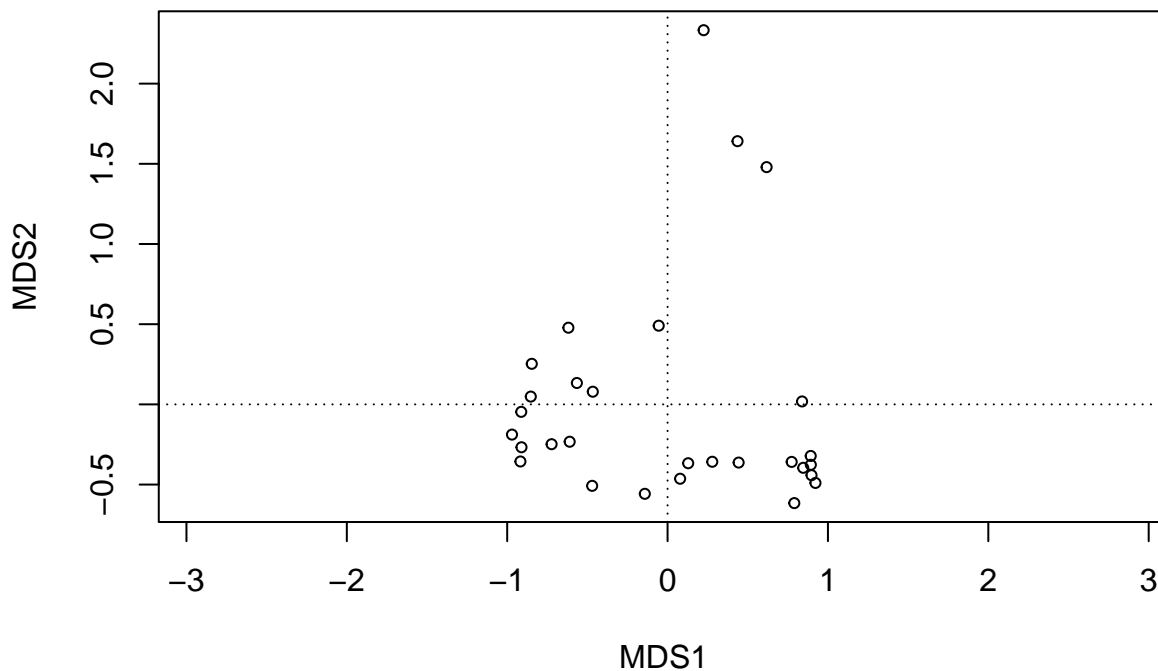


```
psych::corr.test(env.chem)

## Call:psych::corr.test(x = env.chem)
## Correlation matrix
##      pH   har   pho   nit   amm   oxy   bdo
## pH   1.00  0.08 -0.08 -0.04 -0.12  0.19 -0.16
## har   0.08  1.00  0.37  0.53  0.30 -0.37  0.34
## pho  -0.08  0.37  1.00  0.80  0.97 -0.76  0.91
## nit  -0.04  0.53  0.80  1.00  0.80 -0.69  0.68
## amm  -0.12  0.30  0.97  0.80  1.00 -0.75  0.90
## oxy   0.19 -0.37 -0.76 -0.69 -0.75  1.00 -0.84
## bdo  -0.16  0.34  0.91  0.68  0.90 -0.84  1.00
## Sample Size
## [1] 29
## Probability values (Entries above the diagonal are adjusted for multiple tests.)
##      pH   har   pho   nit   amm   oxy   bdo
## pH   0.00  1.00  1.00  1.00  1.00  1.00  1.00
## har  0.66  0.00  0.46  0.03  0.83  0.46  0.59
## pho  0.68  0.05  0.00  0.00  0.00  0.00  0.00
```

```
## nit 0.83 0.00 0.00 0.00 0.00 0.00 0.00
## amm 0.53 0.12 0.00 0.00 0.00 0.00 0.00
## oxy 0.32 0.05 0.00 0.00 0.00 0.00 0.00
## bdo 0.40 0.07 0.00 0.00 0.00 0.00 0.00
##
## To see confidence intervals of the correlations, print with the short=FALSE option
#First we will model only the intercept
doubts.dbrda.mod0 <- dbrda(fish.db ~ 1, as.data.frame(env.chem))

#Note there are no vectors here(we didn't constrain anything)
#Therefore, the axes suggest this is a simple MDS (i.e., PCoA)
ordiplot(doubts.dbrda.mod0)
```



```
#next, we will model the full model with all explanatory variables
doubts.dbrda.mod1 <- dbrda(fish.db ~ ., as.data.frame(env.chem))

#Now we step through all combinations of explanatory variables in our model
#The function returns the model with the lowest AIC value
doubts.dbrda <- ordiR2step(doubts.dbrda.mod0, doubts.dbrda.mod1, perm.max = 200)
```

```
## Step: R2.adj= 0
## Call: fish.db ~ 1
##
##               R2.adjusted
## <All variables> 0.53032584
## + oxy          0.27727176
## + nit          0.25755208
## + bdo          0.17477787
## + pho          0.14568614
## + har          0.14174915
## + amm          0.14142804
## <none>         0.00000000
## + pH          -0.01827054
```

```

##
##          Df      AIC      F Pr(>F)
## + oxy  1 47.939 11.742  0.002 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.2772718
## Call: fish.db ~ oxy
##
##                R2.adjusted
## <All variables>  0.5303258
## + bdo           0.4009000
## + amm           0.3474192
## + pho           0.3452702
## + har           0.3331357
## + nit           0.3316120
## <none>          0.2772718
## + pH           0.2586983
##
##          Df      AIC      F Pr(>F)
## + bdo  1 43.404 6.5716  0.002 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.4009
## Call: fish.db ~ oxy + bdo
##
##                R2.adjusted
## <All variables>  0.5303258
## + nit           0.4980793
## + har           0.4695121
## <none>          0.4009000
## + pho           0.3938042
## + amm           0.3869134
## + pH           0.3865240
##
##          Df      AIC      F Pr(>F)
## + nit  1 39.134 6.034  0.002 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.4980793
## Call: fish.db ~ oxy + bdo + nit
##
##                R2.adjusted
## + amm           0.5415705
## <All variables>  0.5303258
## + pho           0.5277128
## + har           0.5218852
## <none>          0.4980793
## + pH           0.4843267

```

```

#Let's look at the model that was selected
doubts.dbrda$call

```

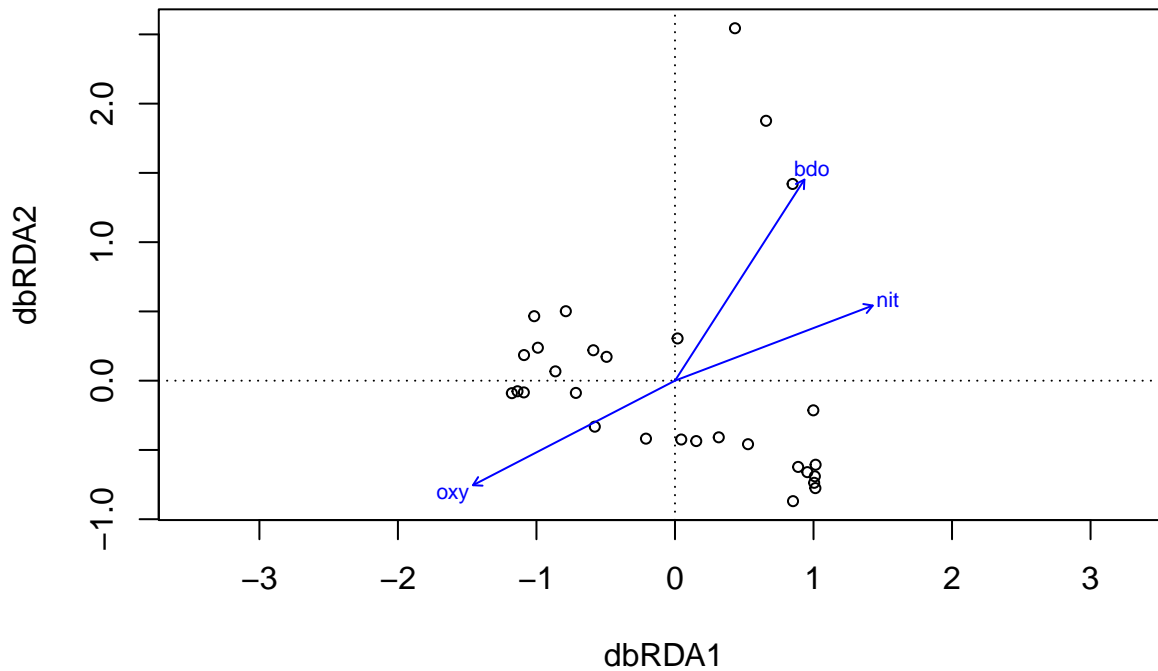


```
## dbrda(formula = fish.db ~ oxy + bdo + nit, data = as.data.frame(env.chem))
```

```
doubs.dbrda$anova
```

```
##
##          R2.adj Df      AIC      F Pr(>F)
## + oxy      0.27727 1 47.939 11.7421 0.002 **
## + bdo      0.40090 1 43.404  6.5716 0.002 **
## + nit      0.49808 1 39.134  6.0340 0.002 **
## <All variables> 0.53033
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

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



```
#Permutation tests to evaluate significance
```

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

```
##
## Permutation test for dbrda under reduced model
##
## Permutation: free
## Number of permutations: 999
##
## Model: dbrda(formula = fish.db ~ oxy + bdo + nit, data =
## as.data.frame(env.chem))
## Permutation test for all constrained eigenvalues
##          Df Inertia      F Pr(>F)
## Model      3  3.7317 10.262 0.001 ***
## Residual 25  3.0304
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
envfit(doubs.dbrda, env.chem[,c(4,6,7)], perm = 999)
```

```
##
```

```

## ***VECTORS
##
##      dbrDA1   dbrDA2    r2 Pr(>r)
## nit  0.87724  0.48005 0.6431  0.001 ***
## oxy -0.82864 -0.55979 0.7656  0.001 ***
## bdo  0.55603  0.83116 0.8939  0.001 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## Permutation: free
## Number of permutations: 999

#Calculate Explained Variation
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

#Define Plot parameters
par(mar = c(5,5,4,4) + 0.1)

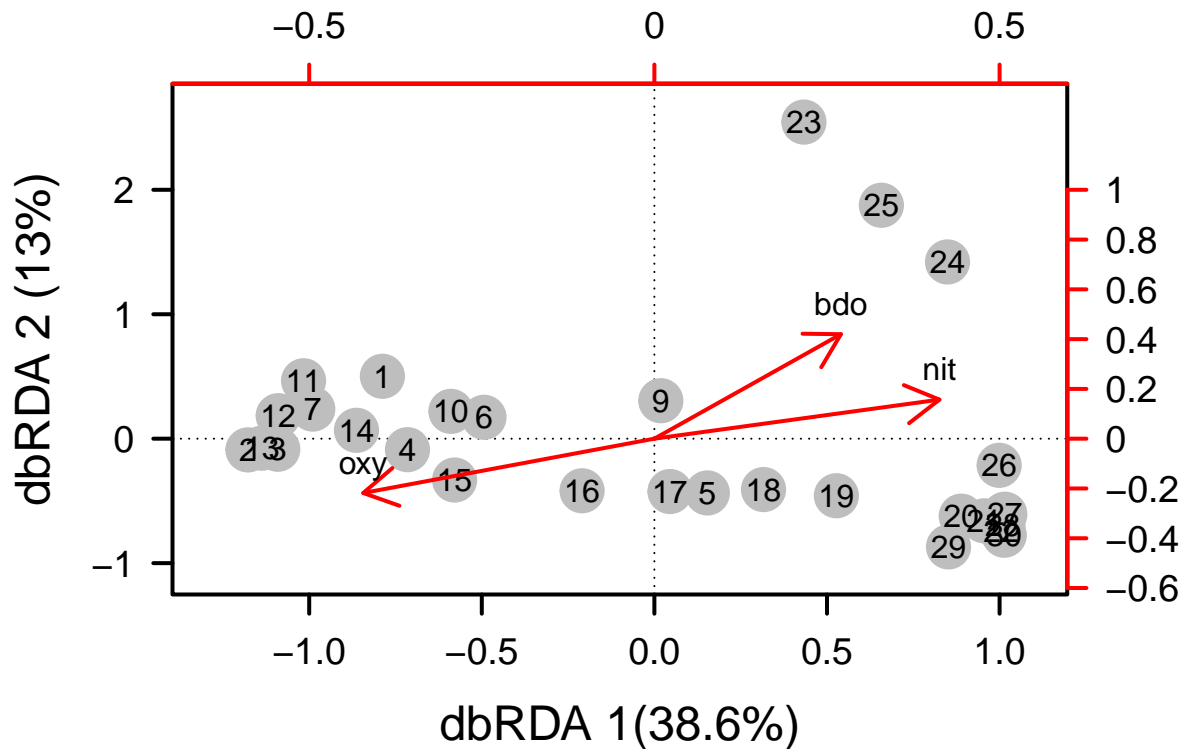
#Initiate Plot
plot(scores(doubs.dbrda, display = "wa"), xlim = c(-1.3, 1.1),
      ylim = c(-1.1, 2.7), xlab = paste("dbRDA 1(", dbrda.explainvar1, "%)",
      sep = ""), ylab = paste("dbRDA 2 (", dbrda.explainvar2, "%)", sep =
      cex.axis = 1.2, axes = FALSE)

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

#Add points & labels
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")))

#Add environmental vectors
vectors <- scores(doubs.dbrda, display = "bp")
#row.names(vectors) <- rownames(vectors)
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])))

```



Question 3: Based on the constrained ordination, what are the environmental variables (or groups of correlated variables) that seem to be contributing to variation in fish community structure?

Answer 3: It seems that nitrogen, oxygen and 'bdo' are contributing the most to variation within fish community structure.

iii. Variation Partitioning

In the code chunk below,

1. Create a matrix model of the selected environmental variables,
2. Create a matrix model of the selected PCNM axes,
3. Perform constrained and partial constrained ordinations using the spatial and environmental models you just created,
4. Test the significance of each of your constrained ordinations using permutation tests,
5. Partition the variation among sites into the relative importance of space, environment, spatially structured environment, and residuals,
6. Plot the variation partitioning output to visualize it.

#Remember, our environmental model uses oxy, bdo, and nit and has R2 = 0.53.

```
doubs.dbrda$anova
```

```
##           R2.adj Df      AIC      F Pr(>F)
## + oxy      0.27727  1 47.939 11.7421  0.002 **
## + bdo      0.40090  1 43.404  6.5716  0.002 **
## + nit      0.49808  1 39.134  6.0340  0.002 **
## <All variables> 0.53033
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

#Let's create a matrix model for our environmental data

```
env.mod <- model.matrix(~ oxy + bdo + nit, as.data.frame(env.chem))[, -1]
```

```
#First, we will weighh each site by its relative abundance
rs <- rowSums(fish)/sum(fish)
```

```
#Next, we will perofmr PCNM
doubts.pcnmw <- pcnm(dist(doubts$xy[-8,]), w = rs, dist.ret = T)
```

```
#PCNM can return negative eigenvalues, but only the eigenvectors associated with the positive eigenvalu
doubts.pcnmw$values > 0
```

```
## [1] TRUE TRUE TRUE TRUE TRUE TRUE TRUE TRUE TRUE TRUE TRUE TRUE
## [13] TRUE TRUE TRUE TRUE TRUE FALSE FALSE FALSE FALSE FALSE FALSE FALSE
## [25] FALSE FALSE
```

```
doubts.space <- as.data.frame(scores(doubts.pcnmw))
doubts.pcnm.mod0 <- dbrda(fish.db ~ 1, doubts.space)
doubts.pcnm.mod1 <- dbrda(fish.db ~ ., doubts.space)
step.pcnm <- ordiR2step(doubts.pcnm.mod0, doubts.pcnm.mod1, perm.max = 200)
```

```
## Step: R2.adj= 0
## Call: fish.db ~ 1
##
##               R2.adjusted
## <All variables> 0.626011301
## + PCNM2        0.235370423
## + PCNM3        0.078394885
## + PCNM13       0.065305668
## + PCNM5        0.046185074
## + PCNM6        0.032809156
## + PCNM16       0.030486700
## + PCNM14       0.029680999
## + PCNM9        0.020357410
## + PCNM15       0.013632610
## + PCNM8        0.009411968
## + PCNM1        0.003986221
## + PCNM17       0.002415012
## + PCNM10       0.001326442
## <none>         0.000000000
## + PCNM7        -0.001861430
## + PCNM11       -0.006841522
## + PCNM4        -0.007089863
## + PCNM12       -0.014396973
##
##           Df      AIC      F Pr(>F)
## + PCNM2   1 49.574 9.619 0.002 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.2353704
## Call: fish.db ~ PCNM2
##
##               R2.adjusted
## <All variables> 0.6260113
## + PCNM3        0.3429270
## + PCNM5        0.3057368
```

```

## + PCNM1          0.2885396
## + PCNM16         0.2786746
## + PCNM14         0.2744520
## + PCNM15         0.2692809
## + PCNM6          0.2659866
## + PCNM13         0.2636194
## + PCNM9          0.2517847
## + PCNM8          0.2496240
## + PCNM10         0.2434688
## + PCNM7          0.2431476
## + PCNM17         0.2404343
## + PCNM11         0.2366833
## <none>           0.2353704
## + PCNM12         0.2288789
## + PCNM4          0.2189522
##
##           Df      AIC      F Pr(>F)
## + PCNM3    1 46.083 5.4196 0.002 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.342927
## Call: fish.db ~ PCNM2 + PCNM3
##
##           R2.adjusted
## <All variables> 0.6260113
## + PCNM5         0.4076020
## + PCNM1         0.3970300
## + PCNM16        0.3853210
## + PCNM15        0.3828748
## + PCNM14        0.3781827
## + PCNM13        0.3770376
## + PCNM6         0.3595644
## + PCNM8         0.3556885
## + PCNM7         0.3541631
## + PCNM10        0.3526775
## + PCNM17        0.3513683
## + PCNM9         0.3433672
## <none>          0.3429270
## + PCNM11        0.3416399
## + PCNM12        0.3396547
## + PCNM4         0.3311509
##
##           Df      AIC      F Pr(>F)
## + PCNM5    1 43.941 3.8385 0.006 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.407602
## Call: fish.db ~ PCNM2 + PCNM3 + PCNM5
##
##           R2.adjusted
## <All variables> 0.6260113
## + PCNM1         0.4721469

```

```

## + PCNM16      0.4631976
## + PCNM15      0.4589111
## + PCNM14      0.4535248
## + PCNM13      0.4511582
## + PCNM6       0.4305640
## + PCNM7       0.4261965
## + PCNM8       0.4224505
## + PCNM17      0.4181666
## + PCNM10      0.4154485
## + PCNM11      0.4112178
## + PCNM9       0.4111995
## + PCNM12      0.4087602
## <none>        0.4076020
## + PCNM4       0.3976526
##
##           Df      AIC      F Pr(>F)
## + PCNM1  1 41.411 4.057 0.008 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.4721469
## Call: fish.db ~ PCNM2 + PCNM3 + PCNM5 + PCNM1
##
##           R2.adjusted
## <All variables> 0.6260113
## + PCNM13      0.5212427
## + PCNM16      0.5208668
## + PCNM15      0.5161770
## + PCNM14      0.5147355
## + PCNM6       0.4999020
## + PCNM7       0.4936559
## + PCNM8       0.4904113
## + PCNM17      0.4856884
## + PCNM10      0.4835952
## + PCNM11      0.4760087
## + PCNM9       0.4751424
## + PCNM12      0.4747221
## <none>        0.4721469
## + PCNM4       0.4651218
##
##           Df      AIC      F Pr(>F)
## + PCNM13  1 39.346 3.4612 0.018 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.5212427
## Call: fish.db ~ PCNM2 + PCNM3 + PCNM5 + PCNM1 + PCNM13
##
##           R2.adjusted
## <All variables> 0.6260113
## + PCNM16      0.5767968
## + PCNM15      0.5715331
## + PCNM14      0.5698343
## + PCNM6       0.5475140

```

```

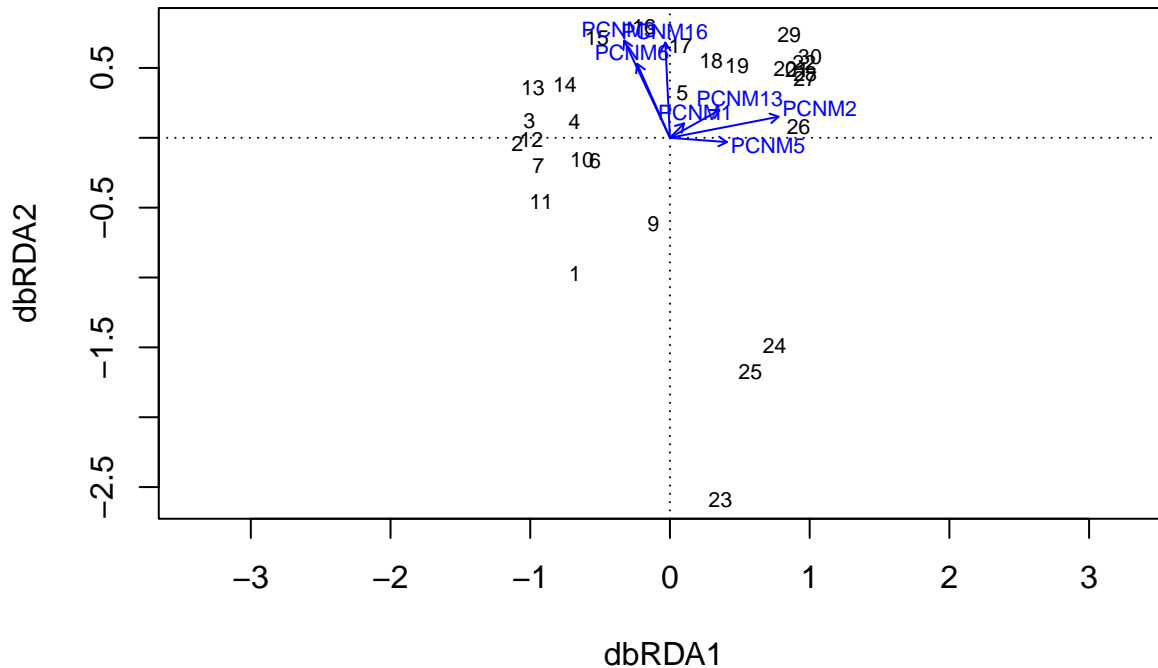
## + PCNM7          0.5392074
## + PCNM8          0.5379134
## + PCNM11         0.5281106
## + PCNM9          0.5267003
## + PCNM10         0.5265029
## + PCNM12         0.5255581
## <none>          0.5212427
## + PCNM17         0.5171800
## + PCNM4          0.5152311
##
##           Df    AIC      F Pr(>F)
## + PCNM16  1 36.48 4.0192 0.004 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.5767968
## Call: fish.db ~ PCNM2 + PCNM3 + PCNM5 + PCNM1 + PCNM13 + PCNM16
##
##           R2.adjusted
## <All variables>  0.6260113
## + PCNM6          0.6043089
## + PCNM8          0.5970286
## + PCNM12         0.5946888
## + PCNM7          0.5946475
## + PCNM9          0.5883735
## + PCNM10         0.5851333
## + PCNM15         0.5846468
## <none>          0.5767968
## + PCNM17         0.5748533
## + PCNM4          0.5733749
## + PCNM11         0.5711176
## + PCNM14         0.5652509
##
##           Df    AIC      F Pr(>F)
## + PCNM6  1 35.182 2.5296 0.034 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Step: R2.adj= 0.6043089
## Call: fish.db ~ PCNM2 + PCNM3 + PCNM5 + PCNM1 + PCNM13 + PCNM16 + PCNM6
##
##           R2.adjusted
## <All variables>  0.6260113
## + PCNM8          0.6248697
## + PCNM12         0.6208788
## + PCNM10         0.6170988
## + PCNM7          0.6142419
## + PCNM15         0.6140369
## + PCNM9          0.6107110
## <none>          0.6043089
## + PCNM17         0.6037430
## + PCNM11         0.5978305
## + PCNM4          0.5963667
## + PCNM14         0.5932113

```

```
##
##           Df      AIC      F Pr(>F)
## + PCNM8   1 34.219 2.151 0.084 .
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

#Because this is another dbRDA, we could visualize the biplot showing how each vector explains variation

```
plot(step.pcnm)
```



#The object 'step.pcnm' now contains the selected model.

```
step.pcnm$anova
```

```
##
##           R2.adj Df      AIC      F Pr(>F)
## + PCNM2       0.23537 1 49.574 9.6190 0.002 **
## + PCNM3       0.34293 1 46.083 5.4196 0.002 **
## + PCNM5       0.40760 1 43.941 3.8385 0.006 **
## + PCNM1       0.47215 1 41.411 4.0570 0.008 **
## + PCNM13      0.52124 1 39.346 3.4612 0.018 *
## + PCNM16      0.57680 1 36.480 4.0192 0.004 **
## + PCNM6       0.60431 1 35.182 2.5296 0.034 *
## <All variables> 0.62601
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

#We can now construct a spatial model using only the selected PCNM axes

```
space.mod <- model.matrix(~ PCNM2 + PCNM3 + PCNM5 + PCNM1 + PCNM13 + PCNM16 + PCNM6, doubles.space)[-1]
```

#First conduct constrained ordinations

```
doubs.total.env <- dbrda(fish.db ~ env.mod)
```

```
doubs.total.space <- dbrda(fish.db ~ space.mod)
```

#Next construct partial constrained ordinations

```
doubs.env.cond.space <- dbrda(fish.db ~ env.mod + Condition(space.mod))
```

```
doubs.space.cond.env <- dbrda(fish.db ~ space.mod + Condition(env.mod))
```



```
#Next test for significance of the dbRDA fractions  
permutest(doubs.env.cond.space, permutations = 999)
```

```
##  
## Permutation test for dbrda under reduced model  
##  
## Permutation: free  
## Number of permutations: 999  
##  
## Model: dbrda(formula = fish.db ~ env.mod + Condition(space.mod))  
## Permutation test for all constrained eigenvalues  
##           Df Inertia      F Pr(>F)  
## Model      3 0.85158 4.423  0.001 ***  
## Residual 18 1.15519  
## ---  
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
permutest(doubs.space.cond.env, permutations = 999)
```

```
##  
## Permutation test for dbrda under reduced model  
##  
## Permutation: free  
## Number of permutations: 999  
##  
## Model: dbrda(formula = fish.db ~ space.mod + Condition(env.mod))  
## Permutation test for all constrained eigenvalues  
##           Df Inertia      F Pr(>F)  
## Model      7 1.8752 4.1741  0.001 ***  
## Residual 18 1.1552  
## ---  
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
permutest(doubs.total.env, permutations = 999)
```

```
##  
## Permutation test for dbrda under reduced model  
##  
## Permutation: free  
## Number of permutations: 999  
##  
## Model: dbrda(formula = fish.db ~ env.mod)  
## Permutation test for all constrained eigenvalues  
##           Df Inertia      F Pr(>F)  
## Model      3 3.7317 10.262  0.001 ***  
## Residual 25 3.0304  
## ---  
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
permutest(doubs.total.space, permutations = 999)
```

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

```

## Number of permutations: 999
##
## Model: dbrda(formula = fish.db ~ space.mod)
## Permutation test for all constrained eigenvalues
##      Df Inertia      F Pr(>F)
## Model      7  4.7553 7.1089  0.001 ***
## Residual 21  2.0068
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

#Using the built-in varpart() function
doubts.varpart <- varpart(fish.db, env.mod, space.mod)
doubts.varpart

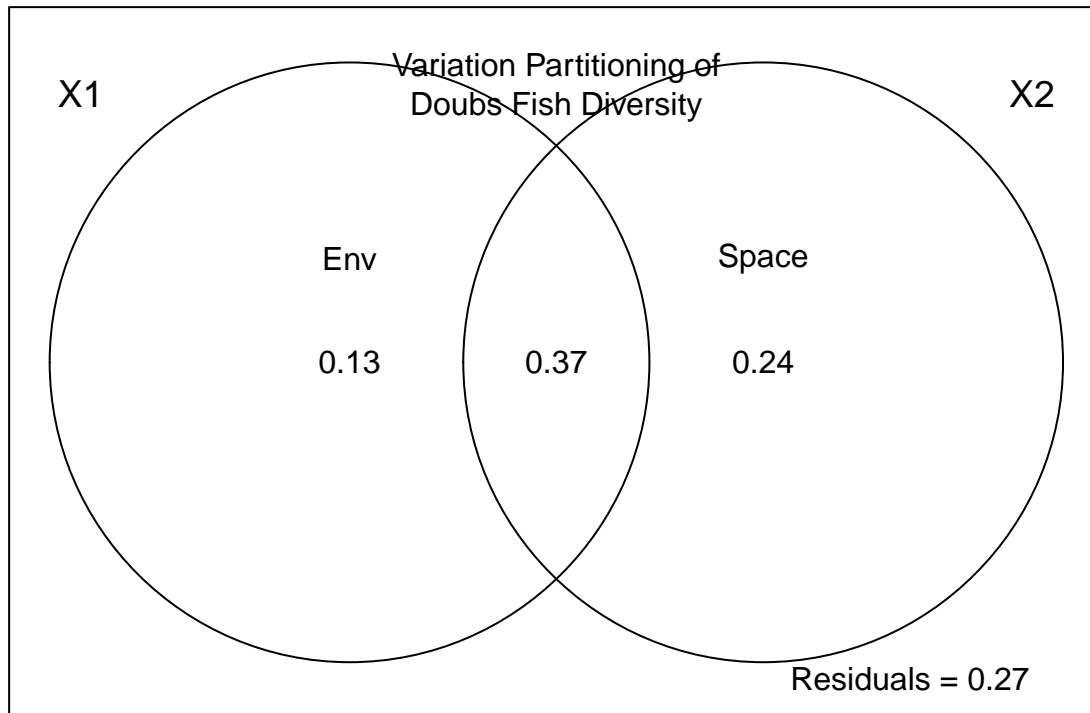
##
## Partition of squared Bray distance in dbRDA
##
## Call: varpart(Y = fish.db, X = env.mod, space.mod)
##
## Explanatory tables:
## X1:  env.mod
## X2:  space.mod
##
## No. of explanatory tables: 2
## Total variation (SS): 6.7621
## No. of observations: 29
##
## Partition table:
##


|                 | Df | R.squared | Adj.R.squared | Testable |
|-----------------|----|-----------|---------------|----------|
| [a+c] = X1      | 3  | 0.55186   | 0.49808       | TRUE     |
| [b+c] = X2      | 7  | 0.70323   | 0.60431       | TRUE     |
| [a+b+c] = X1+X2 | 10 | 0.82917   | 0.73426       | TRUE     |


## Individual fractions
## [a] = X1|X2      3      0.12995      TRUE
## [b] = X2|X1      7      0.23618      TRUE
## [c]              0      0.36813     FALSE
## [d] = Residuals      0.26574     FALSE
## ---
## Use function 'dbrda' to test significance of fractions of interest

par(mar = c(2,2,2,2))
plot(doubts.varpart)
text(1, 0.25, "Space")
text(0, 0.25, "Env")
mtext("Variation Partitioning of\nDoubts Fish Diversity", side = 3, line = -3)

```



Question 4: Interpret the variation partitioning results.

Answer 4: Here we see the percentage in which environment and space play in shaping fish diversity at Doubs. Space explains 24% of the diversity on its own, while Environment explains 13%. Combined they explain 37% of the variation as environmental conditions depend on space. Basically meaning that areas with similar conditions (thus similar space) are more likely to have similar diversity. Based on the residuals, we can see that 27% of variance seems unexplained in this particular case.

SYNTHESIS

Load the dataset from that you and your partner are using for the team project. Use one of the hypothesis-testing tools introduced in the beta diversity module. Interpret the findings of your data with respect to principles of biodiversity.

```
Projdata <- read.csv("/cloud/project/QB2025_Guevara/Week2-Alpha/MAT_fungal_abundances.csv")
str(Projdata)
```

```
## 'data.frame':   704 obs. of  12 variables:
## $ Plot          : chr  "MAT.B1.C" "MAT.B1.C" "MAT.B1.C" "MAT.B1.C" ...
## $ Block         : chr  "B1" "B1" "B1" "B1" ...
## $ Treatment     : chr  "Control" "Control" "Control" "Control" ...
## $ OTU           : chr  "X1" "X2" "X3" "X4" ...
## $ Species       : chr  "Alternaria_alternata" "Articulospora_tetracladia" "Cadophora_finlandica" ...
## $ Genus         : chr  "Alternaria" "Articulospora" "Cadophora" "Cenococcum" ...
## $ Family        : chr  "Pleosporaceae" "Helotiaceae" "Helotiaceae" "Gloniaceae" ...
## $ Order         : chr  "Pleosporales" "Helotiales" "Helotiales" "Mytilinidiales" ...
## $ Phylum      : chr  "Ascomycota" "Ascomycota" "Ascomycota" "Ascomycota" ...
## $ Relative.Abandance: num  0.0345 0 0 0.0345 0 ...
## $ EcM.density    : num  0.679 0.679 0.679 0.679 0.679 ...
## $ Absolute.Abandance: num  0.0234 0 0 0.0234 0 ...
```

```

library(dplyr)

##
## Attaching package: 'dplyr'
## The following objects are masked from 'package:stats':
##
##   filter, lag
## The following objects are masked from 'package:base':
##
##   intersect, setdiff, setequal, union

slim <- Projdata %>%
  select(Plot, Treatment, Species, Relative.Abandance)
#Changing data so that each species is a column, values are the relative.abundance, and rows are each p
library(tidyr)
ExpandProjdata <- slim %>%
  pivot_wider(names_from = Species, values_from = Relative.Abandance, values_fill = 0)
Sorted_ExpandProjdata <- ExpandProjdata %>%
  arrange(Treatment)

#Creating separate vector for plots/treatments and creating new data table that is only numerical
Fungal.treatment <- Sorted_ExpandProjdata[,1]
Rel.abun.fungal <- Sorted_ExpandProjdata [,3:46]
#Fungal.treatment is apparently invalid
str(Sorted_ExpandProjdata$Fungal.treatment)

## Warning: Unknown or uninitialised column: `Fungal.treatment`.
## NULL

#Setting Fungal.treatment as a factor
Fungal.treatment_factor <- as.factor(unlist(Fungal.treatment))
adonis2(Rel.abun.fungal ~ Fungal.treatment_factor, method = "bray", permutations = 999)

## No residual component
##
## adonis2(formula = Rel.abun.fungal ~ Fungal.treatment_factor, permutations = 999, method = "bray")
##           Df SumOfSqs R2 F Pr(>F)
## Model      15  4.6817  1
## Residual    0  0.0000  0
## Total      15  4.6817  1

#I end up with a strange result with no p-value and zero residuals which seems rather odd.
length(unique(Fungal.treatment_factor))

## [1] 16

```

I'm not sure I can interpret these findings as they don't seem to be coming out right for some reason. I've set my treatments/rows as a factor, I removed any non-numeric data from the matrix in order to perform the adonis2 function, but it still is coming out like this. Perhaps it is because the values are relative abundances and thus between 0 and 1 which is affecting the overall PERMANOVA? I'm unsure, I will discuss with Jay and Emma on Friday.