

Funciones automatizadas para visualizar las barras de abundancia, diversidades alfa y beta; a diferentes niveles taxonomicos

Camila Silva

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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/Data1")
outpath = "/home/camila/GIT/Tesis_Maestria/Analisis_Comparativo/Fresa_Solena/Results_img"

fresa_kraken <- import_biom("fresa_kraken.biom")
colnames(fresa_kraken@tax_table@.Data) <- c("Kingdom", "Phylum", "Class", "Order", "Family", "Genus", "Species")
fresa_kraken@tax_table@.Data <- substr(fresa_kraken@tax_table@.Data,4,100)
colnames(fresa_kraken@otu_table@.Data) <- substr(colnames(fresa_kraken@otu_table@.Data),1,6)
metadata_fresa <- read.csv2("/home/camila/GIT/Tesis_Maestria/Data/fresa_solena/Data1/metadata.csv",header=1)
fresa_kraken@sam_data <- sample_data(metadata_fresa)
fresa_kraken@sam_data$Sample <- row.names(fresa_kraken@sam_data)
colnames(fresa_kraken@sam_data) <- c('Treatment', 'Samples')
samples_to_remove <- c("MP2079", "MP2080", "MP2088", "MP2109", "MP2137")
fresa_kraken_fil <- prune_samples(!(sample_names(fresa_kraken) %in% samples_to_remove), fresa_kraken)
percentages_fil <- transform_sample_counts(fresa_kraken_fil, function(x) x*100 / sum(x) )
percentages_df <- psmelt(percentages_fil)
```

Subconjunto de “Eukaryota”

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

Subconjunto de “Bacteria”

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

Funciones

glomToGraph

Crea los subconjuntos de datos necesarios para más adelante

input phy <- es el objeto phyloseq base tax <- rango taxonomico al que queremos recortar output glom <- objeto phyloseq ya recortado percentages <- objeto phyloseq de porcentajes sobre el glom percentages_df <- data frame de porcentajes sobre el glom

```
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))
}
```

Graficar abundancias (stackbar)

```
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=attribute)) +
    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)]
}
```

```

percentages_df$tax[percentages_df$Abundance < abundance_percentage] <- "Others"
percentages_df$tax <- as.factor(percentages_df$tax)
plot_percentages <- ggplot(data=percentages_df, aes_string(x='Sample', y='Abundance', fill='tax', color='tax')) +
  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 diversidad beta

```

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)
}

```

Graficar diversidad alfa

```

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 = attribute)
  return(plot_alpha)
}

```

Graficas

Graficamos las barras de abundancia, las barras de abundancia aglomerando el 10% menos abundantes, alfa y beta diversidad para los siguientes niveles taxonomicos.

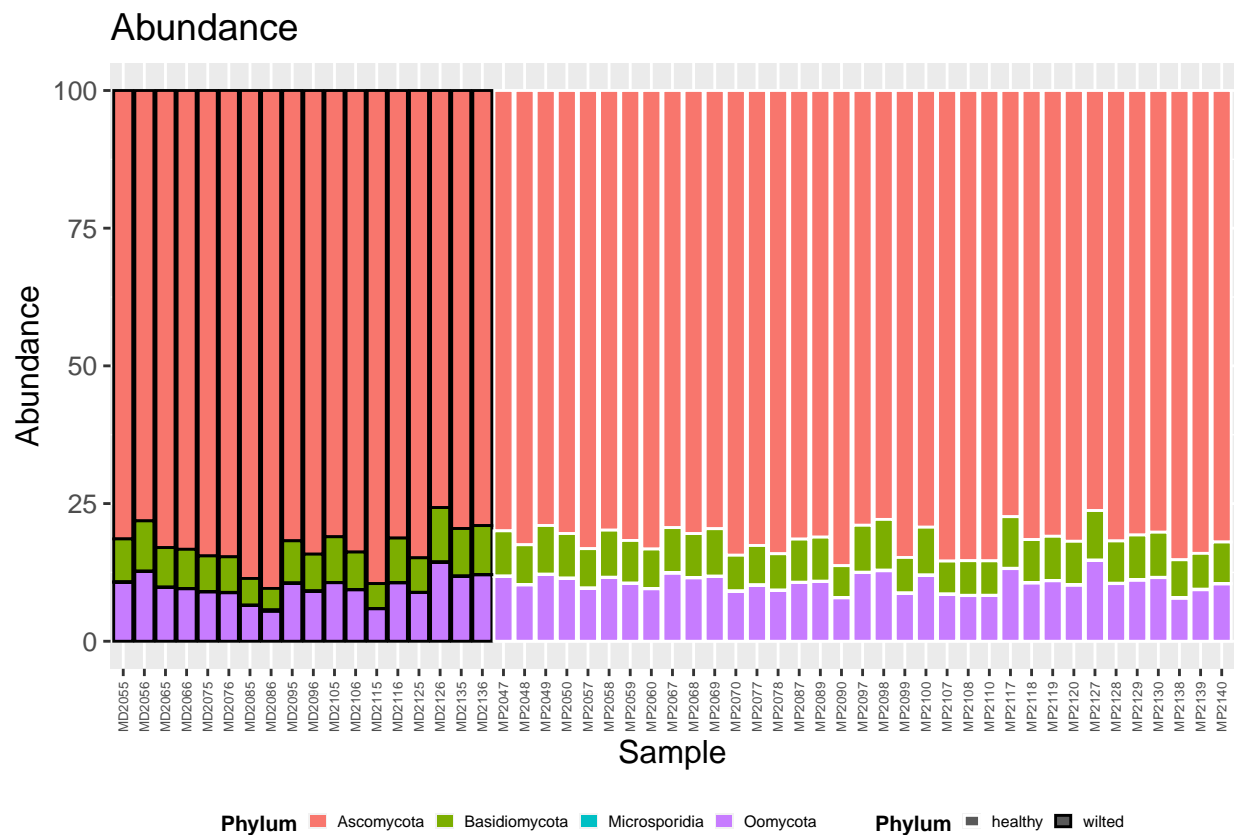
```
## ----- Eukarya by Phylum
```

```
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()'
```

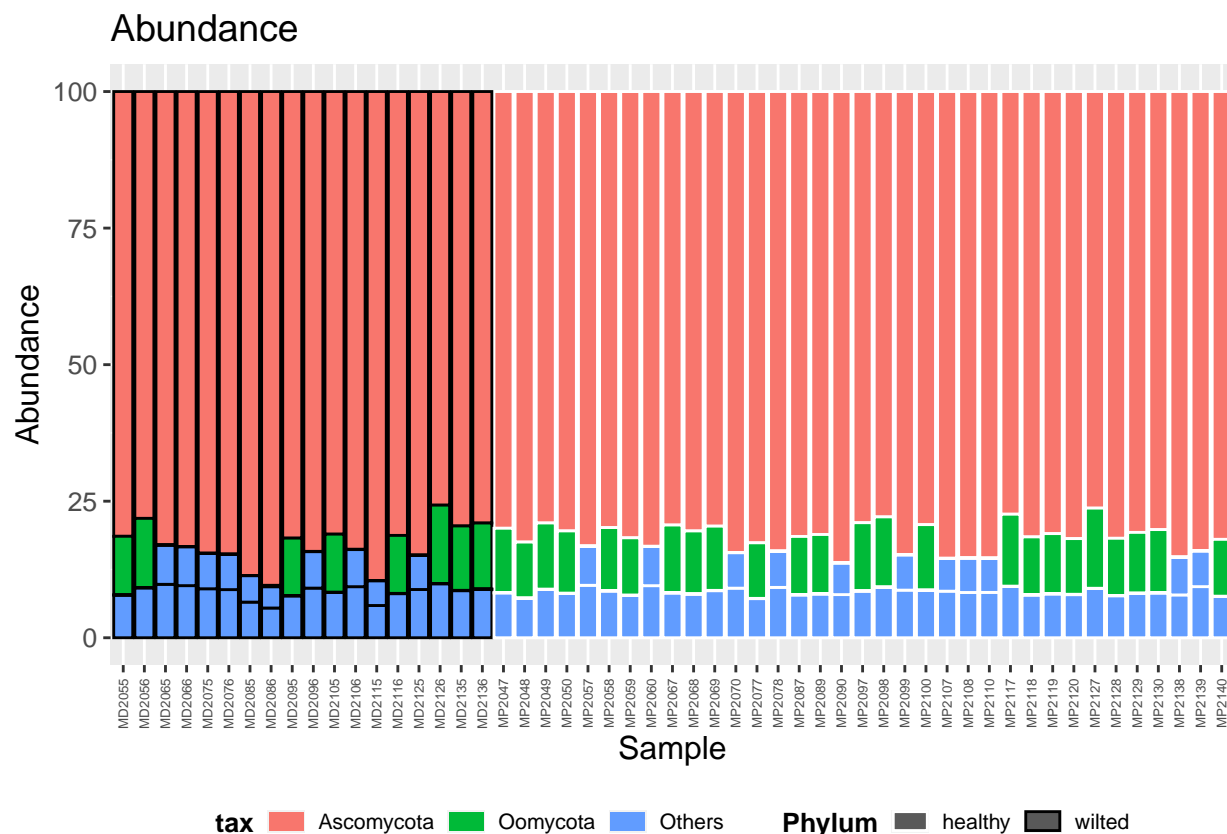
```
Barras_Phylum[1] # normal
```

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



```
Barras_Phylum[2]
```

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



```
Beta_diversity(merge_Eukaryota , 'Phylum' , 'Treatment' , 'bray')
```

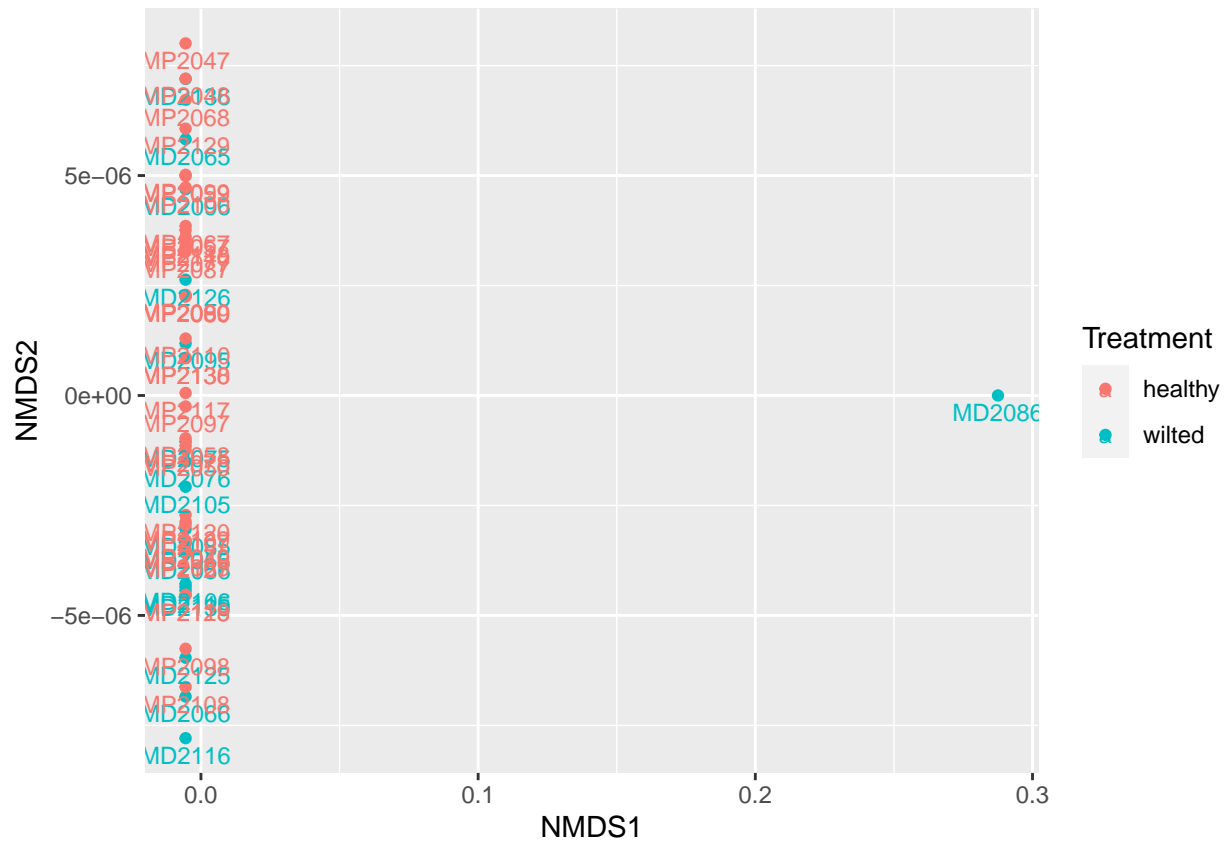
```
## Square root transformation
## Wisconsin double standardization
## Run 0 stress 0.01148681
## Run 1 stress 6.23341e-05
## ... New best solution
## ... Procrustes: rmse 0.03466421 max resid 0.09090798
## Run 2 stress 9.924012e-05
## ... Procrustes: rmse 8.498182e-05 max resid 0.0002174451
## ... Similar to previous best
## Run 3 stress 7.520819e-05
## ... Procrustes: rmse 3.260399e-05 max resid 6.156092e-05
## ... Similar to previous best
## Run 4 stress 8.237494e-05
## ... Procrustes: rmse 3.867383e-05 max resid 0.0001013529
## ... Similar to previous best
## Run 5 stress 9.056216e-05
## ... Procrustes: rmse 3.679735e-05 max resid 0.0001188962
## ... Similar to previous best
## Run 6 stress 9.561394e-05
## ... Procrustes: rmse 0.000151102 max resid 0.0003710265
## ... Similar to previous best
## Run 7 stress 0.0004646677
## ... Procrustes: rmse 0.001372413 max resid 0.003604997
```

```

## ... Similar to previous best
## Run 8 stress 9.29825e-05
## ... Procrustes: rmse 0.0001070232 max resid 0.0002624821
## ... Similar to previous best
## Run 9 stress 9.912037e-05
## ... Procrustes: rmse 0.0001078595 max resid 0.0002706643
## ... Similar to previous best
## Run 10 stress 9.98074e-05
## ... Procrustes: rmse 0.0001723485 max resid 0.0004470798
## ... Similar to previous best
## Run 11 stress 9.90852e-05
## ... Procrustes: rmse 0.0002715707 max resid 0.0007077103
## ... Similar to previous best
## Run 12 stress 9.798793e-05
## ... Procrustes: rmse 7.601213e-05 max resid 0.0001963665
## ... Similar to previous best
## Run 13 stress 8.745481e-05
## ... Procrustes: rmse 3.325777e-05 max resid 8.220807e-05
## ... Similar to previous best
## Run 14 stress 9.958365e-05
## ... Procrustes: rmse 0.0002838867 max resid 0.0007370116
## ... Similar to previous best
## Run 15 stress 0.0002053957
## ... Procrustes: rmse 0.0006058763 max resid 0.001579662
## ... Similar to previous best
## Run 16 stress 9.924389e-05
## ... Procrustes: rmse 0.0001228828 max resid 0.0003199078
## ... Similar to previous best
## Run 17 stress 0.000107705
## ... Procrustes: rmse 0.0003169315 max resid 0.0008220986
## ... Similar to previous best
## Run 18 stress 0.00071692
## Run 19 stress 9.300725e-05
## ... Procrustes: rmse 9.651243e-05 max resid 0.000248036
## ... Similar to previous best
## Run 20 stress 9.965288e-05
## ... Procrustes: rmse 0.0001697613 max resid 0.0004489988
## ... Similar to previous best
## *** Best solution repeated 18 times

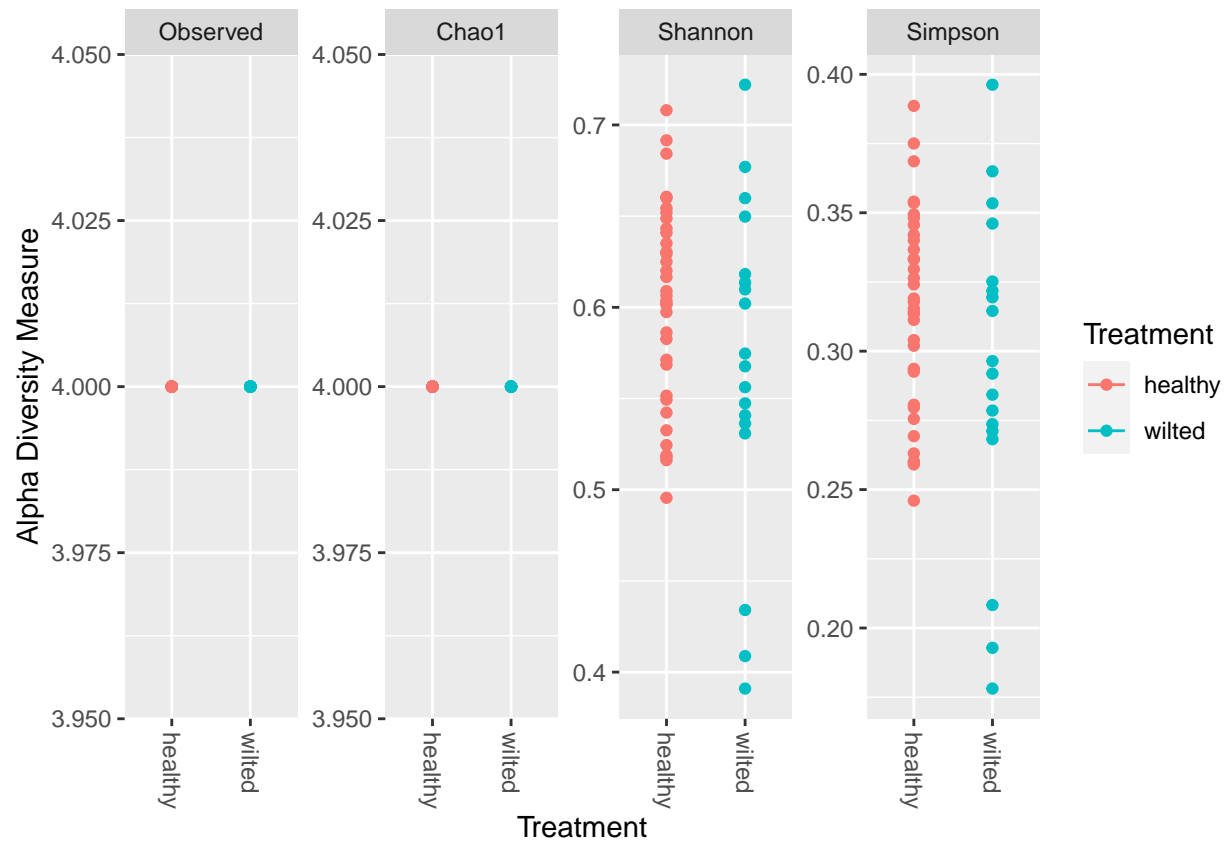
## Warning in metaMDS(veganifyOTU(physeq), distance, ...): stress is (nearly) zero:
## you may have insufficient data

```



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

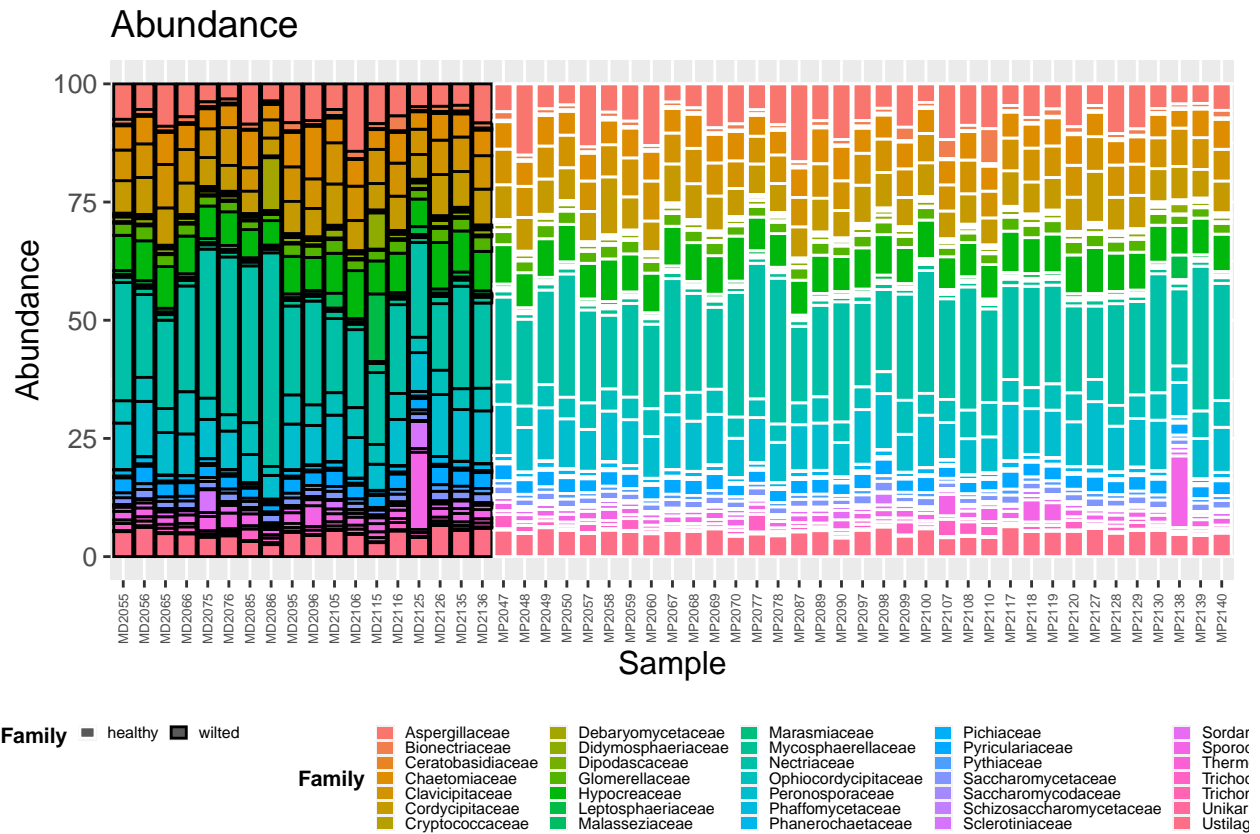
```
## Warning in estimate_richness(physeq, split = TRUE, measures = measures): The data you have provided
## any singletons. This is highly suspicious. Results of richness
## estimates (for example) are probably unreliable, or wrong, if you have already
## trimmed low-abundance taxa from the data.
##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



— Eukarya by Family

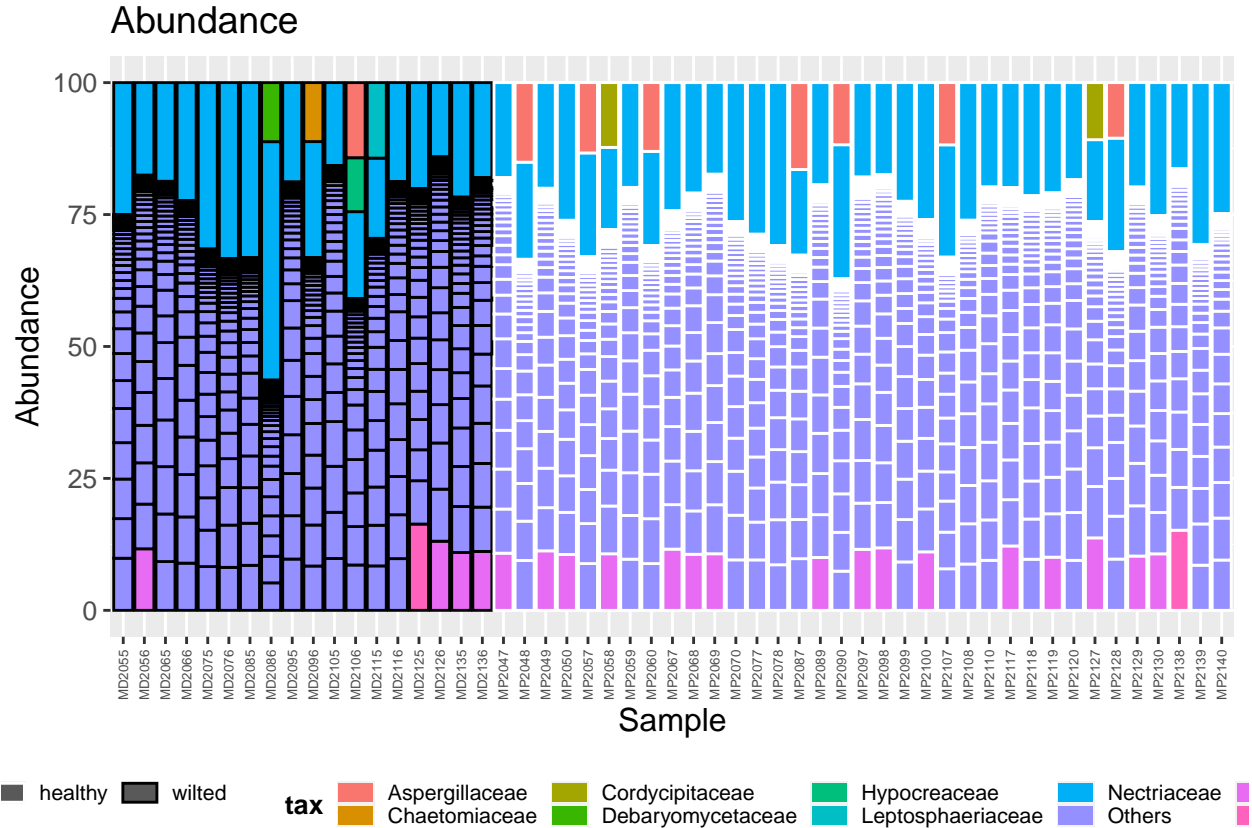
```
Barras_Species <- Abundance_barras(merge_Eukaryota, 'Family', 'Treatment', 10.0)
Barras_Species[1]
```

[[1]]



Barras_Species [2]

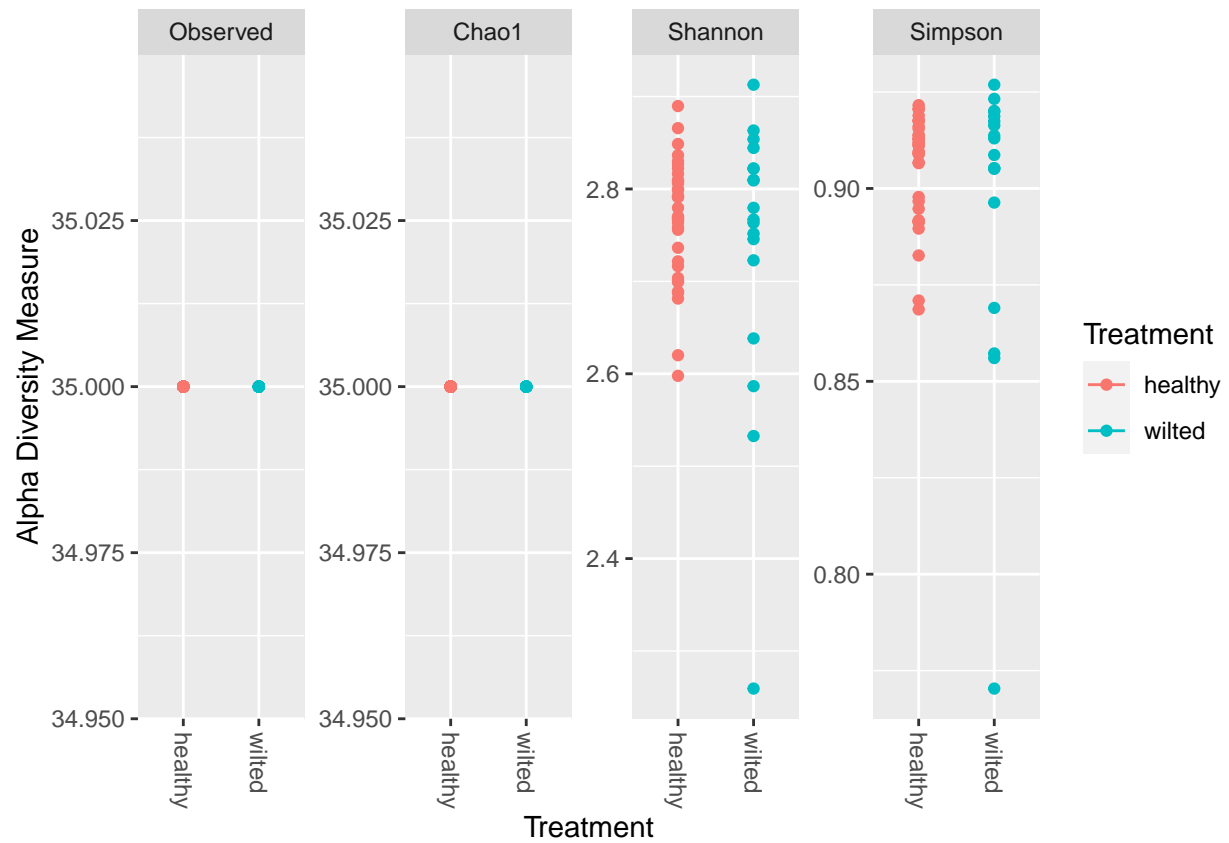
[[1]]



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

```
## Wisconsin double standardization
## Run 0 stress 0.1089081
## Run 1 stress 0.10892
## ... Procrustes: rmse 0.003469475  max resid 0.01752379
## Run 2 stress 0.1090929
## ... Procrustes: rmse 0.005544936  max resid 0.02829244
## Run 3 stress 0.1088917
## ... New best solution
## ... Procrustes: rmse 0.001874879  max resid 0.009516901
## ... Similar to previous best
## Run 4 stress 0.1090933
## ... Procrustes: rmse 0.005791372  max resid 0.02850908
## Run 5 stress 0.1266812
## Run 6 stress 0.1088977
## ... Procrustes: rmse 0.001797222  max resid 0.009009046
## ... Similar to previous best
## Run 7 stress 0.1088875
## ... New best solution
## ... Procrustes: rmse 0.0009644059  max resid 0.005579993
## ... Similar to previous best
## Run 8 stress 0.108929
## ... Procrustes: rmse 0.008413825  max resid 0.05469496
## Run 9 stress 0.1088966
```

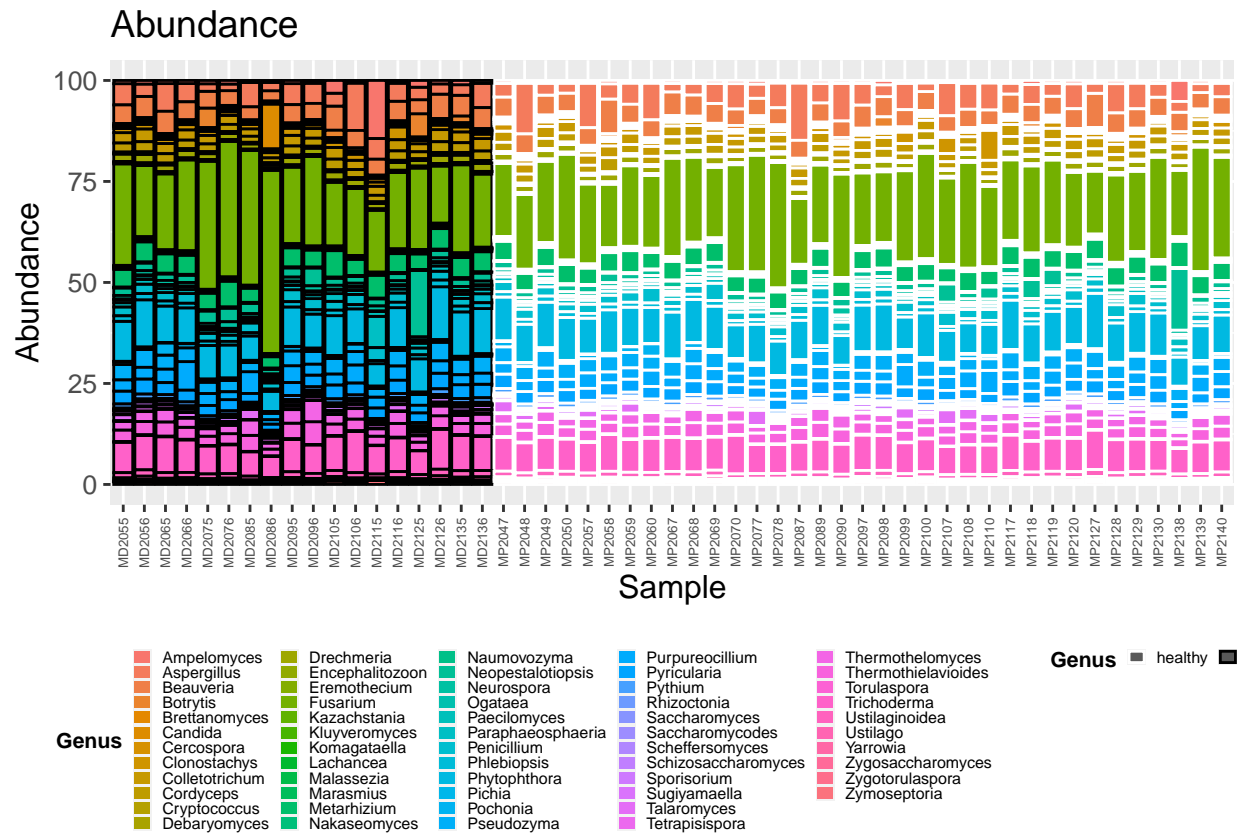
```
## ... Procrustes: rmse 0.001701053  max resid 0.008977046
## ... Similar to previous best
## Run 10 stress 0.1088927
## ... Procrustes: rmse 0.001100288  max resid 0.006369565
## ... Similar to previous best
## Run 11 stress 0.1088875
## ... New best solution
## ... Procrustes: rmse 0.0003458743  max resid 0.001444601
## ... Similar to previous best
## Run 12 stress 0.1090814
## ... Procrustes: rmse 0.007447257  max resid 0.04382634
## Run 13 stress 0.1088878
## ... Procrustes: rmse 0.0004233376  max resid 0.001630264
## ... Similar to previous best
## Run 14 stress 0.1089302
## ... Procrustes: rmse 0.008407645  max resid 0.05481049
## Run 15 stress 0.1089293
## ... Procrustes: rmse 0.008404887  max resid 0.05474606
## Run 16 stress 0.1273453
## Run 17 stress 0.1089711
## ... Procrustes: rmse 0.008789917  max resid 0.05386247
## Run 18 stress 0.1265348
## Run 19 stress 0.1089491
## ... Procrustes: rmse 0.008086409  max resid 0.05430937
## Run 20 stress 0.1089299
## ... Procrustes: rmse 0.008484859  max resid 0.0545113
## *** Best solution repeated 2 times
```

— Eukarya by Genero

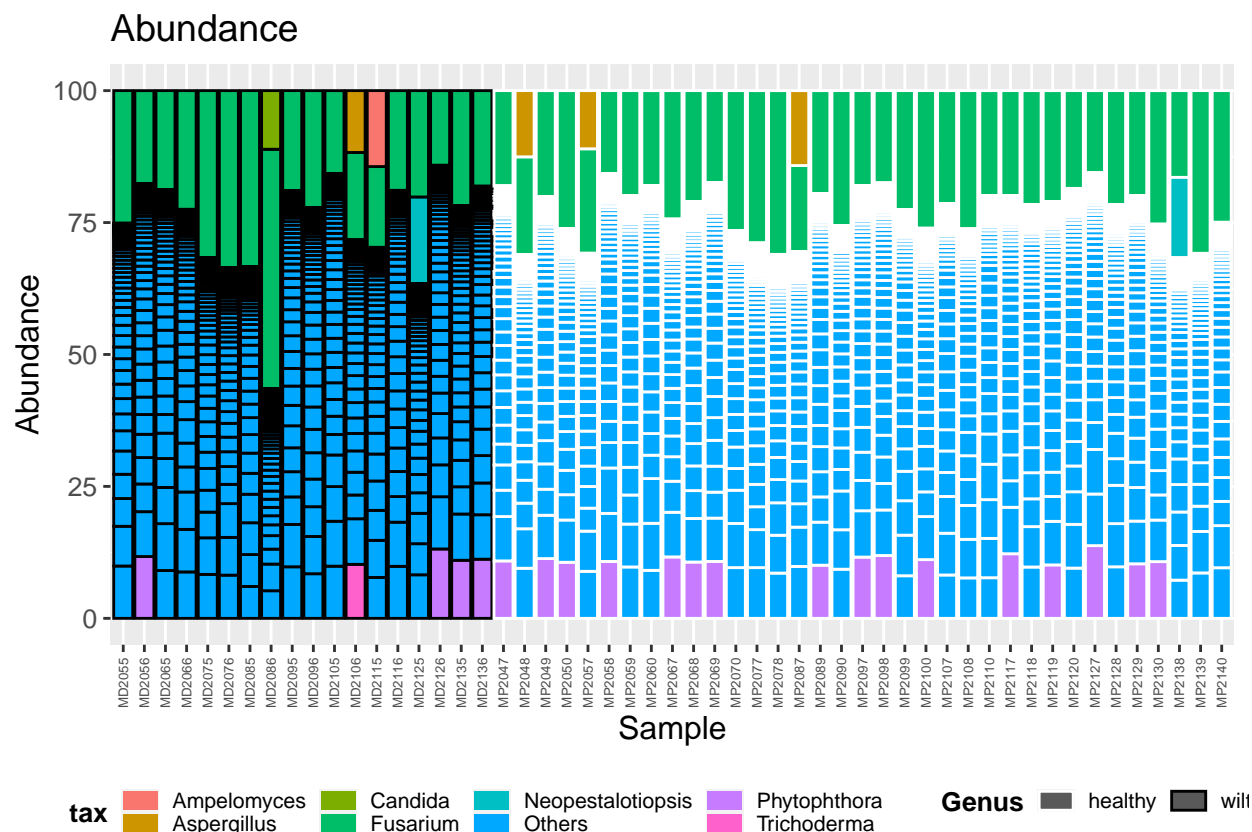
```
Barras_Species <- Abundance_barras(merge_Eukaryota, 'Genus', 'Treatment', 10.0)
Barras_Species[1]
```

[[1]]



Barras_Species [2]

[[1]]



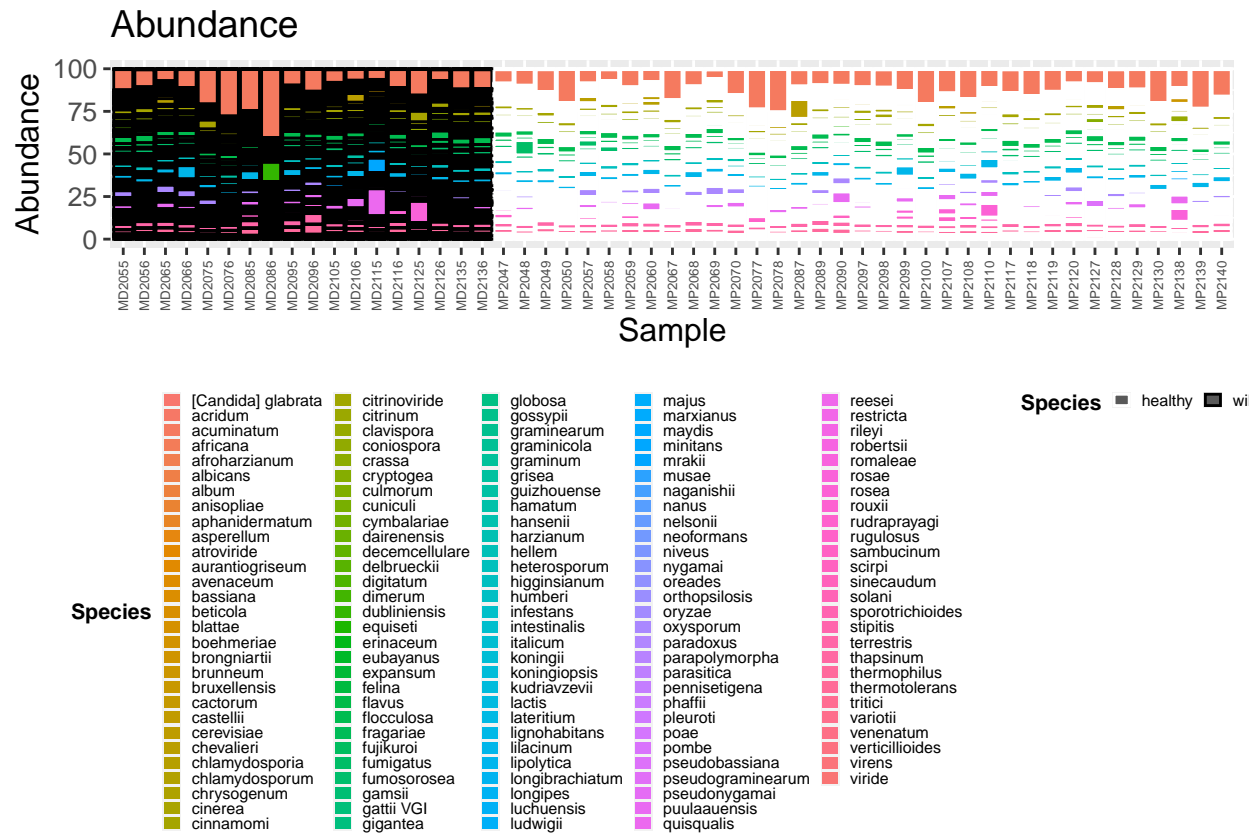
```
Beta_diversity(merge_Eukaryota , 'Genus' , 'Treatment' , 'bray')
```

```
## Wisconsin double standardization
## Run 0 stress 0.110709
## Run 1 stress 0.1119812
## Run 2 stress 0.1236311
## Run 3 stress 0.111981
## Run 4 stress 0.1236303
## Run 5 stress 0.1107095
## ... Procrustes: rmse 0.001124574 max resid 0.006442219
## ... Similar to previous best
## Run 6 stress 0.1107093
## ... Procrustes: rmse 0.001076297 max resid 0.006272975
## ... Similar to previous best
## Run 7 stress 0.1119791
## Run 8 stress 0.1119828
## Run 9 stress 0.1119787
## Run 10 stress 0.1235026
## Run 11 stress 0.1119789
## Run 12 stress 0.1107095
## ... Procrustes: rmse 0.0001517755 max resid 0.0008446115
## ... Similar to previous best
## Run 13 stress 0.1235017
## Run 14 stress 0.1107131
## ... Procrustes: rmse 0.001836109 max resid 0.0104769
```

Treatment

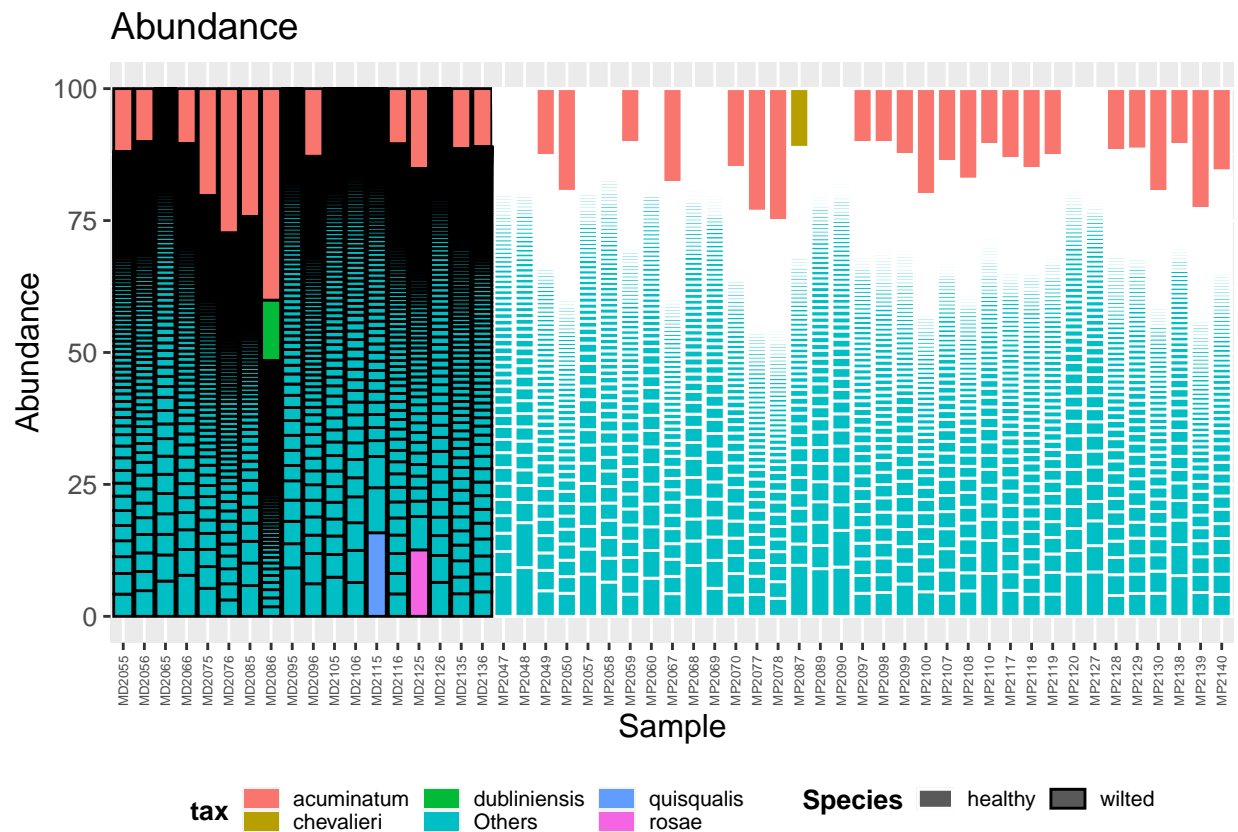
- healthy
- wilted

```
## Warning in estimate_richness(physeq, split = TRUE, measures = measures): The data you have provided contains
## 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.
```

Barras_Species [2]

[[1]]



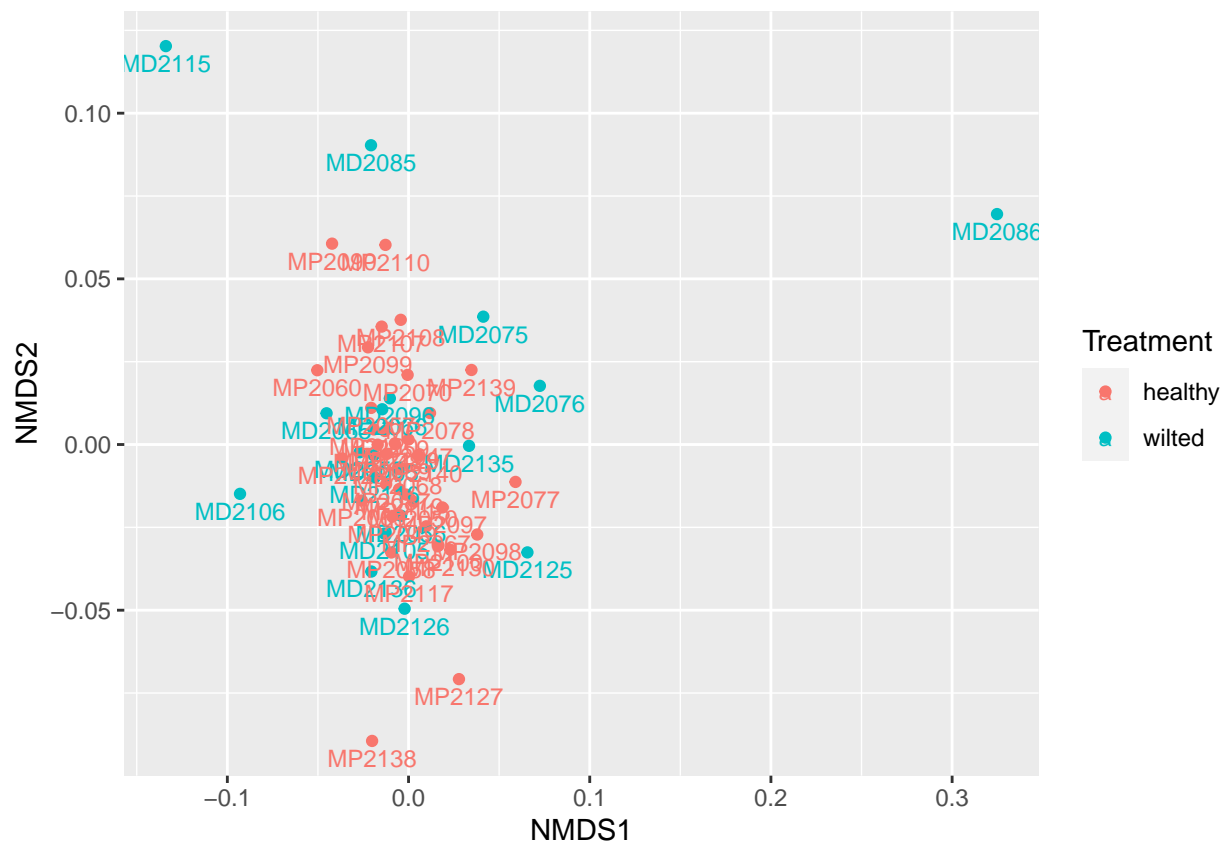
```
Beta_diversity(merge_Eukaryota , 'Species' , 'Treatment', 'bray')
```

```
## Wisconsin double standardization
## Run 0 stress 0.1226512
## Run 1 stress 0.1350917
## Run 2 stress 0.1226512
## ... New best solution
## ... Procrustes: rmse 0.0008641234 max resid 0.004244483
## ... Similar to previous best
## Run 3 stress 0.1226506
## ... New best solution
## ... Procrustes: rmse 0.0006123831 max resid 0.003005903
## ... Similar to previous best
## Run 4 stress 0.1218541
## ... New best solution
## ... Procrustes: rmse 0.0201598 max resid 0.1286162
## Run 5 stress 0.1218545
## ... Procrustes: rmse 0.0002024207 max resid 0.00101306
## ... Similar to previous best
## Run 6 stress 0.1227526
## Run 7 stress 0.1218547
## ... Procrustes: rmse 0.0002533211 max resid 0.001299704
## ... Similar to previous best
## Run 8 stress 0.1218542
## ... Procrustes: rmse 0.000541007 max resid 0.002711941
```

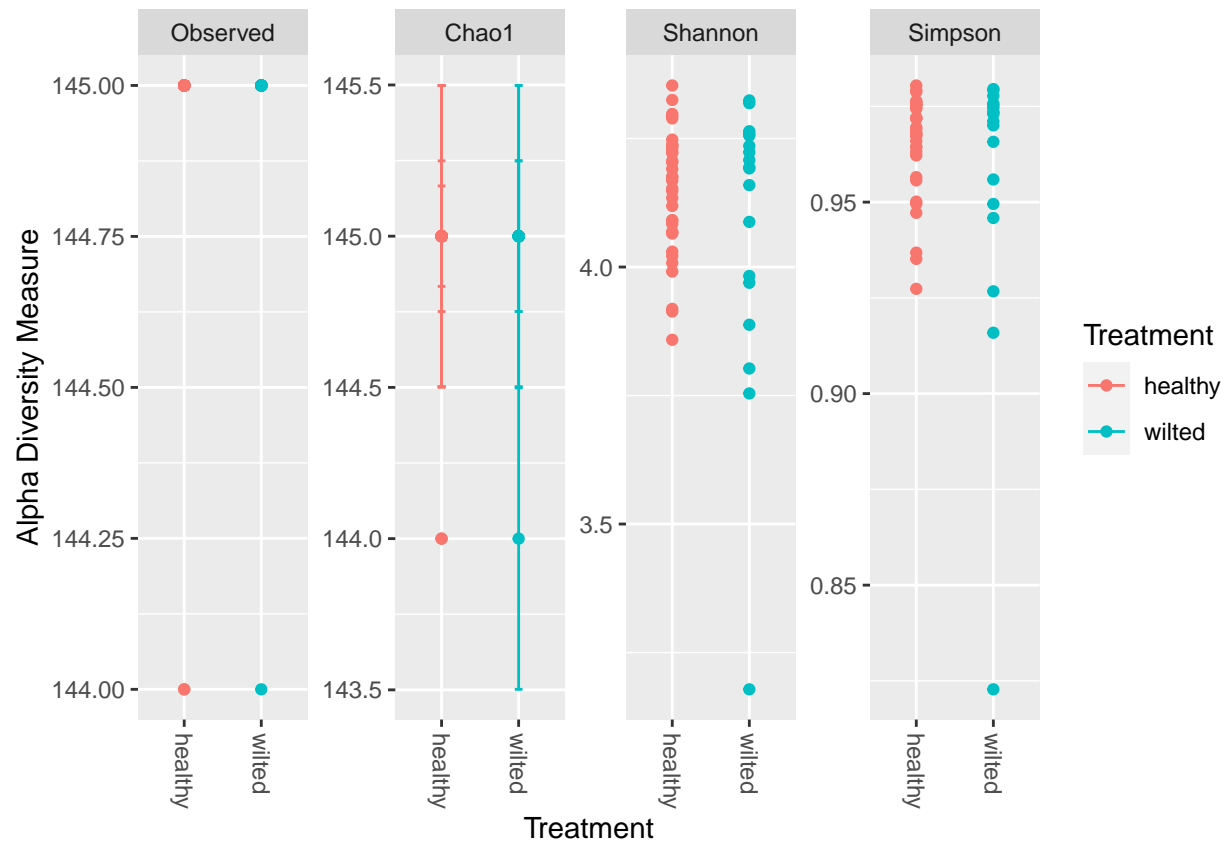
```

## ... Similar to previous best
## Run 9 stress 0.1278288
## Run 10 stress 0.1272837
## Run 11 stress 0.1272842
## Run 12 stress 0.1272836
## Run 13 stress 0.1218539
## ... New best solution
## ... Procrustes: rmse 0.0001043452  max resid 0.0005420929
## ... Similar to previous best
## Run 14 stress 0.1218546
## ... Procrustes: rmse 0.0003174537  max resid 0.001568472
## ... Similar to previous best
## Run 15 stress 0.1218543
## ... Procrustes: rmse 0.0002324318  max resid 0.001171843
## ... Similar to previous best
## Run 16 stress 0.1272842
## Run 17 stress 0.1346961
## Run 18 stress 0.1346963
## Run 19 stress 0.121854
## ... Procrustes: rmse 7.964564e-05  max resid 0.0003900365
## ... Similar to previous best
## Run 20 stress 0.1218546
## ... Procrustes: rmse 0.0006061658  max resid 0.002981664
## ... Similar to previous best
## *** Best solution repeated 5 times

```



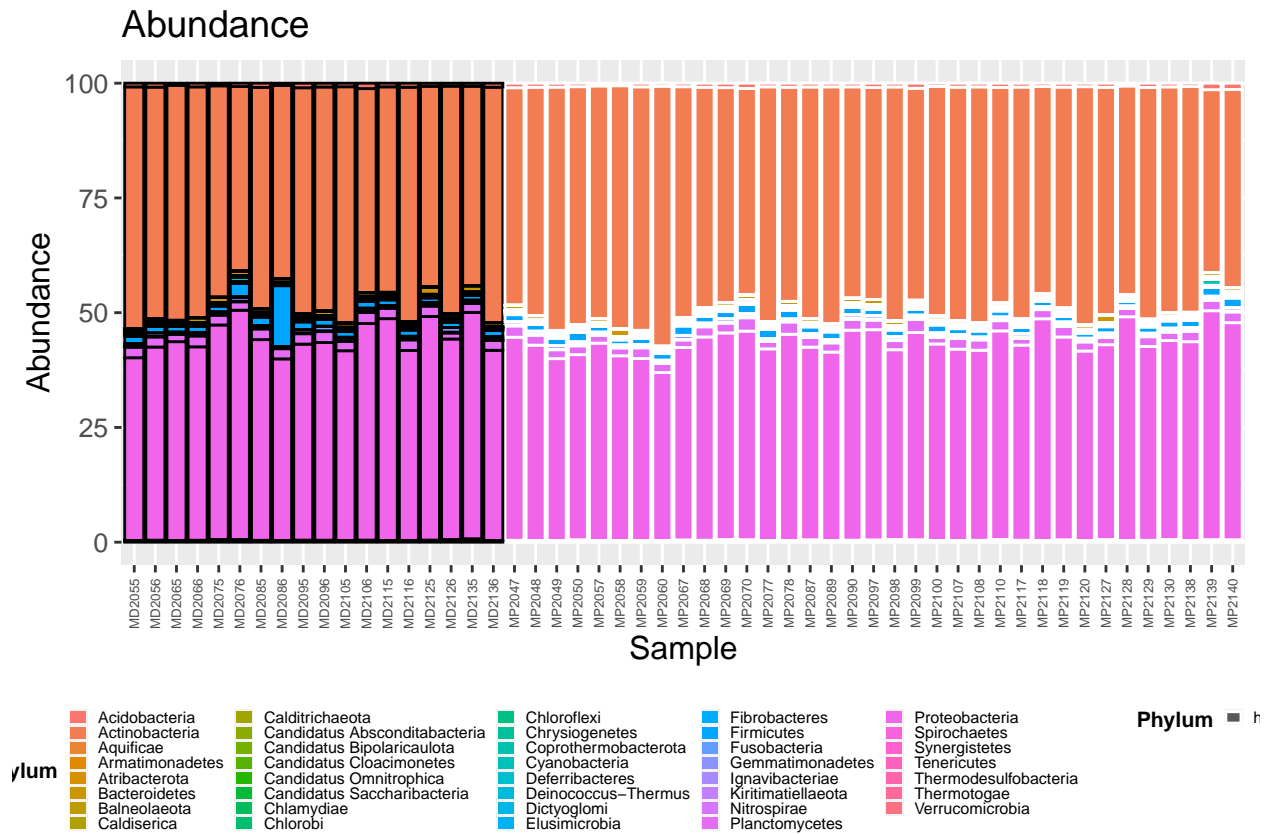
```
Alpha_diversity(merge_Eukaryota , 'Species' , 'Treatment')
```



```
##———Bacteria by Phylum
```

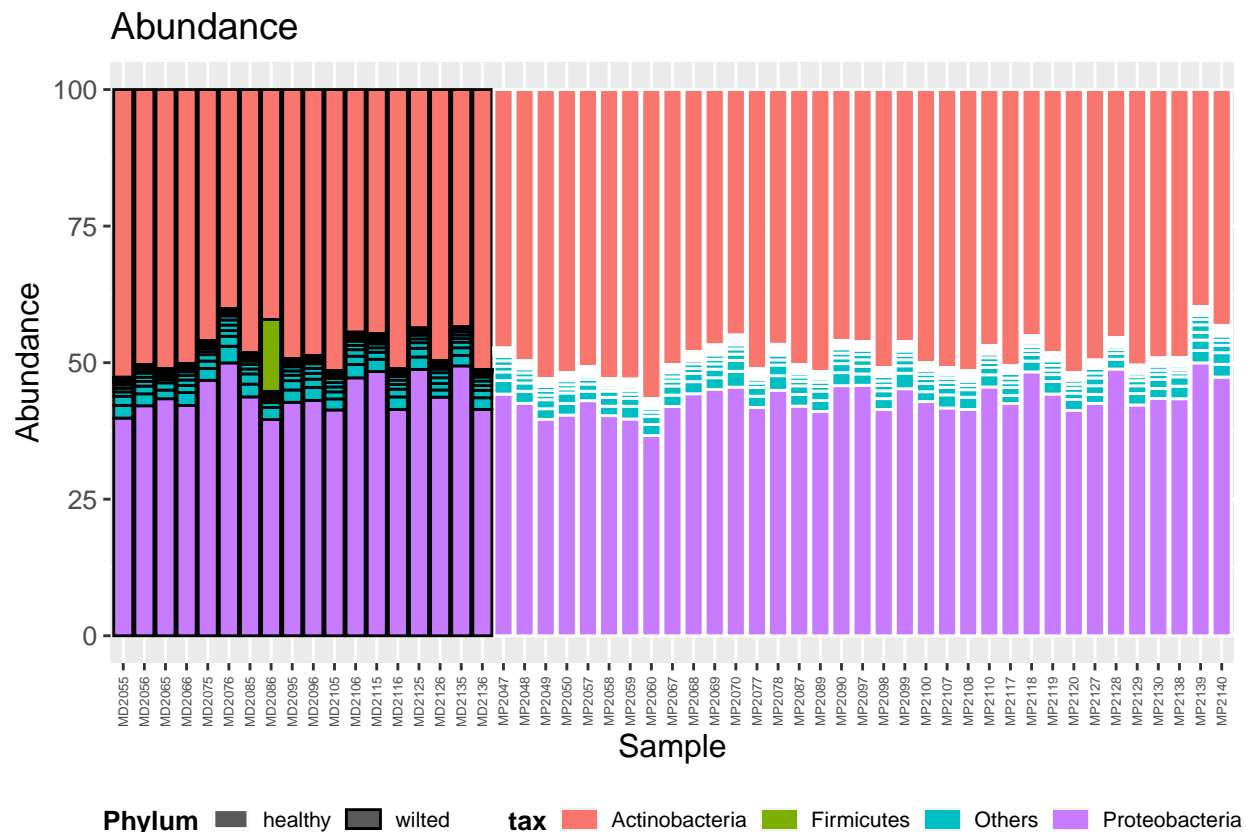
```
Barras_Species <- Abundance_barras(merge_Bacteria,'Phylum','Treatment',10.0)
Barras_Species[1]
```

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



Barras_Species[2]

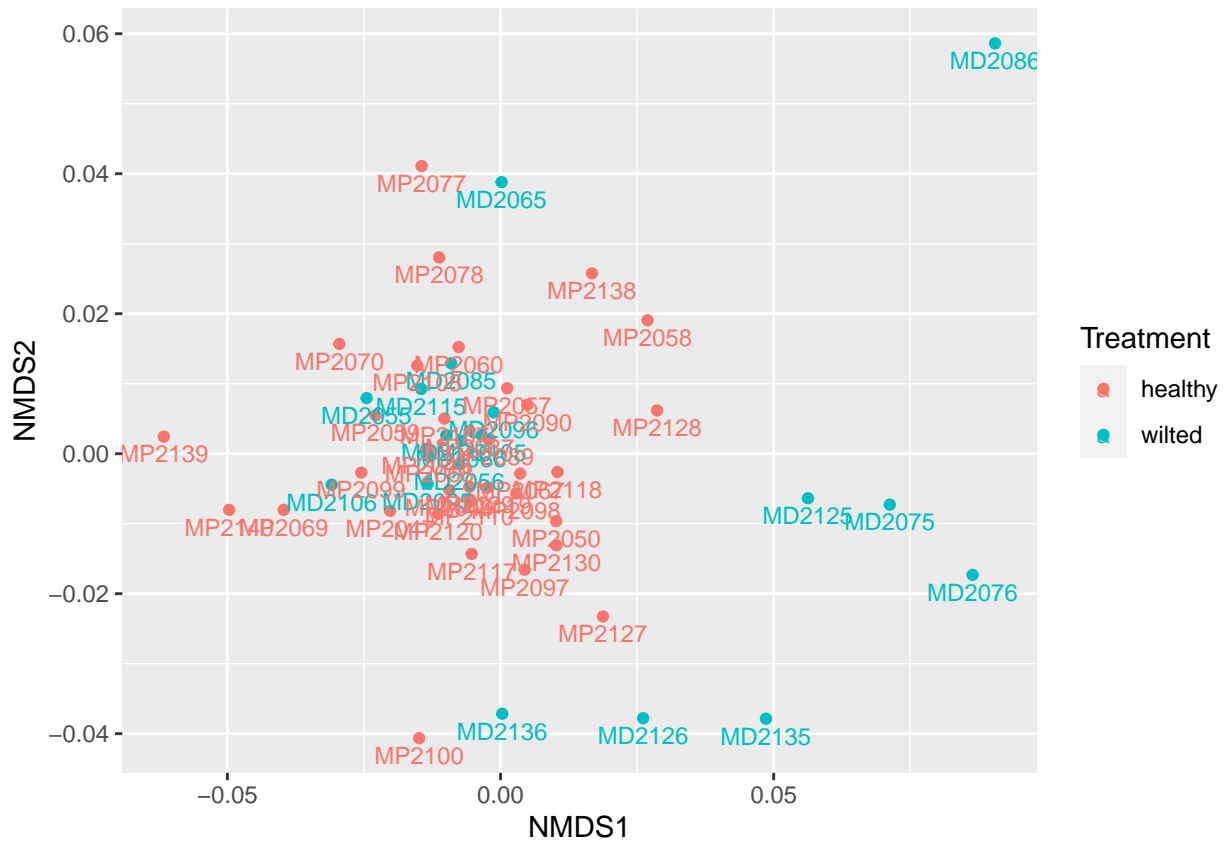
[[1]]



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

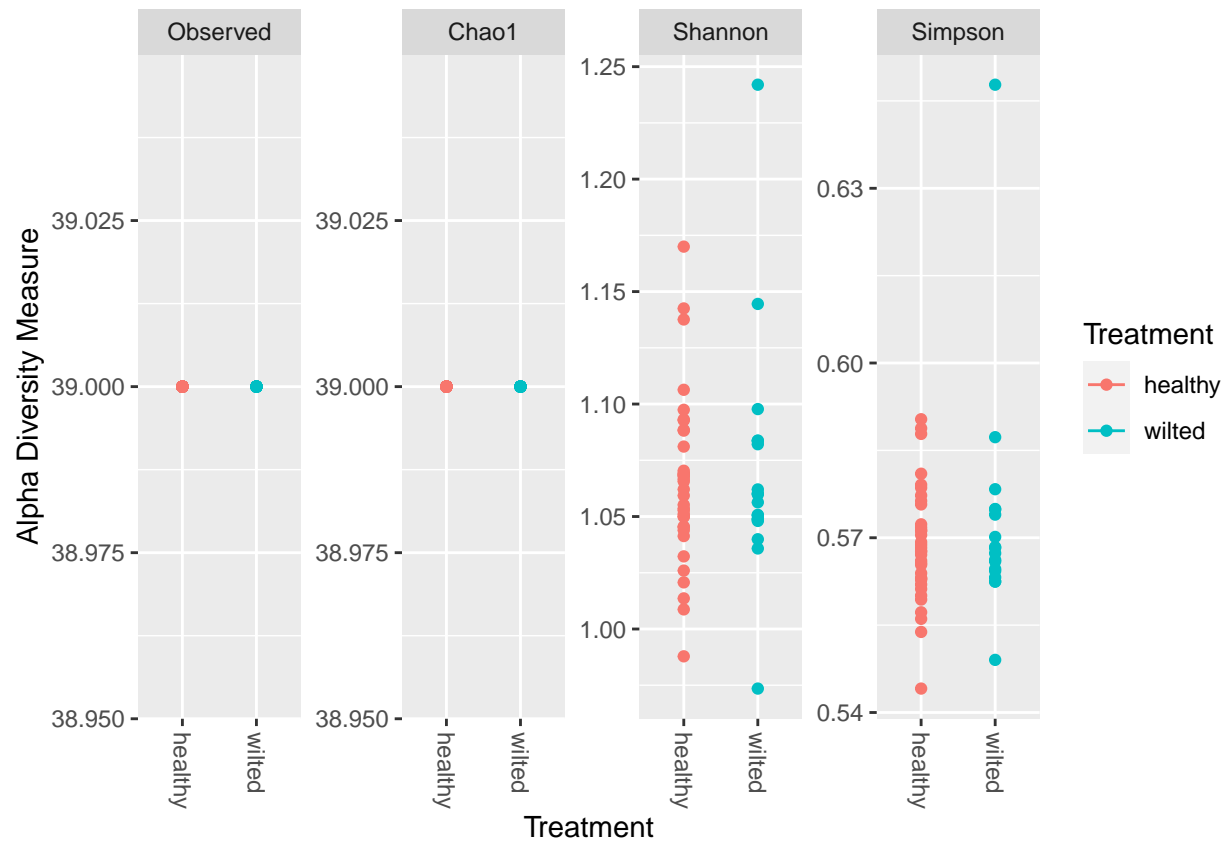
```
## Square root transformation
## Wisconsin double standardization
## Run 0 stress 0.1520004
## Run 1 stress 0.1674646
## Run 2 stress 0.1691779
## Run 3 stress 0.1610424
## Run 4 stress 0.1654743
## Run 5 stress 0.1697232
## Run 6 stress 0.1592903
## Run 7 stress 0.1656345
## Run 8 stress 0.1614665
## Run 9 stress 0.1686077
## Run 10 stress 0.1592034
## Run 11 stress 0.1519726
## ... New best solution
## ... Procrustes: rmse 0.008432635 max resid 0.04770606
## Run 12 stress 0.1524008
## ... Procrustes: rmse 0.04929327 max resid 0.3150102
## Run 13 stress 0.1573638
## Run 14 stress 0.1614019
## Run 15 stress 0.1619357
## Run 16 stress 0.1601871
## Run 17 stress 0.157849
```

```
## Run 18 stress 0.1722158
## Run 19 stress 0.1657873
## Run 20 stress 0.1513902
## ... New best solution
## ... Procrustes: rmse 0.01172811  max resid 0.05381454
## *** Best solution was not repeated -- monoMDS stopping criteria:
##      20: stress ratio > sratmax
```



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

```
## Warning in estimate_richness(physeq, split = TRUE, measures = measures): The data you have provided c
## 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]
```

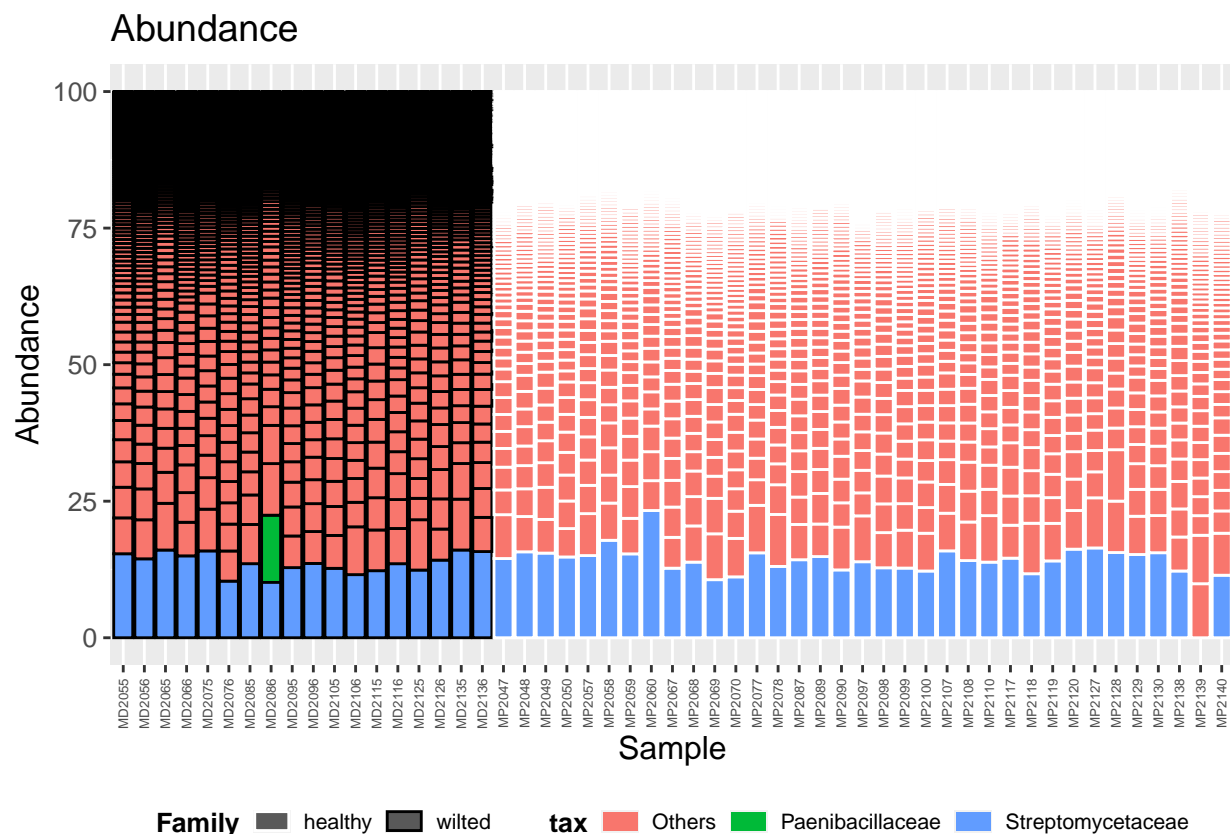
```
## [[1]]
```

Family

Actinomyces	Chloromonadaceae	Halobacteriaceae	Nocardiaceae	St
Actinopolysporaceae	Clostridiaceae	Halobacteroidaceae	Nocardioidaceae	St
Actinopolysporaceae	Clostridiales Family XVI. Incertae Sedis	Halomonadaceae	Nocardiopsaceae	St
Aerococcaceae	Clostridiales Family XVII. Incertae Sedis	Halothiobacillaceae	Nostocaceae	St
Aeromonadaceae	Cohaesibacteraceae	Hapalosiphonaceae	Oceanospirillaceae	St
Akkermansiaceae	Coleofasciculaceae	Helicobacteraceae	Oculatellaceae	St
Alcaligenaceae	Colwelliaceae	Heliothecaceae	Odoribacteraceae	St
Alcanivoracaceae	Comamonadaceae	Holospiraceae	Oleiphilaceae	St
Alicyclobacillaceae	Conexibacteraceae	Hydrogenimonaceae	Opiritaceae	St
Alteromonadaceae	Coprothermobacteraceae	Hydrogenophilaceae	Orbaceae	St
Amoebophilaceae	Coriobacteriaceae	Hydrogenothermaceae	Ornithinimicrobiaceae	Su
Amorphaceae	Corynebacteriaceae	Hyellaceae	Oscillatoriaceae	Su
Anaerohalospiraaceae	Coxiellaceae	Hymenobacteraceae	Oscillospiraceae	Su
Anaerolineaceae	Crocinitomicaceae	Hyphomicrobiaceae	Oxalobacteraceae	Su
Anaeromyxobacteraceae	Cyclobacteriaceae	Hyphomonadaceae	Paenibacillaceae	Su
Anaplasmataceae	Cytophagaceae	Iamaceae	Paludibacteraceae	Sy
Aphanizomenonaceae	Deferribacteraceae	Ichthyobacteriaceae	Parachiamydiaceae	Sy
Aphanothecaceae	Dehalococcoidaceae	Idiomarinaceae	Parvibaculaceae	Sy
Aquificaceae	Deinococcaceae	Ignavibacteriaceae	Parvularculaceae	Sy
Archangiaceae	Demeguinaceae	Ilumatobacteraceae	Pasteurellaceae	Sy
Arcobacteraceae	Dermabacteraceae	Immundisolibacteraceae	Pectobacteriaceae	Sy
Ardenticatenaceae	Dermacoccaceae	Intrasporangiaceae	Pelagibacteraceae	Sy
Atopobiaceae	Dermatophilaceae	Isosphaeraceae	Peptococcaceae	Ta
Atribacteraceae	Dermocarpellaceae	Jatrophihabitantaceae	Peptoniphilaceae	Te
Aurantimonadaceae	Desulfallaceae	Jiangellaceae	Peptostreptococcaceae	Te
Azonexaceae	Desulfarculaceae	Jonesiaceae	Petrotoxiaceae	Te
Azospirillaceae	Desulfatobacteriaceae	Kaistiaceae	Phototrophicaceae	Te
Bacillaceae	Desulfobacteriaceae	Kangiellaceae	Phreatobacteraceae	Te
Bacteriovoracaceae	Desulfobulbaceae	Kineosporiaceae	Phycisphaeraceae	Tr
Bacteroidaceae	Desulfocapsaceae	Kiritimatiellaceae	Phyllobacteriaceae	Tr
Baekduiaceae	Desulfohalobiaceae	Koferiaceae	Pirellulaceae	Tr
Barnesiellaceae	Desulfomicrobiaceae	Koleobacteraceae	Piscirickettsiaceae	Tr
Bartonellaceae	Desulfosudaceae	Kordiimonadaceae	Planctomycetaceae	Tr
Bdellovibrionaceae	Desulfotomaculaceae	Kosmotogaceae	Planococcaceae	Tr
Beijerinckiaceae	Desulfotomaculaceae	Kribbellaceae	Pleomorphomonadaceae	Tr
Bernardetiaceae	Desulfurellaceae	Ktedonosporobacteraceae	Polyangiaceae	Tr
Beutenbergiaceae	Desulfurobacteriaceae	Kytococcaceae	Porphyromonadaceae	Tr
Bifidobacteriaceae	Desulfuromonadaceae	Labilitrachaceae	Prevotellaceae	Tr
Blastochloridaceae	Devosiaceae	Lachnospiraceae	Prochlorococcaceae	Tr
Blattabacteriaceae	Dictyoglomaceae	Lacipirellulaceae	Prochlorotrichaceae	Tr
Bogoriellaceae	Dietziaceae	Lactobacillaceae	Prolixibacteraceae	Tr
Borrelliaceae	Dissulfurispiraceae	Lawsonellaceae	Promicromonosporaceae	Tr
Boseaceae	Dysgonomonadaceae	Legionellaceae	Propionibacteriaceae	Tr
Brachyspiraceae	Ectothiorhodospiraceae	Leptolyngbyaceae	Proteinivoraceae	Tr
Bradymonadaceae	Eggerthellaceae	Leptospiaceae	Pseudanabaenaceae	Tr
Bradyrhizobiaceae	Egibacteraceae	Leptotrichiaceae	Pseudoalteromonadaceae	Tr
Breoghanaceae	Egicoccaceae	Lichenihabitantaceae	Pseudomonadaceae	Tr
Brevibacteriaceae	Elioraeaceae	Limnochordaceae	Pseudonocardaceae	Tr
Brucellaceae	Elusimicrobiaceae	Listeriaceae	Psychromonadaceae	Tr
Bruguierivoracaceae	Emcibacteraceae	Litoricolaceae	Punicococcaceae	Tr
Bryobacteraceae	Endomicrobiaceae	Magnetococcaceae	Rhabdochlamydiaceae	Tr
Budviciaceae	Endozoicomonadaceae	Mangrovivirgaceae	Rhizobiaceae	Tr
Burkholderiaceae	Enterobacteriaceae	Maricaulaceae	Rhodanobacteraceae	Ti
Caedimonadaceae	Enterococcaceae	Marinifilaceae	Rhodobacteraceae	Ti
Caldilineaceae	Entomoplasmataceae	Marinifilaceae	Rhodocyclaceae	Tc
Caldiseriaceae	Erwinaceae	Marinobacteraceae	Rhodospirillaceae	Tr
Calditrichaceae	Erysipelotrichaceae	Mariprofundaceae	Rhodothermaceae	Tr
Calothrixaceae	Erythrobacteraceae	Marinimaceae	Rickettsiaceae	Tr

Barras_Species [2]

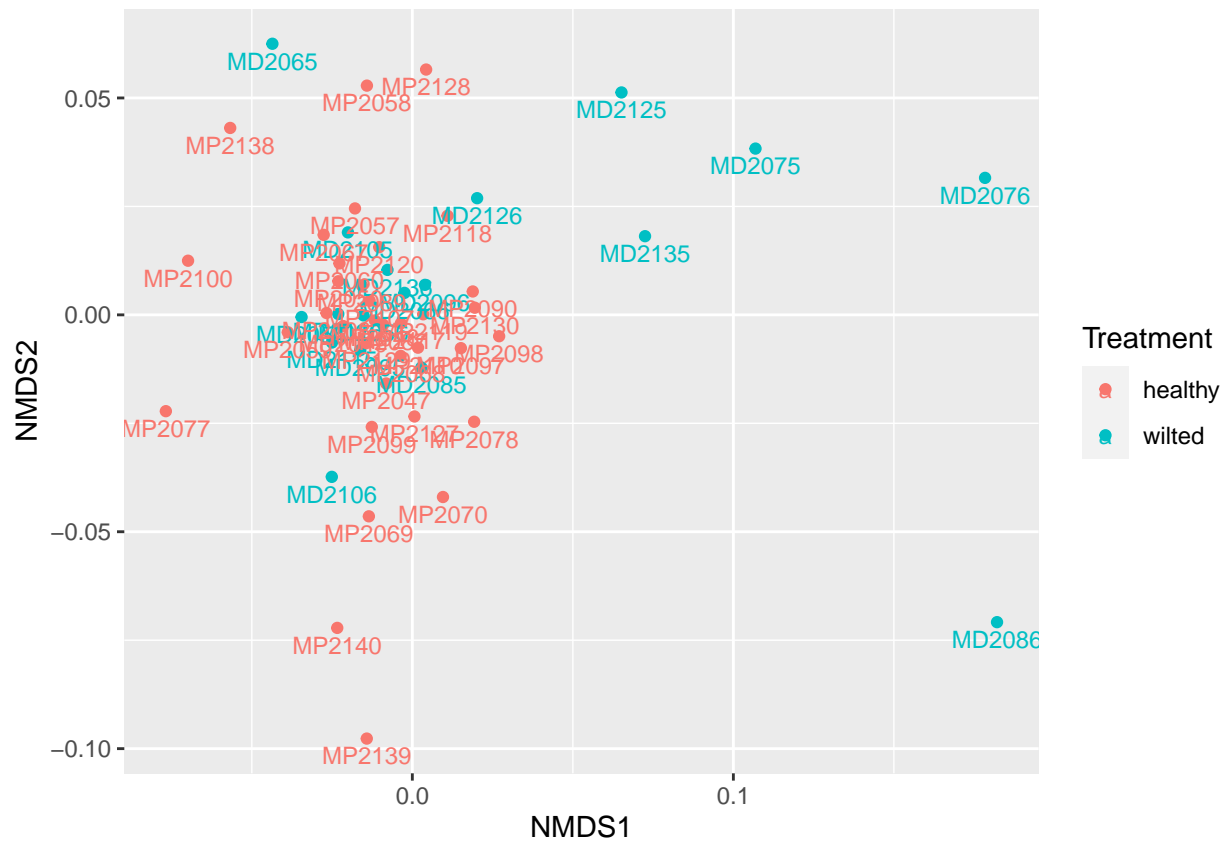
[[1]]



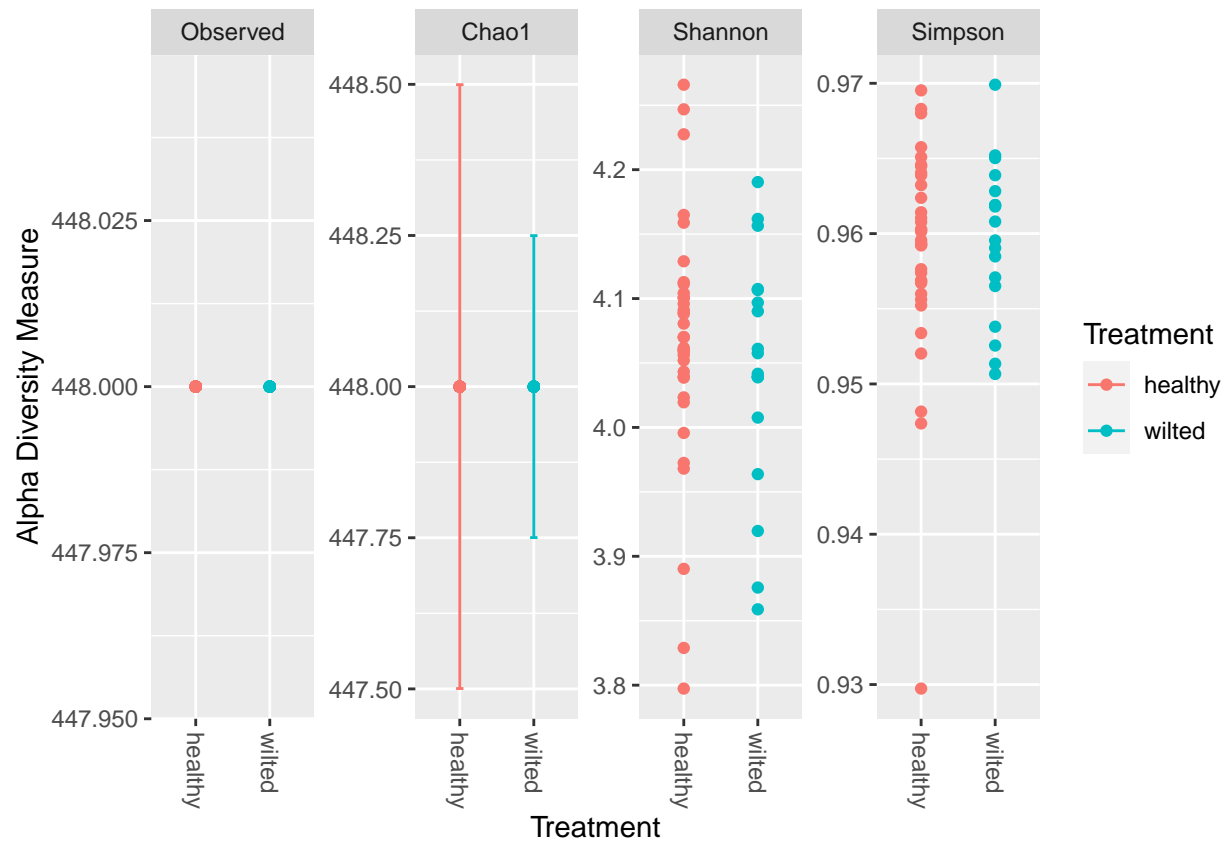
```
Beta_diversity(merge_Bacteria , 'Family' , 'Treatment', 'bray')
```

```
## Wisconsin double standardization
## Run 0 stress 0.1412073
## Run 1 stress 0.1402351
## ... New best solution
## ... Procrustes: rmse 0.0294344 max resid 0.1450997
## Run 2 stress 0.1418149
## Run 3 stress 0.1396507
## ... New best solution
## ... Procrustes: rmse 0.01731879 max resid 0.08105936
## Run 4 stress 0.1728168
## Run 5 stress 0.1412067
## Run 6 stress 0.1825783
## Run 7 stress 0.1521891
## Run 8 stress 0.1400278
## ... Procrustes: rmse 0.0191409 max resid 0.07897066
## Run 9 stress 0.1735308
## Run 10 stress 0.1575206
## Run 11 stress 0.1720444
## Run 12 stress 0.1371727
## ... New best solution
## ... Procrustes: rmse 0.05360706 max resid 0.3267978
## Run 13 stress 0.1518195
## Run 14 stress 0.1370387
```

```
## ... New best solution
## ... Procrustes: rmse 0.0125958  max resid 0.05755869
## Run 15 stress 0.1398013
## Run 16 stress 0.1687823
## Run 17 stress 0.1371929
## ... Procrustes: rmse 0.0113669  max resid 0.05681366
## Run 18 stress 0.1550932
## Run 19 stress 0.1647356
## Run 20 stress 0.1824082
## *** Best solution was not repeated -- monoMDS stopping criteria:
##      7: no. of iterations >= maxit
##     13: stress ratio > sratmax
```



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



```
## ----- Bacteria by Genero
```

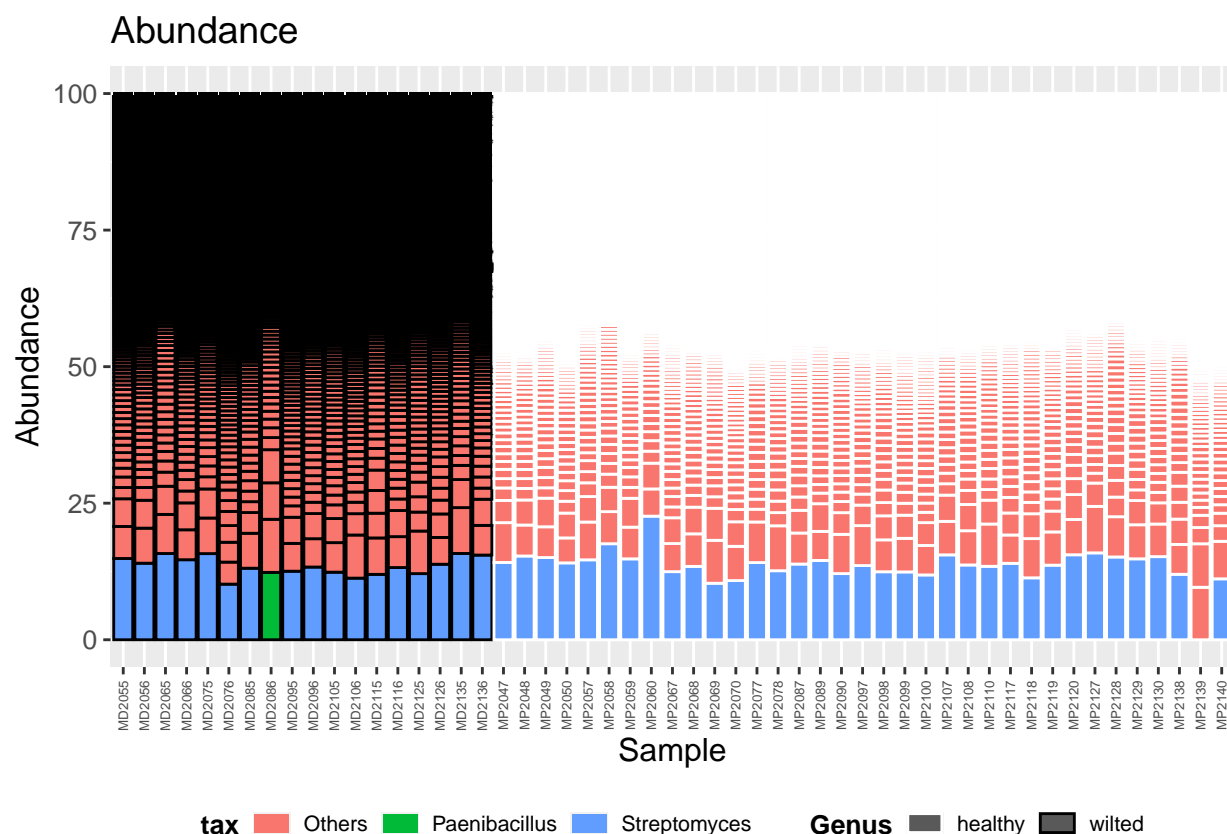
```
Barras_Species <- Abundance_barras(merge_Bacteria, 'Genus', 'Treatment', 10.0)
Barras_Species[1]
```

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

lex	Desulfuricoccus	Labyrinthula	Paramecium	Sulfolobus
flexum	Desulfatibacillum	Labrys	Paroceanicella	Succinivibrio
habitans	Desulfatibacterium	Laceyella	Parolsenella	Suicoccus
luna	Desulfobacca	Lachnoanaerobaculum	Parvibaculum	Sulfidibacter
marina	Desulfobacter	Lachnoclostridium	Parvimonas	Sulfitobacter
ncola	Desulfobacula	Lachnospira	Parvularcula	Sulfuricaulis
rhabdus	Desulfobulbus	Lacibacter	Pasteurella	Sulfuricella
rufa	Desulfocapsa	Lacimicrobium	Paucibacter	Sulfuricurvum
salimonas	Desulfococcus	Lacinutrix	Paucilactobacillus	Sulfuriferula
salinus	Desulfocurvibacter	Lacipirellula	Pauljensenia	Sulfuriflexus
sediminimonas	Desulfofarcimen	Lacrimispora	Pectinatus	Sulfurifustis
sphaera	Desulfoglaeba	Lacticaseibacillus	Pectobacterium	Sulfurihydrogenibium
talea	Desulfoglobium	Lactiplantibacillus	Pediococcus	Sulfurimicrobium
ibacter	Desulfoluna	Lactobacillus	Pedobacter	Sulfurimonas
hidicoccus	Desulfolutivibrio	Lactococcus	Pedococcus	Sulfuriroseicoccus
hnia	Desulfomarina	Lacunisphaera	Pelagerythrobacter	Sulfuritalea
nobacterium	Desulfomicrobium	Lancefieldella	Pelagibacterium	Sulfuritortus
angium	Desulfomonile	Lapidilactobacillus	Pelagovum	Sulfurivermis
bacter	Desulfonema	Laribacter	Pelolistega	Sulfurospirillum
cibacterium	Desulforamulus	Larkinella	Pelobacter	Sulfurovum
ibacter	Desulforapulum	Latilactobacillus	Pelodictyon	Sutcliffeella
imonas	Desulfosarcina	Lautropia	Pelolinea	Sutterella
ratoleum	Desulfoscipio	Lawsonella	Pelosinus	Suttonella
nicrococcus	Desulfosediminicola	Lawsonia	Pengzhengrongella	Swingsia
nophonus	Desulfosporosinus	Leadbetterella	Peptacetobacter	Symbiobacterium
obacter	Desulfosudis	Leclercia	Peptoclostridium	Symmachella
a	Desulfotalea	Lederbergia	Peptoniphilus	Synechococcus
acaulis	Desulfotomaculum	Leeuwenhoekella	Peribacillus	Synechocystis
lassotoga	Desulfovibrio	Legionella	Periweissella	Syntrophobacter
tibacter	Desulfurispirillum	Leifsonia	Permianibacter	Syntrophobotulus
obium	Desulfurivibrio	Leisingera	Persephonella	Syntrophomonas
acter	Desulfurobacterium	Lelliottia	Persicimonas	Syntrophotalea
ntiibacter	Desulfuromonas	Leminorella	Peteryoungia	Syntrophothermus
ntimicrobium	Devosia	Lentibacillus	Petrimonas	Syntrophus
ntimonas	Devriesea	Lentilactobacillus	Petrocella	Tabrizicola
ticoccus	Dialister	Lentilitoribacter	Petrotoga	Tamiana
imonas	Diaminobutyricimonas	Lentzea	Phaeobacter	Tannerella
liella	Diaphorobacter	Leptodesmis	Phascolarctobacterium	Tardibacter
idibacter	Dichelobacter	Leptolyngbya	Pherylobacterium	Tardiphaga
wickia	Dickeya	Leptosira	Phnomibacter	Tateyamaria
acterium	Dictyoglomus	Leptospirillum	Phocaeicola	Tatlockia
rcus	Dietzia	Leptothermofonsia	Phoenicibacter	Tatumella
hizobium	Dinoroseobacter	Leptothrix	Photobacterium	Tautonia
pira	Dissulfurimicrobium	Leptotrichia	Photorhabdus	Taylorella
pirillum	Dissulfurispira	Leucobacter	Phototrophicus	Telmatocola
obacter	Dokdonella	Leuconostoc	Phreatobacter	Tenacibaculum
llus	Dokdonia	Levilactobacillus	Phycococcus	Tenuifilum
eriplanes	Dolichospermum	Liberibacter	Phycisphaera	Tepidanaerobacter
eriovorax	Dolosigranulum	Lichenicola	Phyllobacterium	Tepidibacter
eroides	Dongshaeta	Lichenihabitans	Phytobacter	Tepidiforma
duia	Dorea	Ligilactobacillus	Phytohabitans	Tepidimonas
esiella	Draconibacterium	Lignipirellula	Pigmentiphaga	Tepiditoga
ia	Duganella	Limihaloglobus	Pikeienueella	Terasakiella
lea	Duncaniella	Limnobacter	Pimelobacter	Teredinibacter
lovibrio	Dyadobacter	Limnobaculum	Pirellula	Terribacillus
giatota	Dyella	Limnochorda	Pirellulimonas	Terricaulis
irickia	Dysgonomonas	Limnoglobus	Piscirickettsia	Terriglobus
	Dysnomobacter	Limnolobus	Piscirickettsia	Terrirhabdus

Barras_Species [2]

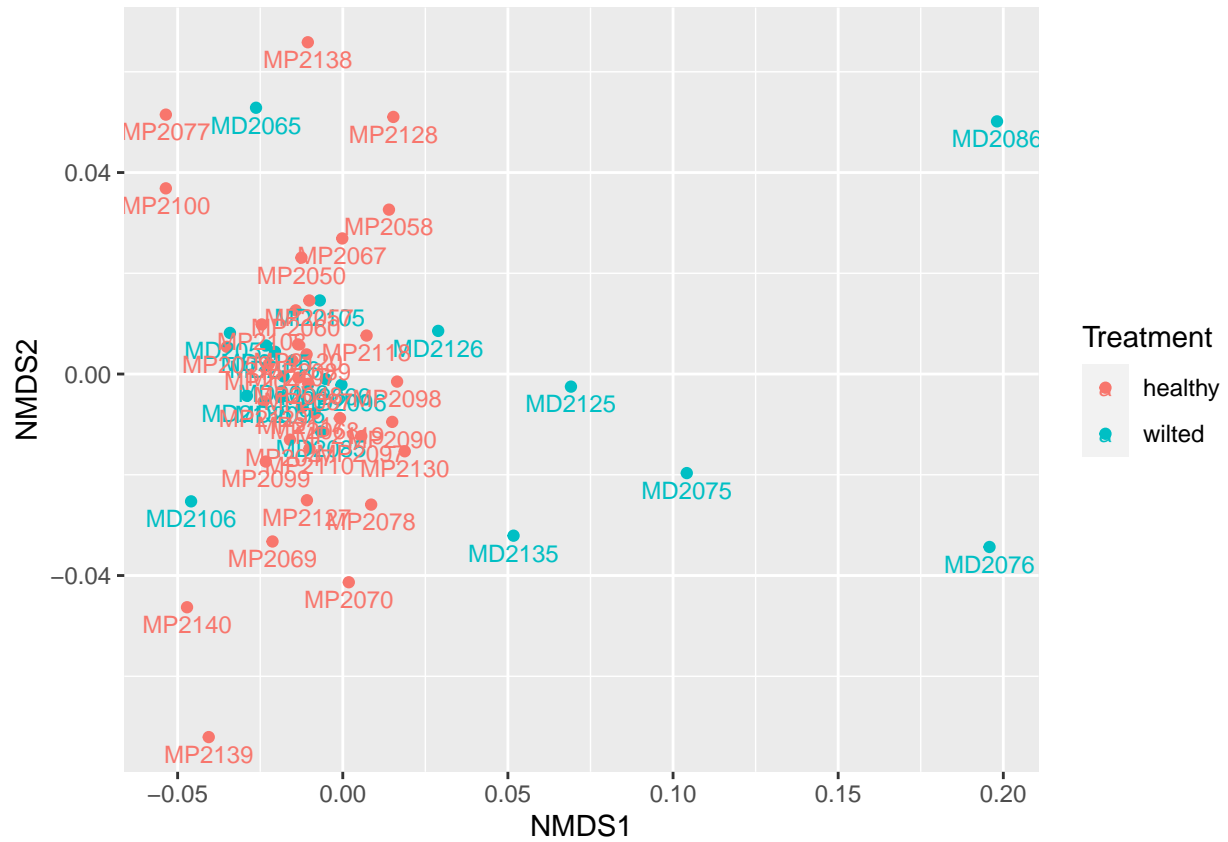
[[1]]



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

```
## Wisconsin double standardization
## Run 0 stress 0.1467945
## Run 1 stress 0.1432834
## ... New best solution
## ... Procrustes: rmse 0.08893595 max resid 0.3293612
## Run 2 stress 0.1432845
## ... Procrustes: rmse 0.0009707145 max resid 0.004281232
## ... Similar to previous best
## Run 3 stress 0.1469874
## Run 4 stress 0.1473387
## Run 5 stress 0.1528047
## Run 6 stress 0.1437863
## Run 7 stress 0.1437853
## Run 8 stress 0.1432832
## ... New best solution
## ... Procrustes: rmse 0.0001931802 max resid 0.0009023473
## ... Similar to previous best
## Run 9 stress 0.1458083
## Run 10 stress 0.1455706
## Run 11 stress 0.1437864
## Run 12 stress 0.1579725
## Run 13 stress 0.1438141
## Run 14 stress 0.1479572
```

```
## Run 15 stress 0.1438153
## Run 16 stress 0.1499361
## Run 17 stress 0.1477432
## Run 18 stress 0.1514192
## Run 19 stress 0.1438482
## Run 20 stress 0.1451995
## *** Best solution repeated 1 times
```

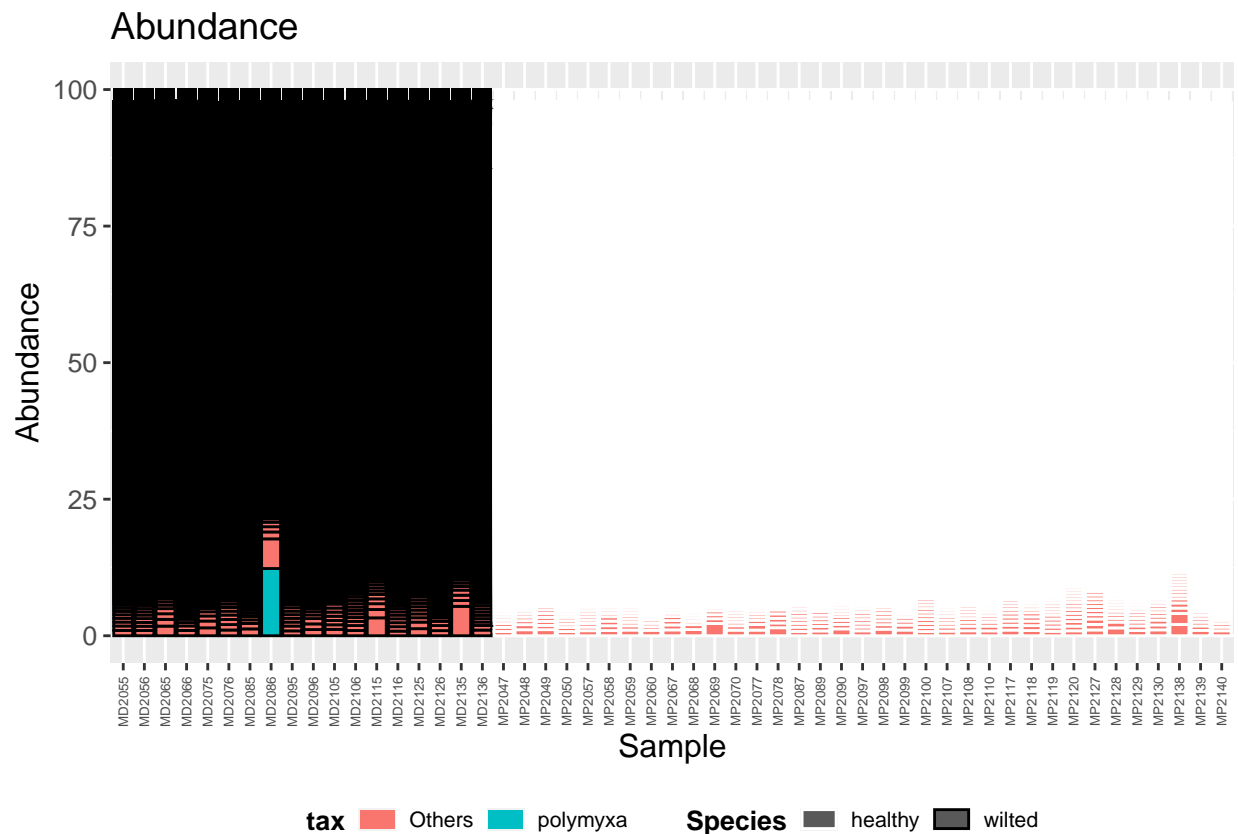


```
Alpha_diversity(merge_Bacteria , 'Genus' , 'Treatment')
```


e		lactamica	sp. 'Soap Lake #7'	sp. KSB-15	sp. X
		lactarius	sp. (Blaberus giganteus)	sp. KSM-R2A25	sp. X
		lactatifermentans	sp. (Blatta orientalis)	sp. KSM-R2A30	sp. X
		lactatiformans	sp. (Blattella germanica)	sp. KT 15	sp. X
		lactis	sp. (Cryptocercus kyebangensis)	sp. KT25b	sp. X
		lactucaae	sp. (Cryptocercus punctulatus) str. Cpu	sp. KTR9	sp. X
		lacunae	sp. (ex Adelges kitamiensis)	sp. KUDC0405	sp. X
		lacus	sp. (ex Biomphalaria glabrata)	sp. KUDC0406	sp. X
		lacuslunae	sp. (Mastotermes darwiniensis)	sp. KUDC1026	sp. X
		lacustris	sp. (Nauphoeta cinerea)	sp. KUDC1714	sp. X
		laguerreae	sp. (Periplaneta americana)	sp. KUIN-1	sp. X
		laidlawii	sp. 001	sp. Kuro-4	sp. X
		laixinhei	sp. 007	sp. KW1	sp. X
		laikuanensis	sp. 008	sp. KX20019	sp. X
		laminatus	sp. 02C 26	sp. KY-GH-1	sp. X
		lanienae	sp. 09C 129	sp. KY-YJ-3	sp. X
		lanii	sp. 09RB8471	sp. KY3	sp. Y
		lansingensis	sp. 09RB8910	sp. KY5	sp. Y
		lanthieri	sp. 1_2014MBL_MicDiv	sp. KY70	sp. Y
		lapagei	sp. 1_2015MBL_MicDiv	sp. KY75	sp. Y
		lapidicaptus	sp. 1-1C	sp. L-07	sp. Y
		lapsinans	sp. 1.5R	sp. L-2-11	sp. Y
		lari	sp. 1(2017)	sp. L-8-10	sp. Y
		lariciata	sp. 103DPR2	sp. L-8-3	sp. Y
		larrymorei	sp. 1063	sp. L-A4	sp. Y
		larvae	sp. 107-1	sp. L12M9	sp. Y
		lata	sp. 1070	sp. L1A13	sp. Y
		latens	sp. 10FS3-1	sp. L1A34	sp. Y
		laterosporus	sp. 10M-3C3	sp. L1A9	sp. Y
		laumondii	sp. 10RB9215	sp. L1139	sp. Y
		lautus	sp. 11-1-2	sp. L1SW	sp. Y
		lavamentivorans	sp. 11-B-312	sp. L2A1	sp. Y
		lavendulae	sp. 113-1-2	sp. L2A11	sp. Y
		leachii	sp. 113-3-3	sp. L3-i22	sp. Y
		leadbetteri	sp. 113-3-9	sp. L3-i23	sp. Y
		leeuwenhoekii	sp. 1137	sp. L3A3	sp. Y
		leguminosarum	sp. 113P3	sp. L3A6	sp. Y
		lehensis	sp. 114	sp. L3A8	sp. Y
		lekithochrous	sp. 11515TR	sp. L5	sp. Y
		lemovicicum	sp. 116-D4	sp. L51/94	sp. Y
		lemurum	sp. 119287	sp. L5B5	sp. Y
		lenghuensis	sp. 11B	sp. L6-1	sp. Y
		lenta	sp. 11kri321	sp. L9-4	sp. Y
		lentiflavum	sp. 12200R-103	sp. LA-2-3-30-S1-D2	sp. Y
		lentis	sp. 13-15	sp. LA112445	sp. Y
		lentocellum	sp. 131	sp. LA31	sp. Y
		lentus	sp. 131-2-1	sp. LAB-08	sp. Y
		leopoldii	sp. 131-2-5	sp. LADL05-105	sp. Y
		leprae	sp. 131-3-5	sp. LAS2	sp. Y
		lepromatosis	sp. 13159349	sp. LB1	sp. Y
		leptomitoformis	sp. 135	sp. LBG001	sp. Y
		lettingae	sp. 140616W15	sp. LBUM 1475	sp. Y
andiella		liangguodongii	sp. 14171R-50	sp. LBUM 1480	sp. Y
ibbosus		liangshanensis	sp. 14181154	sp. LBUM 1482	sp. Y
ifera		liaowanqingii	sp. 143-21	sp. LBUM920	sp. Y
		libanensis	sp. 144S4	sp. LC2018020214	sp. Y
		licheniformis	sp. 15-184	sp. LC6	sp. Y

Barras_Species [2]

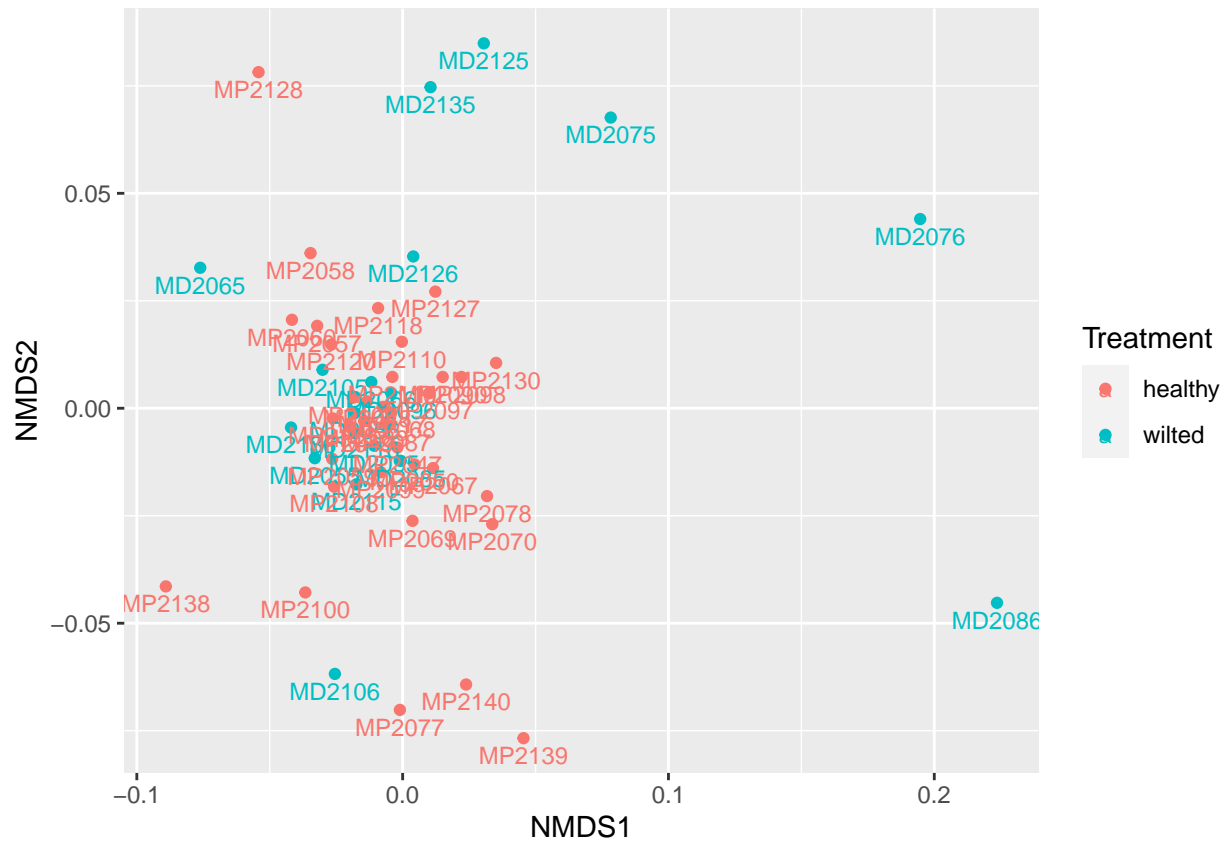
[[1]]



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

```
## Wisconsin double standardization
## Run 0 stress 0.1597841
## Run 1 stress 0.1710249
## Run 2 stress 0.1602923
## Run 3 stress 0.1710683
## Run 4 stress 0.1571545
## ... New best solution
## ... Procrustes: rmse 0.05851399 max resid 0.2993421
## Run 5 stress 0.1633308
## Run 6 stress 0.1603458
## Run 7 stress 0.1568651
## ... New best solution
## ... Procrustes: rmse 0.01116618 max resid 0.05529493
## Run 8 stress 0.1702595
## Run 9 stress 0.1614495
## Run 10 stress 0.1571296
## ... Procrustes: rmse 0.01084661 max resid 0.05428821
## Run 11 stress 0.1630772
## Run 12 stress 0.158739
## Run 13 stress 0.1600774
## Run 14 stress 0.1920147
## Run 15 stress 0.1590775
## Run 16 stress 0.1582729
```

```
## Run 17 stress 0.1755464
## Run 18 stress 0.1582612
## Run 19 stress 0.1746779
## Run 20 stress 0.1710691
## *** Best solution was not repeated -- monoMDS stopping criteria:
##      6: no. of iterations >= maxit
##     14: stress ratio > sratmax
```



```
Alpha_diversity(merge_Bacteria , 'Species' , 'Treatment')
```

