

Beta-diversity

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Load the necessary libraries

```
library(ggpubr)
```

```
## Loading required package: ggplot2
```

```
library(Tjazi)
```

```
library(scater)
```

```
## Loading required package: SingleCellExperiment
```

```
## Loading required package: SummarizedExperiment
```

```
## Loading required package: MatrixGenerics
```

```
## Loading required package: matrixStats
```

```
##
```

```
## Attaching package: 'MatrixGenerics'
```

```
## The following objects are masked from 'package:matrixStats':
```

```
##
```

```
## colAlls, colAnyNAs, colAnys, colAvgPerRowSet, colCollapse,  
## colCounts, colCummaxs, colCummins, colCumprods, colCumsums,  
## colDiffs, colIQRDiffs, colIQRs, colLogSumExps, colMadDiffs,  
## colMads, colMaxs, colMeans2, colMedians, colMins, colOrderStats,  
## colProds, colQuantiles, colRanges, colRanks, colSdDiffs, colSds,  
## colSums2, colTabulates, colVarDiffs, colVars, colWeightedMads,  
## colWeightedMeans, colWeightedMedians, colWeightedSds,  
## colWeightedVars, rowAlls, rowAnyNAs, rowAnys, rowAvgPerColSet,  
## rowCollapse, rowCounts, rowCummaxs, rowCummins, rowCumprods,  
## rowCumsums, rowDiffs, rowIQRDiffs, rowIQRs, rowLogSumExps,  
## rowMadDiffs, rowMads, rowMaxs, rowMeans2, rowMedians, rowMins,  
## rowOrderStats, rowProds, rowQuantiles, rowRanges, rowRanks,  
## rowSdDiffs, rowSds, rowSums2, rowTabulates, rowVarDiffs, rowVars,  
## rowWeightedMads, rowWeightedMeans, rowWeightedMedians,  
## rowWeightedSds, rowWeightedVars
```

```
## Loading required package: GenomicRanges
```

```
## Loading required package: stats4
```

```
## Loading required package: BiocGenerics
```

```
##
```

```
## Attaching package: 'BiocGenerics'
```

```
## The following objects are masked from 'package:stats':
```

```
##
```

```

##      IQR, mad, sd, var, xtabs
## The following objects are masked from 'package:base':
##
##      anyDuplicated, aperm, append, as.data.frame, basename, cbind,
##      colnames, dirname, do.call, duplicated, eval, evalq, Filter, Find,
##      get, grep, grepl, intersect, is.unsorted, lapply, Map, mapply,
##      match, mget, order, paste, pmax, pmax.int, pmin, pmin.int,
##      Position, rank, rbind, Reduce, rownames, sapply, setdiff, sort,
##      table, tapply, union, unique, unsplit, which.max, which.min
## Loading required package: S4Vectors
##
## Attaching package: 'S4Vectors'
## The following object is masked from 'package:utils':
##
##      findMatches
## The following objects are masked from 'package:base':
##
##      expand.grid, I, unname
## Loading required package: IRanges
## Loading required package: GenomeInfoDb
## Loading required package: Biobase
## Welcome to Bioconductor
##
##      Vignettes contain introductory material; view with
##      'browseVignettes()'. To cite Bioconductor, see
##      'citation("Biobase")', and for packages 'citation("pkgname)".
##
## Attaching package: 'Biobase'
## The following object is masked from 'package:MatrixGenerics':
##
##      rowMedians
## The following objects are masked from 'package:matrixStats':
##
##      anyMissing, rowMedians
## Loading required package: scuttle
pseudocount <- 1

#### Niger PCoA analysis ####

Gobero_species <- read.csv("ExportedTables-Gobero-species-samples.qza/Gobero-species.csv",
  row.names = 1, header = TRUE, check.names = FALSE)
metadata2 <- read.table("Library_METADATA.txt", header = TRUE,
  check.names = TRUE)

Gobero_species_pseudo <- Gobero_species + pseudocount

Gobero_sp_clr <- clr_lite(counts = Gobero_species_pseudo, method = "const",

```

```

    samples_are = "cols")

df <- calculateMDS(Gobero_sp_clr, method = "euclidean")

euclidean_pcoa_df <- data.frame(pcoa1 = df[, 1], pcoa2 = df[,
  2])

eucl_Lib_pcoa_df <- cbind(euclidean_pcoa_df, metadata2$LibraryMethod,
  metadata2$ExtractionType)

## Warning in data.frame(..., check.names = FALSE): row names were found from a
## short variable and have been discarded

plot1 <- ggplot(data = eucl_Lib_pcoa_df, aes(x = pcoa1, y = pcoa2,
  color = eucl_Lib_pcoa_df$`metadata2$LibraryMethod`, shape = eucl_Lib_pcoa_df$`metadata2$ExtractionType`,
  geom_point() + labs(x = "Coordinate 1", y = "Coordinate 2",
  title = "PCoA for Niger Samples") + theme(title = element_text(size = 12),
  panel.grid = element_blank()) + scale_color_manual(name = "Library Method",
  values = c(DoubleStranded = "red", SingleStranded = "blue"),
  labels = c("Double Stranded", "Single Stranded")) + scale_shape_manual(name = "Extraction Method",
  values = c(PB_Dabney = 16, QG_Rohland = 17), labels = c("PB method",
    "QG method"))

#### Hungarian PCoA analysis ####

Hungarian_species <- read.csv("Hungary-species-samples.qza.csv",
  row.names = 1, header = TRUE, check.names = FALSE)

Hungarian_species_pseudo <- Hungarian_species + pseudocount

Hungarian_sp_clr <- clr_lite(counts = Hungarian_species_pseudo,
  method = "const", samples_are = "cols")

df2 <- calculateMDS(Hungarian_sp_clr, method = "euclidean")

euclidean_pcoa_df2 <- data.frame(pcoa1 = df2[, 1], pcoa2 = df2[,
  2])

eucl_Lib_pcoa_df2 <- cbind(euclidean_pcoa_df2, metadata2$LibraryMethod,
  metadata2$ExtractionType)

## Warning in data.frame(..., check.names = FALSE): row names were found from a
## short variable and have been discarded

plot2 <- ggplot(data = eucl_Lib_pcoa_df2, aes(x = pcoa1, y = pcoa2,
  color = `metadata2$LibraryMethod`, shape = `metadata2$ExtractionType`)) +
  geom_point() + labs(x = "Coordinate 1", y = "Coordinate 2",
  title = "PCoA for Hungarian Samples") + theme(title = element_text(size = 12),
  panel.grid = element_blank()) + scale_color_manual(name = "Library Method",
  values = c(DoubleStranded = "red", SingleStranded = "blue"),
  labels = c("Double Stranded", "Single Stranded")) + scale_shape_manual(name = "Extraction Method",
  values = c(PB_Dabney = 16, QG_Rohland = 17), labels = c("PB method",
    "QG method"))

```

Plot the results

```
ggarrange(plot1, plot2, labels = c("A", "B"), common.legend = TRUE,
  legend = "bottom", ncol = 2)
```

```
## Warning: Use of `` eucl_Lib_pcoa_df$`metadata2$LibraryMethod` `` is discouraged.
## i Use `metadata2$LibraryMethod` instead.

## Warning: Use of `` eucl_Lib_pcoa_df$`metadata2$ExtractionType` `` is discouraged.
## i Use `metadata2$ExtractionType` instead.

## Warning: Use of `` eucl_Lib_pcoa_df$`metadata2$LibraryMethod` `` is discouraged.
## i Use `metadata2$LibraryMethod` instead.

## Warning: Use of `` eucl_Lib_pcoa_df$`metadata2$ExtractionType` `` is discouraged.
## i Use `metadata2$ExtractionType` instead.
```

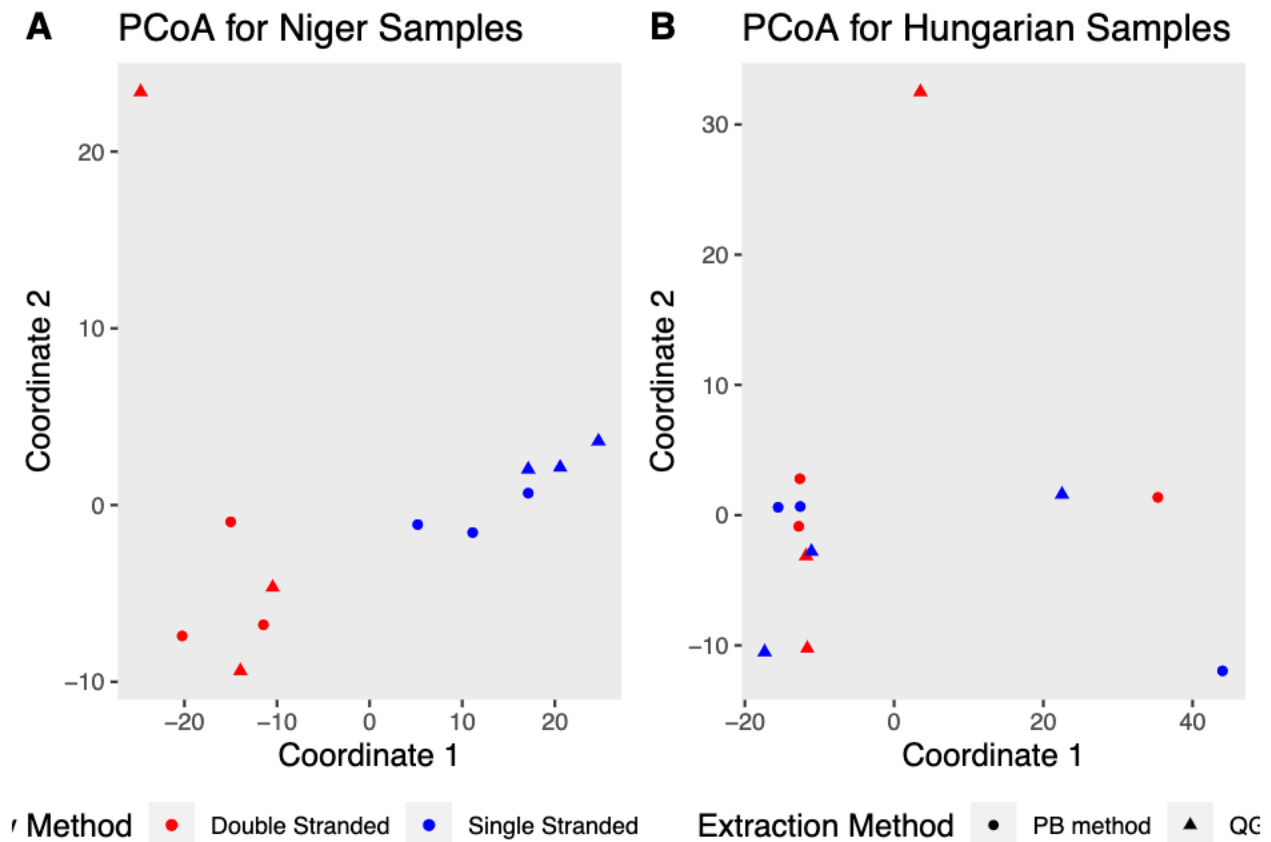


Figure 1: PCoA