6. Worksheet: Among Site (Beta) Diversity – Part 1

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OVERVIEW

In this worksheet, we move beyond the investigation of within-site α-diversity. We will explore β-diversity, which is defined as the diversity that occurs among sites. This requires that we examine the compositional similarity of assemblages that vary in space or time.

After completing this exercise you will know how to:

1. formally quantify β-diversity
2. visualize β-diversity with heatmaps, cluster analysis, and ordination
3. 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, **push** this file to your GitHub repo.
6. For the assignment portion of the worksheet, follow the directions at the bottom of this file.
7. When you are done, **Knit** the text and code into a PDF file.
8. After Knitting, submit the completed exercise by creating a **pull request** via GitHub. Your pull request should include this file (**6.BetaDiversity_1_Worksheet.Rmd**) with all code blocks filled out and questions answered) and the PDF output of `Knitr` (**6.BetaDiversity_1_Worksheet.pdf**).

The completed exercise is due on **Wednesday, February 5th, 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, please provide the code to:

1) Clear your R environment,
2) Print your current working directory,
3) Set your working directory to your `Week3-Beta/` folder folder, and
4) Load the `vegan` R package (be sure to install first if you have not already).

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

```
## [1] "/cloud/project/QB2025_Yoo/Week3-Beta"
```

```
setwd("/cloud/project/QB2025_Yoo/Week3-Beta")

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

```
## This is vegan 2.6-8
```

```
##
## Attaching package: 'gplots'
```

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

2) LOADING DATA

Load dataset

In the R code chunk below, do the following:

1. load the `doubs` dataset from the `ade4` package, and
2. explore the structure of the dataset.

```
# note, please do not print the dataset when submitting
data(doubs)
#str(doubs, max.level = 1)
#head(doubs$env)
#head(doubs$fish)
```

Question 1: Describe some of the attributes of the `doubs` dataset.

a. How many objects are in `doubs`?
b. How many fish species are there in the `doubs` dataset?
c. How many sites are in the `doubs` dataset?

Answer 1a: There are four objects. ***Answer 1b***: 27 species ***Answer 1c***: 30 sites (observations)

Visualizing the Doubs River Dataset

Question 2: Answer the following questions based on the spatial patterns of richness (i.e., α-diversity) and Brown Trout (*Salmo trutta*) abundance in the Doubs River.

a. How does fish richness vary along the sampled reach of the Doubs River?
b. How does Brown Trout (*Salmo trutta*) abundance vary along the sampled reach of the Doubs River?
c. What do these patterns say about the limitations of using richness when examining patterns of biodiversity?

Answer 2a: Downstream sites have greater fish richness than upstream sites. ***Answer 2b***: Downstream sites have less fish richness than upstream sites. ***Answer 2c***: The species richness data does not contain the composition of the community and abundance of each species.

3) QUANTIFYING BETA-DIVERSITY

In the R code chunk below, do the following:

1. write a function (`beta.w()`) to calculate Whittaker's β-diversity (i.e., β_w) that accepts a site-by-species matrix with optional arguments to specify pairwise turnover between two sites, and
2. use this function to analyze various aspects of β-diversity in the Doubs River.

```
beta.w <- function(site.by.species = "", sitenum1 = "", sitenum2 = "", pairwise = TRUE) {
  # ONLY if we specify pairwise as TRUE, do this:
  if(pairwise == TRUE){
    # As a check, let's print an error if we do not provide needed arguments
    if (sitenum1 == "" | sitenum2 == "") {
    print("Error: please specify sites to compare")
    return(NA)}
  # If our function made it this far, let us calculate pairwise beta diversity
    site1 = site.by.species[sitenum1, ]
    site2 = site.by.species[sitenum2, ]
    site1 = subset(site1, select = site1 > 0) # Removes absences
    site2 = subset(site2, select = site2 > 0) # Removes absences
    gamma = union(colnames(site1), colnames(site2)) # Gamma species pool
    s = length(gamma)  # Gamma richness
    a.bar = mean(c(specnumber(site1), specnumber(site2))) # Mean sample richness
    b.w = round(s/a.bar - 1, 3)  # Calculate pairwise beta diversity
return(b.w)
}
  #OTHERWISE pairwise defaults to FALSE, so do this, like before:
  else{
    SbyS.pa <- decostand(site.by.species, method = "pa") # Convert to presence-absence
    S <- ncol(SbyS.pa[, which(colSums(SbyS.pa) > 0)]) # Number of species in the region
    a.bar <- mean(specnumber(SbyS.pa)) # Average richness at each site
    b.w <- round(S/a.bar, 3) # Round to 3 decimal places
    return(b.w)
  }
}

print(beta.w(doubs$fish, 1, 2))
```

```
## [1] 0.5
```

```
print(beta.w(doubs$fish, 1, 10))
```

```
## [1] 0.714
```

Question 3: Using your `beta.w()` function above, answer the following questions:

a. Describe how local richness (α) and turnover (β) contribute to regional (γ) fish diversity in the Doubs.
b. Is the fish assemblage at site 1 more similar to the one at site 2 or site 10?
c. Using your understanding of the equation $\beta_w = \gamma/\alpha$, how would your interpretation of β change if we instead defined beta additively (i.e., $\beta = \gamma - \alpha$)?

Answer 3a: α and β positively contribute to γ. When α or/and β increase, γ increase. ***Answer 3b***: The fish assamblage at site 1 is more similar to site 2 (Whittaker's $\beta = 0.5$) than to site 10 (Whittaker's $\beta = 0.714$). ***Answer 3c***: β will mean the number of species in region subtracted by local diversity instead of relative relationship between # of species in region and a local site.

The Resemblance Matrix

In order to quantify β-diversity for more than two samples, we need to introduce a new primary ecological data structure: the **Resemblance Matrix**.

Question 4: How do incidence- and abundance-based metrics differ in their treatment of rare species?

> ***Answer 4***: Incidence-based matrics treat rare species same as abundant species, while abundacne-based metrics give less weight to rare species by using abundance of each species when calculating the metrics.

In the R code chunk below, do the following:

1. make a new object, `fish`, containing the fish abundance data for the Doubs River,
2. remove any sites where no fish were observed (i.e., rows with sum of zero),
3. construct a resemblance matrix based on Sørensen's Similarity ("fish.ds"), and
4. construct a resemblance matrix based on Bray-Curtis Distance ("fish.db").

```
fish <- doubs$fish
fish <- fish[-8, ]  # Remove site 8 from data

# Calculate Jaccard
fish.dj <- vegdist(fish, method = "jaccard", binary = TRUE)

# Calculate Bray-Curtis
fish.db <- vegdist(fish, method = "bray", diag = TRUE)

# Calculate Sørensen
fish.ds <- vegdist(fish, method = "bray", binary = TRUE, diag = TRUE)

fish.db
```

```
##            1          2          3          4          5          6          7
## 1  0.00000000
## 2  0.60000000 0.00000000
## 3  0.68421053 0.14285714 0.00000000
## 4  0.75000000 0.33333333 0.18918919 0.00000000
## 5  0.89189189 0.69565217 0.68000000 0.49090909 0.00000000
## 6  0.75000000 0.39393939 0.29729730 0.19047619 0.41818182 0.00000000
## 7  0.68421053 0.14285714 0.12500000 0.24324324 0.64000000 0.24324324 0.00000000
## 9  1.00000000 0.69230769 0.73333333 0.65714286 0.58333333 0.54285714 0.66666667
## 10 0.88235294 0.38461538 0.40000000 0.37142857 0.54166667 0.25714286 0.26666667
## 11 0.57142857 0.30434783 0.40740741 0.43750000 0.68888889 0.43750000 0.33333333
## 12 0.71428571 0.20000000 0.23529412 0.33333333 0.69230769 0.38461538 0.17647059
## 13 0.72727273 0.29032258 0.31428571 0.45000000 0.73584906 0.55000000 0.37142857
## 14 0.80645161 0.40000000 0.31818182 0.34693878 0.67741935 0.42857143 0.36363636
## 15 0.83333333 0.51111111 0.46938776 0.40740741 0.55223881 0.37037037 0.38775510
## 16 0.86046512 0.65384615 0.57142857 0.47540984 0.45945946 0.37704918 0.53571429
## 17 0.91489362 0.67857143 0.63333333 0.50769231 0.51282051 0.44615385 0.60000000
## 18 0.95555556 0.74074074 0.72413793 0.58730159 0.50000000 0.52380952 0.68965517
## 19 1.00000000 0.79310345 0.70967742 0.61194030 0.50000000 0.52238806 0.67741935
## 20 1.00000000 0.91176471 0.88888889 0.74025974 0.48888889 0.68831169 0.86111111
## 21 1.00000000 0.94594595 0.92307692 0.78313253 0.50000000 0.73493976 0.89743590
## 22 1.00000000 0.97619048 0.95454545 0.82795699 0.52830189 0.78494624 0.93181818
## 23 1.00000000 1.00000000 1.00000000 0.92000000 0.89473684 0.84000000 0.90000000
## 24 1.00000000 1.00000000 1.00000000 0.88888889 0.79591837 0.77777778 0.93548387
## 25 1.00000000 1.00000000 0.92592593 0.81250000 0.68888889 0.68750000 0.85185185
```

```
## 26 1.00000000 0.96363636 0.93220339 0.78125000 0.55844156 0.68750000 0.89830508
## 27 1.00000000 0.97333333 0.94936709 0.83333333 0.56701031 0.76190476 0.92405063
## 28 1.00000000 0.97560976 0.95348837 0.82417582 0.57692308 0.78021978 0.93023256
## 29 0.97777778 0.93939394 0.92233010 0.81481481 0.53719008 0.77777778 0.90291262
## 30 1.00000000 1.00000000 0.98095238 0.87272727 0.59349593 0.83636364 0.96190476
##             9         10         11         12         13         14         15
## 1
## 2
## 3
## 4
## 5
## 6
## 7
## 9  0.00000000
## 10 0.57142857 0.00000000
## 11 0.76000000 0.44000000 0.00000000
## 12 0.68750000 0.37500000 0.24137931 0.00000000
## 13 0.81818182 0.57575758 0.33333333 0.18918919 0.00000000
## 14 0.76190476 0.47619048 0.43589744 0.21739130 0.19148936 0.00000000
## 15 0.65957447 0.40425532 0.50000000 0.33333333 0.38461538 0.24590164 0.00000000
## 16 0.70370370 0.51851852 0.64705882 0.55172414 0.59322034 0.44117647 0.26027397
## 17 0.68965517 0.51724138 0.63636364 0.58064516 0.61904762 0.50000000 0.40259740
## 18 0.64285714 0.57142857 0.69811321 0.66666667 0.70491803 0.60000000 0.46666667
## 19 0.66666667 0.63333333 0.82456140 0.75000000 0.81538462 0.67567568 0.56962025
## 20 0.68571429 0.77142857 0.91044776 0.89189189 0.92000000 0.83333333 0.70786517
## 21 0.76315789 0.81578947 0.91780822 0.92500000 0.95061728 0.86666667 0.76842105
## 22 0.76744186 0.86046512 0.95180723 0.95555556 0.97802198 0.90000000 0.77142857
## 23 0.77777778 0.88888889 0.86666667 0.90909091 1.00000000 0.93750000 0.94594595
## 24 0.72413793 0.79310345 0.92307692 0.93939394 1.00000000 0.90697674 0.87500000
## 25 0.84000000 0.76000000 0.90909091 0.93103448 1.00000000 0.84615385 0.81818182
## 26 0.71929825 0.82456140 0.92592593 0.93442623 0.96774194 0.85915493 0.76315789
## 27 0.76623377 0.84415584 0.94594595 0.95061728 0.97560976 0.89010989 0.77083333
## 28 0.76190476 0.85714286 0.95061728 0.95454545 0.97752809 0.89795918 0.78640777
## 29 0.78217822 0.84158416 0.89795918 0.90476190 0.90566038 0.84347826 0.73333333
## 30 0.84466019 0.90291262 0.98000000 0.98130841 1.00000000 0.93162393 0.81967213
##            16         17         18         19         20         21         22
## 1
## 2
## 3
## 4
## 5
## 6
## 7
## 9
## 10
## 11
## 12
## 13
## 14
## 15
## 16 0.00000000
## 17 0.26190476 0.00000000
## 18 0.34146341 0.13953488 0.00000000
## 19 0.39534884 0.31111111 0.25000000 0.00000000
```

```
## 20 0.58333333 0.42000000 0.32653061 0.23529412 0.00000000
## 21 0.62745098 0.49056604 0.40384615 0.29629630 0.10169492 0.00000000
## 22 0.66071429 0.55172414 0.47368421 0.38983051 0.18750000 0.10447761 0.00000000
## 23 0.90909091 0.83333333 0.82608696 0.84000000 0.86666667 0.87878788 0.89473684
## 24 0.81818182 0.69491525 0.64912281 0.63934426 0.57746479 0.61038961 0.65517241
## 25 0.76470588 0.74545455 0.66037736 0.61403509 0.67164179 0.69863014 0.73493976
## 26 0.63855422 0.54022989 0.45882353 0.32584270 0.21212121 0.20000000 0.25217391
## 27 0.66990291 0.57009346 0.48571429 0.37614679 0.19327731 0.13600000 0.12592593
## 28 0.69090909 0.57894737 0.50000000 0.41379310 0.22222222 0.16666667 0.12676056
## 29 0.65354331 0.51145038 0.44186047 0.41353383 0.24475524 0.18120805 0.11949686
## 30 0.72093023 0.57894737 0.52671756 0.48148148 0.29655172 0.23178808 0.18012422
##            23         24         25         26         27         28         29
## 1
## 2
## 3
## 4
## 5
## 6
## 7
## 9
## 10
## 11
## 12
## 13
## 14
## 15
## 16
## 17
## 18
## 19
## 20
## 21
## 22
## 23 0.00000000
## 24 0.57894737 0.00000000
## 25 0.46666667 0.46153846 0.00000000
## 26 0.82978723 0.48275862 0.59259259 0.00000000
## 27 0.88059701 0.61538462 0.70270270 0.18867925 0.00000000
## 28 0.89189189 0.64705882 0.72839506 0.23893805 0.09774436 0.00000000
## 29 0.91208791 0.70588235 0.77551020 0.33846154 0.18666667 0.14649682 0.00000000
## 30 0.91397849 0.71153846 0.78000000 0.36363636 0.19736842 0.15723270 0.14772727
##            30
## 1
## 2
## 3
## 4
## 5
## 6
## 7
## 9
## 10
## 11
## 12
## 13
```

```
## 14
## 15
## 16
## 17
## 18
## 19
## 20
## 21
## 22
## 23
## 24
## 25
## 26
## 27
## 28
## 29
## 30 0.00000000
```

```
fish.ds
```

```
##            1          2          3          4          5          6          7
## 1  0.00000000
## 2  0.50000000 0.00000000
## 3  0.60000000 0.14285714 0.00000000
## 4  0.77777778 0.45454545 0.33333333 0.00000000
## 5  0.83333333 0.57142857 0.46666667 0.15789474 0.00000000
## 6  0.81818182 0.53846154 0.42857143 0.11111111 0.04761905 0.00000000
## 7  0.66666667 0.25000000 0.33333333 0.38461538 0.37500000 0.33333333 0.00000000
## 9  1.00000000 0.50000000 0.55555556 0.38461538 0.37500000 0.33333333 0.40000000
## 10 0.71428571 0.33333333 0.40000000 0.28571429 0.29411765 0.25000000 0.09090909
## 11 0.71428571 0.33333333 0.40000000 0.42857143 0.52941176 0.50000000 0.27272727
## 12 0.71428571 0.33333333 0.40000000 0.42857143 0.52941176 0.50000000 0.27272727
## 13 0.71428571 0.33333333 0.40000000 0.57142857 0.64705882 0.62500000 0.45454545
## 14 0.81818182 0.53846154 0.42857143 0.33333333 0.42857143 0.40000000 0.46666667
## 15 0.83333333 0.57142857 0.60000000 0.36842105 0.36363636 0.33333333 0.37500000
## 16 0.88888889 0.70000000 0.61904762 0.36000000 0.28571429 0.25925926 0.54545455
## 17 0.91304348 0.76000000 0.69230769 0.46666667 0.39393939 0.37500000 0.62962963
## 18 0.91666667 0.76923077 0.70370370 0.48387097 0.41176471 0.39393939 0.64285714
## 19 1.00000000 0.84615385 0.77777778 0.54838710 0.41176471 0.45454545 0.71428571
## 20 1.00000000 0.84000000 0.76923077 0.53333333 0.39393939 0.43750000 0.70370370
## 21 1.00000000 0.84615385 0.77777778 0.54838710 0.41176471 0.45454545 0.71428571
## 22 1.00000000 0.92000000 0.84615385 0.60000000 0.45454545 0.50000000 0.77777778
## 23 1.00000000 1.00000000 1.00000000 0.81818182 0.71428571 0.69230769 0.75000000
## 24 1.00000000 1.00000000 1.00000000 0.75000000 0.68421053 0.66666667 0.84615385
## 25 1.00000000 1.00000000 0.83333333 0.62500000 0.36842105 0.44444444 0.69230769
## 26 1.00000000 0.91666667 0.84000000 0.58620690 0.43750000 0.48387097 0.76923077
## 27 1.00000000 0.92000000 0.84615385 0.60000000 0.45454545 0.50000000 0.77777778
## 28 1.00000000 0.92000000 0.84615385 0.60000000 0.45454545 0.50000000 0.77777778
## 29 0.92592593 0.79310345 0.73333333 0.52941176 0.40540541 0.44444444 0.67741935
## 30 1.00000000 1.00000000 0.92000000 0.65517241 0.50000000 0.54838710 0.84615385
##             9         10         11         12         13         14         15
## 1
## 2
## 3
## 4
```

```
## 5
## 6
## 7
## 9  0.00000000
## 10 0.45454545 0.00000000
## 11 0.45454545 0.33333333 0.00000000
## 12 0.45454545 0.33333333 0.00000000 0.00000000
## 13 0.63636364 0.50000000 0.16666667 0.16666667 0.00000000
## 14 0.60000000 0.37500000 0.25000000 0.25000000 0.25000000 0.00000000
## 15 0.50000000 0.29411765 0.29411765 0.29411765 0.29411765 0.14285714 0.00000000
## 16 0.54545455 0.47826087 0.56521739 0.56521739 0.56521739 0.33333333 0.28571429
## 17 0.62962963 0.57142857 0.57142857 0.57142857 0.57142857 0.37500000 0.33333333
## 18 0.64285714 0.58620690 0.58620690 0.58620690 0.58620690 0.39393939 0.35294118
## 19 0.64285714 0.65517241 0.79310345 0.79310345 0.79310345 0.57575758 0.52941176
## 20 0.62962963 0.64285714 0.78571429 0.78571429 0.85714286 0.62500000 0.57575758
## 21 0.64285714 0.65517241 0.79310345 0.79310345 0.86206897 0.63636364 0.58823529
## 22 0.70370370 0.71428571 0.85714286 0.85714286 0.92857143 0.68750000 0.63636364
## 23 0.50000000 0.77777778 0.77777778 0.77777778 1.00000000 0.84615385 0.85714286
## 24 0.69230769 0.71428571 0.85714286 0.85714286 1.00000000 0.77777778 0.78947368
## 25 0.69230769 0.57142857 0.85714286 0.85714286 1.00000000 0.66666667 0.68421053
## 26 0.69230769 0.70370370 0.85185185 0.85185185 0.92592593 0.67741935 0.62500000
## 27 0.70370370 0.71428571 0.85714286 0.85714286 0.92857143 0.68750000 0.63636364
## 28 0.70370370 0.71428571 0.85714286 0.85714286 0.92857143 0.68750000 0.63636364
## 29 0.67741935 0.62500000 0.68750000 0.68750000 0.68750000 0.50000000 0.45945946
## 30 0.76923077 0.77777778 0.92592593 0.92592593 1.00000000 0.74193548 0.68750000
##            16         17         18         19         20         21         22
## 1
## 2
## 3
## 4
## 5
## 6
## 7
## 9
## 10
## 11
## 12
## 13
## 14
## 15
## 16 0.00000000
## 17 0.12820513 0.00000000
## 18 0.15000000 0.02222222 0.00000000
## 19 0.25000000 0.15555556 0.13043478 0.00000000
## 20 0.28205128 0.18181818 0.15555556 0.02222222 0.00000000
## 21 0.30000000 0.20000000 0.17391304 0.04347826 0.02222222 0.00000000
## 22 0.33333333 0.22727273 0.20000000 0.06666667 0.04545455 0.02222222 0.00000000
## 23 0.80000000 0.76000000 0.76923077 0.76923077 0.76000000 0.76923077 0.76000000
## 24 0.68000000 0.60000000 0.54838710 0.48387097 0.46666667 0.48387097 0.46666667
## 25 0.60000000 0.60000000 0.54838710 0.48387097 0.46666667 0.48387097 0.46666667
## 26 0.36842105 0.25581395 0.22727273 0.09090909 0.06976744 0.04545455 0.02325581
## 27 0.33333333 0.22727273 0.20000000 0.06666667 0.04545455 0.02222222 0.00000000
## 28 0.33333333 0.22727273 0.20000000 0.06666667 0.04545455 0.02222222 0.00000000
## 29 0.25581395 0.12500000 0.10204082 0.06122449 0.08333333 0.06122449 0.08333333
```

```
## 30 0.36842105 0.25581395 0.22727273 0.09090909 0.06976744 0.04545455 0.02325581
##            23          24          25          26          27          28          29
## 1
## 2
## 3
## 4
## 5
## 6
## 7
## 9
## 10
## 11
## 12
## 13
## 14
## 15
## 16
## 17
## 18
## 19
## 20
## 21
## 22
## 23 0.00000000
## 24 0.45454545 0.00000000
## 25 0.45454545 0.37500000 0.00000000
## 26 0.75000000 0.44827586 0.44827586 0.00000000
## 27 0.76000000 0.46666667 0.46666667 0.02325581 0.00000000
## 28 0.76000000 0.46666667 0.46666667 0.02325581 0.00000000 0.00000000
## 29 0.79310345 0.52941176 0.52941176 0.10638298 0.08333333 0.08333333 0.00000000
## 30 0.75000000 0.44827586 0.44827586 0.04761905 0.02325581 0.02325581 0.10638298
##            30
## 1
## 2
## 3
## 4
## 5
## 6
## 7
## 9
## 10
## 11
## 12
## 13
## 14
## 15
## 16
## 17
## 18
## 19
## 20
## 21
## 22
## 23
```

```
## 24
## 25
## 26
## 27
## 28
## 29
## 30 0.00000000
```

Question 5: Using the distance matrices from above, answer the following questions:

a. Does the resemblance matrix (`fish.db`) represent similarity or dissimilarity? What information in the resemblance matrix led you to arrive at your answer?
b. Compare the resemblance matrices (`fish.db` or `fish.ds`) you just created. How does the choice of the Sørensen or Bray-Curtis distance influence your interpretation of site (dis)similarity?

Answer 5a: The resemblance matrix represent dissimilarity, because diagonal values are zero.
Answer 5b: The Bray-Curtis distance is based on squared's abundance which leads to the larger impact of abundant species compared to Sørensen distance. Therefore, the values for distance between sites are slightly different. For example, Sørensen distance between site 27 and 28 is 0, but Bray-Curtis distance is 0.098 for same sites.

4) VISUALIZING BETA-DIVERSITY

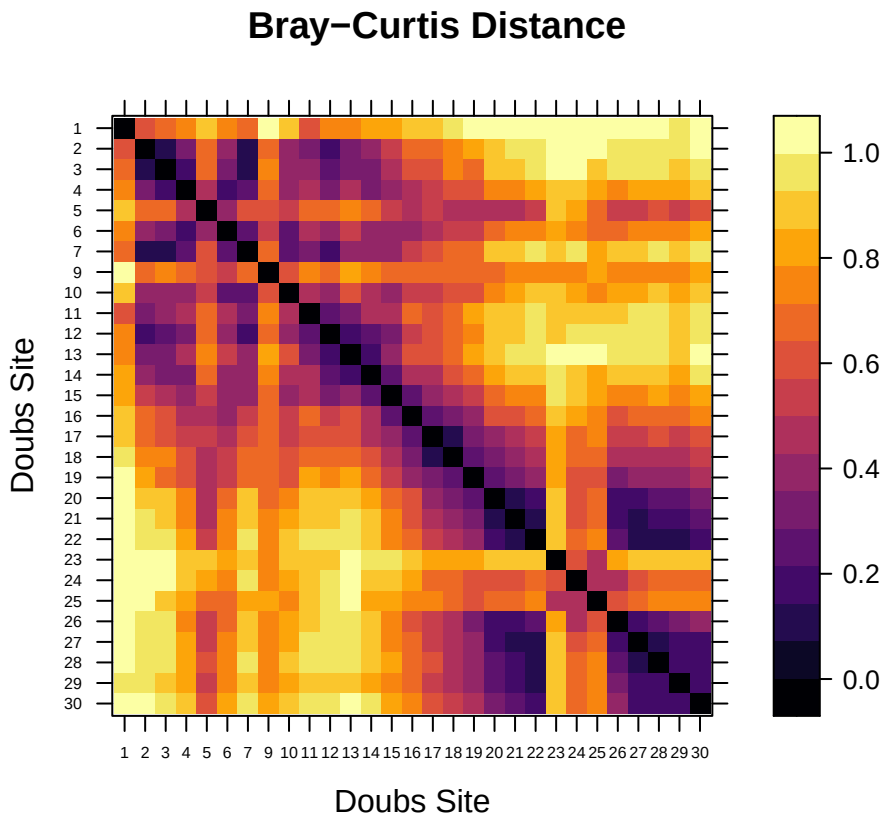
A. Heatmaps

In the R code chunk below, do the following:

1. define a color palette,
2. define the order of sites in the Doubs River, and
3. use the `levelplot()` function to create a heatmap of fish abundances in the Doubs River.

```
# Define Order of Sites
order <- rev(attr(fish.db, "Labels"))

# Plot Heatmap
levelplot(as.matrix(fish.db)[, order],
          aspect = "iso",
          col.regions = inferno,
          xlab = "Doubs Site",
          ylab = "Doubs Site",
          scales = list(cex = 0.5),
          main = "Bray-Curtis Distance")
```



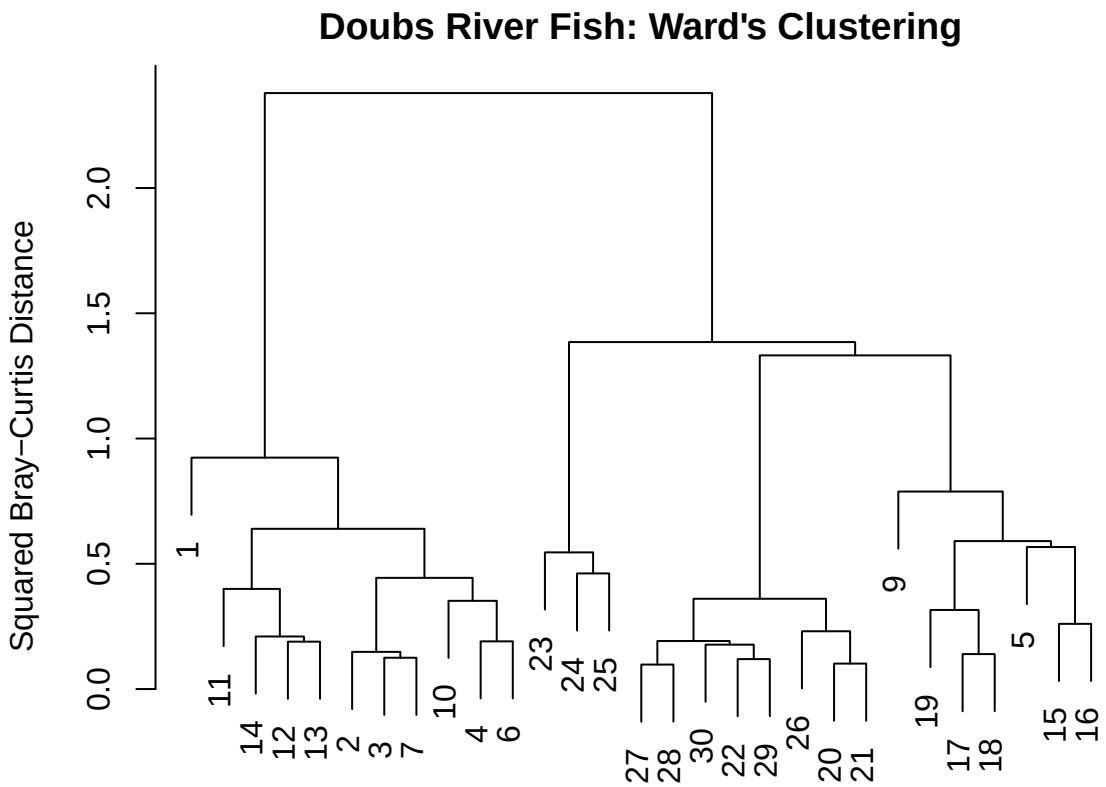
B. Cluster Analysis

In the R code chunk below, do the following:

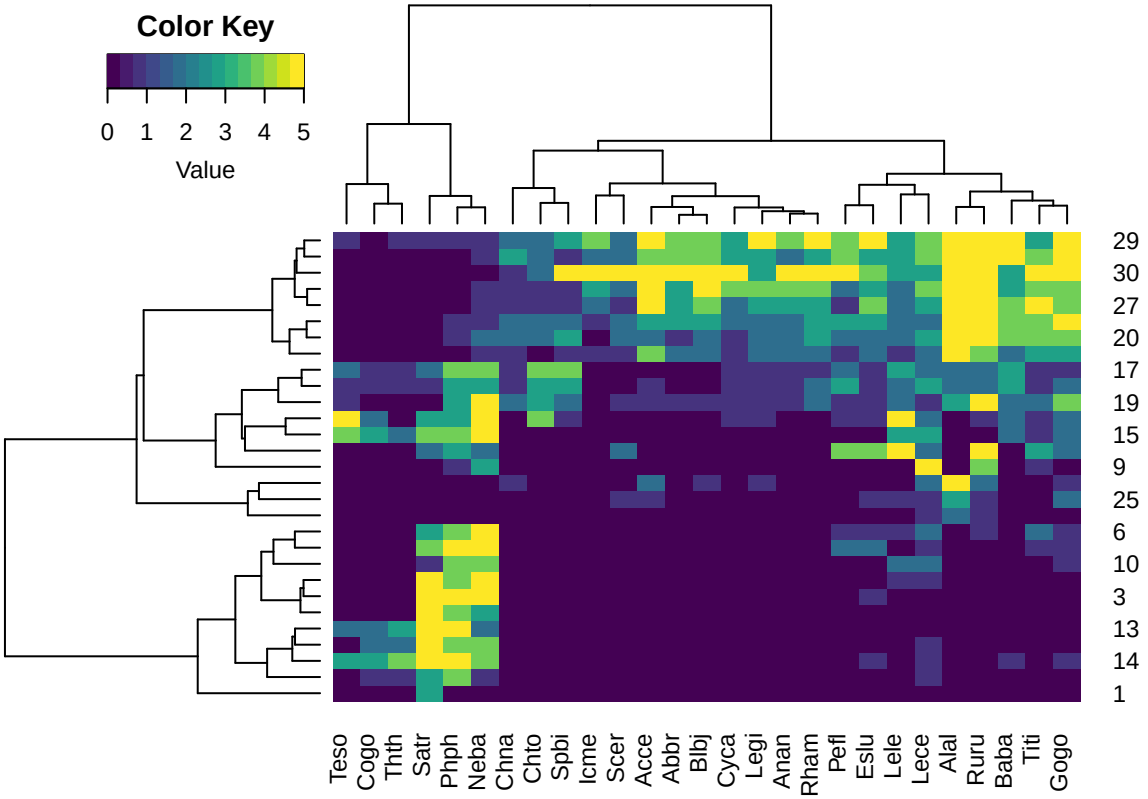
1. perform a cluster analysis using Ward's Clustering, and
2. plot your cluster analysis (use either `hclust` or `heatmap.2`).

```
# Perforem cluster analysis
fish.ward <- hclust(fish.db, method = "ward.D2")

# Plot cluster
par(mar = c(1, 5, 2, 2) + 0.1)
plot(fish.ward, main = "Doubs River Fish: Ward's Clustering",
     ylab = "Squared Bray-Curtis Distance")
```




```
gplots::heatmap.2(as.matrix(fish),
                  distfun = function(x) vegdist(x, method = "bray"),
                  hclustfun = function(x) hclust(x, method = "ward.D2"),
                  col = viridis, trace = "none", density.info = "none")
```



Question 6: Based on cluster analyses and the introductory plots that we generated after loading the data, develop an ecological hypothesis for fish diversity the `doubs` data set?

> ***Answer 6***: THe fish diversity is different in upstreams and downstreams in Doubs river. There are some species more related to downstreams, while others are more related to upstreams.

C. Ordination

Principal Coordinates Analysis (PCoA)

In the R code chunk below, do the following:

1. perform a Principal Coordinates Analysis to visualize beta-diversity
2. calculate the variation explained by the first three axes in your ordination
3. plot the PCoA ordination,
4. label the sites as points using the Doubs River site number, and
5. identify influential species and add species coordinates to PCoA plot.

```
fish.pcoa <- cmdscale(fish.db, eig = TRUE, k = 3)

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

# Define plot parameters
par(mar = c(5, 5, 1, 2) + 0.1)

# Initiate plot
plot(fish.pcoa$points[, 1], fish.pcoa$points[, 2],
     ylim = c(-0.2, 0.7),
```

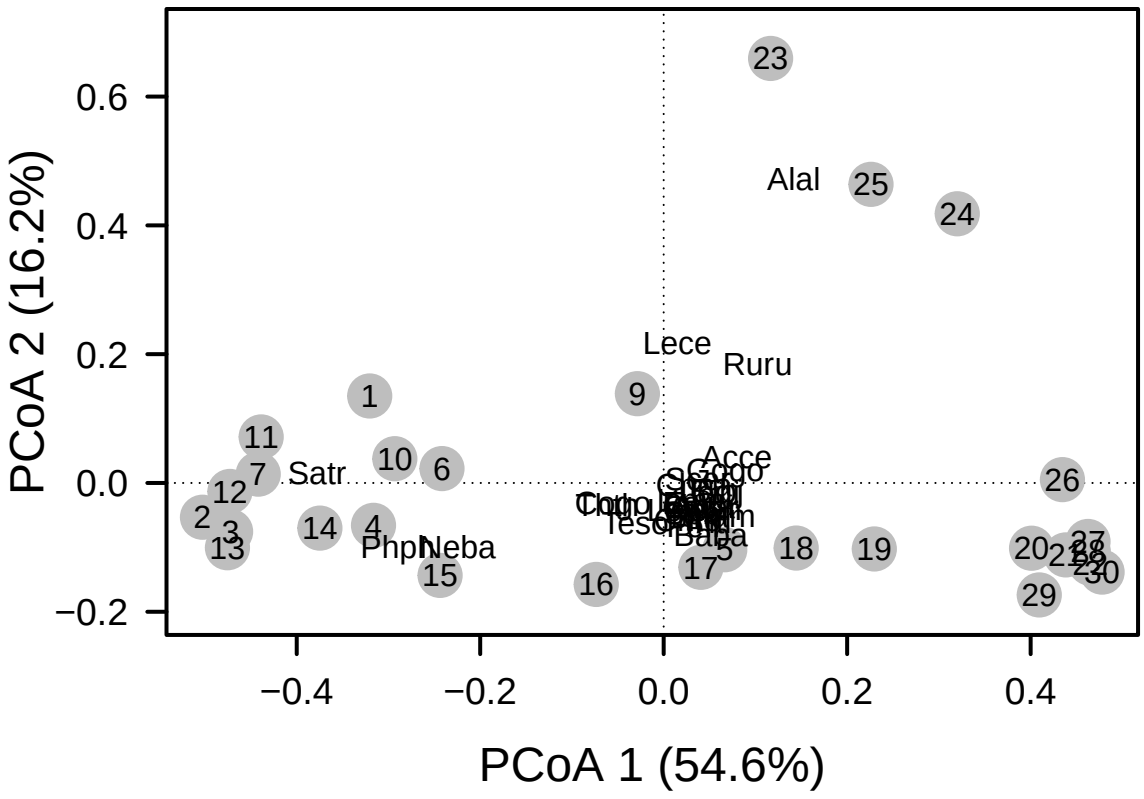
```
     xlab = paste("PCoA 1 (", explainvar1, "%)", sep = ""),
     ylab = paste("PCoA 2 (", explainvar2, "%)", sep = ""),
     pch = 16, cex = 2.0, type = "n", cex.lab = 1.5, cex.axis = 1.2, axes = FALSE)

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

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

# First we calculate the relative abundances of each species at each site
fishREL <- fish
for (i in 1:nrow(fish)) {
    fishREL[i, ] = fish[i, ] / sum(fish[i, ])
    }

# Now, we use this information to calculate and add species scores
fish.pcoa <- add.spec.scores(fish.pcoa, fishREL, method = "pcoa.scores")
text(fish.pcoa$cproj[, 1], fish.pcoa$cproj[, 2],
    labels = row.names(fish.pcoa$cproj), col = "black")
```



In the R code chunk below, do the following:

1. identify influential species based on correlations along each PCoA axis (use a cutoff of 0.70), and
2. use a permutation test (999 permutations) to test the correlations of each species along each axis.

```r
spe.corr <- add.spec.scores(fish.pcoa, fishREL, method = "cor.scores")$cproj
corrcut <- 0.7  # User-defined cutoff
imp.spp <- spe.corr[abs(spe.corr[, 1]) >= corrcut | abs(spe.corr[, 2]) >= corrcut, ]

# Permutation test for species abundances across axes
fit <- envfit(fish.pcoa, fishREL, perm = 999)

imp.spp
```

```
##            Dim1       Dim2        Dim3
## Phph -0.8674640 -0.1699316 -0.12463098
## Neba -0.7674114 -0.1855678 -0.36963830
## Rham  0.8088751 -0.4192567  0.14136301
## Legi  0.8201759 -0.1701803  0.12423941
## Cyca  0.7595122 -0.4442926  0.17313658
## Abbr  0.7704744 -0.3452714  0.29277803
## Acce  0.7635195  0.2155765  0.10288179
## Blbj  0.8118483 -0.1324698  0.25581178
## Alal  0.4471283  0.8119843 -0.05167131
## Anan  0.7974122 -0.3918972  0.20944968
```

```r
fit
```

```
##
## ***VECTORS
##
##          Dim1     Dim2     r2 Pr(>r)
## Cogo -0.83884 -0.54438 0.2982  0.014 *
## Satr -0.99904  0.04371 0.4326  0.003 **
## Phph -0.94110 -0.33813 0.7814  0.001 ***
## Neba -0.91413 -0.40543 0.6234  0.001 ***
## Thth -0.87692 -0.48063 0.2634  0.036 *
## Teso -0.44704 -0.89452 0.1700  0.108
## Chna  0.99707 -0.07644 0.4612  0.001 ***
## Chto  0.42032 -0.90738 0.2579  0.023 *
## Lele  0.33041 -0.94384 0.0495  0.540
## Lece  0.06856  0.99765 0.3399  0.009 **
## Baba  0.54118 -0.84091 0.6752  0.001 ***
## Spbi  0.57341 -0.81927 0.4138  0.003 **
## Gogo  0.97507  0.22188 0.3753  0.002 **
## Eslu  0.72044 -0.69352 0.1673  0.092 .
## Pefl  0.43762 -0.89916 0.3048  0.011 *
## Rham  0.72476 -0.68901 0.8301  0.001 ***
## Legi  0.93461 -0.35568 0.7016  0.001 ***
## Scer  0.98569  0.16858 0.3533  0.005 **
## Cyca  0.68181 -0.73153 0.7743  0.001 ***
## Titi  0.64378 -0.76521 0.4586  0.001 ***
## Abbr  0.77254 -0.63497 0.7128  0.001 ***
## Icme  0.75626 -0.65427 0.5270  0.001 ***
## Acce  0.88799  0.45986 0.6294  0.001 ***
## Ruru  0.48379  0.87518 0.5177  0.002 **
## Blbj  0.95802 -0.28671 0.6766  0.001 ***
## Alal  0.28755  0.95777 0.8592  0.001 ***
## Anan  0.74277 -0.66954 0.7894  0.001 ***
## ---
```

```
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## Permutation: free
## Number of permutations: 999
```

Question 7: Address the following questions about the ordination results of the `doubs` data set:

a. Describe the grouping of sites in the Doubs River based on fish community composition.
b. Generate a hypothesis about which fish species are potential indicators of river quality.

> ***Answer 7a***: Sites 23,24, and 25 are similar in fish community composition, and distinctively different from other sites. Sites 20, 21, 22, 26, 27, 28, 29, and 30 are also similar in fish community composition, and distinctively different from other sites. Other sites are spread in fish community composition without any distict cluster. ***Answer 7b***: Phph, Neba,, Rham, Legi, Cyca Abbr, Acce, Blbj, Alal or Anan could be potential indicators of river quality.

SYNTHESIS

Load the dataset from that you and your partner are using for the team project. Use one of the tools introduced in the beta diversity module to visualize your data. Describe any interesting patterns and identify a hypothesis is relevant to the principles of biodiversity.

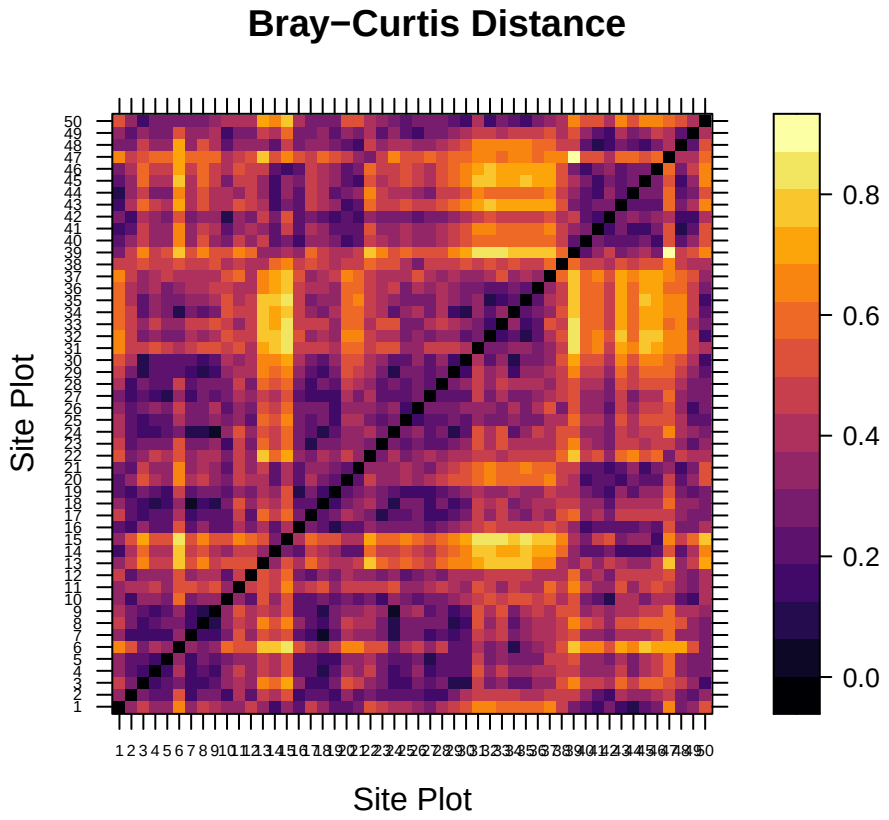
```
library(readr)
setwd("/cloud/project/QB2025_Yoo/Week2-Alpha")
Freq_LDW_final<-read_csv("Freq_LDW_final.csv")
```

```
## Rows: 625 Columns: 27
## -- Column specification --------------------------------------------------------
## Delimiter: ","
## dbl (27): Group, Acer rubrum, Acer saccharum, Carya cordiformis, Carya ovata...
##
## i Use `spec()` to retrieve the full column specification for this data.
## i Specify the column types or set `show_col_types = FALSE` to quiet this message.
```

```
LDW <- Freq_LDW_final[1:50,-1]

# Calculate Bray-Curtis
LDW.db <- vegdist(LDW, method = "bray", diag = TRUE)

# Plot Heatmap
levelplot(as.matrix(LDW.db),
          aspect = "iso",
          col.regions = inferno,
          xlab = "Site Plot",
          ylab = "Site Plot",
          scales = list(cex = 0.5),
          main = "Bray-Curtis Distance")
```


Bray−Curtis Distance
Site Plot
Site Plot
0.0
0.2
0.4
0.6
0.8