# Package 'MetaPhyloTools'

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<b>Description</b> A collection of tools to process and analyse phylogeographic patterns from metabarcoding data, including functions for data cleaning, phylogenetic analysis, and visualization of results
License GPL-3
Imports dplyr, ggplot2, tidyr, phytools, ape, vegan, stringr, readr, purrr, igraph, scatterpie
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edenetwork\_percolation

Calculate Percolation Threshold for Ecological Networks

## Description

Calculates the percolation threshold for ecological networks by identifying the critical distance at which the network fragments into small components. This function implements the percolation analysis described in EDENetworks.

## Usage

edenetwork\_percolation(diss)

## **Arguments**

diss

A distance matrix representing dissimilarities between nodes

#### Details

Percolation threshold is defined as follows in the original reference: from

Arnaud-Haond et al. EDENetworks: Ecological and Evolutionary Networks.

Percolation threshold: When links are removed from a connected network, it eventually fragments into small components. The point where this happens is called the percolation threshold. More accurately, this is the point where the so-called giant component (whose size is of the order of the network size) disappears and there is no long-range connectivity; even before the percolation threshold small disconnected fragments will appear, yet a substantial fraction of nodes belongs to the giant component. The precise location of this percolation point is made using the definition classically proposed for finite systems (Stauffer and Aharony, 1994) by calculating the average proposed for finite systems (Stauffer and Aharony, 1994) by calculating the average size of the clusters excluding the largest one:

```
S = sums < Smax(s^2ns)
```

as a function of the last distance value removed, thr, and identifying the critical distance with the one at which <S>\* has a maximum. N is the total number of nodes not included in the largest cluster and ns is the number of clusters containing s nodes.

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#### Value

```
A list containing:
```

```
graph The original igraph objectS_list Vector of S values for each thresholdcritical_distance The critical distance at percolation threshold
```

#### References

Arnaud-Haond et al. EDENetworks: Ecological and Evolutionary Networks. https://www.researchgate.net/profile/Sophie-Arnaud-Haond/publication/268433406\_EDENetworks\_Ecological\_and\_ Evolutionary\_Networks/links/54f705a30cf28d6dec9c7ad8/EDENetworks-Ecological-and-Evolutionary-Networpdf

## **Examples**

```
# Create example distance matrix
dist_mat <- as.matrix(dist(iris[1:10, 1:4]))
result <- edenetwork_percolation(dist_mat)
print(result$critical_distance)</pre>
```

haplodata4ggplot

haplodata4ggplot

#### **Description**

This function creates a list containing nodes and connections for a haplotype network plot, suitable for use with ggplot2.

#### Usage

```
haplodata4ggplot(haploNet_data_object, method = "pegas")
```

## **Arguments**

haploNet\_data\_object

A list containing the haplotype network data named network, and a dataframe named pieseas. The latter contains the proportions of each sample or group of samples for each haplotype. This object can be generated using the function haploNet\_data.

@param method A string indicating the method to use for plotting the network. Options are "pegas" for the pegas package or "fruchtermanreingold" for the Fruchterman-Reingold algorithm.

haploNet\_data

#### **Details**

This function prepares the data for plotting haplotype networks, using either the pegas package or the Fruchterman-Reingold algorithm to determine node positions. The nodes are represented as pie charts, with sizes proportional to haplotype frequencies. The aim of this function is to facilitate the creation of publication-ready haplotype network plots using ggplot2, addressing common issues such as legend overlap with the network.

#### Value

Returns a list with two data frames: nodes and connections. The nodes data frame contains the x and y coordinates, size, labels, and proportions of each haplotype. The connections data frame contains the coordinates of the connections between nodes.

haploNet\_data

Create haplotype network data for visualization

## **Description**

This function processes MOTU (Molecular Operational Taxonomic Unit) data and metadata to create a haplotype network and associated pie chart data for phylogenetic analysis. It calculates mean abundances across samples grouped by a specified variable and generates network data suitable for haplotype network visualization.

#### Usage

```
haploNet_data(
  motu_tab,
  metadata,
  grouping_col,
  sample_col,
  seq_col,
  id_col,
  size_correction = 1
)
```

#### **Arguments**

motu_tab	A data.frame containing MOTU abundance data. Rows represent MOTUs/haplotypes, columns represent samples. Must include columns specified by id_col and seq_col.
metadata	A data.frame containing sample metadata with grouping information. Must include columns specified by grouping_col and sample_col.
grouping_col	Character string. Name of the column in metadata that contains the grouping variable (e.g., "treatment", "location").
sample_col	Character string. Name of the column in metadata that contains sample identifiers matching column names in motu_tab.

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seq_col	Character string. Name of the column in motu_tab that contains DNA sequences for haplotype analysis.
id_col	Character string. Name of the column in motu_tab that contains unique identifiers for each MOTU/haplotype.
size_correction	1
	Numeric Factor to correct harlotype frequencies for visualization. Default is 1

Numeric. Factor to correct haplotype frequencies for visualization. Default is 1 (no correction).

#### **Details**

The function performs the following steps:

- 1. Groups samples by the specified grouping variable
- 2. Calculates mean abundances for each MOTU across samples within each group
- 3. Filters out MOTUs with zero total abundance
- 4. Converts DNA sequences to DNAbin format for phylogenetic analysis
- 5. Creates haplotypes and constructs a haplotype network
- 6. Attaches frequency information to the network for visualization

#### Value

A list containing two elements:

network

A haplotype network object created by pegas::haploNet(), or NULL if only one haplotype is found. Contains network structure with frequency attributes.

Pieseas

A matrix with haplotypes as rows and groups as columns, containing mean abundances for pie chart visualization.

## Note

- Requires packages ape and pegas
- DNA sequences should be aligned and of equal length
- Sample identifiers in metadata must match column names in motu\_tab
- If only one haplotype is found, network will be NULL with a warning

```
## Not run:
# Example usage
result <- haploNet_data(
   motu_tab = my_motu_data,
   metadata = my_metadata,
   grouping_col = "treatment",
   sample_col = "sample_id",
   seq_col = "sequence",
   id_col = "motu_id",
   size_correction = 100
)</pre>
```

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```
# Access results
network <- result$network
pie_data <- result$pieseas
## End(Not run)</pre>
```

haploNet\_plot

Plot haplotype network with pie charts

## Description

This function creates a publication-ready haplotype network plot with pie charts showing the relative abundance or frequency of haplotypes across different groups. The plot includes a legend and title, and is saved as a PDF file.

#### Usage

```
haploNet_plot(haploNet_data_object, output_file, bg, plot_title)
```

## **Arguments**

haploNet\_data\_object

A list object returned by haploNet\_data() containing network and pie chart data. Must include elements:

- networkA haplotype network object from pegas::haploNet()
- pieseasA matrix with pie chart data for each haplotype

output\_file Character string. I

Character string. File path for the output PDF plot (e.g., "network\_plot.pdf").

bg

plot\_title

A vector of colors for the pie chart segments. Length should match the number of groups in the pie chart data. Used for both pie segments and legend.

Character string. Main title to be displayed at the top of the plot.

## **Details**

The function creates a multi-panel plot with:

- Main haplotype network with pie charts at each node
- Node sizes proportional to haplotype frequencies
- Title at the top of the plot
- Legend at the bottom showing group categories (currently hardcoded as "Inside"/"Outside")

## Plot specifications:

- PDF dimensions: 15 x 15 inches
- High-resolution output suitable for publication
- · Customizable colors and title
- Error handling for plot generation issues

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## Value

No return value. The function creates a PDF file at the specified location.

#### Note

- Requires the pegas package for network plotting
- Legend labels are currently hardcoded as "Inside" and "Outside"
- The function assumes a two-group comparison (modify legend for more groups)
- If plotting fails, an error message is displayed with details

## **Examples**

```
## Not run:
# Create network data first
network_data <- haploNet_data(</pre>
 motu_tab = my_motu_data,
  metadata = my_metadata,
  grouping_col = "location",
  sample_col = "sample_id",
  seq_col = "sequence",
  id_col = "motu_id"
)
# Create the plot
haploNet_plot(
  haploNet_data_object = network_data,
  output_file = "haplotype_network.pdf",
  bg = c("red", "blue"),
  plot_title = "Haplotype Network Analysis"
)
## End(Not run)
```

haplo\_ggplot

Create Haplotype Network Plots with ggplot2

## **Description**

Creates haplotype network visualizations using ggplot2 with customizable legend positioning and scatter pie charts for nodes.

## Usage

```
haplo_ggplot(data, legend_pos = "left_bottom")
```

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## **Arguments**

data A list containing 'nodes' and 'connections' data frames for the network. The

'nodes' data frame should contain x, y coordinates and size information. The

'connections' data frame should specify network edges.

legend\_pos Position for the legend. Options: "left\_bottom", "right\_bottom", "top\_left",

"top\_right", "left", "right", "top", "bottom", "none"

#### **Details**

This function creates publication-ready haplotype network plots with nodes represented as pie charts (using scatterpie) and customizable legend positioning to avoid overlap with the network. The aim of this function is to solve a problem often encountered when plotting haplotype networks with ggplot2, where the legend can overlap with the network itself.

#### Value

A ggplot object representing the haplotype network

## **Examples**

```
# Example with mock network data
nodes <- data.frame(
    x = runif(5),
    y = runif(5),
    size = runif(5, 1, 3),
    pop1 = runif(5),
    pop2 = runif(5)
)
connections <- data.frame(from = c(1,2,3), to = c(2,3,4))
data <- list(nodes = nodes, connections = connections)
plot <- haplo_ggplot(data)</pre>
```

pairwise\_djost

Calculate Pairwise Jost's D Differentiation

#### **Description**

Calculates pairwise Jost's D differentiation indices between samples, optionally grouped by sample groups and per MOTU (Molecular Operational Taxonomic Unit).

## Usage

```
pairwise_djost(
   df,
   sample_names,
   sample_groups = NULL,
   motu_col = "MOTU",
```

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```
rarefy = TRUE,
rarefy_to = min
)
```

#### **Arguments**

df Data frame containing MOTU abundance data

sample\_names Vector of column names representing samples

sample\_groups Optional vector of group assignments for samples. Must be the same length as sample\_names and in the same order. If NULL, each sample is treated as its own group

motu\_col Name of the column containing MOTU identifiers (default: "MOTU")

rarefy Logical, whether to rarefy data within MOTU (default: TRUE)

rarefy\_to Function to determine rarefaction depth (default: min)

#### **Details**

Jost's D is a measure of differentiation that is independent of within-population diversity. This function calculates it for each MOTU separately and can group samples for analysis. The function rarefies data within each MOTU to standardize sampling effort before calculating Jost's D this rarefaction uses the funcion vegan::rrarefy and the specified rarefaction depth defined by the user with the parameter rarefy\_to (default is the minimum sample sum across samples with non-zero counts within each MOTU).

#### Value

A list containing Jost's D matrices for each MOTU, and optionally grouped results if sample\_groups is provided

```
# Example with mock data
df <- data.frame(
   MOTU = rep(c("MOTU1", "MOTU2"), each = 5),
   sample1 = rpois(10, 5),
   sample2 = rpois(10, 3),
   sample3 = rpois(10, 4)
)
result <- pairwise_djost(df, c("sample1", "sample2", "sample3"))</pre>
```

print\_network

print_network	Create and save a haplotype network plot to PDF

## Description

This function is a convenient wrapper that combines haplotype network data generation and plotting into a single step. It processes MOTU data and metadata to create a haplotype network visualization and saves it directly to a PDF file.

## Usage

```
print_network(
  motu_tab,
  output_file = "haplotype_network.pdf",
  metadata,
  grouping_col,
  sample_col,
  seq_col = "sequence",
  id_col = "id",
  size_correction = 1,
  plot_title = "Haplotype Network",
  bg = NULL,
  ...
)
```

## **Arguments**

motu_tab	A data.frame containing MOTU abundance data. Rows represent MOTUs/haplotypes, columns represent samples. Must include columns specified by id_col and seq_col.
output_file	Character string. File path for the output PDF plot. Default is "haplotype_network.pdf".
metadata	A data frame containing sample metadata with grouping information. Must include columns specified by grouping_col and sample_col.
grouping_col	Character string. Name of the column in metadata that contains the grouping variable (e.g., "treatment", "location").
sample_col	Character string. Name of the column in metadata that contains sample identifiers matching column names in motu_tab.
seq_col	Character string. Name of the column in motu_tab that contains DNA sequences for haplotype analysis. Default is "sequence".
id_col	Character string. Name of the column in motu_tab that contains unique identifiers for each MOTU/haplotype. Default is "id".
size_correctio	n
	Numeric. Factor to correct haplotype frequencies for visualization. Default is 1 (no correction).

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plot_title	Character string. Main title to be displayed on the plot. Default is "Haplotype Network".
bg	A vector of colors for the pie chart segments. If NULL, default colors will be used.
	Additional arguments passed to underlying functions.

#### **Details**

This function is a streamlined workflow that:

- 1. Calls haploNet\_data() to process the input data and create network structure
- 2. Calls haploNet\_plot() to generate and save the plot to PDF
- 3. Displays a confirmation message with the output file path

The resulting PDF contains:

- Haplotype network with nodes representing haplotypes
- Pie charts at each node showing group composition
- Node sizes proportional to haplotype frequencies
- · Legend and title
- High-resolution output (15x15 inches) suitable for publication

#### Value

Invisible NULL. The function is called for its side effect of creating a PDF file.

## Note

- This is a convenience function that combines haploNet\_data() and haploNet\_plot()
- Requires packages ape and pegas
- For more control over the plotting process, use haploNet\_data() and haploNet\_plot() separately
- DNA sequences should be aligned and of equal length

## See Also

```
haploNet_data, haploNet_plot, print_network_ggplot
```

```
## Not run:
# Basic usage
print_network(
   motu_tab = my_motu_data,
   metadata = my_metadata,
   grouping_col = "treatment",
   sample_col = "sample_id",
   output_file = "treatment_network.pdf"
```

print\_network\_ggplot

```
# With custom parameters
print_network(
   motu_tab = my_motu_data,
   metadata = my_metadata,
   grouping_col = "location",
   sample_col = "sample_id",
   seq_col = "dna_sequence",
   id_col = "motu_id",
   size_correction = 1000,
   plot_title = "Geographic Distribution",
   bg = c("red", "blue", "green"),
   output_file = "geographic_network.pdf"
)
## End(Not run)
```

print\_network\_ggplot Create haplotype network visualization using ggplot2

## **Description**

This function creates a modern, customizable haplotype network plot using ggplot2. It processes MOTU data and metadata to generate a network visualization with nodes representing haplotypes and edges showing relationships between them.

## Usage

```
print_network_ggplot(
   motu_tab,
   output_file = "haplotype_network.pdf",
   metadata,
   grouping_col,
   sample_col,
   seq_col = "sequence",
   id_col = "id",
   size_correction = 1,
   plot_title = "Haplotype Network",
   bg = NULL,
   method = "pegas",
   legend_pos = "left_bottom",
   ...
)
```

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## **Arguments**

motu_tab	A data.frame containing MOTU abundance data. Rows represent MOTUs/haplotypes, columns represent samples. Must include columns specified by id_col and seq_col.
output_file	Character string. File path for the output PDF plot. Default is "haplotype_network.pdf". Currently not used in this function.
metadata	A data.frame containing sample metadata with grouping information. Must include columns specified by grouping_col and sample_col.
grouping_col	Character string. Name of the column in metadata that contains the grouping variable (e.g., "treatment", "location").
sample_col	Character string. Name of the column in metadata that contains sample identifiers matching column names in motu_tab.
seq_col	Character string. Name of the column in motu_tab that contains DNA sequences for haplotype analysis. Default is "sequence".
id_col	Character string. Name of the column in motu_tab that contains unique identifiers for each MOTU/haplotype. Default is "id".
size_correctio	n
	Numeric. Factor to correct haplotype frequencies for visualization. Default is 1 (no correction).
plot_title	Character string. Main title to be displayed on the plot. Default is "Haplotype Network".
bg	A vector of colors for the groups. If NULL, default colors will be used.
method	Character string. Layout method for the network. Must be either "pegas" (uses pegas package layout) or "fruchtermanreingold" (uses Fruchterman-Reingold layout). Default is "pegas".
legend_pos	Character string. Position of the legend. Default is "left_bottom".
	Additional arguments passed to underlying functions.

## Details

The function performs the following workflow:

- 1. Calls haploNet\_data() to process input data and create network structure
- $2. \ Uses \ haplodata4ggplot() \ to \ convert \ network \ data \ to \ ggplot-compatible \ format$
- 3. Creates the final plot using haplo\_ggplot()

The function supports two layout methods:

- "pegas": Uses the default layout from the pegas package
- "fruchtermanreingold": Uses the Fruchterman-Reingold force-directed layout

## Value

A ggplot object containing the haplotype network visualization.

## Note

- Requires packages ape, pegas, and ggplot2
- This function depends on helper functions: haploNet\_data(), haplodata4ggplot(), and haplo\_ggplot()
- The output\_file parameter is included for consistency but not currently used
- DNA sequences should be aligned and of equal length

#### See Also

haploNet\_data, haploNet\_plot

```
## Not run:
# Basic usage
plot <- print_network_ggplot(</pre>
 motu_tab = my_motu_data,
 metadata = my_metadata,
  grouping_col = "treatment",
  sample_col = "sample_id",
  plot_title = "Treatment Comparison"
)
# With custom parameters
plot <- print_network_ggplot(</pre>
  motu_tab = my_motu_data,
  metadata = my_metadata,
  grouping_col = "location",
  sample_col = "sample_id",
  seq_col = "dna_sequence",
  id_col = "motu_id",
  size_correction = 100,
  method = "fruchtermanreingold",
  bg = c("#FF6B6B", "#4ECDC4", "#45B7D1"),
  legend_pos = "right"
# Display the plot
print(plot)
# Save the plot
ggsave("my_network.pdf", plot, width = 12, height = 10)
## End(Not run)
```

rarefy\_within\_motu 15

rarefy\_within\_motu Rarefy Data Within MOTUs

**Description** 

Performs rarefaction of sample data within each MOTU (Molecular Operational Taxonomic Unit) to standardize sampling effort.

## Usage

```
rarefy_within_motu(
   df,
   sample_names,
   motu_col = "MOTU",
   rarefy = TRUE,
   rarefy_to = min,
   rel_abund = TRUE
)
```

## **Arguments**

df	Data frame containing MOTU abundance data
sample_names	Vector of column names representing samples
motu_col	Name of the column containing MOTU identifiers (default: "MOTU")
rarefy	Logical, whether to perform rarefaction (default: TRUE)
rarefy_to	Function or numeric value to determine rarefaction depth (default: min function)
rel_abund	Logical, whether to convert to relative abundance after rarefaction (default: TRUE)

## **Details**

This function rarefies data within each MOTU separately, which is important for metabarcoding data where different MOTUs may have very different abundance ranges.

#### Value

A data frame with rarefied abundance data

```
# Example with mock data
df <- data.frame(
   MOTU = rep(c("MOTU1", "MOTU2"), each = 3),
   sample1 = rpois(6, 10),
   sample2 = rpois(6, 15)
)
result <- rarefy_within_motu(df, c("sample1", "sample2"))</pre>
```

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