

# 5. Worksheet: Alpha Diversity

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## OVERVIEW

In this exercise, we will explore aspects of local or site-specific diversity, also known as alpha ( $\alpha$ ) diversity. First we will quantify two of the fundamental components of ( $\alpha$ ) diversity: **richness** and **evenness**. From there, we will then discuss ways to integrate richness and evenness, which will include univariate metrics of diversity along with an investigation of the **species abundance distribution (SAD)**.

## Directions:

1. In the Markdown version of this document in your cloned repo, change “Student Name” on line 3 (above) to 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 the proper scripting needed to carry out the exercise.
4. Answer questions in the worksheet. Space for your answer is provided in this document and 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 `AlphaDiversity_Worskheet.Rmd` and the PDF output of Knitr (`AlphaDiversity_Worskheet.pdf`).

## 1) R SETUP

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 5.AlphaDiversity folder, and 4) Load the **vegan** R package (be sure to install first if you haven’t already).

```
remove(list=ls())
getwd()
```

```
## [1] "/Users/mark/Box Sync/Courses/Quantitative Biodiversity/QB2019_Hibbins/2.Worksheets/5.AlphaDiversity"
setwd('/Users/mark/Box Sync/Courses/Quantitative Biodiversity/QB2019_Hibbins/2.Worksheets/5.AlphaDiversity')
library(vegan)
```

```
## Warning: package 'vegan' was built under R version 3.4.4
## Loading required package: permute
## Loading required package: lattice
## This is vegan 2.5-3
```

## 2) LOADING DATA

In the R code chunk below, do the following: 1) Load the BCI dataset, and 2) Display the structure of the dataset (if the structure is long, use the `max.level = 0` argument to show the basic information).

```
data(BCI)
str(BCI, max.level = 0)

## 'data.frame':   50 obs. of  225 variables:
##  - attr(*, "original.names")= chr  "Abarema.macradenium" "Acacia.melanoceras" "Acalypha.diversifolia"
```

## 3) SPECIES RICHNESS

**Species richness (S)** refers to the number of species in a system or the number of species observed in a sample.

### Observed richness

In the R code chunk below, do the following:

1. Write a function called `S.obs` to calculate observed richness
2. Use your function to determine the number of species in `site1` of the BCI data set, and
3. Compare the output of your function to the output of the `specnumber()` function in `vegan`.

```
S.obs <- function(x){
  rowSums(x > 0) * 1
}
```

```
S.obs(BCI[1,])
```

```
## 1
## 93
```

```
specnumber(BCI[1,])
```

```
## 1
## 93
```

```
specnumber(BCI[2,])
```

```
## 2
## 84
```

```
specnumber(BCI[3,])
```

```
## 3
## 90
```

```
specnumber(BCI[4,])
```

```
## 4
## 94
```

**Question 1:** Does `specnumber()` from `vegan` return the same value for observed richness in `site1` as our function `S.obs`? What is the species richness of the first four sites (i.e., rows) of the BCI matrix?

**Answer 1:** `specnumber()` and the `S.obs` function return the same value (as they should). The richness for the first four sites are 93, 84, 90, and 94.

### Coverage: How well did you sample your site?

In the R code chunk below, do the following:

1. Write a function to calculate Good's Coverage, and
2. Use that function to calculate coverage for all sites in the BCI matrix.

```
C = function(x) {
  1 - (sum(x == 1) / sum(x))
}

apply(BCI, 1, C)
```

```
##           1           2           3           4           5           6           7
## 0.9308036 0.9287356 0.9200864 0.9468504 0.9287129 0.9174757 0.9326923
##           8           9          10          11          12          13          14
## 0.9443155 0.9095355 0.9275362 0.9152120 0.9071038 0.9242054 0.9132420
##          15          16          17          18          19          20          21
## 0.9350649 0.9267735 0.8950131 0.9193084 0.8891455 0.9114219 0.8946078
##          22          23          24          25          26          27          28
## 0.9066986 0.8705882 0.9030612 0.9095023 0.9115479 0.9088729 0.9198966
##          29          30          31          32          33          34          35
## 0.8983516 0.9221053 0.9382423 0.9411765 0.9220183 0.9239374 0.9267887
##          36          37          38          39          40          41          42
## 0.9186047 0.9379310 0.9306488 0.9268868 0.9386503 0.8880597 0.9299517
##          43          44          45          46          47          48          49
## 0.9140049 0.9168704 0.9234234 0.9348837 0.8847059 0.9228916 0.9086651
##          50
## 0.9143519
```

**Question 2:** Answer the following questions about coverage:

- a. What is the range of values that can be generated by Good's Coverage?
- b. What would we conclude from Good's Coverage if  $n_i$  equaled  $N$ ?
- c. What portion of taxa in `site1` was represented by singletons?
- d. Make some observations about coverage at the BCI plots.

**Answer 2a:** Because Good's Coverage is calculated as a proportion, it should be a value between 0 and 1.

**Answer 2b:** If  $n_i$  equals  $N$ , then Good's coverage will equal 0, meaning every individual in the sample belongs to its own species that was only sampled once. This is not good sampling coverage.

**Answer 2c:** Approximately 7% ( $1 - 0.93$ )

**Answer 2d:** The values of coverage for this dataset are in the range of 85-95%, meaning that the majority of individuals at each site belong to a species that is sampled at least twice.

### Estimated richness

In the R code chunk below, do the following:

1. Load the microbial dataset (located in the `5.AlphaDiversity/data` folder),

2. Transform and transpose the data as needed (see handout),
3. Create a new vector (`soilbac1`) by indexing the bacterial OTU abundances of any site in the dataset,
4. Calculate the observed richness at that particular site, and
5. Calculate coverage of that site

```
soilbac <- read.table('data/soilbac.txt', sep = '\t', header = TRUE, row.names = 1)

soilbac_transpose <- as.data.frame(t(soilbac))
soilbac3 <- soilbac_transpose[3,]

sum(soilbac3)
```

```
## [1] 2533
```

```
specnumber(soilbac3)
```

```
## T1_3
```

```
## 1174
```

```
C(soilbac3)
```

```
## [1] 0.6735097
```

**Question 3:** Answer the following questions about the soil bacterial dataset.

- a. How many sequences did we recover from the sample `soilbac1`, i.e.  $N$ ?
- b. What is the observed richness of `soilbac1`?
- c. How does coverage compare between the BCI sample (`site1`) and the KBS sample (`soilbac1`)?

**Answer 3a:** 2533

**Answer 3b:** 1174

**Answer 3c:** The coverage for the KBS sample is much lower (67.3% vs. 93%)

## Richness estimators

In the R code chunk below, do the following:

1. Write a function to calculate **Chao1**,
2. Write a function to calculate **Chao2**,
3. Write a function to calculate **ACE**, and
4. Use these functions to estimate richness at `site1` and `soilbac1`.

```
S.chao1 <- function(x){
  specnumber(x) + (sum(x == 1)^2) / (2 * sum(x == 2))
}
```

```
S.chao2 <- function(site, SbyS){
  SbyS = as.data.frame(SbyS)
  x = SbyS[site,]
  SbyS_presenceabsence <- (SbyS > 0) * 1
  Q1 = sum(colSums(SbyS_presenceabsence) == 1)
  Q2 = sum(colSums(SbyS_presenceabsence) == 2)
  S.chao2 <- S.obs(x) + (Q1^2)/(2*Q2)
  return(S.chao2)
```

```

}

S.ace <- function(x, threshold){
  x <- x[x>0]
  S_abund <- length(which(x > threshold))
  S_rare <- length(which(x <= threshold))
  singletons <- length(which(x == 1))
  N_rare <- sum(x[which(x <= threshold)])
  C_ace <- 1 - (singletons / N_rare)
  i <- c(1:threshold)
  count <- function(i, y){
    length(y[y == i])
  }
  a_1 <- sapply(i, count, x)
  f_1 <- (i * (i-1)) * a_1
  G_ace <- (S_rare/C_ace)*(sum(f_1)/(N_rare*(N_rare-1)))
  S_ace <- S_abund + (S_rare/C_ace) + (singletons/C_ace) * max(G_ace,0)
  return(S_ace)
}

site1 <- BCI[1,]

S.chao1(site1)

##          1
## 119.6944

S.chao1(soilbac3)

##      T1_3
## 3042.658

S.chao2(site = 1, SbyS = BCI)

##          1
## 104.6053

S.chao2(site = 3, SbyS = soilbac_transpose)

##      T1_3
## 21155.39

S.ace(site1, 10)

## [1] 159.3404

S.ace(soilbac3, 10)

## [1] 5090.28

```

**Question 4:** What is the difference between ACE and the Chao estimators? Do the estimators give consistent results? Which one would you choose to use and why?

**Answer 4:** The ACE estimator defines a threshold for comparison of rare vs. common species, while the Chao estimators use singletons and doubletons. The Chao 1 and ACE estimators seem to give roughly similar estimates, while the Chao 2 estimator reports a much larger value for soilbac3. This is probably because the Chao 2 estimator reports richness across multiple sites, and sampling coverage for an individual site in the soilbac data is lower. All the estimators seem to operate best under different circumstances. but the ACE estimator is less intuitive and also

cannot be effectively used in datasets where there are many species with few samples. Therefore, for most situations I would probably use one of the Chao estimators.

## Rarefaction

In the R code chunk below, please do the following:

1. Calculate observed richness for all samples in `soilbac`,
2. Determine the size of the smallest sample,
3. Use the `rarefy()` function to rarefy each sample to this level,
4. Plot the rarefaction results, and
5. Add the 1:1 line and label.

## 4) SPECIES EVENNESS

Here, we consider how abundance varies among species, that is, **species evenness**.

### Visualizing evenness: the rank abundance curve (RAC)

One of the most common ways to visualize evenness is in a **rank-abundance curve** (sometime referred to as a rank-abundance distribution or Whittaker plot). An RAC can be constructed by ranking species from the most abundant to the least abundant without respect to species labels (and hence no worries about ‘ties’ in abundance).

In the R code chunk below, do the following:

1. Write a function to construct a RAC,
2. Be sure your function removes species that have zero abundances,
3. Order the vector (RAC) from greatest (most abundant) to least (least abundant), and
4. Return the ranked vector

Now, let’s examine the RAC for `site1` of the BCI data set.

In the R code chunk below, do the following:

1. Create a sequence of ranks and plot the RAC with natural-log-transformed abundances,
2. Label the x-axis “Rank in abundance” and the y-axis “log(abundance)”

**Question 5:** What effect does visualizing species abundance data on a log-scaled axis have on how we interpret evenness in the RAC?

**Answer 5:**

Now that we have visualized unevenness, it is time to quantify it using Simpson’s evenness ( $E_{1/D}$ ) and Smith and Wilson’s evenness index ( $E_{var}$ ).

### Simpson’s evenness ( $E_{1/D}$ )

In the R code chunk below, do the following:

1. Write the function to calculate  $E_{1/D}$ , and

2. Calculate  $E_{1/D}$  for `site1`.

### Smith and Wilson's evenness index ( $E_{var}$ )

In the R code chunk below, please do the following:

1. Write the function to calculate  $E_{var}$ ,
2. Calculate  $E_{var}$  for `site1`, and
3. Compare  $E_{1/D}$  and  $E_{var}$ .

**Question 6:** Compare estimates of evenness for `site1` of BCI using  $E_{1/D}$  and  $E_{var}$ . Do they agree? If so, why? If not, why? What can you infer from the results.

**Answer 6:**

## 5) INTEGRATING RICHNESS AND EVENNESS: DIVERSITY METRICS

So far, we have introduced two primary aspects of diversity, i.e., richness and evenness. Here, we will use popular indices to estimate diversity, which explicitly incorporate richness and evenness. We will write our own diversity functions and compare them against the functions in `vegan`.

### Shannon's diversity (a.k.a., Shannon's entropy)

In the R code chunk below, please do the following:

1. Provide the code for calculating  $H'$  (Shannon's diversity),
2. Compare this estimate with the output of `vegan`'s diversity function using `method = "shannon"`.

### Simpson's diversity (or dominance)

In the R code chunk below, please do the following:

1. Provide the code for calculating  $D$  (Simpson's diversity),
2. Calculate both the inverse ( $1/D$ ) and  $1 - D$ ,
3. Compare this estimate with the output of `vegan`'s diversity function using `method = "simp"`.

**Question 7:** Compare estimates of evenness for `site1` of BCI using  $E_{H'}$  and  $E_{var}$ . Do they agree? If so, why? If not, why? What can you infer from the results.

**Answer 7:**

### Fisher's $\alpha$

In the R code chunk below, please do the following:

1. Provide the code for calculating Fisher's  $\alpha$ ,
2. Calculate Fisher's  $\alpha$  for `site1` of BCI.

**Question 8:** How is Fisher's  $\alpha$  different from  $E_{H'}$  and  $E_{var}$ ? What does Fisher's  $\alpha$  take into account that  $E_{H'}$  and  $E_{var}$  do not?

**Answer 8:**

## 6) MOVING BEYOND UNIVARIATE METRICS OF $\alpha$ DIVERSITY

The diversity metrics that we just learned about attempt to integrate richness and evenness into a single, univariate metric. Although useful, information is invariably lost in this process. If we go back to the rank-abundance curve, we can retrieve additional information – and in some cases – make inferences about the processes influencing the structure of an ecological system.

### Species abundance models

The RAC is a simple data structure that is both a vector of abundances. It is also a row in the site-by-species matrix (minus the zeros, i.e., absences).

Predicting the form of the RAC is the first test that any biodiversity theory must pass and there are no less than 20 models that have attempted to explain the uneven form of the RAC across ecological systems.

In the R code chunk below, please do the following:

1. Use the `radfit()` function in the `vegan` package to fit the predictions of various species abundance models to the RAC of `site1` in BCI,
2. Display the results of the `radfit()` function, and
3. Plot the results of the `radfit()` function using the code provided in the handout.

**Question 9:** Answer the following questions about the rank abundance curves: a) Based on the output of `radfit()` and plotting above, discuss which model best fits our rank-abundance curve for `site1`? b) Can we make any inferences about the forces, processes, and/or mechanisms influencing the structure of our system, e.g., an ecological community?

**Answer 9a:**

**Answer 9b:**

**Question 10:** Answer the following questions about the preemption model: a. What does the preemption model assume about the relationship between total abundance ( $N$ ) and total resources that can be preempted? b. Why does the niche preemption model look like a straight line in the RAD plot?

**Answer 10a:**

**Answer 10b:**

**Question 11:** Why is it important to account for the number of parameters a model uses when judging how well it explains a given set of data?

**Answer 11:**

## SYNTHESIS

1. As stated by Magurran (2004) the  $D = \sum p_i^2$  derivation of Simpson's Diversity only applies to communities of infinite size. For anything but an infinitely large community, Simpson's Diversity index is calculated as  $D = \sum \frac{n_i(n_i-1)}{N(N-1)}$ . Assuming a finite community, calculate Simpson's  $D$ ,  $1 - D$ , and Simpson's inverse (i.e.  $1/D$ ) for `site 1` of the BCI site-by-species matrix.
2. Along with the rank-abundance curve (RAC), another way to visualize the distribution of abundance among species is with a histogram (a.k.a., frequency distribution) that shows the frequency of different abundance classes. For example, in a given sample, there may be 10 species represented by a single individual, 8 species with two individuals, 4 species with three individuals, and so on. In fact, the rank-abundance curve and the frequency distribution are the two most common ways to visualize the species-abundance distribution (SAD) and to test species abundance models and biodiversity theories.



To address this homework question, use the R function **hist()** to plot the frequency distribution for **site 1** of the BCI site-by-species matrix, and describe the general pattern you see.

3. We asked you to find a biodiversity dataset with your partner. This data could be one of your own or it could be something that you obtained from the literature. Load that dataset. How many sites are there? How many species are there in the entire site-by-species matrix? Any other interesting observations based on what you learned this week?

## SUBMITTING YOUR ASSIGNMENT

Use Knitr to create a PDF of your completed `alpha_assignment.Rmd` document, push it to GitHub, and create a pull request. Please make sure your updated repo include both the HTML and RMarkdown files.

Unless otherwise noted, this assignment is due on **Wednesday, January 23<sup>rd</sup>, 2017 at 12:00 PM (noon)**.