8. 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, it is *imperative* that you **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 '8.BetaDiversity' folder.
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 (**8.BetaDiversity_2_Worksheet.Rmd**) with all code blocks filled out and questions answered) and the PDF output of `Knitr` (**8.BetaDiversity_2_Worksheet.pdf**).

The completed exercise is due on **Wednesday, April 23rd, 2021 before 09:00 AM**.

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 "*/8.BetaDiversity*" folder, and

4. load the **vegan** R package (be sure to install if needed).

```
rm(list = ls())
getwd()
```

```
## [1] "C:/Users/joshu/quantbio/QB2021_Jones/2.Worksheets/8.BetaDiversity"
```

```
library(vegan)
```

```
## Warning: package 'vegan' was built under R version 4.0.4
```

```
## Loading required package: permute
```

```
## Warning: package 'permute' was built under R version 4.0.4
```

```
## Loading required package: lattice
```

```
## This is vegan 2.5-7
```

2) LOADING DATA

Load dataset

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

```
# note, pleae do not print the dataset when submitting
library('indicspecies')
```

```
## Warning: package 'indicspecies' was built under R version 4.0.5
```

```
library(ade4)
```

```
## Warning: package 'ade4' was built under R version 4.0.5
```

```
data("doubs")

#Importing site-by-species matrix from list
fish <- doubs$fish

#Removing all rows with a total of 0 total observations
fish <-fish[rowSums(fish !=0) > 0,]
```

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.

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

#Run PERMANOVA with adonis function
adonis(fish ~ quality, method = "bray", permutations = 999)
```

```
##
## Call:
## adonis(formula = fish ~ quality, permutations = 999, method = "bray")
##
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##           Df SumsOfSqs MeanSqs F.Model      R2 Pr(>F)
## quality    2    3.0947 1.54733   10.97 0.45765  0.001 ***
## Residuals 26    3.6674 0.14105         0.54235
## Total     28    6.7621                 1.00000
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
#Indicator Value
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   0.016 *
##
##  Group HQ+MQ  #sps.  2
##       stat p.value
## Satr 0.860   0.004 **
## Phph 0.859   0.009 **
##
##  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   0.001 ***
## Cyca 0.866   0.001 ***
## Acce 0.866   0.001 ***
## Lele 0.863   0.006 **
## Titi 0.853   0.007 **
## Chto 0.829   0.002 **
## Rham 0.829   0.001 ***
## Anan 0.829   0.001 ***
## Eslu 0.827   0.026 *
## Pefl 0.806   0.021 *
## Blbj 0.791   0.002 **
## Scer 0.766   0.006 **
## Abbr 0.750   0.005 **
## Icme 0.661   0.023 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

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

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***: From these analysis there seems to be a significant and consistent relationship between habitat quality and fish diversity. The Pearson correlation, which provides relationships between individual species and the habitat quality, does seem to somewhat match what we saw in the figures from last week, except that there seems to be more spies associated with low quality environments than was observed in the PCoA.

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.

•

```
# Creating 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%
## 0.0911 0.1354 0.1716 0.1983
## 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***: That there is an overall correlation between differences in environmental condition and fish diversity, or as environmental condition changes along the river the fish community also seems to change. This supports the hypothesis that stream quality influences fish communities.

ii. Constrained Ordination

In the R code chunk below, do the following:

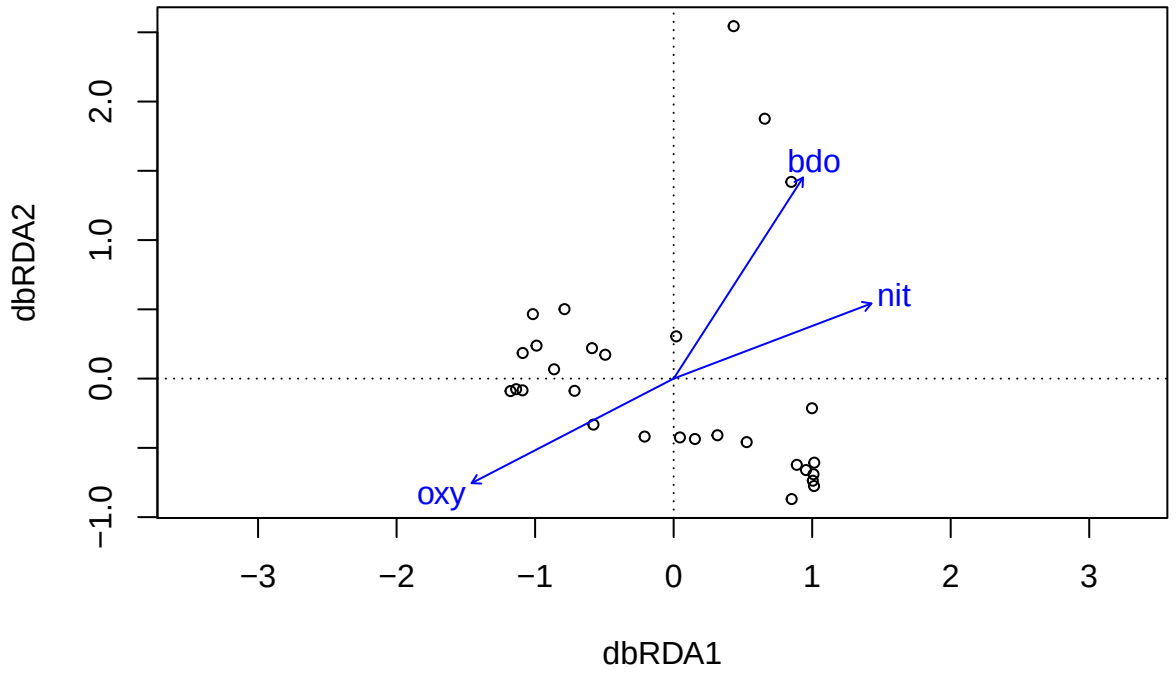
1. create an environmental matrix of the water chemistry data included in the `doubs` dataset using forward and reverse selection of variables,
2. conduct a redundancy analysis on the fish assemblages of the Doubs River,
3. use a permutation test to determine the significance of the constrained analysis,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

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

#selecting for variables
fish.db <- vegdist(fish, method = "bray")
doubs.dbrda.mod0 <- dbrda(fish.db ~ 1, as.data.frame(env.chem))
doubs.dbrda.mod1 <- dbrda(fish.db ~ ., as.data.frame(env.chem))
doubs.dbrda <- ordiR2step(doubs.dbrda.mod0, doubs.dbrda.mod1, perm.max = 999)
```

```
## 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
```

```
#Performing distance-based Redundancy Analysis (dbRDA)
ordiplot(doubs.dbrda)
```



```r
#Permutation test for significance and correlation
permutest(doubs.dbrda, permutations = 999)
```

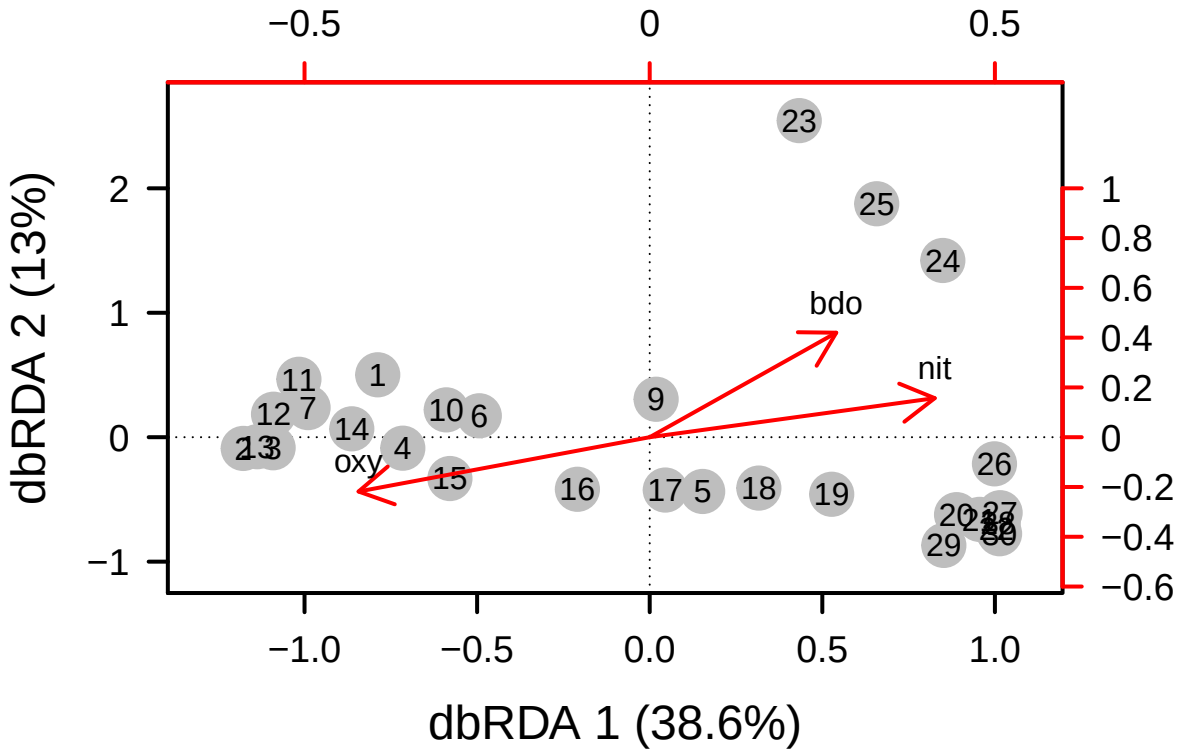
```
##
## Permutation test for dbrda under reduced model
##
## Permutation: free
## Number of permutations: 999
##
## Model: dbrda(formula = fish.db ~ oxy + bdo + nit, data =
## as.data.frame(env.chem))
## Permutation test for all constrained eigenvalues
##          Df Inertia      F Pr(>F)
## Model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

```r
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
```

```r
#Calculating explained variation on axes
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

#Plotting constrained ordination results
par(mar = c(5,5,4,4) + .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")))

#Plotting vectors for enfluence of environmental factors
vectors <- scores(doubs.dbrda, display = "bp")
arrows(0, 0, vectors[,1], vectors[,2],
       lwd = 2, lty = 1, length = .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***: All of the environmental variables seem to have strong contributions with fish community structure, especially oxtgen, nitrogren, and the variables within 'bdo'. pH and hardness of water seem to have a small effect far less than these other variables.

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.

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

#Matrix model of spacial data
rs <- rowSums(fish)/sum(fish)
doubs.pcnmw <- pcnm(dist(doubs$xy[-8,]), w = rs, dist.ret = T)
doubs.pcnmw$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
```

```
doubs.space <- as.data.frame(scores(doubs.pcnmw))
doubs.pcnm.mod0 <- dbrda(fish.db ~ 1, doubs.space)
doubs.pcnm.mod1 <- dbrda(fish.db ~ ., doubs.space)
step.pcnm <- ordiR2step(doubs.pcnm.mod0, doubs.pcnm.mod1, perm.max = 999)
```

```
## 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  0.008 **
## ---
## 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  0.004 **
## ---
## 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  0.016 *
## ---
## 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  0.022 *
## ---
## 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  0.058 .
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

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

```r
#Performing constrained and partial constrained ordinations using spatial and environmental models
doubs.total.env <- dbrda(fish.db ~ env.mod)
doubs.total.space <- dbrda(fish.db ~ space.mod)

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


#Permutations to test for significance
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
```

```r
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
```

```r
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
```

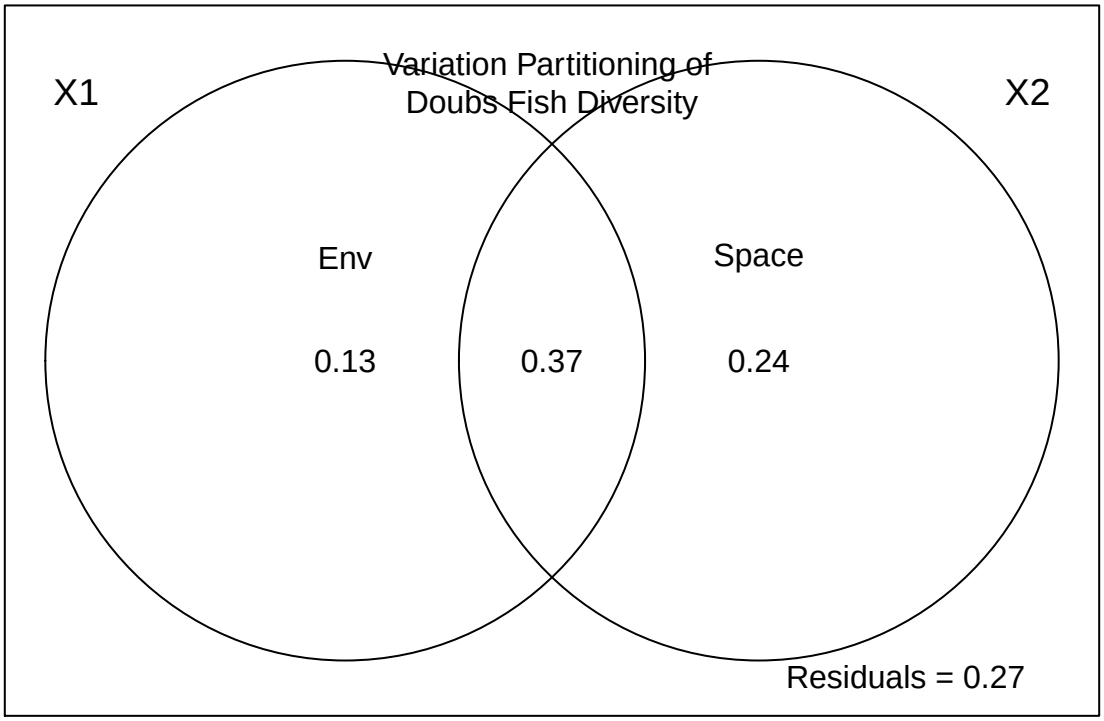
```
permutest(doubs.total.space, permutations = 999)
```

```
##
## 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
```

```
#Partitioning drivers of variation
doubs.varpart <- varpart(fish.db, env.mod, space.mod)
doubs.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+b] = 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]                    0                 0.36813    FALSE
## [c] = X2|X1            7                 0.23618     TRUE
## [d] = Residuals                          0.26574    FALSE
## ---
## Use function 'dbrda' to test significance of fractions of interest
```

```r
#Visualizing
par(mar = c(2,2,2,2))
plot(doubs.varpart)
text(1, .25, "Space")
text(0, .25, "Env")
mtext("Variation Partitioning of \nDoubs Fish Diversity", side = 3, line = -3)
```



Question 4: Interpret the variation partitioning results.

> ***Answer 4***: The majority of the explained variation in fish diversity is from environmental variation across space, followed by space, with environment having the least power in explaining variation. But there is a large portion of the variance not explained by any of these variables.

SYNTHESIS

As in the previous worksheet, use the `mobsim` package from the DataWrangling module to simulate two local communities each containing 1000 individuals (N) and 25 species (S), but with one having a random spatial distribution and the other having a patchy spatial distribution. Take ten (10) subsamples from each site using the quadrat function and answer the following questions:

1) Perform a PERMANOVA to test whether or not the spatial distribution of species affects species composition.

```
library(mobsim)
```

```
## Warning: package 'mobsim' was built under R version 4.0.4
```

```
# simulate community with `mobsim`
com1 <- sim_poisson_community(s_pool = 25, n_sim = 1000, sad_type = "lnorm",
        sad_coef = list("meanlog" = 2, "sdlog" = 1))

# Lay down sampling quadrats on the community
comm_mat1 <- sample_quadrats(com1, n_quadrats = 10, quadrat_area = 0.01,
             method = "random", avoid_overlap = T)
```

```
# Rename sampled areas as quadrats
quads <- c("site1", "site2", "site3", "site4", "site5", "site6", "site7",
           "site8", "site9", "site10")
row.names(comm_mat1$xy_dat) <- quads
row.names(comm_mat1$spec_dat) <- quads
randdots <- comm_mat1$spec_dat

# simulate community with `mobsim`
com2 <- sim_thomas_community(s_pool = 25, n_sim = 1000, sad_type = "lnorm",
        sad_coef = list("meanlog" = 2, "sdlog" = 1))

# Lay down sampling quadrats on the community
comm_mat2 <- sample_quadrats(com2, n_quadrats = 10, quadrat_area = 0.01,
             method = "random", avoid_overlap = T)
```

```
# Rename sampled areas as quadrats
quads <- c("site1", "site2", "site3", "site4", "site5", "site6", "site7",
           "site8", "site9", "site10")
row.names(comm_mat2$xy_dat) <- quads
row.names(comm_mat2$spec_dat) <- quads
patchdots <- comm_mat2$spec_dat

dots <- rbind(randdots, patchdots)
distribution <- c(rep("random", 10), rep("patchy", 10))

#PERMANOVA
adonis(dots ~ distribution, method = "bray", permutations = 999)
```

```
##
## Call:
## adonis(formula = dots ~ distribution, permutations = 999, method = "bray")
##
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##              Df SumsOfSqs MeanSqs F.Model      R2 Pr(>F)
## distribution  1    0.7506 0.75058  2.1599 0.10714  0.014 *
## Residuals    18    6.2550 0.34750         0.89286
```

```
## Total        19    7.0056                 1.00000
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

2) Load the dataset you are using for your Team Project. Perform an ordination to visualize your dataset. Using this ordination, develop some hypotheses relevant to β-diversity. Use a statistic to test one of these hypotheses. Succinctly explain the finding and its relevance to your system.

Importing data into R

```
site_species <- read.csv("OTU_table.csv", header = TRUE)
site_species.t <- t(site_species)
```

#Rarefaction

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

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

```
#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,])
  }
}
```

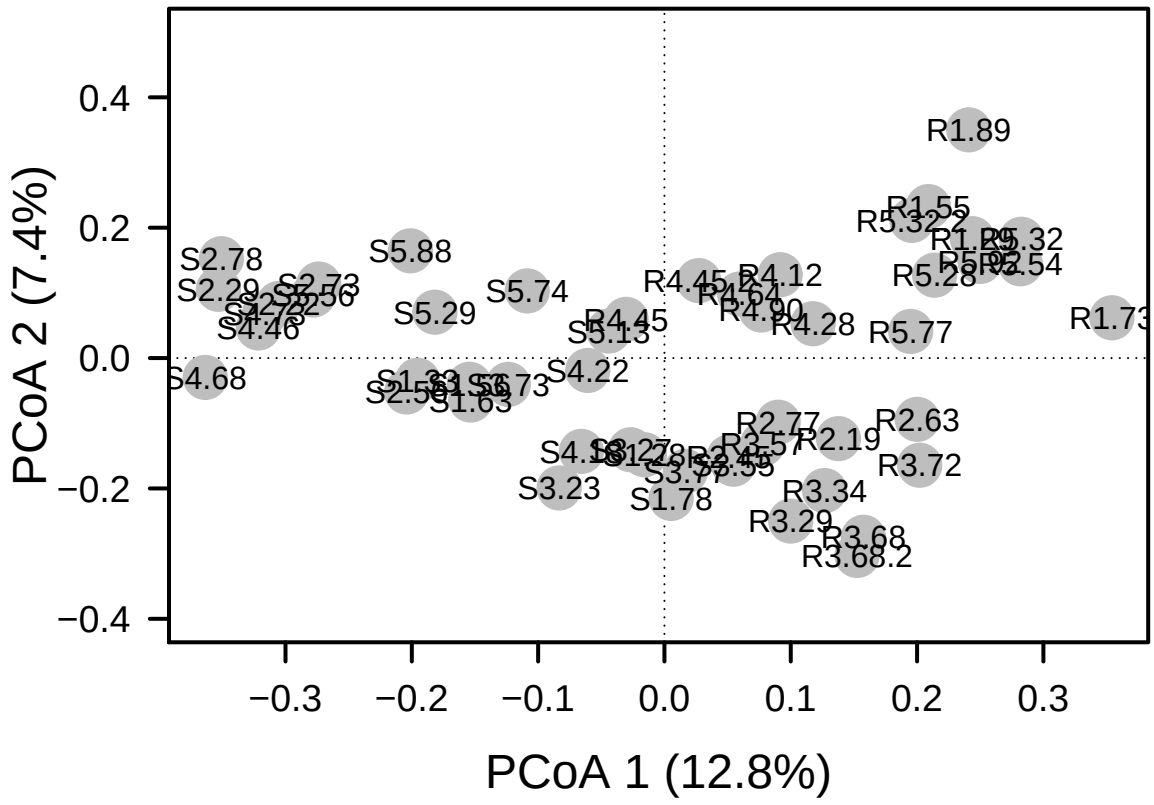
#Calculating Sorensen Beta-Diversity

```
fungalBC <- vegdist(rarefied_site_species, method = "bray", binary = "TRUE")
```

PCoA of Fungal Communities

```
fungal.pcoa <- cmdscale(fungalBC, eig = TRUE, k = 3)

explainvar1 <- round(fungal.pcoa$eig[1]/sum(fungal.pcoa$eig), 3) * 100
explainvar2 <- round(fungal.pcoa$eig[2]/sum(fungal.pcoa$eig), 3) * 100
explainvar3 <- round(fungal.pcoa$eig[3]/sum(fungal.pcoa$eig), 3) * 100
sum.eig <- sum(explainvar1, explainvar2, explainvar3)

#Define Plot Parameters
par(mar = c(5,5,1,2), .1)
```

```
#Initiate Plot
plot(fungal.pcoa$points[,1], fungal.pcoa$points[,2], ylim = c(-.4, .5),
     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)

#Add axis
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)

#Add Points & Labels
points(fungal.pcoa$points[,1], fungal.pcoa$points[,2],
       pch = 19, cex = 3, bg = "gray", col = "gray")
text(fungal.pcoa$points[,1], fungal.pcoa$points[,2],
     labels = row.names(fungal.pcoa$points))
```



Hypothesis: The taxa found within the site can be clearly defined based on the location of the sampling site (Ridge (R) vs Snowbed (S))

#Hypothesis testing

```
location <- c(rep("Snowbed", 26), rep("Ridge", 29))

adonis(site_species.r ~ location, binary = TRUE, permutations = 999)
```

```
##
## Call:
## adonis(formula = site_species.r ~ location, permutations = 999,      binary = TRUE)
##
## Permutation: free
## Number of permutations: 999
##
## Terms added sequentially (first to last)
##
##           Df SumsOfSqs MeanSqs F.Model      R2 Pr(>F)
## location   1    1.6082 1.60815  5.6823 0.09683  0.001 ***
## Residuals 53   14.9996 0.28301         0.90317
## Total     54   16.6078                 1.00000
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Conclusion: Location of sampling site does significantly account for a portion of the differences in taxa across samples but the majority of the differences are still within the risiduals. At this point I would hypothesize that a lot of the differences across samples will also be as a result of different sites and different plant populations within the sites.

```
sessionInfo()
```

```
## R version 4.0.3 (2020-10-10)
## Platform: x86_64-w64-mingw32/x64 (64-bit)
## Running under: Windows 10 x64 (build 19042)
##
## Matrix products: default
##
## locale:
## [1] LC_COLLATE=English_United States.1252
## [2] LC_CTYPE=English_United States.1252
## [3] LC_MONETARY=English_United States.1252
## [4] LC_NUMERIC=C
## [5] LC_TIME=English_United States.1252
##
## attached base packages:
## [1] stats     graphics  grDevices utils     datasets  methods   base
##
## other attached packages:
## [1] mobsim_0.2.0       ade4_1.7-16        indicspecies_1.7.9 vegan_2.5-7
## [5] lattice_0.20-41    permute_0.9-5
##
## loaded via a namespace (and not attached):
##  [1] Rcpp_1.0.6        knitr_1.31        cluster_2.1.0     magrittr_2.0.1
##  [5] hms_1.0.0         splines_4.0.3     progress_1.2.2    MASS_7.3-53
##  [9] R6_2.5.0          rlang_0.4.10      highr_0.8         stringr_1.4.0
## [13] tools_4.0.3       parallel_4.0.3    grid_4.0.3        nlme_3.1-149
```

```
## [17] mgcv_1.8-33       xfun_0.20         ellipsis_0.3.1    htmltools_0.5.1.1
## [21] yaml_2.2.1        digest_0.6.27     lifecycle_1.0.0   crayon_1.4.1
## [25] Matrix_1.2-18     vctrs_0.3.6       evaluate_0.14     rmarkdown_2.6
## [29] stringi_1.5.3     compiler_4.0.3    prettyunits_1.1.1 pkgconfig_2.0.3
```