Community ecology - Computer lab I - AB332

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> This is an R Markdown Notebook. When you execute code within the notebook, the results appear beneath the code. Try executing this chunk by clicking the *Run* button within the chunk or by placing your cursor inside it and pressing *Cmd+Shift+Enter*.
>
> Add a new chunk by clicking the *Insert Chunk* button on the toolbar or by pressing *Cmd+Option+I*.
>
> When you save the notebook, an HTML file containing the code and output will be saved alongside it (click the *Preview* button or press *Cmd+Shift+K* to preview the HTML file).
>
> The preview shows you a rendered HTML copy of the contents of the editor. Consequently, unlike *Knit*, *Preview* does not run any R code chunks. Instead, the output of the chunk when it was last run in the editor is displayed.

It's time for you to try and do the same analysis as was shown in the lecture but using a different dataset: Make sure you have installed all packages!

Load Packages

Starting community ecology analyses

Read the data from the github page:

```
otu.tab <- read_tsv("https://raw.githubusercontent.com/krabberod/UNIS_AB332_2023/main/computer_lab/data/
```

```
## Rows: 3697 Columns: 83
## -- Column specification --------------------------------------------------------
## Delimiter: "\t"
## chr  (1): OTUNumber
## dbl (82): Isa_111214, Isa_120117, Isa_120128, Isa_120209, Isa_120216, Isa_12...
##
## 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.
```

First, get to know the data: - *How many samples and how many OTUs are in the dataset?* - *What do the numbers in the sample names mean?*

```
head(otu.tab)
```

```
## # A tibble: 6 x 83
##   OTUNumber Isa_111214 Isa_120117 Isa_120128 Isa_120209 Isa_120216 Isa_120223
##   <chr>          <dbl>      <dbl>      <dbl>      <dbl>      <dbl>      <dbl>
## 1 OTU1             436        348        236        139        260        446
## 2 OTU10           2308       2537       1599       1956        328        668
## 3 OTU100            83         36         21         30          1         21
## 4 OTU1000            0          0          0          0          0          0
## 5 OTU1001            1          0          0          0          0          2
## 6 OTU1002            0          2          0          1          0          0
## # i 76 more variables: Isa_120301 <dbl>, Isa_120308 <dbl>, Isa_120320 <dbl>,
## #   Isa_120321 <dbl>, Isa_120322 <dbl>, Isa_120323 <dbl>, Isa_120329 <dbl>,
## #   Isa_120403 <dbl>, Isa_120411 <dbl>, Isa_120416 <dbl>, Isa_120419 <dbl>,
## #   Isa_120423 <dbl>, Isa_120426 <dbl>, Isa_120430 <dbl>, Isa_120503 <dbl>,
## #   Isa_120507 <dbl>, Isa_120508 <dbl>, Isa_120509 <dbl>, Isa_120510 <dbl>,
## #   Isa_120516 <dbl>, Isa_120524 <dbl>, Isa_120621 <dbl>, Isa_120706 <dbl>,
## #   Isa_120806 <dbl>, Isa_120823 <dbl>, Isa_120906 <dbl>, Isa_120918 <dbl>, ...
```

```
dim(otu.tab)
```

```
## [1] 3697   83
```

You can look at a given selection of the table by specifying a range of rows and columns:

```
otu.tab[5:15, 1:5] # The first 10 rows, and the first 5 columns
```

```
## # A tibble: 11 x 5
##    OTUNumber Isa_111214 Isa_120117 Isa_120128 Isa_120209
##    <chr>          <dbl>      <dbl>      <dbl>      <dbl>
##  1 OTU1001            1          0          0          0
##  2 OTU1002            0          2          0          1
##  3 OTU1003            3          0          0          0
##  4 OTU1004            9          2          5          7
##  5 OTU1005            1          0          2          0
##  6 OTU1006            0          2          0          4
##  7 OTU1007            0          0          0          0
##  8 OTU1008            0          0          0          0
##  9 OTU1009            0          0          0          1
## 10 OTU101             3          0          0          0
## 11 OTU1010            1          3          0          3
```

You can also see the entire table with the `View()` function:

```
View(otu.tab)
```

- *See if you can choose a different subset. For instance samples 6-12 and OTUs 20-26:*

We can assign OTU-numbers as rownames

```
otu.tab <- column_to_rownames(otu.tab, var = "OTUNumber")
```

Let's check the names

```
head(rownames(otu.tab))
```

```
## [1] "OTU1"    "OTU10"   "OTU100"  "OTU1000" "OTU1001" "OTU1002"
```

```
dim(otu.tab)
```

```
## [1] 3697   82
```

For simplicity, I have included only 25 samples in the rest of the tutorial. As an exercise, *you should redo the analysis with the full dataset*. I.e. remove the part of the code that selects particular samples in the following chunk.
This way your numbers will differ from the pdf, and you can also see the effect of a different dataset.s

```
otu.tab.red <- otu.tab[, 6:30]
```

The data needs to be transposed since this is how Vegan likes it (i.e. Vegan prefers OTUs as columns, and Samples as rows).

```
otu.tab.trans <- t(otu.tab.red)
otu.tab.trans[1:5, 1:5]
```

```
##            OTU1 OTU10 OTU100 OTU1000 OTU1001
## Isa_120223  446   668     21       0       2
## Isa_120301  149   551     23       1       1
## Isa_120308  321  1462     99      11       0
## Isa_120320  204   896     75       1       0
## Isa_120321  442   646     61       4       0
```

You can get the total number of reads for each sample using rowSums(). and the total reads per OTU with colSums().
- *Do you understand what these two functions do just by looking at their names?*

```
rowSums(otu.tab.trans)
```

```
## Isa_120223 Isa_120301 Isa_120308 Isa_120320 Isa_120321 Isa_120322 Isa_120323
##      13682      15783      32833      19361      17110      13658      16251
## Isa_120329 Isa_120403 Isa_120411 Isa_120416 Isa_120419 Isa_120423 Isa_120426
##      17002      13551      25606      29877      16194      17161      22305
## Isa_120430 Isa_120503 Isa_120507 Isa_120508 Isa_120509 Isa_120510 Isa_120516
##      19942      25237      27389      19969      34666      17682      25256
## Isa_120524 Isa_120621 Isa_120706 Isa_120806
##      40267      24636      23156      20390
```

```
head(colSums(otu.tab.trans)) # Too many to show them all.
```

```
##    OTU1   OTU10  OTU100 OTU1000 OTU1001 OTU1002
##  100420    7814     786      54       4      28
```

- *Can you figure out a way to view only the last part of the the list of total reads per OTU? Hint: look up the help file for the head() function*:

```
?head
```

Since I have selected only a few of the samples is possible that some of the OTU's are left with a total abundance of zero. In R it is possible to have functions within functions so the following will print the number of columns in the data set that has a sum equal to 0:

```
length(which(colSums(otu.tab.trans) == 0))
```

```
## [1] 1163
```

This code is nested so that R reads evaluates the innermost function first (i.e `colSums()`), then applies the next function to that result (i.e. `which (...) ==0`), then finally the outermost function `lenght()`. In "normal" language the nested function asks: what is the length of the list of column-sums which are exactly zero.

We can use the same idea of a function within a function to exclude the OTUs with a total number of 0. In the next chunk we use the square brackets to make a selection in the dataframe (as before), and add a '-' to get the opposite of what is evaluated by the function.

```
otu.tab.trans <- otu.tab.trans[, -(which(colSums(otu.tab.trans) == 0))]
```

Now how many are 0?

```
length(which(colSums(otu.tab.trans) == 0))
```

```
## [1] 0
```

How many have more than 0 reads?

```
length(which(colSums(otu.tab.trans) > 0))
```

```
## [1] 2534
```

Can you find how many OTU's that have more than 10 reads (in total)?

Common metrics and methods

The following calculations make use of functions in the vegan package written by Jari Oksanen.
Vegan is an R package for community ecologists. It contains the most popular methods of multivariate analysis needed in analyzing ecological communities, and tools for diversity analysis, and other potentially useful functions. If you want to learn more about the vegan package you can check out the vignette (a form for introduction) by running: `browseVignettes("vegan")`

Richness estimations

Now lets do some ecology:

```
richness <- estimateR(otu.tab.trans)
richness
```

```
##          Isa_120223 Isa_120301 Isa_120308 Isa_120320 Isa_120321 Isa_120322
## S.obs     701.00000  902.00000 1042.00000  952.00000  897.00000  858.00000
## S.chao1  1100.22581 1260.69343 1355.30612 1316.81633 1200.76642 1337.40517
## se.chao1   66.62128   54.17742   47.66939   53.77784   47.47591   72.14089
## S.ACE    1036.24030 1249.23447 1337.63473 1341.50902 1197.73116 1278.33686
## se.ACE     17.06514   18.59112   18.50405   19.72472   17.80976   19.74198
##          Isa_120323 Isa_120329 Isa_120403 Isa_120411 Isa_120416 Isa_120419
## S.obs     959.00000  942.00000  848.00000 1088.00000  992.00000  450.00000
## S.chao1  1377.95000 1397.55469 1155.65000 1517.36486 1287.23077  731.44286
## se.chao1   60.96351   67.07142   47.65508   61.13897   44.73270   54.74508
## S.ACE    1365.23150 1356.87666 1181.99016 1480.20490 1295.47022  759.36594
## se.ACE     19.72705   19.88369   18.05970   20.02279   18.55676   16.47752
##          Isa_120423 Isa_120426 Isa_120430 Isa_120503 Isa_120507 Isa_120508
## S.obs     384.00000  374.00000  304.00000  420.00000  272.00000 219.000000
## S.chao1   693.25532  511.60000  438.63830  654.23077  447.77778 324.636364
## se.chao1   67.46164   32.24236   34.05155   48.23404   45.97426  31.165443
## S.ACE     652.06713  539.26272  435.54374  684.48661  432.26064 329.851638
## se.ACE     14.78726   12.98133   10.86998   15.37044   11.73649   9.569067
##          Isa_120509 Isa_120510 Isa_120516 Isa_120524 Isa_120621 Isa_120706
## S.obs    205.000000  224.00000  252.00000  335.00000  417.00000  414.00000
## S.chao1  363.052632  371.17143  369.00000  528.75000  618.88235  582.52174
## se.chao1  51.971891   40.07574   33.24605   48.16268   46.10172   36.35228
## S.ACE    304.844016  395.87277  364.99135  514.52628  590.13947  615.28756
## se.ACE     9.305907   12.00009   10.37553   12.39624   13.01145   13.57143
##          Isa_120806
## S.obs     605.00000
## S.chao1   873.07692
## se.chao1   50.85980
## S.ACE     851.28562
## se.ACE     15.83698
```

Above we have the estimators Chao and ACE as well as the species number. What do the numbers mean?

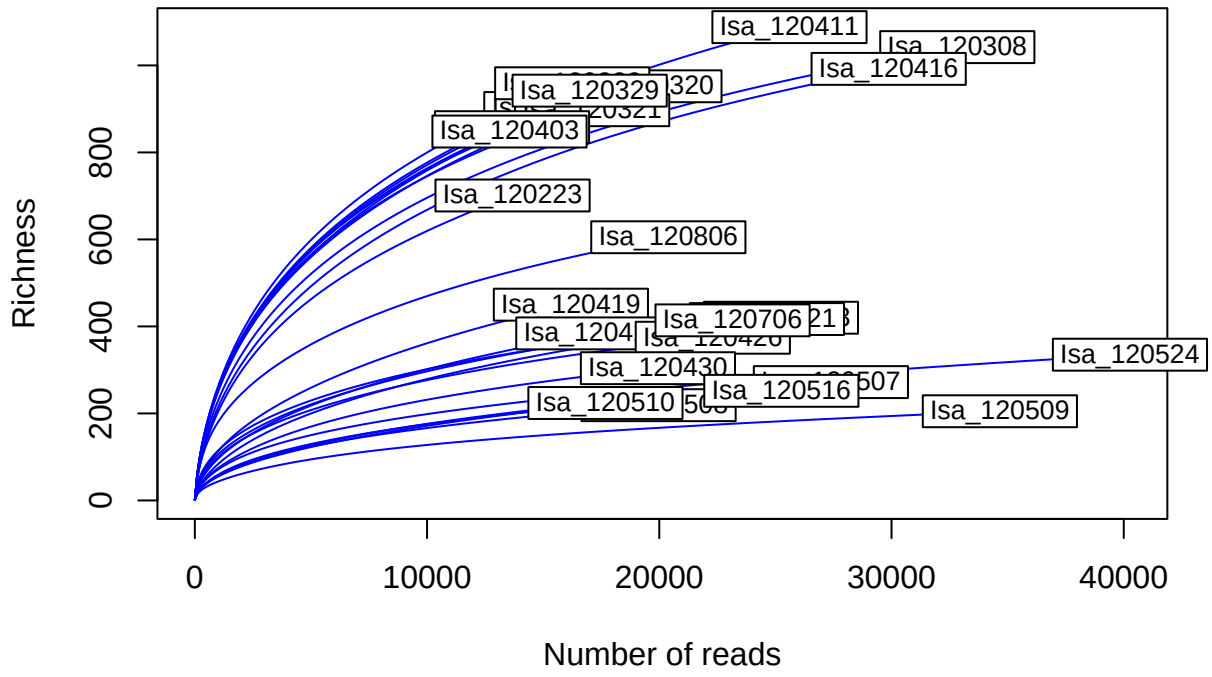
Rarefaction

Let's calculate the number of reads per sample as reefaction curves:

```
rowSums(otu.tab.trans)
```

```
## Isa_120223 Isa_120301 Isa_120308 Isa_120320 Isa_120321 Isa_120322 Isa_120323
##      13682      15783      32833      19361      17110      13658      16251
## Isa_120329 Isa_120403 Isa_120411 Isa_120416 Isa_120419 Isa_120423 Isa_120426
##      17002      13551      25606      29877      16194      17161      22305
## Isa_120430 Isa_120503 Isa_120507 Isa_120508 Isa_120509 Isa_120510 Isa_120516
##      19942      25237      27389      19969      34666      17682      25256
## Isa_120524 Isa_120621 Isa_120706 Isa_120806
##      40267      24636      23156      20390
```

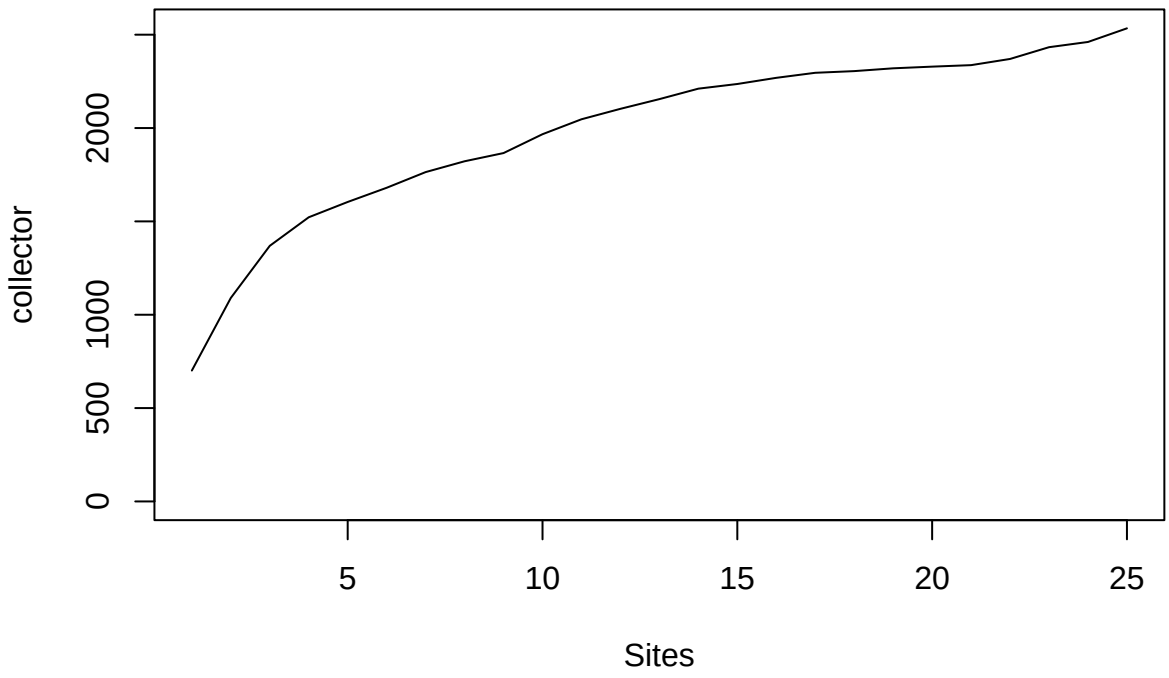
```
rarecurve(otu.tab.trans, step = 100, xlab = "Number of reads", ylab = "Richness", col = "blue")
```



How do you interpret these curves? Which samples have the lowest number of total reads? Which are the highest?

Accumulation curves

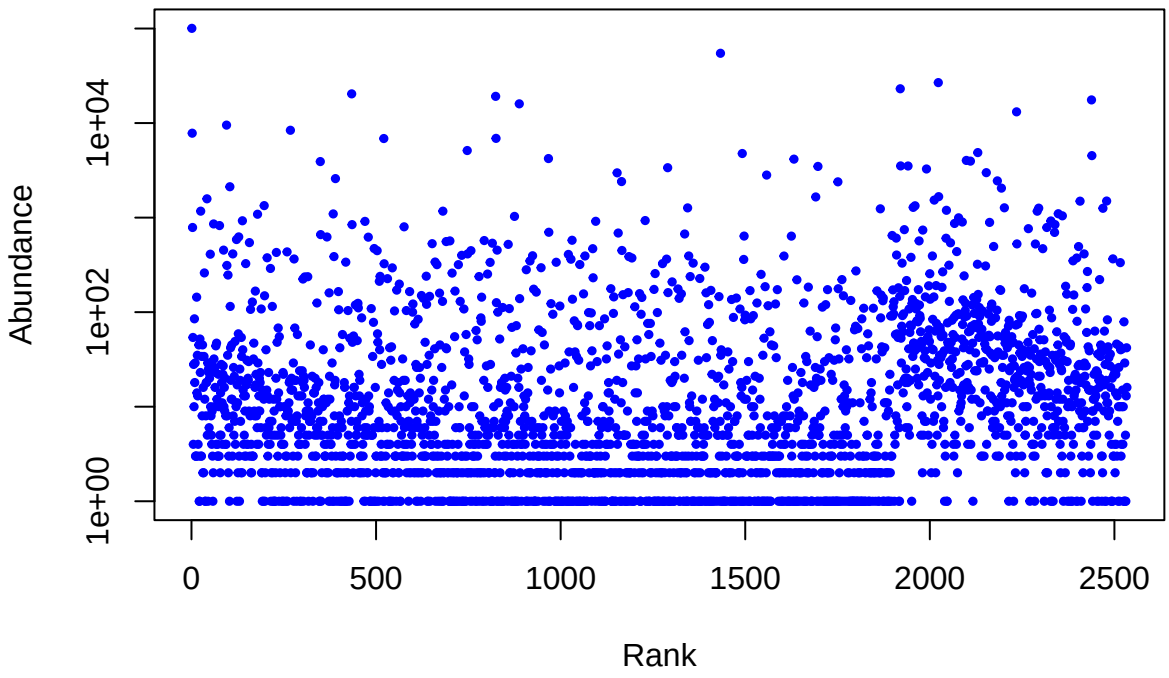
```
accum.curve <- specaccum(otu.tab.trans, method = "collector")
plot(accum.curve)
```



What does this curve represent? How do you interpret it?

Evenness

```
plot(colSums(otu.tab.trans), log = "y", xlab = "Rank", ylab = "Abundance", pch = 19, cex = 0.5, col = "
```



Fitting rank-abundance distribution models to the data

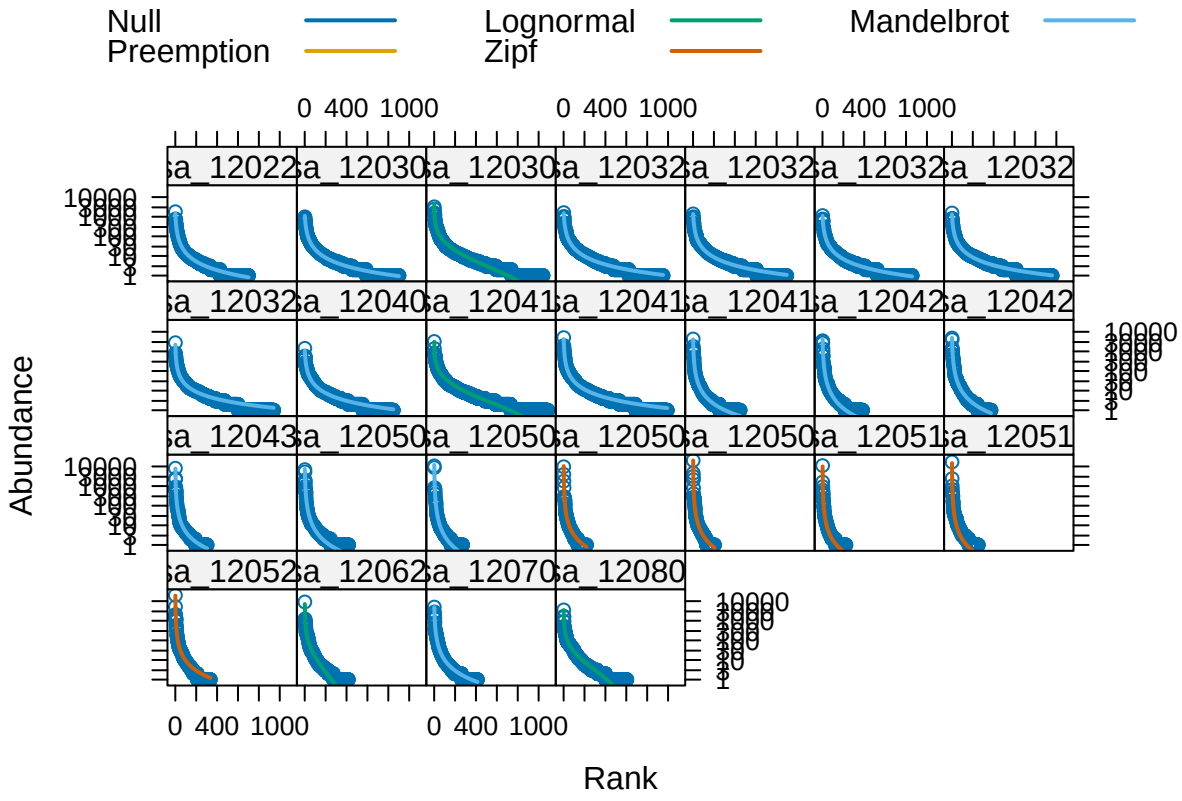
```
mod <- radfit(otu.tab.trans)
```

```
## Error in glm.fit(x = structure(c(1, 1, 1, 1, 1, 1, 1, 1, 1, 1, 1, 1, 1,  :
##   NA/NaN/Inf in 'x'
```

```
## Warning: glm.fit: algorithm did not converge
```

```
## Warning: glm.fit: algorithm did not converge
```

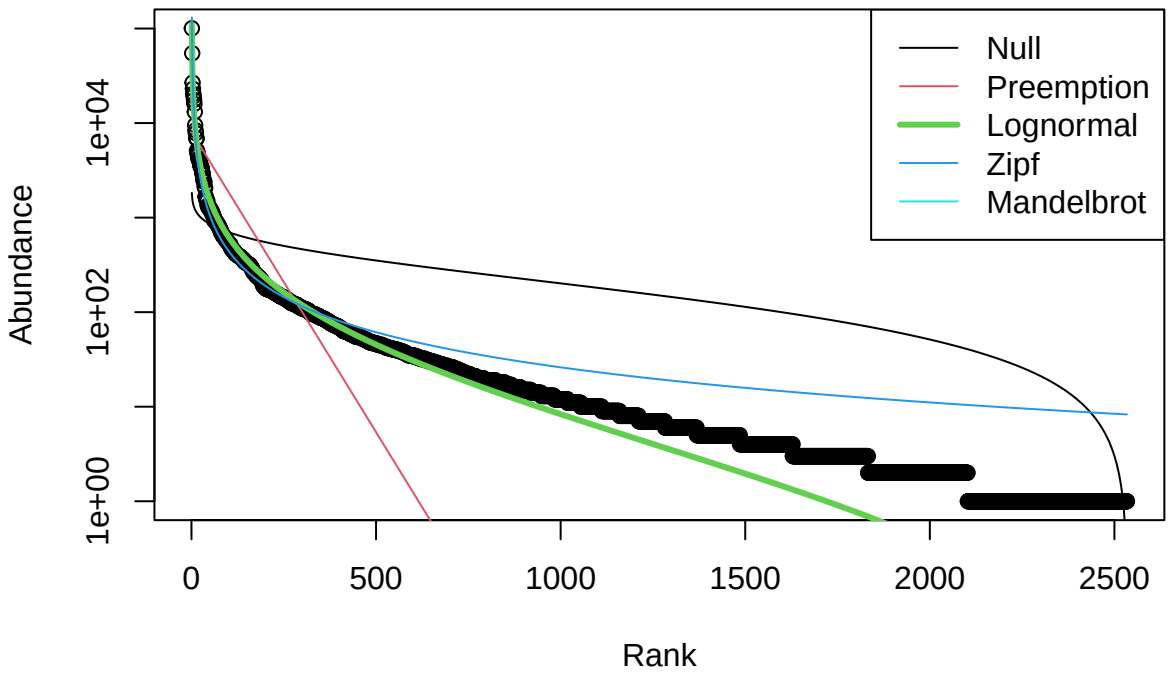
```
plot(mod)
```




```
mod.all <- radfit(colSums(otu.tab.trans))
```

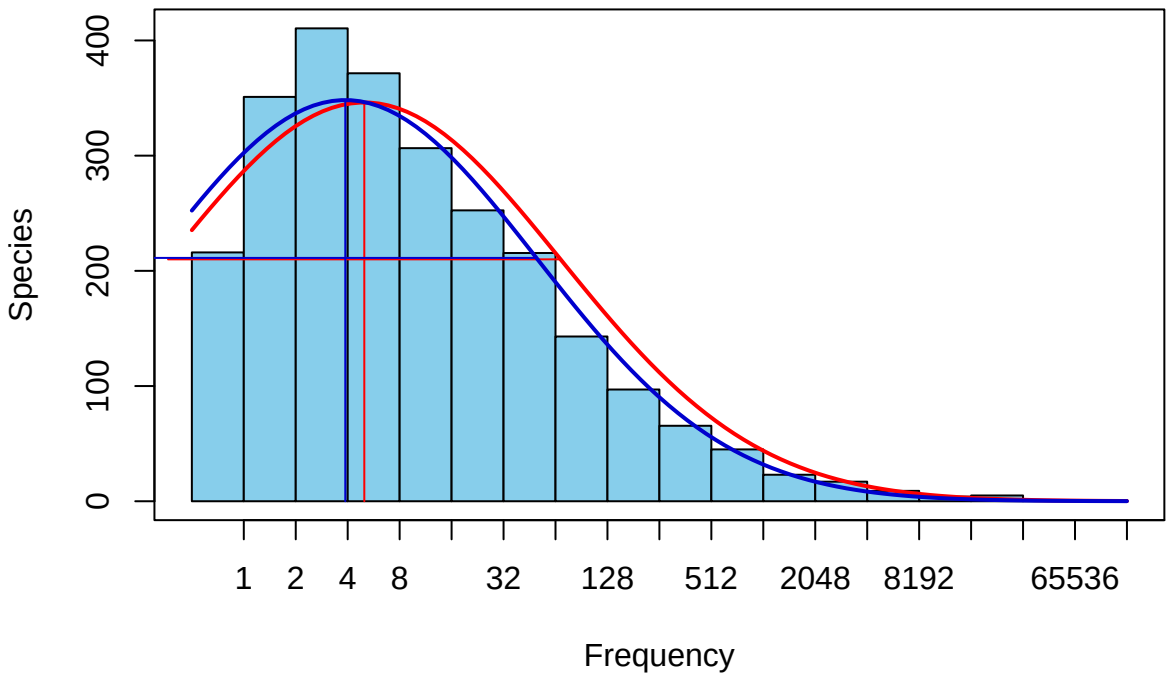
```
## Error in glm.fit(x = structure(c(1, 1, 1, 1, 1, 1, 1, 1, 1, 1, 1, 1, 1,  :
##   NA/NaN/Inf in 'x'
```

```
plot(mod.all)
```



Fitting data to the Preston model

```
preston <- prestonfit(colSums(otu.tab.trans))
preston.dist <- prestondistr(colSums(otu.tab.trans))
plot(preston)
lines(preston.dist, line.col = "blue3")
```



Extrapolated richness

```
veiledspec(preston)
```

```
## Extrapolated     Observed       Veiled
##    3278.6837    2534.0000     744.6837
```

```
veiledspec(preston.dist)
```

```
## Extrapolated     Observed       Veiled
##    3211.9212    2534.0000     677.9212
```

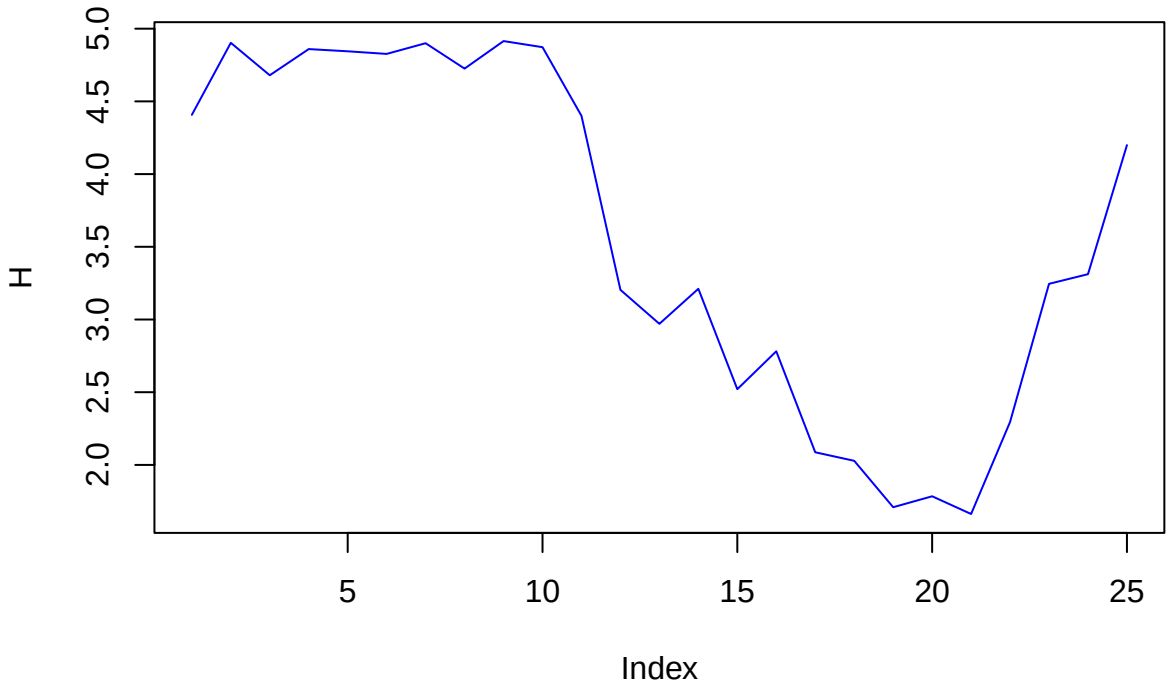
Shannon H index (considers richness and evenness)

```
H <- diversity(otu.tab.trans, index = "shannon")
H
```

```
## Isa_120223 Isa_120301 Isa_120308 Isa_120320 Isa_120321 Isa_120322 Isa_120323
##   4.407409   4.902868   4.679546   4.859799   4.844499   4.826550   4.900137
## Isa_120329 Isa_120403 Isa_120411 Isa_120416 Isa_120419 Isa_120423 Isa_120426
```

```
##   4.725430   4.914670   4.872817   4.400562   3.203907   2.970289   3.211268
## Isa_120430 Isa_120503 Isa_120507 Isa_120508 Isa_120509 Isa_120510 Isa_120516
##   2.520753   2.781324   2.086866   2.028217   1.709035   1.784380   1.662585
## Isa_120524 Isa_120621 Isa_120706 Isa_120806
##   2.295421   3.245357   3.311757   4.199123
```

```
plot(H, type = "l", col = "blue")
```



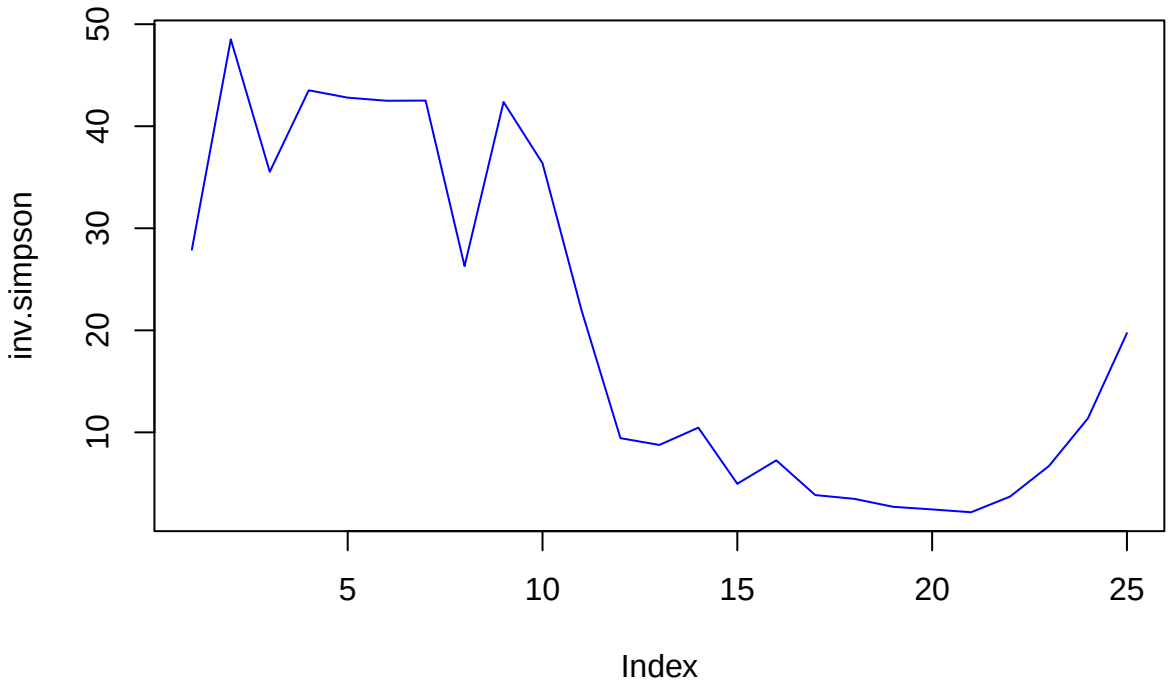
Pielou's index of evenness (range 0-1, 1 = maximum evenness)

```
J=H/Hmax
J=Shannon (H) / log(S=species richness)
```

```
J <- H / log(rowSums(otu.tab.trans > 0))
```

Inverse Simpson's D index (richness+evenness. Larger values, larger diversity)

```
inv.simpson <- diversity(otu.tab.trans, "invsimpson")
plot(inv.simpson, type = "l", col = "blue")
```



Beta diversity

We rarefy all samples to the same sequencing depth, to reduce biases.

```
min(rowSums(otu.tab.trans)) # We calculate the sample with the minimum amount of reads
```

```
## [1] 13551
```

```
otu.tab.trans.ss <- rrarefy(otu.tab.trans, min(rowSums(otu.tab.trans))) # Samples are rarefied to lowes
rowSums(otu.tab.trans.ss)
```

```
## Isa_120223 Isa_120301 Isa_120308 Isa_120320 Isa_120321 Isa_120322 Isa_120323
##      13551      13551      13551      13551      13551      13551      13551
## Isa_120329 Isa_120403 Isa_120411 Isa_120416 Isa_120419 Isa_120423 Isa_120426
##      13551      13551      13551      13551      13551      13551      13551
## Isa_120430 Isa_120503 Isa_120507 Isa_120508 Isa_120509 Isa_120510 Isa_120516
##      13551      13551      13551      13551      13551      13551      13551
## Isa_120524 Isa_120621 Isa_120706 Isa_120806
##      13551      13551      13551      13551
```

What is the number of reads these samples have been rarified to? What does it imply, do you understand how it is done?

Check that the number of OTUs are the same in the new table

```
dim(otu.tab.trans)
```

```
## [1]   25 2534
```

```
dim(otu.tab.trans.ss)
```

```
## [1]   25 2534
```

The tables have the same size, but, after removing reads, OTUs might be left with zero read abundance. These are typically those with very low abundace to begin with, and (hopefully) do not play an important in the system we are studing.

```
length(which(colSums(otu.tab.trans) == 0))
```

```
## [1] 0
```

```
length(which(colSums(otu.tab.trans.ss) == 0))
```

```
## [1] 195
```

```
head(which(colSums(otu.tab.trans.ss) == 0)) # Show the OTUs and the position in the table that have 0 a
```

```
## OTU1016 OTU1017 OTU1212 OTU1213 OTU1240 OTU1250
##      21      22     191     192     215     223
```

- *How many OTUs are empty after rarefaction?*

We can compare the number of reads for a selected OTU (in this case the 13th OTU in the list) between the origianl and the subsampled OTU table:

```
colnames(otu.tab.trans)[13]
```

```
## [1] "OTU1009"
```

```
otu.tab.trans[, 13] # This gives the abundance of the OTU1009  across the different samples in the tabl
```

```
## Isa_120223 Isa_120301 Isa_120308 Isa_120320 Isa_120321 Isa_120322 Isa_120323
##          0          0          0          0          0          0          0
## Isa_120329 Isa_120403 Isa_120411 Isa_120416 Isa_120419 Isa_120423 Isa_120426
##          0          0          0          0          0          0          0
## Isa_120430 Isa_120503 Isa_120507 Isa_120508 Isa_120509 Isa_120510 Isa_120516
##          0          0          0          0          0          0          0
## Isa_120524 Isa_120621 Isa_120706 Isa_120806
##          0          0          0          3
```

```
otu.tab.trans.ss[, 13] # # This gives the abundance of the OTU1009  across the different samples in the
```

```
## Isa_120223 Isa_120301 Isa_120308 Isa_120320 Isa_120321 Isa_120322 Isa_120323
##          0          0          0          0          0          0          0
## Isa_120329 Isa_120403 Isa_120411 Isa_120416 Isa_120419 Isa_120423 Isa_120426
##          0          0          0          0          0          0          0
## Isa_120430 Isa_120503 Isa_120507 Isa_120508 Isa_120509 Isa_120510 Isa_120516
##          0          0          0          0          0          0          0
## Isa_120524 Isa_120621 Isa_120706 Isa_120806
##          0          0          0          2
```

We can remove the OTUs with zero abundance with a similar command as we used at the beginning of the lab:

```
otu.tab.trans.ss.nozero <- otu.tab.trans.ss[, -(which(colSums(otu.tab.trans.ss) == 0))] # Removes OTUs
length(which(colSums(otu.tab.trans.ss.nozero) == 0)) # Check that no zero abundance OTUs are left
```

```
## [1] 0
```

Let's check dimensions of the tables:

```
dim(otu.tab.trans.ss)
```

```
## [1]   25 2534
```

```
dim(otu.tab.trans.ss.nozero)
```

```
## [1]   25 2339
```

-How many OTUs gave been removed?
There are other ways to transform and normalise the data, but we will not go into the details in this course. Here's an example for those interested:

> Replace zeros (problems with log calculations) with pseudo-counts
>
> ```
> otu.tab.trans.gbm <- zCompositions::cmultRepl(t(otu.tab.trans), output = "p-counts")
> ```
>
> ```
> ## Warning in zCompositions::cmultRepl(t(otu.tab.trans), output = "p-counts"): Column 12 containing
> ## Column 13 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ## Column 14 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ## Column 15 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ## Column 16 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ## Column 17 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ## Column 18 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ## Column 19 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ## Column 20 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ## Column 21 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ## Column 22 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ## Column 23 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ## Column 24 containing more than 80% zeros/unobserved values was deleted (pre-check out using func
> ```

```
## Warning in zCompositions::cmultRepl(t(otu.tab.trans), output = "p-counts"): Row 5 containing mor
## Row 12 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 13 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 21 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 22 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 31 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
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## Row 37 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 38 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 45 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 55 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 57 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 58 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 59 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 69 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 71 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 78 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 89 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 92 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 93 containing more than 80% zeros/unobserved values was deleted (pre-check out using functio
## Row 100 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 101 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 102 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 103 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 104 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 111 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 113 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 117 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 118 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 119 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 120 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 123 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 126 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 130 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 133 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 134 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 136 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 137 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 140 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 141 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 146 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 148 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 151 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 152 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 158 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 159 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 161 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 163 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 166 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 167 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 168 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 170 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 182 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
## Row 187 containing more than 80% zeros/unobserved values was deleted (pre-check out using functi
```

```
## No. adjusted imputations:  5870
```

```
otu.tab.trans.gbm[1:5, 1:5] # We have a look to the replaced values
```

```
##           Isa_120223 Isa_120301 Isa_120308 Isa_120320 Isa_120321
## OTU1    446.00000000        149        321        204        442
## OTU10   668.00000000        551       1462        896        646
## OTU100   21.00000000         23         99         75         61
## OTU1000   0.01660788          1         11          1          4
## OTU1002   0.01012178          2          7          2          2
```

centered log-ratio (clr) transformation

```
otu.tab.trans.gbm.clr <- compositions::clr(otu.tab.trans.gbm) # We apply a centered log-ratio (clr)
otu.tab.trans.gbm.clr[1:5, 1:5] # Values now look different than counts.
```

```
##          Isa_120223 Isa_120301 Isa_120308 Isa_120320 Isa_120321
## OTU1      0.4060384 -0.6903342 0.07716061 -0.3761605  0.3970294
## OTU10     0.2201524  0.0275990 1.00342483  0.5138046  0.1866637
## OTU100   -0.8803293 -0.7893575 0.67026812  0.3926364  0.1860221
## OTU1000 -4.8510572 -0.7531793 1.64471597 -0.7531793  0.6331151
## OTU1002 -4.2328733  1.0533392 2.30610215  1.0533392  1.0533392
```

Distance metrics

Let's calculate the Bray Curtis dissimilarities for the rarefied dataset

```
otu.tab.trans.ss.nozero.bray <- vegdist(otu.tab.trans.ss.nozero, method = "bray")
as.matrix(otu.tab.trans.ss.nozero.bray)[1:5, 1:5]
```

```
##             Isa_120223 Isa_120301 Isa_120308 Isa_120320 Isa_120321
## Isa_120223  0.0000000  0.3078002  0.2692052  0.2929673  0.2954763
## Isa_120301  0.3078002  0.0000000  0.2426389  0.2056675  0.2056675
## Isa_120308  0.2692052  0.2426389  0.0000000  0.2666224  0.2876540
## Isa_120320  0.2929673  0.2056675  0.2666224  0.0000000  0.1408752
## Isa_120321  0.2954763  0.2056675  0.2876540  0.1408752  0.0000000
```

- *What kind of data is the Bray-Curtis dissimilarity suitable for?*
- *How can you change the dissimilarity index in the previous chunk of code?*

Phew That was Part I. Now before you have a break save the data so it can be loaded if you want to use some of the same data for the next session. This way you don't have to redo all analysis for the next lab.

```
save.image("AB332_lab_I.RData")
```