# 230306\_Reporte1Exploracion

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```
library("phyloseq")
library("ggplot2")
library("vegan")

## Loading required package: permute

## Loading required package: lattice

## This is vegan 2.6-4

library("RColorBrewer")
library("stringi")
```

#### Cargado de datos originales

```
setwd("/home/camila/GIT/Tesis_Maestria/Data/fresa_solena/Data_all")
fresa_kraken <- import_biom("fresa_kraken_all.biom")</pre>
class(fresa kraken)
## [1] "phyloseq"
## attr(,"package")
## [1] "phyloseq"
## como tenemos diferencias de longitud en los nombres de las muestras, al cortar los nombres, quedan l
## usando lalibreria stringi
sample_names(fresa_kraken)<-stri_replace_all_regex(sample_names(fresa_kraken),'\\.','A')</pre>
## renombrar las columnas del tax_table
colnames(fresa_kraken@tax_table@.Data) <- c("Kingdom", "Phylum", "Class", "Order", "Family", "Genus", "</pre>
## quitar los primeros caracteres de los nombres del tax_table
fresa_kraken@tax_table@.Data <- substr(fresa_kraken@tax_table@.Data,4,100)</pre>
## recortar los nombres de las muestras
colnames(fresa_kraken@otu_table@.Data) <- substr(colnames(fresa_kraken@otu_table@.Data),1,6)</pre>
## cargar los metadatos
metadata_fresa <- read.csv2("/home/camila/GIT/Tesis_Maestria/Data/fresa_solena/Data_all/metadata.csv",h</pre>
rownames(metadata_fresa) <- sample_names(fresa_kraken)</pre>
## unir los metadatos al objeto phyloseq
fresa_kraken@sam_data <- sample_data(metadata_fresa)</pre>
```

```
# filtro de calidad
samples_to_remove <- c("MP2079","MP2080","MP2088","MP2109","MP2137")
samples<-!(sample_names(fresa_kraken) %in% samples_to_remove)
names(samples)<-as.character(sample_names(fresa_kraken))
fresa_kraken_fil <- prune_samples(samples, fresa_kraken)
nsamples(fresa_kraken) # 85

## [1] 85

msamples(fresa_kraken_fil) # 80</pre>
## [1] 80
```

### Subconjunto de "Eukaryota"

```
merge_Eukaryota<-subset_taxa(fresa_kraken_fil,Kingdom=="Eukaryota")</pre>
```

### Subconjunto de "Bacteria"

```
merge_Bacteria<-subset_taxa(fresa_kraken_fil,Kingdom=="Bacteria")</pre>
```

### **Funciones**

Crea los subconjuntos de datos input phy phyliseq total tax rango al que queremos recortar output

```
glomToGraph<-function(phy,tax){
    ## creamos el subconjunto dependiendo del linaje taxonomico deseado
    glom <- tax_glom(phy, taxrank = tax)
    ## sacamos los porcentajes
    percentages <- transform_sample_counts(glom, function(x) x*100 / sum(x) )
    percentages_df <- psmelt(percentages)
    return(list(glom,percentages,percentages_df))
}</pre>
```

#### Graficar abundancias stackbar

input entra el percentages df

```
Abundance_barras <- function(phy,tax,attribute,abundance_percentage){
    ##llamar funcion de datos
    Data <- glomToGraph(phy,tax)
    glom <- Data[[1]] #phyloseq
    percentages <- Data[[2]] #phyloseq
    percentages_df <- Data[[3]] # dataframe
    ## Graficamos para cada subconjunto las barras de abundancia
```

```
plot_barras <- ggplot(data=percentages_df, aes_string(x='Sample', y='Abundance', fill=tax ,color=attr
  scale_colour_manual(values=c('white','black')) +
  geom_bar(aes(), stat="identity", position="stack") +
  labs(title = "Abundance", x='Sample', y='Abundance', color = tax) +
  theme(legend.key.size = unit(0.2, "cm"),
        legend.key.width = unit(0.25, "cm"),
        legend.position = "bottom",
        legend.direction = "horizontal",
        legend.title=element_text(size=8, face = "bold"),
        legend.text=element_text(size=6),
        text = element_text(size=12),
        axis.text.x = element_text(angle=90, size=5, hjust=1, vjust=0.5))
percentages_df$tax<-percentages_df[,ncol(percentages_df)]</pre>
percentages_df$tax[percentages_df$Abundance < abundance_percentage] <- "abundance_percentage"
percentages_df$tax <- as.factor(percentages_df$tax)</pre>
plot_percentages <- ggplot(data=percentages_df, aes_string(x='Sample', y='Abundance', fill='tax', col
  scale_colour_manual(values=c('white','black')) +
  geom_bar(aes(), stat="identity", position="stack") +
 labs(title = "Abundance", x='Sample', y='Abundance', color = tax) +
  theme(legend.key.size = unit(0.3, "cm"),
        legend.key.width = unit(0.5, "cm"),
        legend.position = "bottom",
        legend.direction = "horizontal",
        legend.title=element_text(size=10, face = "bold"),
        legend.text=element_text(size=8),
        text = element_text(size=12),
        axis.text.x = element_text(angle=90, size=5, hjust=1, vjust=0.5))
return(list(plot_barras,plot_percentages))
```

### Graficar betadiversity

```
Beta_diversity <- function(phy,tax,attribute,distance) {
   Data <- glomToGraph(phy,tax)
   glom <- Data[[1]]
#CREAR UN GLOM AL 10%
   percentages <- Data[[2]]
   percentages_df <- Data[[3]]
## Beta diversidad
meta_ord <- ordinate(physeq = percentages, method = "NMDS", distance = distance)
   plot_beta <- plot_ordination(physeq = percentages, ordination = meta_ord, color = attribute) +
        geom_text(mapping = aes(label = colnames(phy@otu_table@.Data)), size = 3, vjust = 1.5)
   return(plot_beta)
}</pre>
```

### Graficar alphadiversity

```
Alpha_diversity <- function(phy,tax,attribute){
## llamamos la funcion que crea los dataset
```

```
Data <- glomToGraph(phy,tax)
glom <- Data[[1]]

percentages <- Data[[2]]
percentages_df <- Data[[3]]
## Alfa diversidad
plot_alpha <- plot_richness(physeq = glom, measures = c("Observed", "Chao1", "Shannon", "simpson"), x = a
return(plot_alpha)
}

# Eukarya by Phylum

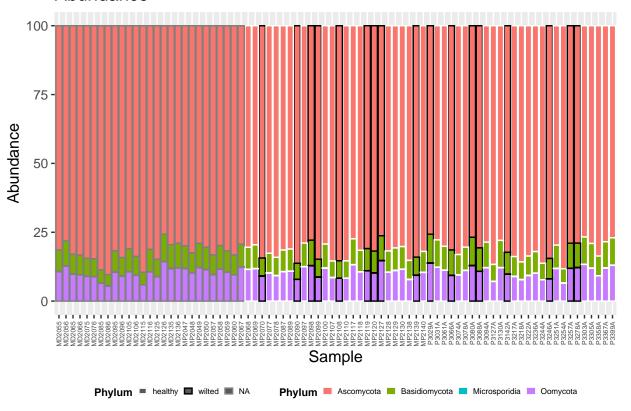
Barras_Phylum <- Abundance_barras(merge_Eukaryota, 'Phylum' , 'Treatment', 10.0)

## Warning: 'aes_string()' was deprecated in ggplot2 3.0.0.
## i Please use tidy evaluation ideoms with 'aes()'

Barras_Phylum[1] # normal</pre>
```

## [[1]]

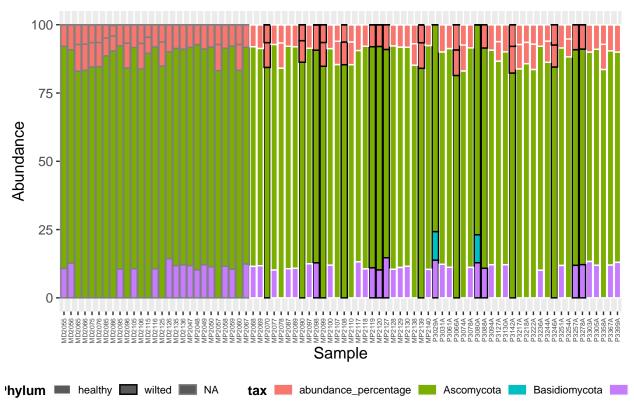
# Abundance



Barras\_Phylum[2]

#### ## [[1]]

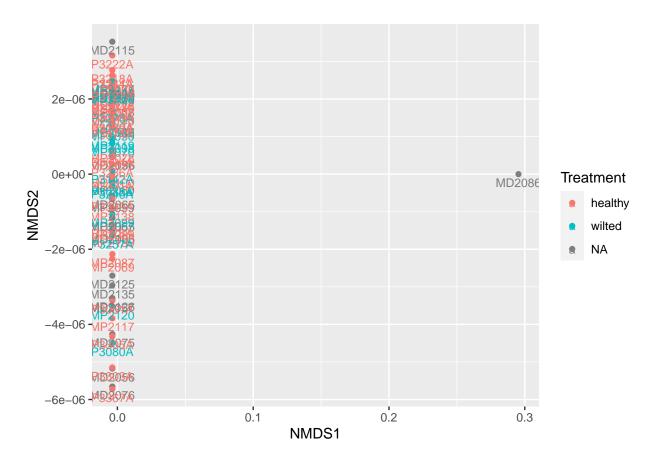
### Abundance



Beta\_diversity(merge\_Eukaryota , 'Phylum' , 'Treatment', 'bray')

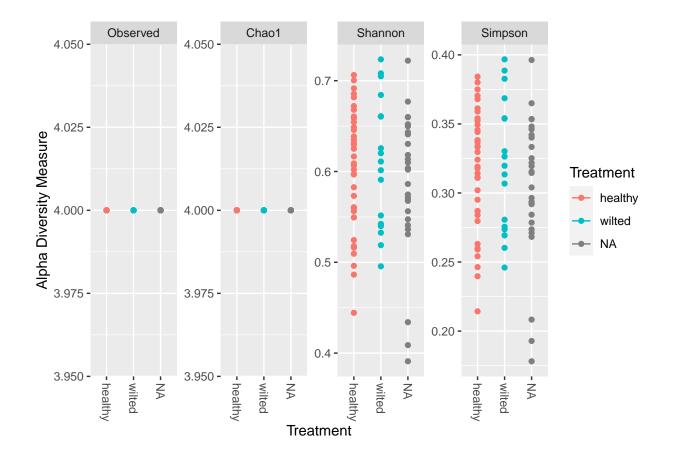
```
## Square root transformation
## Wisconsin double standardization
## Run 0 stress 0.01092347
## Run 1 stress 9.734801e-05
  ... New best solution
  ... Procrustes: rmse 0.02230386 max resid 0.05599817
## Run 2 stress 9.871748e-05
  ... Procrustes: rmse 0.0001228726 max resid 0.0003500669
## ... Similar to previous best
## Run 3 stress 9.819744e-05
## ... Procrustes: rmse 9.857134e-05 max resid 0.0002705246
## ... Similar to previous best
## Run 4 stress 0.001167197
## Run 5 stress 0.0001449023
## ... Procrustes: rmse 0.0002674023 max resid 0.0006336985
## ... Similar to previous best
## Run 6 stress 9.814744e-05
## ... Procrustes: rmse 2.469828e-06 max resid 1.147763e-05
## ... Similar to previous best
## Run 7 stress 5.445962e-05
## ... New best solution
```

```
## ... Procrustes: rmse 0.0001020114 max resid 0.0002247589
## ... Similar to previous best
## Run 8 stress 9.522178e-05
## ... Procrustes: rmse 2.77495e-05 max resid 5.373292e-05
## ... Similar to previous best
## Run 9 stress 9.928758e-05
## ... Procrustes: rmse 9.8982e-05 max resid 0.0002412854
## ... Similar to previous best
## Run 10 stress 9.567226e-05
## ... Procrustes: rmse 9.071014e-05 max resid 0.0002201253
## ... Similar to previous best
## Run 11 stress 9.546742e-05
## ... Procrustes: rmse 0.000115702 max resid 0.0002830266
## ... Similar to previous best
## Run 12 stress 9.916533e-05
## ... Procrustes: rmse 8.248923e-05 max resid 0.0001793601
## ... Similar to previous best
## Run 13 stress 6.239085e-05
## ... Procrustes: rmse 1.963294e-05 max resid 4.511147e-05
## ... Similar to previous best
## Run 14 stress 9.930775e-05
## ... Procrustes: rmse 6.361762e-05 max resid 0.0001497356
## ... Similar to previous best
## Run 15 stress 0.0001609772
## ... Procrustes: rmse 0.0003290465 max resid 0.000790323
## ... Similar to previous best
## Run 16 stress 0.0001376356
## ... Procrustes: rmse 0.0002811157 max resid 0.0006678026
## ... Similar to previous best
## Run 17 stress 9.688643e-05
## ... Procrustes: rmse 8.353791e-05 max resid 0.0001828628
## ... Similar to previous best
## Run 18 stress 4.265144e-05
## ... New best solution
## ... Procrustes: rmse 1.546682e-05 max resid 4.158713e-05
## ... Similar to previous best
## Run 19 stress 9.586864e-05
## ... Procrustes: rmse 2.010005e-05 max resid 0.0001050013
## ... Similar to previous best
## Run 20 stress 9.040026e-05
## ... Procrustes: rmse 3.165034e-05 max resid 8.116249e-05
## ... Similar to previous best
## *** Best solution repeated 3 times
## Warning in metaMDS(veganifyOTU(physeq), distance, ...): stress is (nearly) zero:
## you may have insufficient data
```



```
Alpha_diversity(merge_Eukaryota , 'Phylum' , 'Treatment')
```

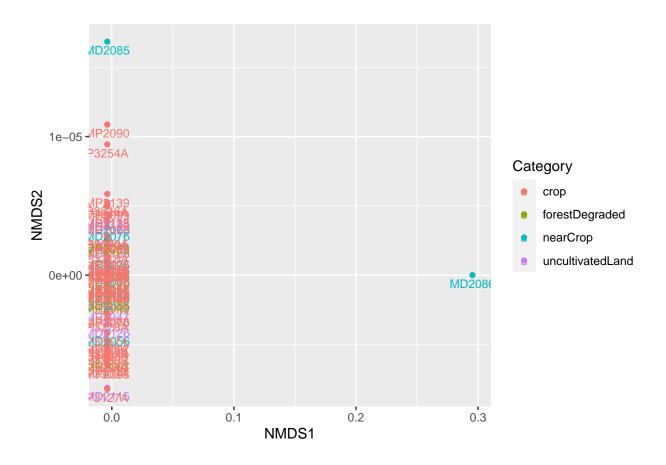
```
## Warning in estimate_richness(physeq, split = TRUE, measures = measures): The data you have provided
## any singletons. This is highly suspicious. Results of richness
## estimates (for example) are probably unreliable, or wrong, if you have already
## trimmed low-abundance taxa from the data.
##
## We recommended that you find the un-trimmed data and retry.
```



Beta\_diversity(merge\_Eukaryota , 'Phylum' , 'Category', 'bray')

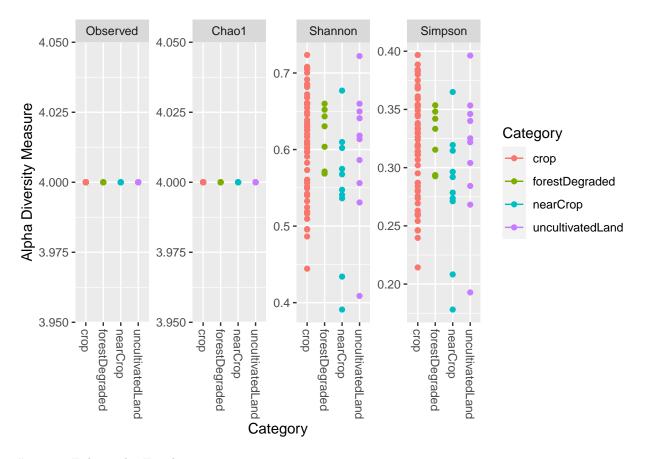
```
## Square root transformation
## Wisconsin double standardization
## Run 0 stress 0.01092347
## Run 1 stress 9.335705e-05
## ... New best solution
## ... Procrustes: rmse 0.0223114 max resid 0.05607995
## Run 2 stress 9.05933e-05
## ... New best solution
## ... Procrustes: rmse 9.334717e-05 max resid 0.0002510353
## ... Similar to previous best
## Run 3 stress 8.295263e-05
## ... New best solution
## ... Procrustes: rmse 0.0001070632 max resid 0.0002674128
## ... Similar to previous best
## Run 4 stress 9.459767e-05
## ... Procrustes: rmse 2.538734e-05 max resid 7.767238e-05
## ... Similar to previous best
## Run 5 stress 6.333525e-05
## ... New best solution
## ... Procrustes: rmse 2.811014e-05 max resid 7.472418e-05
## ... Similar to previous best
## Run 6 stress 8.947329e-05
## ... Procrustes: rmse 3.872243e-05 max resid 0.0001050772
```

```
## ... Similar to previous best
## Run 7 stress 9.420623e-05
## ... Procrustes: rmse 4.92327e-05 max resid 0.000135095
## ... Similar to previous best
## Run 8 stress 9.691644e-05
## ... Procrustes: rmse 0.0001108939 max resid 0.0002399508
## ... Similar to previous best
## Run 9 stress 9.770556e-05
## ... Procrustes: rmse 9.135358e-05 max resid 0.0001946187
## ... Similar to previous best
## Run 10 stress 6.32783e-05
## ... New best solution
## ... Procrustes: rmse 2.9795e-05 max resid 0.0001080258
## ... Similar to previous best
## Run 11 stress 9.297361e-05
## ... Procrustes: rmse 8.763944e-05 max resid 0.0002378802
## ... Similar to previous best
## Run 12 stress 9.554237e-05
## ... Procrustes: rmse 6.618609e-05 max resid 0.0002140865
## ... Similar to previous best
## Run 13 stress 9.838965e-05
## ... Procrustes: rmse 6.238972e-05 max resid 0.000184277
## ... Similar to previous best
## Run 14 stress 9.005017e-05
## ... Procrustes: rmse 5.407641e-05 max resid 0.0001296094
## ... Similar to previous best
## Run 15 stress 9.72548e-05
## ... Procrustes: rmse 6.160573e-05 max resid 0.0002030988
## ... Similar to previous best
## Run 16 stress 9.854852e-05
## ... Procrustes: rmse 3.23828e-05 max resid 8.60709e-05
## ... Similar to previous best
## Run 17 stress 7.382101e-05
## ... Procrustes: rmse 3.769402e-05 max resid 0.0001091618
## ... Similar to previous best
## Run 18 stress 9.911898e-05
## ... Procrustes: rmse 0.0001054754 max resid 0.0003025607
## ... Similar to previous best
## Run 19 stress 9.742706e-05
## ... Procrustes: rmse 3.960747e-05 max resid 7.743144e-05
## ... Similar to previous best
## Run 20 stress 9.181024e-05
## ... Procrustes: rmse 6.712639e-05 max resid 0.0002175394
## ... Similar to previous best
## *** Best solution repeated 11 times
## Warning in metaMDS(veganifyOTU(physeq), distance, ...): stress is (nearly) zero:
## you may have insufficient data
```



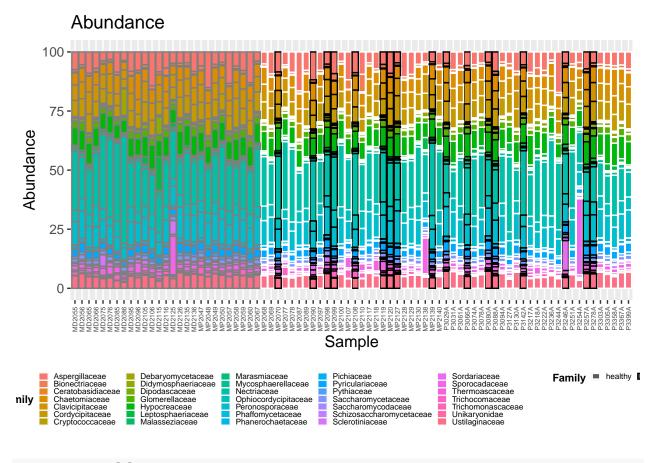
```
Alpha_diversity(merge_Eukaryota , 'Phylum' , 'Category')
```

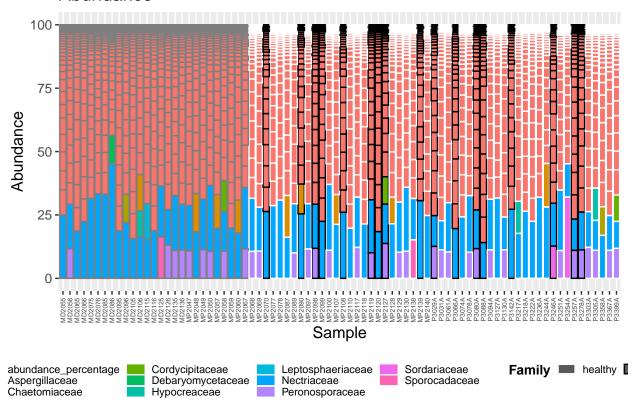
```
## Warning in estimate_richness(physeq, split = TRUE, measures = measures): The data you have provided
## any singletons. This is highly suspicious. Results of richness
## estimates (for example) are probably unreliable, or wrong, if you have already
## trimmed low-abundance taxa from the data.
##
## We recommended that you find the un-trimmed data and retry.
```



#——Eukarya by Family

Barras\_Species <- Abundance\_barras(merge\_Eukaryota, 'Family', 'Treatment', 10.0)
Barras\_Species[1]</pre>





### Beta\_diversity(merge\_Eukaryota , 'Family' , 'Treatment', 'bray')

```
## Wisconsin double standardization
## Run 0 stress 0.136765
## Run 1 stress 0.1371023
## ... Procrustes: rmse 0.008825574 max resid 0.07706356
## Run 2 stress 0.1375698
## Run 3 stress 0.1367639
## ... New best solution
  ... Procrustes: rmse 0.001002787 max resid 0.00643578
  ... Similar to previous best
## Run 4 stress 0.1382601
## Run 5 stress 0.1414158
## Run 6 stress 0.1424672
## Run 7 stress 0.1448042
## Run 8 stress 0.1443076
## Run 9 stress 0.1370173
  ... Procrustes: rmse 0.01446446 max resid 0.1233838
## Run 10 stress 0.1456562
## Run 11 stress 0.1388323
## Run 12 stress 0.155677
## Run 13 stress 0.1367785
  ... Procrustes: rmse 0.001519871 max resid 0.00796185
## ... Similar to previous best
## Run 14 stress 0.1412243
```

```
## Run 15 stress 0.1371028

## ... Procrustes: rmse 0.008985293 max resid 0.07707036

## Run 16 stress 0.1376853

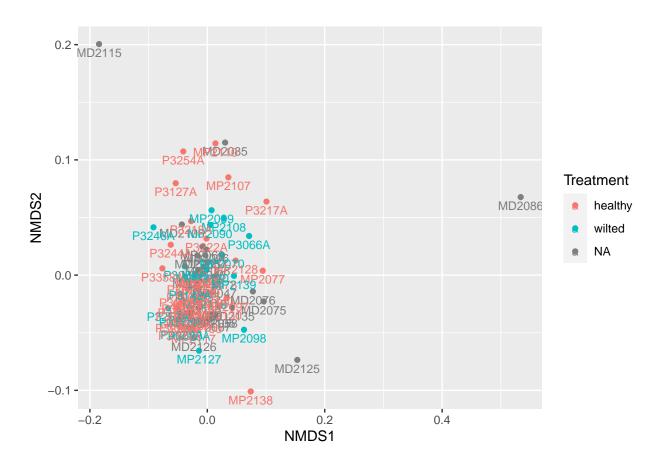
## Run 17 stress 0.152514

## Run 18 stress 0.4090803

## Run 19 stress 0.1441295

## Run 20 stress 0.1388286

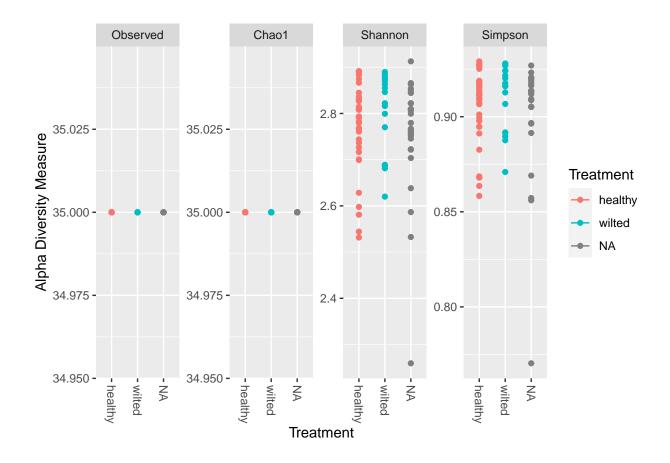
## *** Best solution repeated 2 times
```



```
Alpha_diversity(merge_Eukaryota , 'Family' , 'Treatment')
```

## We recommended that you find the un-trimmed data and retry.

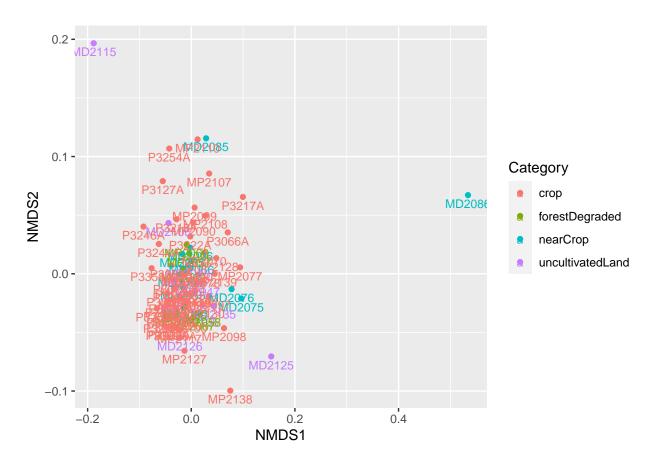
```
## Warning in estimate_richness(physeq, split = TRUE, measures = measures): The data you have provided
## any singletons. This is highly suspicious. Results of richness
## estimates (for example) are probably unreliable, or wrong, if you have already
## trimmed low-abundance taxa from the data.
##
```



# Beta\_diversity(merge\_Eukaryota , 'Family' , 'Category', 'bray')

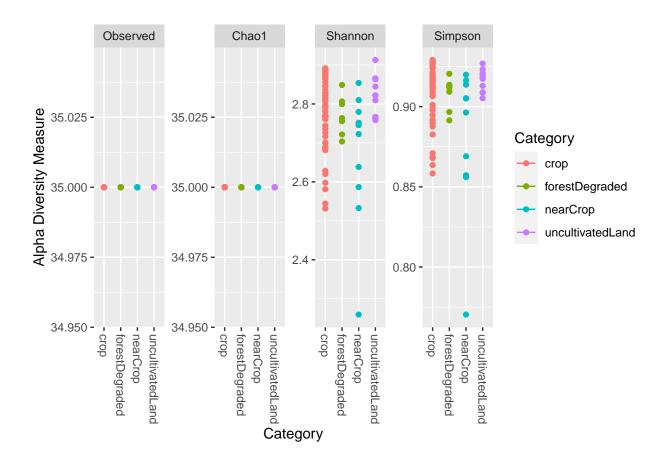
```
## Wisconsin double standardization
## Run 0 stress 0.136765
## Run 1 stress 0.1426953
## Run 2 stress 0.1375597
## Run 3 stress 0.1426931
## Run 4 stress 0.158903
## Run 5 stress 0.1481642
## Run 6 stress 0.1371009
## ... Procrustes: rmse 0.008803992 max resid 0.07709067
## Run 7 stress 0.137101
## ... Procrustes: rmse 0.008804207 max resid 0.07708579
## Run 8 stress 0.1428083
## Run 9 stress 0.1370891
## ... Procrustes: rmse 0.01445755 max resid 0.1215379
## Run 10 stress 0.1371022
## ... Procrustes: rmse 0.008824543 max resid 0.07706358
## Run 11 stress 0.141876
## Run 12 stress 0.1496806
## Run 13 stress 0.1385872
## Run 14 stress 0.1428099
## Run 15 stress 0.1640928
## Run 16 stress 0.1424547
## Run 17 stress 0.1375701
```

```
## Run 18 stress 0.1430875
## Run 19 stress 0.150898
## Run 20 stress 0.1388863
## *** Best solution was not repeated -- monoMDS stopping criteria:
## 7: no. of iterations >= maxit
## 13: stress ratio > sratmax
```



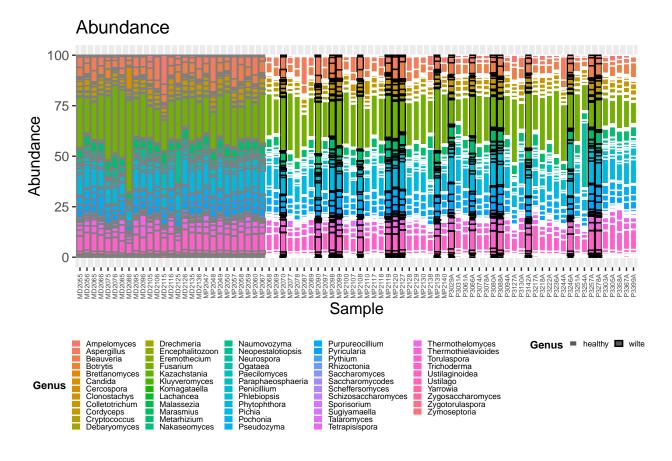
Alpha\_diversity(merge\_Eukaryota , 'Family' , 'Category')

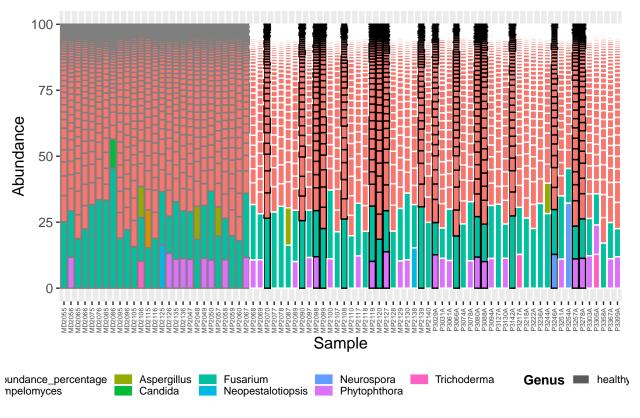
```
## Warning in estimate_richness(physeq, split = TRUE, measures = measures): The data you have provided of
## any singletons. This is highly suspicious. Results of richness
## estimates (for example) are probably unreliable, or wrong, if you have already
## trimmed low-abundance taxa from the data.
##
## We recommended that you find the un-trimmed data and retry.
```



#——Eukarya by Genero

Barras\_Species <- Abundance\_barras(merge\_Eukaryota,'Genus','Treatment',10.0)
Barras\_Species[1]</pre>

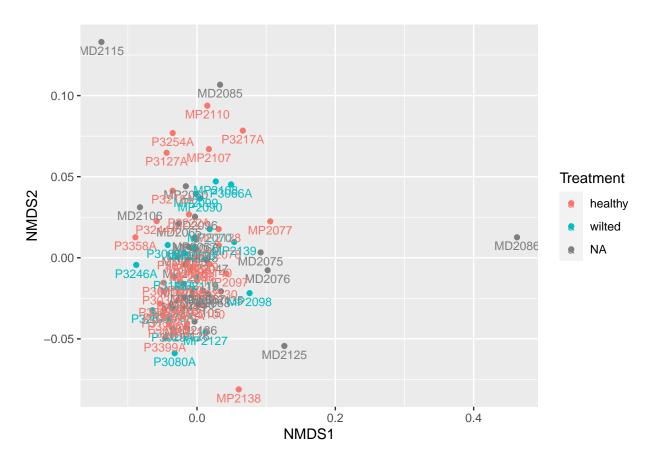




#### Beta\_diversity(merge\_Eukaryota , 'Genus' , 'Treatment', 'bray')

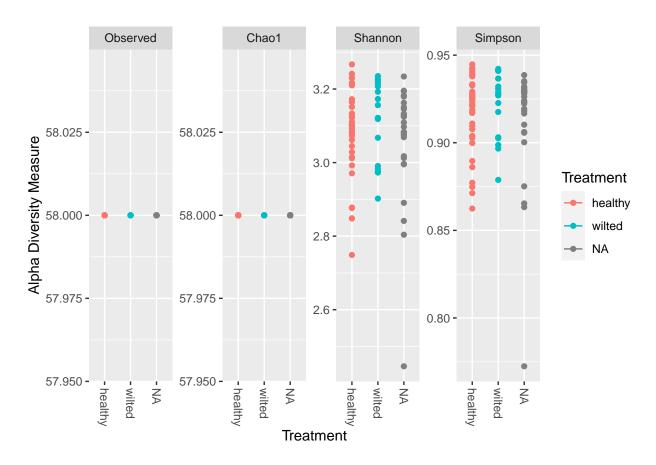
```
## Wisconsin double standardization
## Run 0 stress 0.1425943
## Run 1 stress 0.1556068
## Run 2 stress 0.157223
## Run 3 stress 0.1430761
  ... Procrustes: rmse 0.02756276 max resid 0.1227383
## Run 4 stress 0.1535271
## Run 5 stress 0.1545259
## Run 6 stress 0.1430704
  ... Procrustes: rmse 0.01572318 max resid 0.08365698
## Run 7 stress 0.1527883
## Run 8 stress 0.1406338
  ... New best solution
## ... Procrustes: rmse 0.02876197 max resid 0.1880479
## Run 9 stress 0.1650957
## Run 10 stress 0.1489875
## Run 11 stress 0.1554675
## Run 12 stress 0.1523229
## Run 13 stress 0.1398565
  ... New best solution
## ... Procrustes: rmse 0.01919991 max resid 0.08750119
## Run 14 stress 0.1463
## Run 15 stress 0.1404287
```

```
## Run 16 stress 0.1477418
## Run 17 stress 0.1400933
## ... Procrustes: rmse 0.01598115 max resid 0.08878172
## Run 18 stress 0.1399938
## ... Procrustes: rmse 0.003567462 max resid 0.02256952
## Run 19 stress 0.1462764
## Run 20 stress 0.1433316
## *** Best solution was not repeated -- monoMDS stopping criteria:
## 4: no. of iterations >= maxit
## 16: stress ratio > sratmax
```



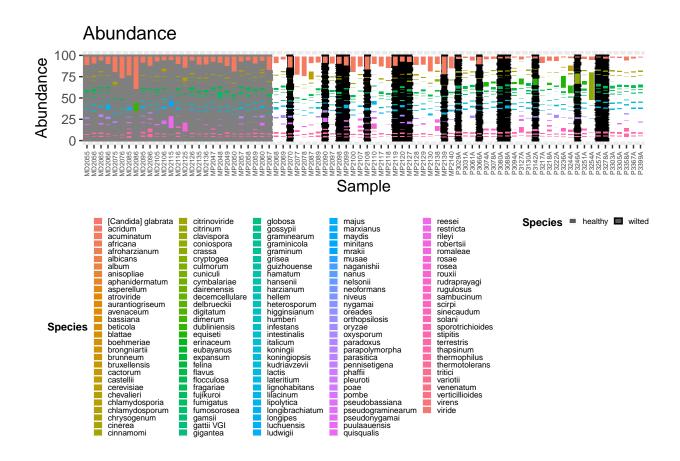
Alpha\_diversity(merge\_Eukaryota , 'Genus' , 'Treatment')

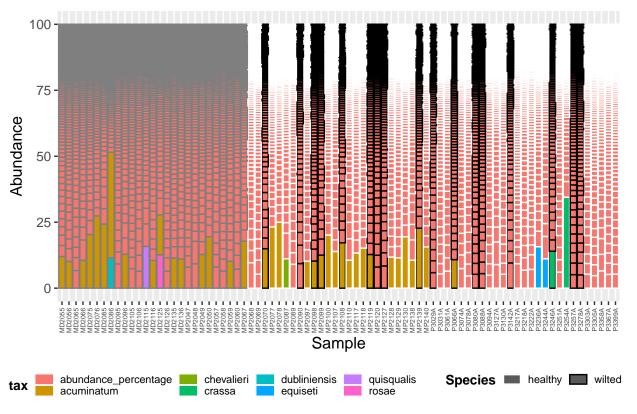
```
## Warning in estimate_richness(physeq, split = TRUE, measures = measures): The data you have provided of
## any singletons. This is highly suspicious. Results of richness
## estimates (for example) are probably unreliable, or wrong, if you have already
## trimmed low-abundance taxa from the data.
##
## We recommended that you find the un-trimmed data and retry.
```



#——Eukarya by Species

Barras\_Species <- Abundance\_barras(merge\_Eukaryota, 'Species', 'Treatment', 10.0)
Barras\_Species[1]</pre>

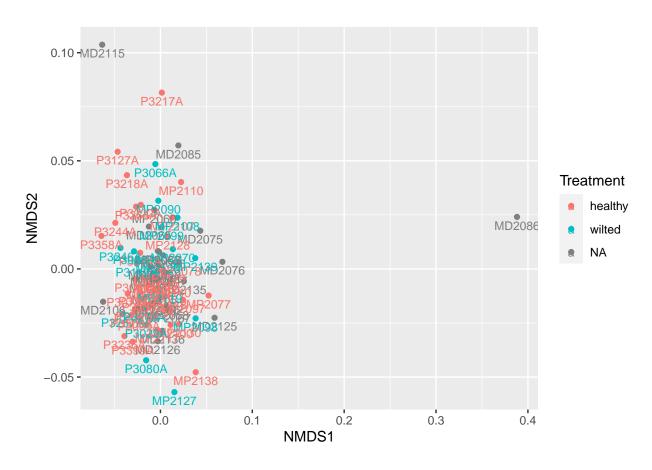




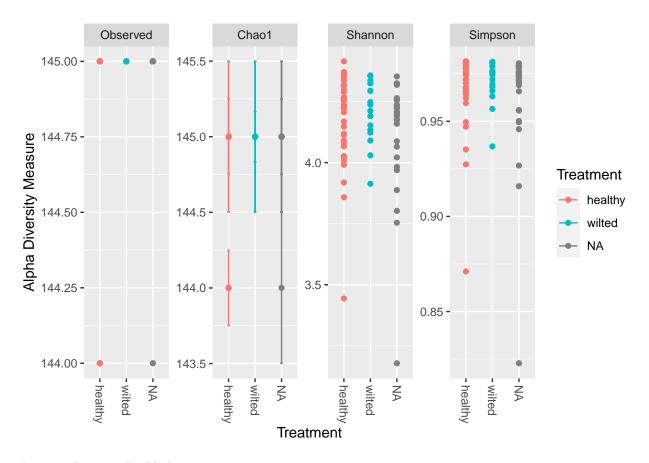
Beta\_diversity(merge\_Eukaryota , 'Species' , 'Treatment', 'bray')

```
## Wisconsin double standardization
## Run 0 stress 0.1394648
## Run 1 stress 0.1462513
## Run 2 stress 0.1394572
## ... New best solution
## ... Procrustes: rmse 0.002366879 max resid 0.01554427
## Run 3 stress 0.1388445
## ... New best solution
## ... Procrustes: rmse 0.01768916 max resid 0.1452212
## Run 4 stress 0.1388439
## ... New best solution
## ... Procrustes: rmse 0.001111111 max resid 0.006376012
## ... Similar to previous best
## Run 5 stress 0.1394655
## Run 6 stress 0.1394572
## Run 7 stress 0.1394575
## Run 8 stress 0.1394567
## Run 9 stress 0.1388431
## ... New best solution
## ... Procrustes: rmse 0.0003275494 max resid 0.001879371
## ... Similar to previous best
## Run 10 stress 0.139457
## Run 11 stress 0.144448
```

```
## Run 12 stress 0.1394575
## Run 13 stress 0.1394577
## Run 14 stress 0.1394569
## Run 15 stress 0.1394577
## Run 16 stress 0.1394573
## Run 17 stress 0.1394571
## Run 18 stress 0.1388439
## ... Procrustes: rmse 0.0003470212 max resid 0.001990059
## ... Similar to previous best
## Run 19 stress 0.1388444
## ... Procrustes: rmse 0.0004392323 max resid 0.002517098
## ... Similar to previous best
## Run 20 stress 0.1394611
## *** Best solution repeated 3 times
```

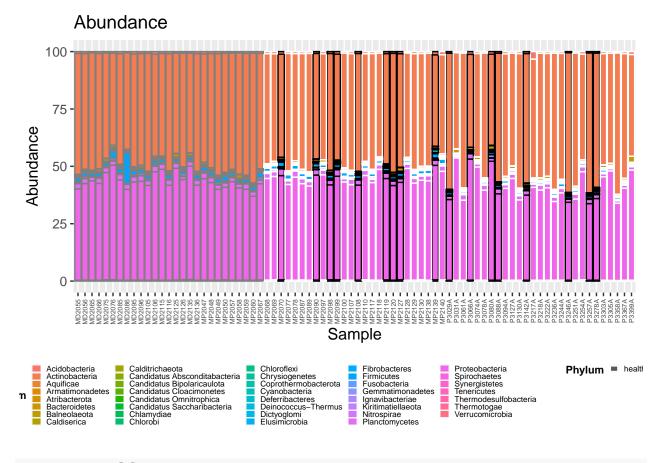


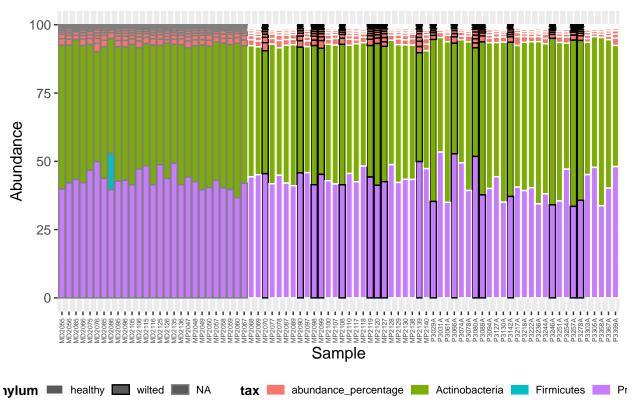
Alpha\_diversity(merge\_Eukaryota , 'Species' , 'Treatment')



#——Bacteria by Phylum

Barras\_Species <- Abundance\_barras(merge\_Bacteria, 'Phylum', 'Treatment', 10.0)
Barras\_Species[1]</pre>

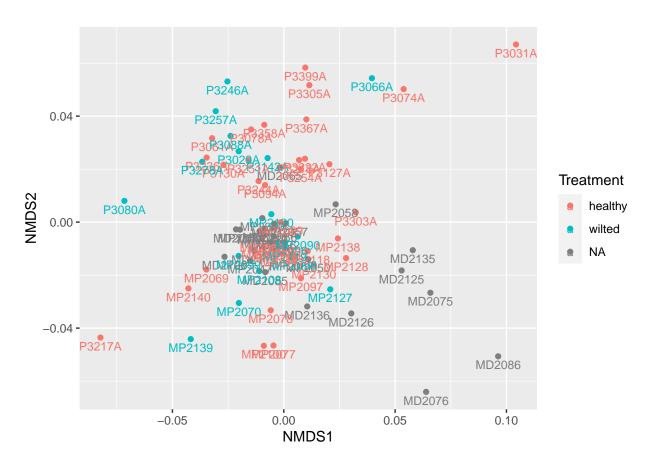




Beta\_diversity(merge\_Bacteria , 'Phylum' , 'Treatment', 'bray')

```
## Square root transformation
## Wisconsin double standardization
## Run 0 stress 0.1704433
## Run 1 stress 0.1708577
## ... Procrustes: rmse 0.01316983 max resid 0.06454964
## Run 2 stress 0.1712858
## Run 3 stress 0.1716125
## Run 4 stress 0.169571
## ... New best solution
## ... Procrustes: rmse 0.02946184 max resid 0.2259082
## Run 5 stress 0.1696863
## ... Procrustes: rmse 0.0166448 max resid 0.09703316
## Run 6 stress 0.1704877
## Run 7 stress 0.1779854
## Run 8 stress 0.1683153
## ... New best solution
## ... Procrustes: rmse 0.01148517 max resid 0.06885598
## Run 9 stress 0.1683139
## ... New best solution
## ... Procrustes: rmse 0.001687889 max resid 0.01121499
## Run 10 stress 0.1796243
## Run 11 stress 0.1683149
## ... Procrustes: rmse 0.001764232 max resid 0.01125403
```

```
## Run 12 stress 0.1713043
## Run 13 stress 0.1704412
## Run 14 stress 0.1683134
## ... New best solution
## ... Procrustes: rmse 0.0003057321 max resid 0.002361624
## ... Similar to previous best
## Run 15 stress 0.1713142
## Run 16 stress 0.1683016
## ... New best solution
## ... Procrustes: rmse 0.005655508 max resid 0.04443228
## Run 17 stress 0.1704576
## Run 18 stress 0.1682886
## ... New best solution
## ... Procrustes: rmse 0.001710559 max resid 0.01114268
## Run 19 stress 0.1713528
## Run 20 stress 0.1822664
  *** Best solution was not repeated -- monoMDS stopping criteria:
##
        1: no. of iterations >= maxit
##
       19: stress ratio > sratmax
```



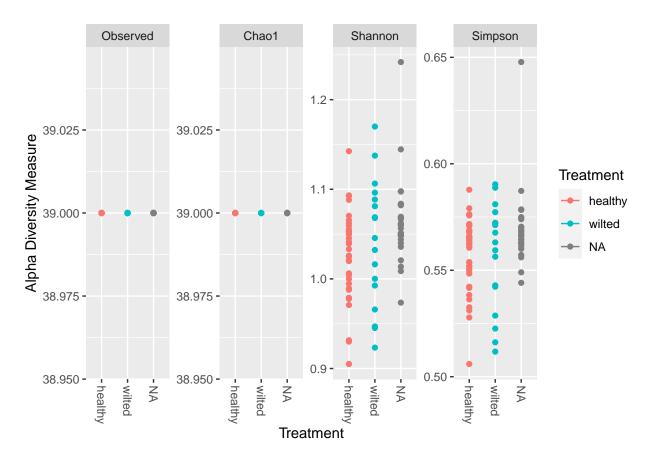
Alpha\_diversity(merge\_Bacteria , 'Phylum' , 'Treatment')

```
## Warning in estimate_richness(physeq, split = TRUE, measures = measures): The data you have provided of
## any singletons. This is highly suspicious. Results of richness
## estimates (for example) are probably unreliable, or wrong, if you have already
```

## trimmed low-abundance taxa from the data.

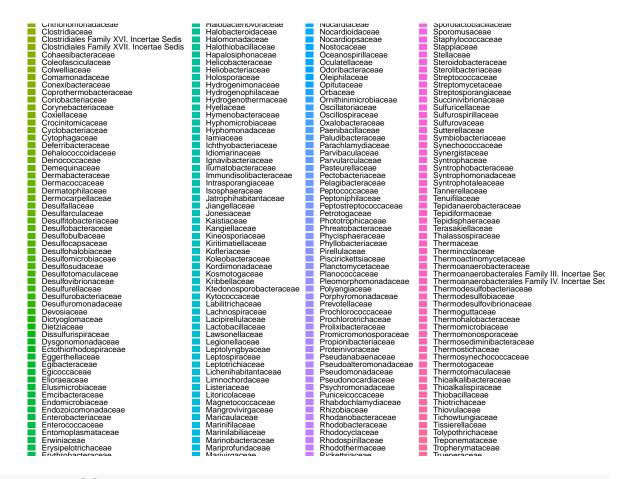
##

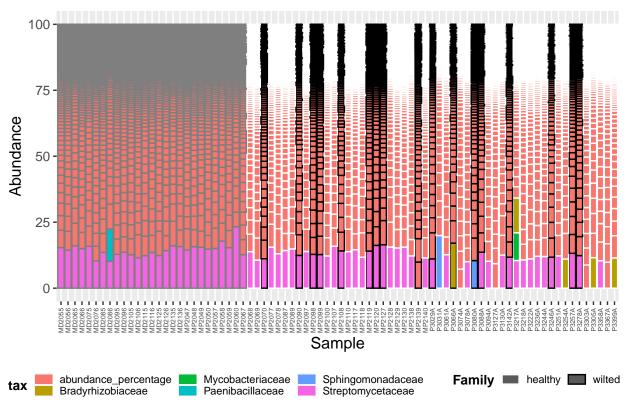
## We recommended that you find the un-trimmed data and retry.



#——Bacteria by Familia

Barras\_Species <- Abundance\_barras(merge\_Bacteria, 'Family', 'Treatment',10.0)
Barras\_Species[1]</pre>

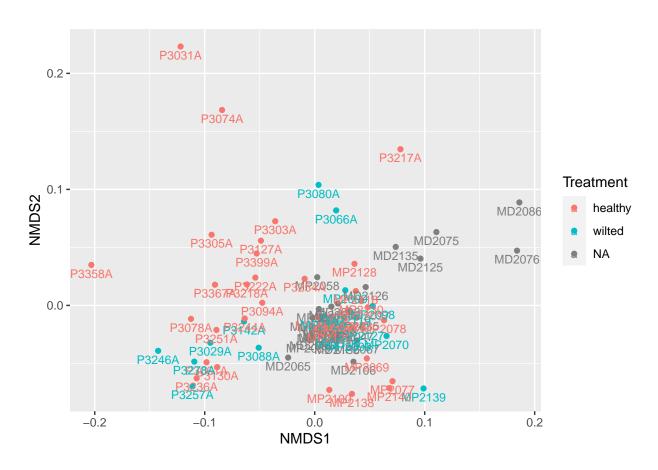




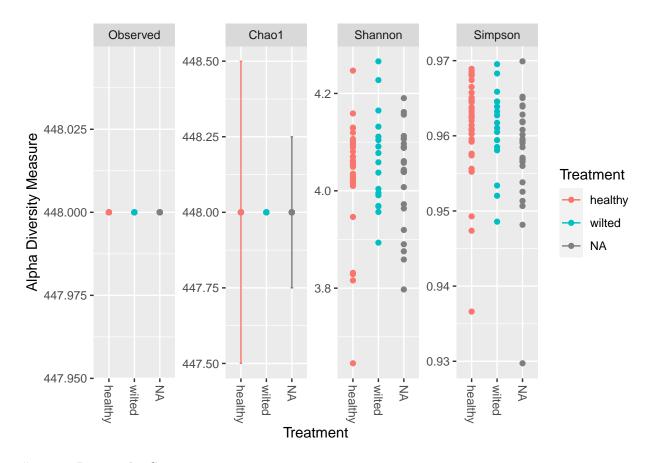
```
Beta_diversity(merge_Bacteria , 'Family' , 'Treatment', 'bray')
```

```
## Wisconsin double standardization
## Run 0 stress 0.132377
## Run 1 stress 0.1405609
## Run 2 stress 0.1391985
## Run 3 stress 0.1519021
## Run 4 stress 0.1527065
## Run 5 stress 0.1432062
## Run 6 stress 0.1705558
## Run 7 stress 0.1395452
## Run 8 stress 0.1391766
## Run 9 stress 0.163441
## Run 10 stress 0.1411044
## Run 11 stress 0.1637101
## Run 12 stress 0.1444844
## Run 13 stress 0.1566534
## Run 14 stress 0.1486548
## Run 15 stress 0.1337582
## Run 16 stress 0.141046
## Run 17 stress 0.155059
## Run 18 stress 0.1626394
## Run 19 stress 0.1405614
## Run 20 stress 0.1622084
## *** Best solution was not repeated -- monoMDS stopping criteria:
```

## 2: no. of iterations >= maxit
## 18: stress ratio > sratmax

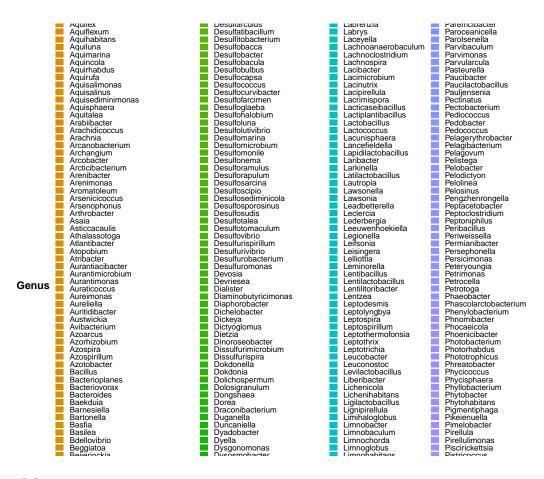


Alpha\_diversity(merge\_Bacteria , 'Family' , 'Treatment')

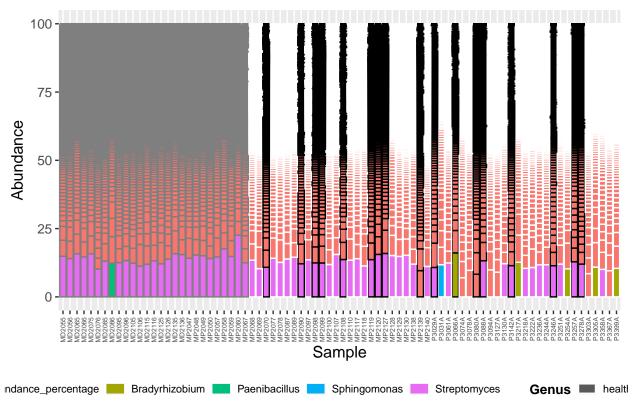


#——Bacteria by Genero

Barras\_Species <- Abundance\_barras(merge\_Bacteria, 'Genus', 'Treatment', 10.0)
Barras\_Species[1]</pre>



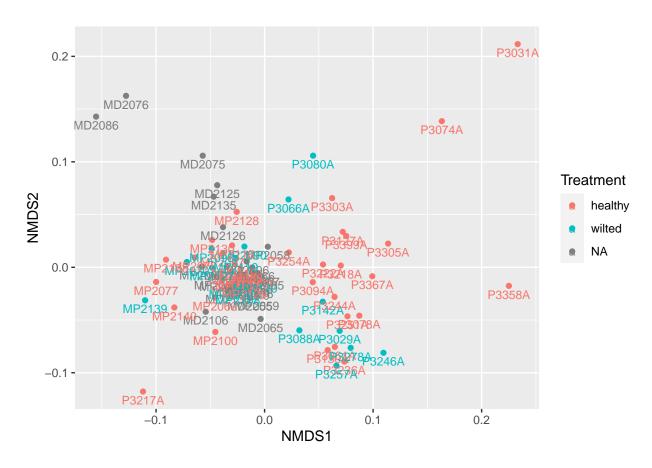




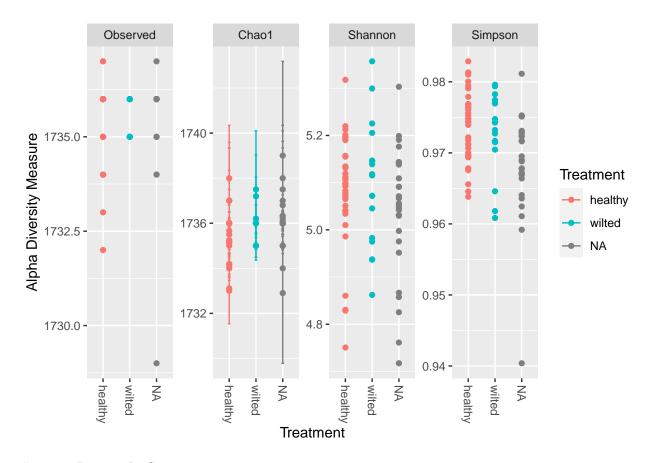
# Beta\_diversity(merge\_Bacteria , 'Genus' , 'Treatment', 'bray')

```
## Wisconsin double standardization
## Run 0 stress 0.1323538
## Run 1 stress 0.1318739
## ... New best solution
## ... Procrustes: rmse 0.04483879 max resid 0.3308961
## Run 2 stress 0.1320811
## ... Procrustes: rmse 0.04682465 max resid 0.3312729
## Run 3 stress 0.1322392
## ... Procrustes: rmse 0.01733447 max resid 0.1405044
## Run 4 stress 0.1383274
## Run 5 stress 0.1328271
## Run 6 stress 0.1266989
## ... New best solution
## ... Procrustes: rmse 0.02281745 max resid 0.1891363
## Run 7 stress 0.1412309
## Run 8 stress 0.1321914
## Run 9 stress 0.148244
## Run 10 stress 0.1420427
## Run 11 stress 0.1467141
## Run 12 stress 0.1323434
## Run 13 stress 0.1324735
## Run 14 stress 0.148499
## Run 15 stress 0.1330579
```

```
## Run 16 stress 0.1682506
## Run 17 stress 0.1489298
## Run 18 stress 0.1517771
## Run 19 stress 0.1554539
## Run 20 stress 0.1333587
## *** Best solution was not repeated -- monoMDS stopping criteria:
## 1: no. of iterations >= maxit
## 19: stress ratio > sratmax
```

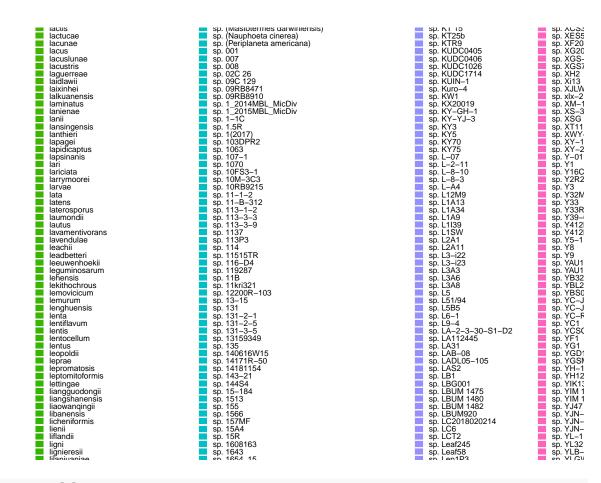


Alpha\_diversity(merge\_Bacteria , 'Genus' , 'Treatment')



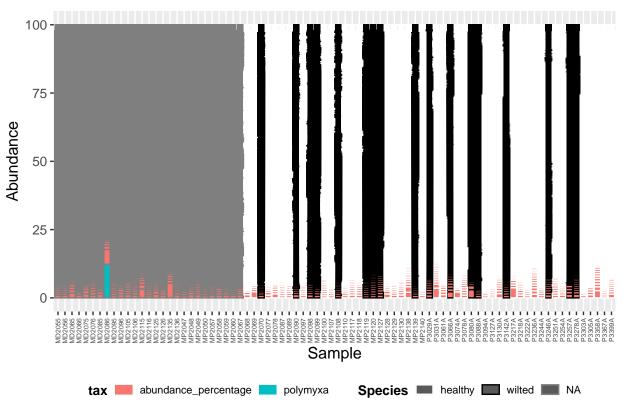
#——Bacteria by Species

Barras\_Species <- Abundance\_barras(merge\_Bacteria, 'Species', 'Treatment',10.0)
Barras\_Species[1]</pre>



## [[1]]

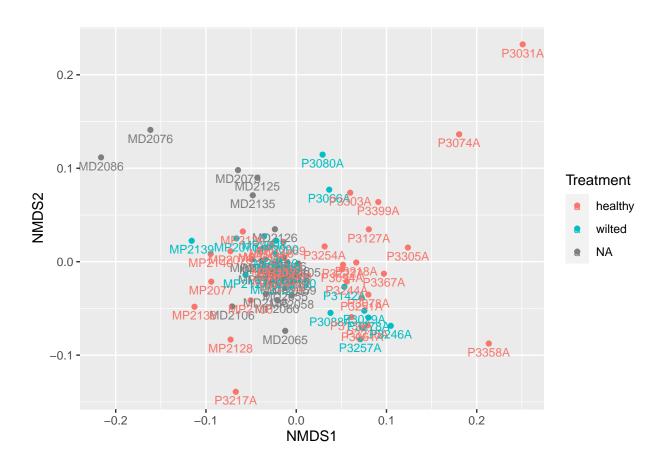
ella sus



Beta\_diversity(merge\_Bacteria , 'Species' , 'Treatment', 'bray')

```
## Wisconsin double standardization
## Run 0 stress 0.1456926
## Run 1 stress 0.1551344
## Run 2 stress 0.153833
## Run 3 stress 0.1545858
## Run 4 stress 0.1462371
## Run 5 stress 0.1568186
## Run 6 stress 0.1398761
## ... New best solution
## ... Procrustes: rmse 0.03959896 max resid 0.3261456
## Run 7 stress 0.1490098
## Run 8 stress 0.1445075
## Run 9 stress 0.1575396
## Run 10 stress 0.1591801
## Run 11 stress 0.1482487
## Run 12 stress 0.151126
## Run 13 stress 0.1706569
## Run 14 stress 0.1634706
## Run 15 stress 0.1430293
## Run 16 stress 0.1497951
## Run 17 stress 0.1407344
## Run 18 stress 0.1577634
## Run 19 stress 0.1481045
```

```
## Run 20 stress 0.1739684
## *** Best solution was not repeated -- monoMDS stopping criteria:
## 2: no. of iterations >= maxit
## 18: stress ratio > sratmax
```



Alpha\_diversity(merge\_Bacteria , 'Species' , 'Treatment')

