

# Package ‘microeco’

May 17, 2022

**Type** Package

**Title** Microbial Community Ecology Data Analysis

**Version** 0.9.0

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**Description** A series of statistical and plotting approaches in microbial community ecology based on the R6 class. The classes are designed for data preprocessing, taxa abundance plotting, alpha diversity analysis, beta diversity analysis, differential abundance test, null model analysis, network analysis, machine learning, environmental data analysis and functional analysis.

**URL** <https://github.com/ChiLiubio/microeco>

**Depends** R (>= 3.5.0)

**Imports** R6, stats, ape, vegan, rlang, data.table, magrittr, dplyr,  
tibble, scales, grid, ggplot2, RColorBrewer, reshape2

**Suggests** GUniFrac, MASS, ggpubr, randomForest, ggdendro, ggrepel,  
agricolae, gridExtra, picante, pheatmap, igraph, rgexf, mice

**License** GPL-3

**LazyData** true

**Encoding** UTF-8

**NeedsCompilation** no

**Repository** CRAN

**Date/Publication** 2022-05-17 09:10:16 UTC

**RoxygenNote** 7.1.2

**R topics documented:**

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---

clone

*Copy an R6 class object completely*


---

**Description**

Copy an R6 class object completely

**Usage**

```
clone(x, deep = TRUE)
```

**Arguments**

x                    R6 class object  
deep                default TRUE; deep copy

**Value**

identical but unrelated R6 object.

**Examples**

```
data("dataset")  
clone(dataset)
```

---

color\_palette\_20            *A color palette for 20 elements.*

---

**Description**

This is one palette option for users who have  $\leq 20$  elements to plot. The palletes of RColorBrewer package provide at most 12 discrete colors, such as "Set3" and "Paired". This palette is adapted from D3.js library and can be used as a supplement.

**Usage**

```
color_palette_20
```

**Format**

An object of class character of length 20.

---

dataset                    *The dataset in the microeco package*

---

**Description**

The dataset is structured with microtable class for the demonstration of examples and tutorials.

**Usage**

```
data(dataset)
```

**Format**

An R6 class object

**Details**

- `sample_table`: sample information table
- `otu_table`: species-community abundance table
- `tax_table`: taxonomic table
- `phylo_tree`: phylogenetic tree
- `taxa_abund`: taxa abundance list with several tables for Phylum...Genus
- `alpha_diversity`: alpha diversity table
- `beta_diversity`: list with several beta diversity distance matrix

---

<code>dropallfactors</code>	<i>Remove all factors in a data frame</i>
-----------------------------	---

---

**Description**

Remove all factors in a data frame

**Usage**

```
dropallfactors(x, unfac2num = FALSE, char2num = FALSE)
```

**Arguments**

<code>x</code>	data frame
<code>unfac2num</code>	default FALSE; whether try to convert all character to numeric; if FALSE, only try to convert column with factor attribute. Note that this can only transform the columns that may be transformed to numeric without using factor.
<code>char2num</code>	default FALSE; whether force all the character to be numeric class by using factor as an intermediate.

**Value**

data frame without factor

**Examples**

```
data("taxonomy_table_16S")
taxonomy_table_16S[, 1] <- as.factor(taxonomy_table_16S[, 1])
str(dropallfactors(taxonomy_table_16S))
```

---

env_data_16S	<i>The environmental factors for the 16S dataset in the microeco package</i>
--------------	--

---

**Description**

The environmental factors for the 16S dataset in the microeco package

**Usage**

```
data(env_data_16S)
```

---

fungi_func_FungalTraits	<i>The FungalTraits database for fungi trait identification in the microeco package</i>
-------------------------	---

---

**Description**

The FungalTraits database for fungi trait identification in the microeco package

**Usage**

```
data(fungi_func_FungalTraits)
```

---

fungi_func_FUNGuild	<i>The FUNGuild database for fungi trait identification in the microeco package</i>
---------------------	---

---

**Description**

The FUNGuild database for fungi trait identification in the microeco package

**Usage**

```
data(fungi_func_FUNGuild)
```

---

microtable

*Create microtable object to store and manage all the basic files.*

---

## Description

This class is a wrapper for a series of operations on the original files and the basic manipulations, including the microtable object creation, data reduction, data rarefaction based on Paul et al. (2013) <doi:10.1371/journal.pone.0061217>, taxa abundance calculation, alpha and beta diversity calculation based on the An et al. (2019) <doi:10.1016/j.geoderma.2018.09.035> and Lozupone et al. (2005) <doi:10.1128/AEM.71.12.8228–8235.2005> and other basic operations.

The tutorial website: [https://chiliubio.github.io/microeco\\_tutorial/](https://chiliubio.github.io/microeco_tutorial/) or [http://chiliubio.gitee.io/microeco\\_tutorial/](http://chiliubio.gitee.io/microeco_tutorial/)

## Format

microtable.

## Methods

### Public methods:

- `microtable$new()`
- `microtable$print()`
- `microtable$filter_pollution()`
- `microtable$rarefy_samples()`
- `microtable$tidy_dataset()`
- `microtable$add_rownames2taxonomy()`
- `microtable$cal_abund()`
- `microtable$save_abund()`
- `microtable$sample_sums()`
- `microtable$taxa_sums()`
- `microtable$sample_names()`
- `microtable$taxa_names()`
- `microtable$rename_taxa()`
- `microtable$merge_samples()`
- `microtable$merge_taxa()`
- `microtable$cal_alphadiv()`
- `microtable$save_alphadiv()`
- `microtable$cal_betadiv()`
- `microtable$save_betadiv()`
- `microtable$clone()`

### Method `new()`:

*Usage:*

```

microtable$new(
  otu_table,
  sample_table = NULL,
  tax_table = NULL,
  phylo_tree = NULL,
  rep_fasta = NULL,
  auto_tidy = FALSE
)

```

*Arguments:*

`otu_table` data.frame; necessary; The feature abundance table, rows are features, e.g. species, cols are samples.

`sample_table` data.frame; default NULL; The sample information table, rows are samples, cols are sample metadata; If not provided, the function can generate a table automatically according to the sample names in `otu_table`.

`tax_table` data.frame; default NULL; The taxonomic information table, rows are species, cols are taxonomic classes.

`phylo_tree` phylo; default NULL; The phylogenetic tree; use `read.tree` function in `ape` package for input.

`rep_fasta` list or `DNASTringSet`; default NULL; The representative sequences; use `read.fasta` function in `seqinr` package or `readDNASTringSet` function in `Biostrings` package for input.

`auto_tidy` default FALSE; Whether trim the files in dataset automatically. If TRUE, all other operations that

*Returns:* an object of class "microtable" with the following components:

`sample_table` The sample information table.

`otu_table` The OTU table.

`tax_table` The taxonomic table.

`phylo_tree` The phylogenetic tree.

`rep_fasta` The representative sequence.

`taxa_abund` default NULL; use `cal_abund` function to calculate.

`alpha_diversity` default NULL; use `cal_alphadiv` function to calculate.

`beta_diversity` default NULL; use `cal_betadiv` function to calculate.

*Examples:*

```

data(otu_table_16S)
data(taxonomy_table_16S)
data(sample_info_16S)
data(phylo_tree_16S)
dataset <- microtable$new(otu_table = otu_table_16S)
dataset <- microtable$new(sample_table = sample_info_16S, otu_table = otu_table_16S,
  tax_table = taxonomy_table_16S, phylo_tree = phylo_tree_16S)
# trim the files in the dataset
dataset$tidy_dataset()

```

**Method** `print()`: Print the microtable object.

*Usage:*

```
microtable$print()
```

**Method** `filter_pollution()`: Filter the taxa considered as pollution from `tax_table`. This operation will remove any line of the `tax_table` containing any the word in `taxa` parameter regardless of word case.

*Usage:*

```
microtable$filter_pollution(taxa = c("mitochondria", "chloroplast"))
```

*Arguments:*

`taxa` default: `c("mitochondria", "chloroplast")`; filter mitochondria and chloroplast, or others as needed.

*Returns:* None

*Examples:*

```
dataset$filter_pollution(taxa = c("mitochondria", "chloroplast"))
```

**Method** `rarefy_samples()`: Rarefy communities to make all samples have same species number. See also `rrarefy` for the alternative method.

*Usage:*

```
microtable$rarefy_samples(sample.size = NULL, rngseed = 123, replace = TRUE)
```

*Arguments:*

`sample.size` default: NULL; species number, If not provided, use minimum number of all samples.

`rngseed` random seed; default: 123.

`replace` default: TRUE; See [sample](#) for the random sampling.

*Returns:* None; rarefied dataset.

*Examples:*

```
\donttest{
dataset$rarefy_samples(sample.size = min(dataset$sample_sums()), replace = TRUE)
}
```

**Method** `tidy_dataset()`: Tidy the object of `microtable` Class. Trim files in the object to make `taxa` and `samples` consistent across all files in the object. So the results are intersections.

*Usage:*

```
microtable$tidy_dataset(main_data = FALSE)
```

*Arguments:*

`main_data` default FALSE; if TRUE, only basic files in `microtable` object is trimmed. Otherwise, all files, including `taxa_abund`, `alpha_diversity` and `beta_diversity`, are all trimmed.

*Returns:* None, Object of `microtable` itself cleaned up.

*Examples:*

```
dataset$tidy_dataset(main_data = TRUE)
```

**Method** `add_rownames2taxonomy()`: Add the rownames of `tax_table` as the last column of `tax_table`. This is especially useful when the rownames of `tax_table` are required as a taxonomic level for the following `taxa_abund` calculation and biomarker identification.

*Usage:*



```
microtable$add_rownames2taxonomy(use_name = "OTU")
```

*Arguments:*

`use_name` default "OTU"; The column name used in the `tax_table`.

*Returns:* new `tax_table` stored in object.

*Examples:*

```
\donttest{
dataset$add_rownames2taxonomy()
}
```

**Method** `cal_abund()`: Calculate the taxonomic abundance at each taxonomic rank.

*Usage:*

```
microtable$cal_abund(
  select_cols = NULL,
  rel = TRUE,
  split_group = FALSE,
  split_by = "&&",
  split_column = NULL
)
```

*Arguments:*

`select_cols` default NULL; numeric vector or character vector of colnames of `tax_table`; used to select columns to merge and calculate abundances. This is very useful if there are commented columns or some columns with multiple structure that cannot be used directly.

`rel` default TRUE; if TRUE, relative abundance is used; if FALSE, absolute abundance will be summed.

`split_group` default FALSE; if TRUE, split the rows to multiple rows according to one or more columns in `tax_table`. Very useful when multiple mapping info exist.

`split_by` default "&&"; Separator delimiting collapsed values; only useful when `split_group == TRUE`; see `sep` in `separate_rows` function.

`split_column` default NULL; character vector or list; only useful when `split_group == TRUE`; character vector: fixed column or columns used for the splitting in `tax_table` in each abundance calculation; list: containing more character vectors to assign the column names to each calculation, such as `list(c("Phylum"), c("Phylum", "Class"))`.

*Returns:* `taxa_abund` in object.

*Examples:*

```
\donttest{
dataset$cal_abund()
}
```

**Method** `save_abund()`: Save taxonomic abundance to the computer local place.

*Usage:*

```
microtable$save_abund(dirpath = "taxa_abund")
```

*Arguments:*

`dirpath` default "taxa\_abund"; directory name to save the taxonomic abundance files.

*Examples:*

```
\dontrun{
dataset$save_abund(dirpath = "taxa_abund")
}
```

**Method** `sample_sums()`: Sum the species number for each sample.

*Usage:*

```
microtable$sample_sums()
```

*Returns:* species number of samples.

*Examples:*

```
\donttest{
dataset$sample_sums()
}
```

**Method** `taxa_sums()`: Sum the species number for each taxa.

*Usage:*

```
microtable$taxa_sums()
```

*Returns:* species number of taxa.

*Examples:*

```
\donttest{
dataset$taxa_sums()
}
```

**Method** `sample_names()`: Show sample names.

*Usage:*

```
microtable$sample_names()
```

*Returns:* sample names.

*Examples:*

```
\donttest{
dataset$sample_names()
}
```

**Method** `taxa_names()`: Show taxa names of `tax_table`.

*Usage:*

```
microtable$taxa_names()
```

*Returns:* taxa names.

*Examples:*

```
\donttest{
dataset$taxa_names()
}
```

**Method** `rename_taxa()`: Rename the taxa, including the rownames of `otu_table`, rownames of `tax_table`, tip labels of phylogenetic tree and representative sequences.

*Usage:*

```
microtable$rename_taxa(newname_prefix = "ASV_")
```

*Arguments:*

`newname_prefix` default "ASV\_"; the prefix of new names; new names will be `newname_prefix` + numbers according to the `rowname` order of `otu_table`.

*Returns:* renamed dataset.

*Examples:*

```
\donttest{  
dataset$rename_taxa()  
}
```

**Method** `merge_samples()`: Merge samples according to specific group to generate a new microtable.

*Usage:*

```
microtable$merge_samples(use_group)
```

*Arguments:*

`use_group` the group column in `sample_table`.

*Returns:* a new merged microtable object.

*Examples:*

```
\donttest{  
dataset$merge_samples(use_group = "Group")  
}
```

**Method** `merge_taxa()`: Merge taxa according to specific taxonomic rank to generate a new microtable.

*Usage:*

```
microtable$merge_taxa(taxa = "Genus")
```

*Arguments:*

`taxa` the specific rank in `tax_table`.

*Returns:* a new merged microtable object.

*Examples:*

```
\donttest{  
dataset$merge_taxa(taxa = "Genus")  
}
```

**Method** `cal_alphadiv()`: Calculate alpha diversity in microtable object.

*Usage:*

```
microtable$cal_alphadiv(measures = NULL, PD = FALSE)
```

*Arguments:*

`measures` default NULL; one or more indexes from "Observed", "Coverage", "Chao1", "ACE", "Shannon", "Simpson", "InvSimpson", "Fisher", "PD"; If null, use all those measures.

`PD` TRUE or FALSE, whether phylogenetic tree should be calculated, default FALSE.

*Returns:* alpha\_diversity stored in object.

*Examples:*

```
\donttest{
dataset$cal_alphadiv(measures = NULL, PD = FALSE)
class(dataset$alpha_diversity)
}
```

**Method** save\_alphadiv(): Save alpha diversity table to the computer.

*Usage:*

```
microtable$save_alphadiv(dirpath = "alpha_diversity")
```

*Arguments:*

dirpath default "alpha\_diversity"; directory name to save the alpha\_diversity.csv file.

**Method** cal\_betadiv(): Calculate beta diversity in microtable object, including Bray-Curtis, Jaccard, and UniFrac. See An et al. (2019) <doi:10.1016/j.geoderma.2018.09.035> and Lozupone et al. (2005) <doi:10.1128/AEM.71.12.8228–8235.2005>.

*Usage:*

```
microtable$cal_betadiv(method = NULL, unifrac = FALSE, binary = FALSE, ...)
```

*Arguments:*

method default NULL; a character vector with one or more elements; If default, "bray" and "jaccard" will be used; see [vegdist](#) function and method parameter in vegan package.

unifrac default FALSE; TRUE or FALSE, whether unifrac index should be calculated.

binary default FALSE; TRUE is used for jaccard and unweighted unifrac; optional for other indexes.

... parameters passed to [vegdist](#) function.

*Returns:* beta\_diversity stored in object.

*Examples:*

```
\donttest{
dataset$cal_betadiv(unifrac = FALSE)
class(dataset$beta_diversity)
}
```

**Method** save\_betadiv(): Save beta diversity matrix to the computer.

*Usage:*

```
microtable$save_betadiv(dirpath = "beta_diversity")
```

*Arguments:*

dirpath default "beta\_diversity"; directory name to save the beta diversity matrix files.

**Method** clone(): The objects of this class are cloneable with this method.

*Usage:*

```
microtable$clone(deep = FALSE)
```

*Arguments:*

deep Whether to make a deep clone.

**Examples**

```
## -----  
## Method `microtable$new`  
## -----  
  
data(otu_table_16S)  
data(taxonomy_table_16S)  
data(sample_info_16S)  
data(phylo_tree_16S)  
dataset <- microtable$new(otu_table = otu_table_16S)  
dataset <- microtable$new(sample_table = sample_info_16S, otu_table = otu_table_16S,  
  tax_table = taxonomy_table_16S, phylo_tree = phylo_tree_16S)  
# trim the files in the dataset  
dataset$tidy_dataset()  
  
## -----  
## Method `microtable$filter_pollution`  
## -----  
  
dataset$filter_pollution(taxa = c("mitochondria", "chloroplast"))  
  
## -----  
## Method `microtable$rarefy_samples`  
## -----  
  
dataset$rarefy_samples(sample.size = min(dataset$sample_sums()), replace = TRUE)  
  
## -----  
## Method `microtable$tidy_dataset`  
## -----  
  
dataset$tidy_dataset(main_data = TRUE)  
  
## -----  
## Method `microtable$add_rownames2taxonomy`  
## -----  
  
dataset$add_rownames2taxonomy()  
  
## -----  
## Method `microtable$scal_abund`  
## -----  
  
dataset$scal_abund()
```

```
## -----  
## Method `microtable$save_abund`  
## -----  
  
## Not run:  
dataset$save_abund(dirpath = "taxa_abund")  
  
## End(Not run)  
  
## -----  
## Method `microtable$sample_sums`  
## -----  
  
dataset$sample_sums()  
  
## -----  
## Method `microtable$taxa_sums`  
## -----  
  
dataset$taxa_sums()  
  
## -----  
## Method `microtable$sample_names`  
## -----  
  
dataset$sample_names()  
  
## -----  
## Method `microtable$taxa_names`  
## -----  
  
dataset$taxa_names()  
  
## -----  
## Method `microtable$rename_taxa`  
## -----  
  
dataset$rename_taxa()  
  
## -----  
## Method `microtable$merge_samples`  
## -----
```

```

dataset$merge_samples(use_group = "Group")

## -----
## Method `microtable$merge_taxa`
## -----

dataset$merge_taxa(taxa = "Genus")

## -----
## Method `microtable$cal_alphadiv`
## -----

dataset$cal_alphadiv(measures = NULL, PD = FALSE)
class(dataset$alpha_diversity)

## -----
## Method `microtable$cal_betadiv`
## -----

dataset$cal_betadiv(unifrac = FALSE)
class(dataset$beta_diversity)

```

---

otu\_table\_16S

*The OTU table of the 16S dataset in the microeco package*


---

### Description

The OTU table of the 16S dataset in the microeco package

### Usage

```
data(otu_table_16S)
```

---

otu\_table\_ITS

*The OTU table of the ITS dataset in the microeco package*


---

### Description

The OTU table of the ITS dataset in the microeco package

**Usage**

```
data(otu_table_ITS)
```

---

phylo_tree_16S	<i>The phylogenetic tree of 16S dataset in the microeco package</i>
----------------	---

---

**Description**

The phylogenetic tree of 16S dataset in the microeco package

**Usage**

```
data(phylo_tree_16S)
```

---

prok_func_FAPROTAX	<i>The modified FAPROTAX trait database in the microeco package</i>
--------------------	---

---

**Description**

The modified FAPROTAX trait database in the microeco package

**Usage**

```
data(prok_func_FAPROTAX)
```

---

prok_func_NJC19_list	<i>The modified NJC19 database in the microeco package</i>
----------------------	--

---

**Description**

The modified NJC19 database in the microeco package

**Usage**

```
data(prok_func_NJC19_list)
```



---

rep_fasta_16S	<i>The fasta file of 16S dataset used in tax4fun2 method.</i>
---------------	---

---

**Description**

See the document of microtable class for more details. This file is with read.fasta function in seqinr package.

**Usage**

```
data(rep_fasta_16S)
```

---

sample_info_16S	<i>The sample information of 16S dataset in the microeco package</i>
-----------------	--

---

**Description**

The sample information of 16S dataset in the microeco package

**Usage**

```
data(sample_info_16S)
```

---

sample_info_ITS	<i>The sample information of ITS dataset in the microeco package</i>
-----------------	--

---

**Description**

The sample information of ITS dataset in the microeco package

**Usage**

```
data(sample_info_ITS)
```

---

Tax4Fun2_KEGG	<i>The KEGG data files used in the cal_tax4fun2 function of trans_func class.</i>
---------------	---

---

**Description**

The KEGG data files used in the cal\_tax4fun2 function of trans\_func class.

**Usage**

```
data(Tax4Fun2_KEGG)
```

---

taxonomy_table_16S	<i>The taxonomic information of 16S dataset in the microeco package</i>
--------------------	---

---

**Description**

The taxonomic information of 16S dataset in the microeco package

**Usage**

```
data(taxonomy_table_16S)
```

---

taxonomy_table_ITS	<i>The taxonomic information of ITS dataset in the microeco package</i>
--------------------	---

---

**Description**

The taxonomic information of ITS dataset in the microeco package

**Usage**

```
data(taxonomy_table_ITS)
```

---

tidy_taxonomy	<i>Clean up the taxonomic table to make taxonomic assignments consistent.</i>
---------------	---

---

**Description**

Clean up the taxonomic table to make taxonomic assignments consistent.

**Usage**

```
tidy_taxonomy(  
  taxonomy_table,  
  column = "all",  
  pattern = c(".*uncultur.*", ".*unknown.*", ".*unidentif.*", ".*unclassified.*",  
    ".*No blast hit.*", ".*sp\\.$", ".*metagenome.*", ".*cultivar.*", ".*archaeon$",  
    "__synthetic.*", ".*\\sbacterium$", ".*bacterium\\s.*"),  
  replacement = "",  
  ignore.case = TRUE,  
  na_fill = ""  
)
```

**Arguments**

taxonomy_table	a data.frame with taxonomic information.
column	default "all"; "all" or a number; 'all' represents cleaning up all the columns; a number represents cleaning up this column.
pattern	default see the function parameter; the characters (regular expression) to be cleaned up or replaced; cleaned up when parameter replacement = "", replaced when parameter replacement has something; Note that the capital and small letters are not distinguished.
replacement	default ""; the characters used to replace the character in pattern parameter.
ignore.case	default TRUE; if FALSE, the pattern matching is case sensitive and if TRUE, case is ignored during matching.
na_fill	default ""; used to replace the NA.

**Format**

`data.frame` object.

**Value**

taxonomic table.

**Examples**

```
data("taxonomy_table_16S")
tidy_taxonomy(taxonomy_table_16S)
```

---

trans_abund	<i>Create trans_abund object to transform taxonomic abundance for plotting.</i>
-------------	---

---

**Description**

This class is a wrapper for the taxonomic abundance transformations and plotting. The transformed data style is the long-format for ggplot2 plotting. The plotting methods include bar plot, boxplot, heatmap and pie chart.

**Methods****Public methods:**

- `trans_abund$new()`
- `trans_abund$plot_bar()`
- `trans_abund$plot_heatmap()`
- `trans_abund$plot_box()`
- `trans_abund$plot_line()`
- `trans_abund$plot_pie()`

- `trans_abund$print()`
- `trans_abund$clone()`

**Method new():**

*Usage:*

```
trans_abund$new(
  dataset = NULL,
  taxrank = "Phylum",
  show = 0,
  ntaxa = 10,
  groupmean = NULL,
  delete_full_prefix = TRUE,
  delete_part_prefix = FALSE,
  prefix = NULL,
  use_percentage = TRUE,
  input_taxaname = NULL
)
```

*Arguments:*

`dataset` default NULL; the object of `microtable` class.

`taxrank` default "Phylum"; taxonomic rank.

`show` default 0; the relative abundance threshold used for filtering.

`ntaxa` default 10; how many taxa will be used, ordered by abundance from high to low; this parameter does not conflict with the parameter `show`; both can be used.

`groupmean` default NULL; calculating mean abundance for each group; select a group column name in `sample_table`.

`delete_full_prefix` default TRUE; whether delete both the prefix of taxonomy and the character in front of them.

`delete_part_prefix` default FALSE; whether only delete the prefix of taxonomy.

`prefix` default NULL; character string; can be used when `delete_full_prefix = T` or `delete_part_prefix = T`; default NULL represents using the "letter+\_\_", e.g. "k\_\_" for Phylum level; Please alter this parameter when the prefix is not standard.

`use_percentage` default TRUE; show the abundance percentage.

`input_taxaname` default NULL; character vector; if some taxa are selected, input taxa names.

*Returns:* `data_abund` stored in the object for plotting.

*Examples:*

```
\donttest{
data(dataset)
t1 <- trans_abund$new(dataset = dataset, taxrank = "Phylum", ntaxa = 10)
}
```

**Method plot\_bar():** Bar plot.

*Usage:*

```
trans_abund$plot_bar(
  color_values = RColorBrewer::brewer.pal(12, "Paired"),
  bar_type = "full",
```

```

others_color = "grey90",
facet = NULL,
facet2 = NULL,
order_facet = NULL,
x_axis_name = NULL,
order_x = NULL,
barwidth = NULL,
use_alluvium = FALSE,
clustering = FALSE,
facet_color = "grey95",
strip_text = 11,
legend_text_italic = FALSE,
xtext_type_hor = TRUE,
xtext_size = 10,
xtext_keep = TRUE,
xtitle_keep = TRUE,
ytitle_size = 17,
ylab_title = NULL
)

```

*Arguments:*

`color_values` default `RColorBrewer::brewer.pal(12, "Paired")`; colors palette for the plotting.  
`bar_type` default "full"; "full" or "notfull"; if full, the total abundance sum to 1 or 100 percent-age.

`others_color` default "grey90"; the color for "others" taxa.

`facet` default NULL; if using facet, providing a group column name of `sample_table`, such as, "Group".

`facet2` default NULL; the second facet, used with `facet` parameter together; `facet2` should have a finer scale; use this parameter, please first install package `ggh4x` using `install.packages("ggh4x")`

`order_facet` NULL; vector; used to order the facet, such as, `c("Group1", "Group3", "Group2")`.

`x_axis_name` NULL; a character string; a column name of `sample_table` used to show the sample names in x axis.

`order_x` default NULL; vector; used to order the sample names in x axis; must be the samples vector, such as, `c("S1", "S3", "S2")`.

`barwidth` default NULL; bar width, see width in [geom\\_bar](#).

`use_alluvium` default FALSE; whether add alluvium plot

`clustering` default FALSE; whether order samples by the clustering

`facet_color` default "grey95"; facet background color.

`strip_text` default 11; facet text size.

`legend_text_italic` default FALSE; whether use italic in legend.

`xtext_type_hor` default TRUE; x axis text horizontal, if FALSE; text slant.

`xtext_size` default 10; x axis text size.

`xtext_keep` default TRUE; whether retain x text.

`xtitle_keep` default TRUE; whether retain x title.

`ytitle_size` default 17; y axis title size.

`ylab_title` default NULL; y axis title.

*Returns:* ggplot2 plot.

*Examples:*

```
\donttest{
t1$plot_bar(facet = "Group", xtext_keep = FALSE)
}
```

**Method** plot\_heatmap(): Plot the heatmap.

*Usage:*

```
trans_abund$plot_heatmap(
  color_values = rev(RColorBrewer::brewer.pal(n = 11, name = "RdYlBu")),
  facet = NULL,
  order_facet = NULL,
  x_axis_name = NULL,
  order_x = NULL,
  withmargin = TRUE,
  plot_numbers = FALSE,
  plot_text_size = 4,
  plot_breaks = NULL,
  margincolor = "white",
  plot_colorscale = "log10",
  min_abundance = 0.01,
  max_abundance = NULL,
  strip_text = 11,
  xtext_size = 10,
  ytext_size = 11,
  xtext_keep = TRUE,
  xtitle_keep = TRUE,
  grid_clean = TRUE,
  xtext_type_hor = TRUE,
  pheatmap = FALSE,
  ...
)
```

*Arguments:*

`color_values` default `rev(RColorBrewer::brewer.pal(n = 11, name = "RdYlBu"))`; colors palette for the plotting.

`facet` default `NULL`; a character string; if using facet, provide a column name in `sample_table`, such as "Group".

`order_facet` `NULL`; vector; used to order the facet, such as, `c("Group1", "Group3", "Group2")`.

`x_axis_name` `NULL`; a character string; a column name of `sample_table` used to show the sample names in x axis.

`order_x` default `NULL`; vector; used to order the sample names in x axis; must be the samples vector, such as, `c("S1", "S3", "S2")`.

`withmargin` default `TRUE`; whether retain the tile margin.

`plot_numbers` default `FALSE`; whether plot the number in heatmap.

`plot_text_size` default 4; If `plot_numbers` `TRUE`, text size in plot.

`plot_breaks` default `NULL`; The legend breaks.

margincolor default "white"; If withmargin TRUE, use this as the margin color.  
 plot\_colorscale default "log10"; color scale.  
 min\_abundance default .01; the minimum abundance percentage in plot.  
 max\_abundance default NULL; the maximum abundance percentage in plot, NULL represent the max percentage.  
 strip\_text default 11; facet text size.  
 xtext\_size default 10; x axis text size.  
 ytext\_size default 11; y axis text size.  
 xtext\_keep default TRUE; whether retain x text.  
 xtitle\_keep default TRUE; whether retain x title.  
 grid\_clean default TRUE; whether remove grid lines.  
 xtext\_type\_hor default TRUE; x axis text horizontal, if FALSE; text slant.  
 pheatmap default FALSE; whether use pheatmap package to plot the heatmap.  
 ... parameters pass to pheatmap when pheatmap = TRUE.

*Returns:* ggplot2 plot or grid plot based on pheatmap.

*Examples:*

```
\donttest{
t1 <- trans_abund$new(dataset = dataset, taxrank = "Genus", ntaxa = 40)
t1$plot_heatmap(facet = "Group", xtext_keep = FALSE, withmargin = FALSE)
}
```

**Method** plot\_box(): Box plot.

*Usage:*

```
trans_abund$plot_box(
  color_values = RColorBrewer::brewer.pal(8, "Dark2"),
  group = NULL,
  show_point = FALSE,
  point_color = "black",
  point_size = 3,
  point_alpha = 0.3,
  plot_flip = FALSE,
  boxfill = TRUE,
  middlecolor = "grey95",
  middlesize = 1,
  xtext_type_hor = FALSE,
  xtext_size = 10,
  xtext_keep = TRUE,
  xtitle_keep = TRUE,
  ytitle_size = 17,
  ...
)
```

*Arguments:*

color\_values default RColorBrewer::brewer.pal(8, "Dark2"); colors palette for the plotting.  
 group default NULL; a column name of sample table to show abundance across groups.  
 show\_point default FALSE; whether show points in plot.

point\_color default "black"; If show\_point TRUE; use the color  
 point\_size default 3; If show\_point TRUE; use the size  
 point\_alpha default .3; If show\_point TRUE; use the transparency.  
 plot\_flip default FALSE; Whether rotate plot.  
 boxfill default TRUE; Whether fill the box with colors.  
 middlecolor default "grey95"; The middle line color.  
 middlesize default 1; The middle line size.  
 xtext\_type\_hor default TRUE; x axis text horizontal, if FALSE; text slant.  
 xtext\_size default 10; x axis text size.  
 xtext\_keep default TRUE; whether retain x text.  
 xtitle\_keep default TRUE; whether retain x title.  
 ytitle\_size default 17; y axis title size.  
 ... parameters pass to [geom\\_boxplot](#).

*Returns:* ggplot2 plot.

*Examples:*

```

\donttest{
t1$plot_box(group = "Group")
}

```

**Method** plot\_line(): Plot the line chart.

*Usage:*

```

trans_abund$plot_line(
  color_values = RColorBrewer::brewer.pal(8, "Dark2"),
  plot_SE = TRUE,
  position = position_dodge(0.1),
  errorbar_size = 1,
  point_size = 3,
  point_alpha = 0.8,
  line_size = 0.8,
  line_alpha = 0.8,
  line_type = 1,
  xtext_type_hor = FALSE,
  xtext_size = 10,
  ytitle_size = 17
)

```

*Arguments:*

color\_values default RColorBrewer::brewer.pal(8, "Dark2"); colors palette for the plotting.  
 plot\_SE default TRUE; TRUE: plot the errorbar with mean±se; FALSE: plot the errorbar with mean±sd.  
 position default position\_dodge(0.1); Position adjustment, either as a string (such as "identity"), or the result of a call to a position adjustment function.  
 errorbar\_size default 1; errorbar size.  
 point\_size default 3; point size for taxa.  
 point\_alpha default 0.8; point transparency.



line\_size default 0.8; line size.  
 line\_alpha default 0.8; line transparency.  
 line\_type default 1; an integer; line type.  
 xtext\_type\_hor default TRUE; x axis text horizontal, if FALSE; text slant.  
 xtext\_size default 10; x axis text size.  
 ytitle\_size default 17; y axis title size.

*Returns:* ggplot2 plot.

*Examples:*

```
\donttest{
t1 <- trans_abund$new(dataset = dataset, taxrank = "Genus", ntaxa = 5)
t1$plot_line(point_size = 3)
t1 <- trans_abund$new(dataset = dataset, taxrank = "Genus", ntaxa = 5, groupmean = "Group")
t1$plot_line(point_size = 5, errorbar_size = 1, xtext_type_hor = TRUE)
}
```

**Method** plot\_pie(): Plot pie chart.

*Usage:*

```
trans_abund$plot_pie(
  color_values = RColorBrewer::brewer.pal(8, "Dark2"),
  facet_nrow = 1,
  strip_text = 11,
  legend_text_italic = FALSE
)
```

*Arguments:*

color\_values default RColorBrewer::brewer.pal(8, "Dark2"); colors palette for the plotting.  
 facet\_nrow default 1; how many rows in the plot.  
 strip\_text default 11; sample title size.  
 legend\_text\_italic default FALSE; whether use italic in legend.

*Returns:* ggplot2 plot.

*Examples:*

```
\donttest{
t1 <- trans_abund$new(dataset = dataset, taxrank = "Phylum", ntaxa = 6, groupmean = "Group")
t1$plot_pie(facet_nrow = 1)
}
```

**Method** print(): Print the trans\_abund object.

*Usage:*

```
trans_abund$print()
```

**Method** clone(): The objects of this class are cloneable with this method.

*Usage:*

```
trans_abund$clone(deep = FALSE)
```

*Arguments:*

deep Whether to make a deep clone.

**Examples**

```

## -----
## Method `trans_abund$new`
## -----

data(dataset)
t1 <- trans_abund$new(dataset = dataset, taxrank = "Phylum", ntaxa = 10)

## -----
## Method `trans_abund$plot_bar`
## -----

t1$plot_bar(facet = "Group", xtext_keep = FALSE)

## -----
## Method `trans_abund$plot_heatmap`
## -----

t1 <- trans_abund$new(dataset = dataset, taxrank = "Genus", ntaxa = 40)
t1$plot_heatmap(facet = "Group", xtext_keep = FALSE, withmargin = FALSE)

## -----
## Method `trans_abund$plot_box`
## -----

t1$plot_box(group = "Group")

## -----
## Method `trans_abund$plot_line`
## -----

t1 <- trans_abund$new(dataset = dataset, taxrank = "Genus", ntaxa = 5)
t1$plot_line(point_size = 3)
t1 <- trans_abund$new(dataset = dataset, taxrank = "Genus", ntaxa = 5, groupmean = "Group")
t1$plot_line(point_size = 5, errorbar_size = 1, xtext_type_hor = TRUE)

## -----
## Method `trans_abund$plot_pie`
## -----

```

```
t1 <- trans_abund$new(dataset = dataset, taxrank = "Phylum", ntaxa = 6, groupmean = "Group")
t1$plot_pie(facet_nrow = 1)
```

---

trans\_alpha

Create trans\_alpha object for alpha diversity statistics and plotting.

---

## Description

This class is a wrapper for a series of alpha diversity related analysis, including the statistics and plotting based on An et al. (2019) <doi:10.1016/j.geoderma.2018.09.035> and Paul et al. (2013) <doi:10.1371/journal.pone.0061217>.

## Methods

### Public methods:

- `trans_alpha$new()`
- `trans_alpha$cal_diff()`
- `trans_alpha$plot_alpha()`
- `trans_alpha$print()`
- `trans_alpha$clone()`

### Method new():

*Usage:*

```
trans_alpha$new(dataset = NULL, group = NULL, order_x = NULL)
```

*Arguments:*

dataset the object of `microtable` Class.

group default NULL; the sample column used for the statistics; If provided, can return data\_stat.

order\_x default NULL; sample\_table column name or a vector containing sample names; if provided, order samples by using factor.

*Returns:* data\_alpha and data\_stat stored in the object.

*Examples:*

```
\donttest{
data(dataset)
t1 <- trans_alpha$new(dataset = dataset, group = "Group")
}
```

### Method cal\_diff(): Test the difference of alpha diversity across groups.

*Usage:*

```
trans_alpha$cal_diff(
  method = c("KW", "KW_dunn", "wilcox", "t.test", "anova")[1],
  measure = NULL,
  p_adjust_method = "fdr",
  anova_set = NULL,
  ...
)
```

*Arguments:*

method default "KW"; see the following available options:

'KW' KW: Kruskal-Wallis Rank Sum Test for all groups ( $\geq 2$ )

'KW\_dunn' Dunn's Kruskal-Wallis Multiple Comparisons, see dunnTest function in FSA package

'wilcox' Wilcoxon Rank Sum and Signed Rank Tests for all paired groups

't.test' Student's t-Test for all paired groups

'anova' Duncan's multiple range test for anova

measure default NULL; a vector; if null, all indexes will be calculated; see names of microtable\$alpha\_diversity, e.g. Observed, Chao1, ACE, Shannon, Simpson, InvSimpson, Fisher, Coverage, PD.

p\_adjust\_method default "fdr"; p.adjust method; see method parameter of p.adjust function for available options; NULL can disuse the p value adjustment.

anova\_set default NULL; specified group set for anova, such as 'block + N\*P\*K', see aov.

... parameters passed to kruskal.test or wilcox.test function (method = "KW") or dunnTest function of FSA package (method = "KW\_dunn") or agricolae::duncan.test (method = "anova").

*Returns:* res\_diff in object. A data.frame generally. A list for anova when anova\_set is assigned. In the data frame, 'Group' column means that the group has the maximum median or mean value across the test groups; For non-parametric methods, maximum median value; For t.test, maximum mean value.

*Examples:*

```
\donttest{
t1$cal_diff(method = "KW")
t1$cal_diff(method = "KW_dunn")
t1$cal_diff(method = "anova")
}
```

**Method** plot\_alpha(): Plotting the alpha diversity.

*Usage:*

```
trans_alpha$plot_alpha(
  use_diff = TRUE,
  color_values = RColorBrewer::brewer.pal(8, "Dark2"),
  measure = "Shannon",
  group = NULL,
  add_label = "Significance",
  use_boxplot = TRUE,
  boxplot_color = TRUE,
  boxplot_add = "jitter",
  order_x_mean = FALSE,
  y_start = 1.01,
  y_increase = 0.05,
  xtext_angle = NULL,
  xtext_size = 15,
  ytitle_size = 17,
  ...
)
```

*Arguments:*

use\_diff default TRUE; wheter add significance label using the result of cal\_diff function, i.e. object\$res\_diff; This is manily designed to add post hoc test of anova or Dunn's Kruskal-Wallis Multiple Comparisons to make the label adding easy.  
 color\_values default RColorBrewer::brewer.pal(8, "Dark2"); color pallete for groups.  
 measure default Shannon; alpha diveristy measurement; see names of alpha\_diversity of dataset, e.g., Observed, Chao1, ACE, Shannon, Simpson, InvSimpson, Fisher, Coverage, PD.  
 group default NULL; group name used for the plot.  
 add\_label default "Significance"; select a colname of object\$res\_diff for the label text, such as 'P.adj' or 'Significance'.  
 use\_boxplot default TRUE; TRUE: boxplot; FALSE: mean-se plot.  
 boxplot\_color default TRUE; TRUE: use color\_values, FALSE: use "black".  
 boxplot\_add default "jitter"; points type, see the add parameter in ggpubr::ggboxplot.  
 order\_x\_mean default FALSE; whether order x axis by the means of groups from large to small.  
 y\_start default 1.01; the y axis value from which to add the label; the default 1.01 means  $1.01 * \max(\text{values})$ .  
 y\_increase default 0.05; the increasing y axia space to add label; the default 0.05 means  $0.05 * y\_start$ ; this parameter is also used to label the letters of anova result with the fixed  $(1 + y\_increase) * y\_start$  space.  
 xtext\_angle default NULL; number (e.g. 30) used to make x axis text generate angle.  
 xtext\_size default 15; x axis text size.  
 ytitle\_size default 17; y axis title size.  
 ... parameters pass to ggpubr::ggboxplot function.

*Returns:* ggplot.

*Examples:*

```

\donttest{
t1$plot_alpha(measure = "Shannon", group = "Group")
}

```

**Method** print(): Print the trans\_alpha object.

*Usage:*

```
trans_alpha$print()
```

**Method** clone(): The objects of this class are cloneable with this method.

*Usage:*

```
trans_alpha$clone(deep = FALSE)
```

*Arguments:*

deep Whether to make a deep clone.

**Examples**

```
## -----
```

```

## Method `trans_alpha$new`
## -----

data(dataset)
t1 <- trans_alpha$new(dataset = dataset, group = "Group")

## -----
## Method `trans_alpha$cal_diff`
## -----

t1$cal_diff(method = "KW")
t1$cal_diff(method = "KW_dunn")
t1$cal_diff(method = "anova")

## -----
## Method `trans_alpha$plot_alpha`
## -----

t1$plot_alpha(measure = "Shannon", group = "Group")

```

---

trans\_beta

---

*Create trans\_beta object for the analysis of distance matrix of beta-diversity.*


---

## Description

This class is a wrapper for a series of beta-diversity related analysis, including several ordination calculations and plotting based on An et al. (2019) <doi:10.1016/j.geoderma.2018.09.035>, group distance comparison, clustering, perMANOVA based on Anderson al. (2008) <doi:10.1111/j.1442-9993.2001.01070.pp.x> and PERMDISP. Please also cite the original paper: An et al. (2019). Soil bacterial community structure in Chinese wetlands. *Geoderma*, 337, 290-299.

## Methods

### Public methods:

- `trans_beta$new()`
- `trans_beta$cal_ordination()`
- `trans_beta$plot_ordination()`
- `trans_beta$cal_manova()`
- `trans_beta$cal_betadisper()`
- `trans_beta$cal_group_distance()`
- `trans_beta$plot_group_distance()`

- `trans_beta$plot_clustering()`
- `trans_beta$clone()`

**Method** `new()`:

*Usage:*

```
trans_beta$new(dataset = NULL, measure = NULL, group = NULL)
```

*Arguments:*

`dataset` the object of `microtable` Class.

`measure` default NULL; bray, jaccard, wei\_unifrac or unwei\_unifrac, or other name of matrix you add; beta diversity index used for ordination, manova or group distance.

`group` default NULL; sample group used for manova, betadisper or group distance.

*Returns:* parameters stored in the object.

*Examples:*

```
data(dataset)
```

```
t1 <- trans_beta$new(dataset = dataset, measure = "bray", group = "Group")
```

**Method** `cal_ordination()`: Ordination based on An et al. (2019) <doi:10.1016/j.geoderma.2018.09.035>.

*Usage:*

```
trans_beta$cal_ordination(
  ordination = "PCoA",
  ncomp = 3,
  trans_otu = FALSE,
  scale_species = FALSE
)
```

*Arguments:*

`ordination` default "PCoA"; "PCA", "PCoA" or "NMDS". PCA: principal component analysis; PCoA: principal coordinates analysis; NMDS: non-metric multidimensional scaling.

`ncomp` default 3; the returned dimensions.

`trans_otu` default FALSE; whether species abundance will be square transformed, used for PCA.

`scale_species` default FALSE; whether species loading in PCA will be scaled.

*Returns:* `res_ordination` stored in the object.

*Examples:*

```
t1$cal_ordination(ordination = "PCoA")
```

**Method** `plot_ordination()`: Plotting the ordination result based on An et al. (2019) <doi:10.1016/j.geoderma.2018.09.035>.

*Usage:*

```
trans_beta$plot_ordination(
  plot_type = "point",
  color_values = RColorBrewer::brewer.pal(8, "Dark2"),
  shape_values = c(16, 17, 7, 8, 15, 18, 11, 10, 12, 13, 9, 3, 4, 0, 1, 2, 14),
  plot_color = NULL,
  plot_shape = NULL,
  plot_group_order = NULL,
```

```

add_sample_label = NULL,
point_size = 3,
point_alpha = 0.8,
centroid_segment_alpha = 0.6,
centroid_segment_size = 1,
centroid_segment_linetype = 3,
ellipse_chull_fill = TRUE,
ellipse_chull_alpha = 0.1,
ellipse_level = 0.9,
ellipse_type = "t"
)

```

*Arguments:*

`plot_type` default "point"; one or more elements of "point", "ellipse", "chull" and "centroid".

**'point'** add point

**'ellipse'** add confidence ellipse for points of each group

**'chull'** add convex hull for points of each group

**'centroid'** add centroid line of each group

`color_values` default `RColorBrewer::brewer.pal(8, "Dark2")`; colors palette for different groups.

`shape_values` default `c(16, 17, 7, 8, 15, 18, 11, 10, 12, 13, 9, 3, 4, 0, 1, 2, 14)`; a vector for point shape types of groups, see `ggplot2` tutorial.

`plot_color` default `NULL`; a colname of `sample_table` to assign colors to different groups in plot.

`plot_shape` default `NULL`; a colname of `sample_table` to assign shapes to different groups in plot.

`plot_group_order` default `NULL`; a vector used to order the groups in the legend of plot.

`add_sample_label` default `NULL`; the column name in sample table, if provided, show the point name in plot.

`point_size` default 3; point size in plot when "point" is in `plot_type`.

`point_alpha` default .8; point transparency in plot when "point" is in `plot_type`.

`centroid_segment_alpha` default 0.6; segment transparency in plot when "centroid" is in `plot_type`.

`centroid_segment_size` default 1; segment size in plot when "centroid" is in `plot_type`.

`centroid_segment_linetype` default 3; the line type related with centroid in plot when "centroid" is in `plot_type`.

`ellipse_chull_fill` default `TRUE`; whether fill colors to the area of ellipse or chull.

`ellipse_chull_alpha` default 0.1; color transparency in the ellipse or convex hull depending on whether "ellipse" or "centroid" is in `plot_type`.

`ellipse_level` default .9; confidence level of ellipse when "ellipse" is in `plot_type`.

`ellipse_type` default "t"; ellipse type when "ellipse" is in `plot_type`; see type in [stat\\_ellipse](#).

*Returns:* `ggplot`.

*Examples:*

```

t1$plot_ordination(plot_type = "point")
t1$plot_ordination(plot_color = "Group", plot_shape = "Group", plot_type = "point")
t1$plot_ordination(plot_color = "Group", plot_type = c("point", "ellipse"))

```



```
t1$plot_ordination(plot_color = "Group", plot_type = c("point", "chull"))
t1$plot_ordination(plot_color = "Group", plot_type = c("point", "centroid"),
  centroid_segment_linetype = 1)
```

**Method** `cal_manova()`: Calculate perMANOVA based on Anderson al. (2008) <doi:10.1111/j.1442-9993.2001.01070.pp.x> and R `vegan` `adonis2` function.

*Usage:*

```
trans_beta$cal_manova(
  manova_all = TRUE,
  manova_set = NULL,
  group = NULL,
  p_adjust_method = "fdr",
  ...
)
```

*Arguments:*

`manova_all` default TRUE; TRUE represents test for all the groups, i.e. the overall test; FALSE represents test for all the paired groups.

`manova_set` default NULL; other specified group set for manova, such as "Group + Type" and "Group\*Type"; see also `adonis2`.

`group` default NULL; a column name of `sample_table` used for manova. If NULL, search group stored in the object.

`p_adjust_method` default "fdr"; p.adjust method when `manova_all = FALSE`; see method parameter of `p.adjust` function for available options.

... parameters passed to `adonis2` function of `vegan` package.

*Returns:* `res_manova` stored in object.

*Examples:*

```
t1$cal_manova(manova_all = TRUE)
```

**Method** `cal_betadisper()`: A wrapper for `betadisper` function in `vegan` package for multivariate homogeneity test of groups dispersions.

*Usage:*

```
trans_beta$cal_betadisper(...)
```

*Arguments:*

... parameters passed to `betadisper` function.

*Returns:* `res_betadisper` stored in object.

*Examples:*

```
t1$cal_betadisper()
```

**Method** `cal_group_distance()`: Transform sample distances within groups or between groups.

*Usage:*

```
trans_beta$cal_group_distance(within_group = TRUE)
```

*Arguments:*

`within_group` default TRUE; whether transform sample distance within groups, if FALSE, transform sample distance between any two groups.

*Returns:* res\_group\_distance stored in object.

*Examples:*

```
\donttest{
t1$cal_group_distance(within_group = TRUE)
}
```

**Method** plot\_group\_distance(): Plotting the distance between samples within or between groups.

*Usage:*

```
trans_beta$plot_group_distance(
  plot_group_order = NULL,
  color_values = RColorBrewer::brewer.pal(8, "Dark2"),
  distance_pair_stat = FALSE,
  hide_ns = FALSE,
  hide_ns_more = NULL,
  pair_compare_filter_match = NULL,
  pair_compare_filter_select = NULL,
  pair_compare_method = "wilcox.test",
  plot_distance_xtype = NULL
)
```

*Arguments:*

plot\_group\_order default NULL; a vector used to order the groups in the plot.

color\_values colors for presentation.

distance\_pair\_stat default FALSE; whether do the paired comparisons.

hide\_ns default FALSE; whether hide the "ns" pairs, i.e. non significant comparisons.

hide\_ns\_more default NULL; character vector; available when hide\_ns = TRUE; if provided, used for the specific significance filtering, such as c("ns", "\*").

pair\_compare\_filter\_match default NULL; only available when hide\_ns = FALSE; if provided, remove the matched groups; use the regular express to match the paired groups.

pair\_compare\_filter\_select default NULL; numeric vector; only available when hide\_ns = FALSE; if provided, only select those input groups. This parameter must be a numeric vector used to select the paired combination of groups. For example, pair\_compare\_filter\_select = c(1, 3) can be used to select "CW"- "IW" and "IW"- "TW" from all the three pairs "CW"- "IW", "CW"- "TW" and "IW"- "TW" of ordered groups ("CW", "IW", "TW"). The parameter pair\_compare\_filter\_select and pair\_compare\_filter\_match can not be both used together.

pair\_compare\_method default wilcox.test; wilcox.test, kruskal.test, t.test or anova.

plot\_distance\_xtype default NULL; number used to make x axis text generate angle.

*Returns:* ggplot.

*Examples:*

```
\donttest{
t1$plot_group_distance(distance_pair_stat = TRUE)
t1$plot_group_distance(distance_pair_stat = TRUE, hide_ns = TRUE)
t1$plot_group_distance(distance_pair_stat = TRUE, hide_ns = TRUE, hide_ns_more = c("ns", "*"))
t1$plot_group_distance(distance_pair_stat = TRUE, pair_compare_filter_select = 3)
}
```

**Method** `plot_clustering()`: Plotting clustering result. Require `ggdendro` package.

*Usage:*

```
trans_beta$plot_clustering(
  use_colors = RColorBrewer::brewer.pal(8, "Dark2"),
  measure = NULL,
  group = NULL,
  replace_name = NULL
)
```

*Arguments:*

`use_colors` colors for presentation.

`measure` default NULL; beta diversity index; If NULL, using the measure when creating object

`group` default NULL; if provided, use this group to assign color.

`replace_name` default NULL; if provided, use this as label.

*Returns:* `ggplot`.

*Examples:*

```
t1$plot_clustering(group = "Group", replace_name = c("Saline", "Type"))
```

**Method** `clone()`: The objects of this class are cloneable with this method.

*Usage:*

```
trans_beta$clone(deep = FALSE)
```

*Arguments:*

`deep` Whether to make a deep clone.

## Examples

```
## -----
## Method `trans_beta$new`
## -----

data(dataset)
t1 <- trans_beta$new(dataset = dataset, measure = "bray", group = "Group")

## -----
## Method `trans_beta$cal_ordination`
## -----

t1$cal_ordination(ordination = "PCoA")

## -----
## Method `trans_beta$plot_ordination`
## -----

t1$plot_ordination(plot_type = "point")
t1$plot_ordination(plot_color = "Group", plot_shape = "Group", plot_type = "point")
t1$plot_ordination(plot_color = "Group", plot_type = c("point", "ellipse"))
t1$plot_ordination(plot_color = "Group", plot_type = c("point", "chull"))
```

```

t1$plot_ordination(plot_color = "Group", plot_type = c("point", "centroid"),
  centroid_segment_linetype = 1)

## -----
## Method `trans_beta$cal_manova`
## -----

t1$cal_manova(manova_all = TRUE)

## -----
## Method `trans_beta$cal_betadisper`
## -----

t1$cal_betadisper()

## -----
## Method `trans_beta$cal_group_distance`
## -----

t1$cal_group_distance(within_group = TRUE)

## -----
## Method `trans_beta$plot_group_distance`
## -----

t1$plot_group_distance(distance_pair_stat = TRUE)
t1$plot_group_distance(distance_pair_stat = TRUE, hide_ns = TRUE)
t1$plot_group_distance(distance_pair_stat = TRUE, hide_ns = TRUE, hide_ns_more = c("ns", "*"))
t1$plot_group_distance(distance_pair_stat = TRUE, pair_compare_filter_select = 3)

## -----
## Method `trans_beta$plot_clustering`
## -----

t1$plot_clustering(group = "Group", replace_name = c("Saline", "Type"))

```

---

trans_classifier	<i>Create trans_classifier object for machine-learning-based model prediction.</i>
------------------	--

---

## Description

This class is a wrapper for methods of machine-learning-based classification models, including data pre-processing, feature selection, data split, model training, prediction, confusionMatrix and ROC (Receiver Operator Characteristic) or PR (Precision-Recall) curve.

Author(s): Felipe Mansoldo and Chi Liu

## Methods

### Public methods:

- `trans_classifier$new()`
- `trans_classifier$cal_preProcess()`
- `trans_classifier$cal_feature_sel()`
- `trans_classifier$cal_split()`
- `trans_classifier$set_trainControl()`
- `trans_classifier$cal_train()`
- `trans_classifier$cal_feature_imp()`
- `trans_classifier$cal_predict()`
- `trans_classifier$plot_confusionMatrix()`
- `trans_classifier$cal_ROC()`
- `trans_classifier$plot_ROC()`
- `trans_classifier$clone()`

**Method** `new()`: Create the `trans_classifier` object.

*Usage:*

```
trans_classifier$new(
  dataset = NULL,
  x.predictors = "all",
  y.response = NULL,
  n.cores = 1
)
```

*Arguments:*

`dataset` the object of `microtable` Class.

`x.predictors` default "all"; character string or data.frame; a character string represents selecting the corresponding data from `microtable$taxa_abund`; data.frame represents other customized data. See the following available options:

**'all'** use all the taxa stored in `microtable$taxa_abund`

**'Genus'** use Genus level table in `microtable$taxa_abund`, or other specific taxonomic rank, e.g. 'Phylum'

**other input** must be a data.frame; It should have the same format with the data.frame in `microtable$taxa_abund`, i.e. rows are features; cols are samples with same names in `sample_table`

`y.response` default NULL; the response variable in `sample_table`.

`n.cores` default 1; the CPU thread used.

*Returns:* `data_feature` and `data_response` in the object.

*Examples:*

```
\donttest{
data(dataset)
t1 <- trans_classifier$new(
dataset = dataset,
x.predictors = "Genus",
y.response = "Group")
}
```

**Method** `cal_preProcess()`: Pre-process (centering, scaling etc.) of the feature data based on the `caret::preProcess` function. See <https://topepo.github.io/caret/pre-processing.html> for more details.

*Usage:*

```
trans_classifier$cal_preProcess(...)
```

*Arguments:*

... parameters pass to `preProcess` function of `caret` package.

*Returns:* converted `data_feature` in the object.

*Examples:*

```
\dontrun{
t1$cal_preProcess(method = c("center", "scale", "nzv"))
}
```

**Method** `cal_feature_sel()`: Perform feature selection. See <https://topepo.github.io/caret/feature-selection-overview.html> for more details.

*Usage:*

```
trans_classifier$cal_feature_sel(
  boruta.maxRuns = 300,
  boruta.pValue = 0.01,
  boruta.repetitions = 4,
  ...
)
```

*Arguments:*

`boruta.maxRuns` default 300; maximal number of importance source runs; passed to the `maxRuns` parameter in `Boruta` function of `Boruta` package.

`boruta.pValue` default 0.01; p value passed to the `pValue` parameter in `Boruta` function of `Boruta` package.

`boruta.repetitions` default 4; repetition runs for the feature selection.

... parameters pass to `Boruta` function of `Boruta` package.

*Returns:* optimized `data_feature` in the object.

*Examples:*

```
\donttest{
t1$cal_feature_sel(boruta.maxRuns = 300, boruta.pValue = 0.01)
}
```

**Method** `cal_split()`: Split data for training and testing.

*Usage:*

```
trans_classifier$cal_split(prop.train = 3/4)
```

*Arguments:*

`prop.train` default 3/4; the ratio of the dataset used for the training.

*Returns:* `data_train` and `data_test` in the object.

*Examples:*

```
\donttest{
t1$cal_split(prop.train = 3/4)
}
```

**Method** `set_trainControl()`: Control parameters for the following training. See `trainControl` function of `caret` package for details.

*Usage:*

```
trans_classifier$set_trainControl(
  method = "repeatedcv",
  classProbs = TRUE,
  savePredictions = TRUE,
  ...
)
```

*Arguments:*

`method` default 'repeatedcv'; 'repeatedcv': Repeated k-Fold cross validation; see `method` parameter in `trainControl` function of `caret` package for available options.

`classProbs` default TRUE; should class probabilities be computed for classification models?; see `classProbs` parameter in `caret::trainControl` function.

`savePredictions` default TRUE; see `savePredictions` parameter in `caret::trainControl` function  
... parameters pass to `trainControl` function of `caret` package.

*Returns:* `trainControl` in the object.

*Examples:*

```
\dontrun{
t1$set_trainControl(method = 'repeatedcv')
}
```

**Method** `cal_train()`: Run the model training.

*Usage:*

```
trans_classifier$cal_train(
  method = "rf",
  metric = "Accuracy",
  max.mtry = 2,
  max.ntree = 200,
  ...
)
```

*Arguments:*

`method` default "rf"; "rf": random forest; see `method` in `caret::train` function for other options.

`metric` default "Accuracy"; see `metric` in `caret::train` function for other options.

`max.mtry` default 2; for `method = "rf"`; maximum `mtry` used for the `tuneGrid` to do hyperparameter tuning to optimize the model.

`max.ntree` default 200; for `method = "rf"`; maximum number of trees used to optimize the model.

... parameters pass to `train` function of `caret` package.

*Returns:* `res_train` in the object.

*Examples:*

```
\dontrun{
# random forest
t1$cal_train(method = "rf")
# Support Vector Machines with Radial Basis Function Kernel
t1$cal_train(method = "svmRadial", tuneLength = 15)
}
```

**Method** `cal_feature_imp()`: Get feature importance from the training model.

*Usage:*

```
trans_classifier$cal_feature_imp(...)
```

*Arguments:*

... parameters pass to `varImp` function of `caret` package.

*Returns:* `res_feature_imp` in the object. One row for each predictor variable. The column(s) are different importance measures.

*Examples:*

```
\dontrun{
t1$cal_feature_imp()
}
```

**Method** `cal_predict()`: Run the prediction.

*Usage:*

```
trans_classifier$cal_predict(positive_class = NULL)
```

*Arguments:*

`positive_class` default `NULL`; see `positive` parameter in `confusionMatrix` function of `caret` package; If `positive_class` is `NULL`, use the first group in data as the positive class automatically.

*Returns:* `res_predict`, `res_confusion_fit` and `res_confusion_stats` stored in the object.

*Examples:*

```
\dontrun{
t1$cal_predict()
}
```

**Method** `plot_confusionMatrix()`: Plot the cross-tabulation of observed and predicted classes with associated statistics.

*Usage:*

```
trans_classifier$plot_confusionMatrix(
  plot_confusion = TRUE,
  plot_statistics = TRUE
)
```

*Arguments:*

`plot_confusion` default `TRUE`; whether plot the confusion matrix.

`plot_statistics` default `TRUE`; whether plot the statistics.



*Returns:* ggplot object.

*Examples:*

```
\dontrun{
t1$plot_confusionMatrix()
}
```

**Method** `cal_ROC()`: Get ROC (Receiver Operator Characteristic) curve data and the performance data.

*Usage:*

```
trans_classifier$cal_ROC(input = "pred")
```

*Arguments:*

`input` default "pred"; 'pred' or 'train'; 'pred' represents using prediction results; 'train' represents using training results.

*Returns:* a list `res_ROC` stored in the object.

*Examples:*

```
\dontrun{
t1$cal_ROC()
}
```

**Method** `plot_ROC()`: Plot ROC curve.

*Usage:*

```
trans_classifier$plot_ROC(
  plot_type = c("ROC", "PR")[1],
  plot_group = "all",
  color_values = RColorBrewer::brewer.pal(8, "Dark2"),
  add_AUC = TRUE,
  plot_method = FALSE,
  ...
)
```

*Arguments:*

`plot_type` default `c("ROC", "PR")[1]`; 'ROC' represents ROC (Receiver Operator Characteristic) curve; 'PR' represents PR (Precision-Recall) curve.

`plot_group` default "all"; 'all' represents all the classes in the model; 'add' represents all adding micro-average and macro-average results, see [https://scikit-learn.org/stable/auto\\_examples/model\\_selection/p](https://scikit-learn.org/stable/auto_examples/model_selection/p) other options should be one or more class names, same with the names in Group column of `res_ROC$res_roc` from `cal_ROC` function.

`color_values` default `RColorBrewer::brewer.pal(8, "Dark2")`; colors used in the plot.

`add_AUC` default TRUE; whether add AUC in the legend.

`plot_method` default FALSE; If TRUE, show the method in the legend though only one method is found.

... parameters pass to `geom_path` function of `ggplot2` package.

*Returns:* ggplot2 object.

*Examples:*

```

\dontrun{
t1$plot_ROC(size = 1, alpha = 0.7)
}

```

**Method** clone(): The objects of this class are cloneable with this method.

*Usage:*

```
trans_classifier$clone(deep = FALSE)
```

*Arguments:*

deep Whether to make a deep clone.

## Examples

```

## -----
## Method `trans_classifier$new`
## -----

data(dataset)
t1 <- trans_classifier$new(
  dataset = dataset,
  x.predictors = "Genus",
  y.response = "Group")

## -----
## Method `trans_classifier$cal_preProcess`
## -----

## Not run:
t1$cal_preProcess(method = c("center", "scale", "nzv"))

## End(Not run)

## -----
## Method `trans_classifier$cal_feature_sel`
## -----

t1$cal_feature_sel(boruta.maxRuns = 300, boruta.pValue = 0.01)

## -----
## Method `trans_classifier$cal_split`
## -----

t1$cal_split(prop.train = 3/4)

## -----

```

```
## Method `trans_classifier$set_trainControl`  
## -----  
  
## Not run:  
t1$set_trainControl(method = 'repeatedcv')  
  
## End(Not run)  
  
## -----  
## Method `trans_classifier$cal_train`  
## -----  
  
## Not run:  
# random forest  
t1$cal_train(method = "rf")  
# Support Vector Machines with Radial Basis Function Kernel  
t1$cal_train(method = "svmRadial", tuneLength = 15)  
  
## End(Not run)  
  
## -----  
## Method `trans_classifier$cal_feature_imp`  
## -----  
  
## Not run:  
t1$cal_feature_imp()  
  
## End(Not run)  
  
## -----  
## Method `trans_classifier$cal_predict`  
## -----  
  
## Not run:  
t1$cal_predict()  
  
## End(Not run)  
  
## -----  
## Method `trans_classifier$plot_confusionMatrix`  
## -----  
  
## Not run:  
t1$plot_confusionMatrix()  
  
## End(Not run)  
  
## -----  
## Method `trans_classifier$cal_ROC`  
## -----  
  
## Not run:  
t1$cal_ROC()
```

```
## End(Not run)

## -----
## Method `trans_classifier$plot_ROC`
## -----

## Not run:
t1$plot_ROC(size = 1, alpha = 0.7)

## End(Not run)
```

---

trans_diff	<i>Create trans_diff object for the differential analysis on the taxonomic abundance.</i>
------------	---

---

## Description

This class is a wrapper for a series of differential abundance test and indicator analysis methods, including LEfSe based on the Segata et al. (2011) <doi:10.1186/gb-2011-12-6-r60>, random forest <doi:10.1016/j.geoderma.2018.09.035>, metastat based on White et al. (2009) <doi:10.1371/journal.pcbi.1000352>, the method in R package metagenomeSeq Paulson et al. (2013) <doi:10.1038/nmeth.2658>, non-parametric Kruskal-Wallis Rank Sum Test, Dunn's Kruskal-Wallis Multiple Comparisons based on the FSA package, Wilcoxon Rank Sum and Signed Rank Tests, t test and anova.

Authors: Chi Liu, Yang Cao, Chenhao Li

## Methods

### Public methods:

- `trans_diff$new()`
- `trans_diff$plot_diff_abund()`
- `trans_diff$plot_diff_bar()`
- `trans_diff$plot_diff_cladogram()`
- `trans_diff$clone()`

### Method `new()`:

*Usage:*

```
trans_diff$new(
  dataset = NULL,
  method = c("lefse", "rf", "metastat", "mseq", "KW", "KW_dunn", "wilcox", "t.test",
    "anova")[1],
  group = NULL,
  taxa_level = "all",
  filter_thres = 0,
  alpha = 0.05,
  p_adjust_method = "fdr",
```

```

lefse_subgroup = NULL,
lefse_min_subsam = 10,
lefse_norm = 1e+06,
nresam = 0.6667,
boots = 30,
rf_ntree = 1000,
group_choose_paired = NULL,
mseq_count = 1,
...
)

```

*Arguments:*

`dataset` the object of `microtable` Class.

`method` default "lefse"; see the following available options:

**'lefse'** LEfSe method based on Segata et al. (2011) <doi:10.1186/gb-2011-12-6-r60>

**'rf'** random forest and non-parametric test method based on An et al. (2019) <doi:10.1016/j.geoderma.2018.09.035>

**'metastat'** Metastat method for all paired groups based on White et al. (2009) <doi:10.1371/journal.pcbi.1000352>

**'mseq'** zero-inflated log-normal model-based differential test method from metagenome-Seq package.

**'KW'** KW: Kruskal-Wallis Rank Sum Test for all groups ( $\geq 2$ )

**'KW\_dunn'** Dunn's Kruskal-Wallis Multiple Comparisons when group number  $> 2$ ; see `dunnTest` function in FSA package

**'wilcox'** Wilcoxon Rank Sum and Signed Rank Tests for all paired groups

**'t.test'** Student's t-Test for all paired groups

**'anova'** Duncan's multiple range test for anova

`group` default NULL; sample group used for the comparison; a colname of `microtable$sample_table`.

`taxa_level` default "all"; 'all' represents using abundance data at all taxonomic ranks; For testing at a specific rank, provide taxonomic rank name, such as "Genus"; this parameter can be applied when `method != "mseq"`; 'mseq' method is performed on the feature abundance, i.e. `microtable$otu_table`.

`filter_thres` default 0; the relative abundance threshold used for `method != "metastat" or "mseq"`.

`alpha` default 0.05; differential significance threshold for `method = "lefse" or "rf"`; used to select taxa with significance across groups.

`p_adjust_method` default "fdr"; p.adjust method; see `method` parameter of `p.adjust` function for other available options; NULL can disuse the p value adjustment.

`lefse_subgroup` default NULL; sample sub group used for sub-comparison in `lefse`; Segata et al. (2011) <doi:10.1186/gb-2011-12-6-r60>.

`lefse_min_subsam` default 10; sample numbers required in the subgroup test.

`lefse_norm` default 1000000; scale value in `lefse`.

`nresam` default 0.6667; sample number ratio used in each bootstrap for `method = "lefse" or "rf"`.

`boots` default 30; bootstrap test number for `method = "lefse" or "rf"`.

`rf_ntree` default 1000; see `ntree` in `randomForest` function of `randomForest` package when `method = "rf"`.

group\_choose\_paired default NULL; a vector used for selecting the required groups for paired testing, only used for method = "metastat" or "mseq".

mseq\_count default 1; Filter features to have at least 'counts' counts.; see the count parameter in MRcoefs function of metagenomeSeq package.

... parameters passed to cal\_diff function of trans\_alpha class when method is one of "KW", "KW\_dunn", "wilcox", "t.test" and "anova".

Returns: res\_diff and res\_abund.

**res\_abund** includes mean abundance of each taxa (Mean), standard deviation (SD), standard error (SE) and sample number (N) in the group (Group).

**res\_diff** is the detailed differential test result, containing:

**"Comparison"**: The groups for the comparison, maybe all groups or paired groups. If this column is not found, all groups used;

**"Group"**: Which group has the maximum median or mean value across the test groups; For non-parametric methods, median value; For t.test, mean value;

**"Taxa"**: which taxa is used in this comparison;

**"Method"**: Test method used in the analysis depending on the method input;

**"LDA" or "MeanDecreaseGini"**: LDA: linear discriminant score in LefSe; MeanDecreaseGini: mean decreasing gini index in random forest;

**"Punadj" and "P.adj"**: raw p value; P.adj: adjusted p value;

**"qvalue"**: qvalue for metastat analysis.

Examples:

```
\donttest{
data(dataset)
t1 <- trans_diff$new(dataset = dataset, method = "lefse", group = "Group")
t1 <- trans_diff$new(dataset = dataset, method = "rf", group = "Group")
t1 <- trans_diff$new(dataset = dataset, method = "metastat", group = "Group", taxa_level = "Genus")
t1 <- trans_diff$new(dataset = dataset, method = "wilcox", group = "Group")
t1 <- trans_diff$new(dataset = dataset, method = "KW_dunn", group = "Group", taxa_level = "Phylum")
}
```

**Method** plot\_diff\_abund(): Plotting the abundance of differential taxa.

Usage:

```
trans_diff$plot_diff_abund(
  use_number = 1:20,
  color_values = RColorBrewer::brewer.pal(8, "Dark2"),
  select_group = NULL,
  select_taxa = NULL,
  simplify_names = TRUE,
  keep_prefix = TRUE,
  group_order = NULL,
  barwidth = 0.9,
  use_se = TRUE,
  add_sig = FALSE,
  add_sig_label = "Significance",
  add_sig_label_color = "black",
  add_sig_tip_length = 0.01,
  y_start = 1.01,
```

```

    y_increase = 0.05,
    text_y_size = 10,
    coord_flip = TRUE,
    ...
)

```

*Arguments:*

`use_number` default 1:20; numeric vector; the taxa numbers (1:n) used in the plot; If the n is larger than the number of total significant taxa, automatically use all the taxa.

`color_values` default RColorBrewer::brewer.pal(8, "Dark2"); colors palette.

`select_group` default NULL; this is used to select the paired groups. This parameter is especially useful when the comparison methods is applied to paired groups; The input `select_group` must be one of `object$res_diff$Comparison`.

`select_taxa` default NULL; character vector to provide taxa names. The taxa names should be same with the names shown in the plot, not the 'Taxa' column names in `object$res_diff$Taxa`.

`simplify_names` default TRUE; whether use the simplified taxonomic name.

`keep_prefix` default TRUE; whether retain the taxonomic prefix.

`group_order` default NULL; a vector to order groups, i.e. reorder the legend and colors in plot; If NULL, the function can first check whether the `group` column of `sample_table` is factor. If yes, use the levels in it. If provided, overlook the levels in the group of `sample_table`.

`barwidth` default 0.9; the bar width in plot.

`use_se` default TRUE; whether use SE in plot, if FALSE, use SD.

`add_sig` default FALSE; whether add the significance label to the plot.

`add_sig_label` default "Significance"; select a colname of `object$res_diff` for the label text, such as 'P.adj' or 'Significance'.

`add_sig_label_color` default "black"; the color for the label text when `add_sig = TRUE`.

`add_sig_tip_length` default 0.01; the tip length for the added line when `add_sig = TRUE`.

`y_start` default 1.01; the y axis position from which to add the label; the default 1.01 means  $1.01 * \text{Value}$ ; For method  $\neq$  "anova", all the start positions are same, i.e.  $\text{Value} = \max(\text{Mean} + \text{SD} \text{ or } \text{Mean} + \text{SE})$ ; For method = "anova"; the stat position is calculated for each point, i.e.  $\text{Value} = \text{Mean} + \text{SD} \text{ or } \text{Mean} + \text{SE}$ .

`y_increase` default 0.05; the increasing y axis space to add label for paired groups; the default 0.05 means  $0.05 * y\_start * \text{Value}$ ; In addition, this parameter is also used to label the letters of anova result with the fixed  $(1 + y\_increase) * y\_start * \text{Value}$ .

`text_y_size` default 10; the size for the y axis text.

`coord_flip` default TRUE; whether flip cartesian coordinates so that horizontal becomes vertical, and vertical, horizontal.

... parameters passed to `ggsignif::stat_signif` when `add_sig = TRUE`.

*Returns:* ggplot.

*Examples:*

```

\donttest{
t1 <- trans_diff$new(dataset = dataset, method = "anova", group = "Group", taxa_level = "Genus")
t1$plot_diff_abund(use_number = 1:10)
t1$plot_diff_abund(use_number = 1:10, add_sig = TRUE)
t1 <- trans_diff$new(dataset = dataset, method = "wilcox", group = "Group")
}

```

```

t1$plot_diff_abund(use_number = 1:20)
t1$plot_diff_abund(use_number = 1:20, add_sig = TRUE)
t1 <- trans_diff$new(dataset = dataset, method = "lefse", group = "Group")
t1$plot_diff_abund(use_number = 1:20)
t1$plot_diff_abund(use_number = 1:20, add_sig = TRUE)
}

```

**Method** `plot_diff_bar()`: Bar plot for LDA score.

*Usage:*

```

trans_diff$plot_diff_bar(
  color_values = RColorBrewer::brewer.pal(8, "Dark2"),
  use_number = 1:10,
  threshold = NULL,
  select_group = NULL,
  simplify_names = TRUE,
  keep_prefix = TRUE,
  group_order = NULL,
  axis_text_y = 12,
  plot_vertical = TRUE,
  ...
)

```

*Arguments:*

`color_values` default `RColorBrewer::brewer.pal(8, "Dark2")`; colors palette for different groups.

`use_number` default `1:10`; numeric vector; the taxa numbers used in the plot, i.e. `1:n`.

`threshold` default `NULL`; threshold value for selecting taxa, such as 3 for LDA score of LEfSe.

`select_group` default `NULL`; this is used to select the paired group when multiple comparisons are generated; The input `select_group` must be one of `object$res_diff$Comparison`.

`simplify_names` default `TRUE`; whether use the simplified taxonomic name.

`keep_prefix` default `TRUE`; whether retain the taxonomic prefix.

`group_order` default `NULL`; a vector to order the legend and colors in plot; If `NULL`, the function can first check whether the group column of `sample_table` is factor. If yes, use the levels in it. If provided, this parameter can overwrite the levels in the group of `sample_table`.

`axis_text_y` default `12`; the size for the y axis text.

`plot_vertical` default `TRUE`; whether use vertical bar plot or horizontal.

... parameters pass to `geom_bar`

*Returns:* `ggplot`.

*Examples:*

```

\donttest{
t1$plot_diff_bar(use_number = 1:20)
}

```

**Method** `plot_diff_cladogram()`: Plot the cladogram using taxa with significant difference.

*Usage:*



```

trans_diff$plot_diff_cladogram(
  color = RColorBrewer::brewer.pal(8, "Dark2"),
  use_taxa_num = 200,
  filter_taxa = NULL,
  use_feature_num = NULL,
  group_order = NULL,
  clade_label_level = 4,
  select_show_labels = NULL,
  only_select_show = FALSE,
  sep = "|",
  branch_size = 0.2,
  alpha = 0.2,
  clade_label_size = 2,
  clade_label_size_add = 5,
  clade_label_size_log = exp(1),
  node_size_scale = 1,
  node_size_offset = 1,
  annotation_shape = 22,
  annotation_shape_size = 5
)

```

*Arguments:*

`color` default `RColorBrewer::brewer.pal(8, "Dark2")`; color palette used in the plot.

`use_taxa_num` default 200; integer; The taxa number used in the background tree plot; select the taxa according to the mean abundance .

`filter_taxa` default `NULL`; The mean relative abundance used to filter the taxa with low abundance.

`use_feature_num` default `NULL`; integer; The feature number used in the plot; select the features according to the LDA score (method = "lfe") or MeanDecreaseGini (method = "rf") from high to low.

`group_order` default `NULL`; a vector to order the legend and colors in plot; If `NULL`, the function can first check whether the group column of `sample_table` is factor. If yes, use the levels in it. If provided, this parameter can overwrite the levels in the group of `sample_table`.

`clade_label_level` default 4; the taxonomic level for marking the label with letters, root is the largest.

`select_show_labels` default `NULL`; character vector; The features to show in the plot with full label names, not the letters.

`only_select_show` default `FALSE`; whether only use the the select features in the parameter `select_show_labels`.

`sep` default "|"; the separate character in the taxonomic information.

`branch_size` default 0.2; numeric, size of branch.

`alpha` default 0.2; shading of the color.

`clade_label_size` default 2; basic size for the clade label; please also see `clade_label_size_add` and `clade_label_size_log`

`clade_label_size_add` default 5; added basic size for the clade label; see the formula in `clade_label_size_log` parameter.

`clade_label_size_log` default `exp(1)`; the base of log function for added size of the clade label; the size formula: `clade_label_size + log(clade_label_level + clade_label_size_add,`

base = clade\_label\_size\_log); so use clade\_label\_size\_log, clade\_label\_size\_add and clade\_label\_size can totally control the label size for different taxonomic levels.

node\_size\_scale default 1; scale for the node size.

node\_size\_offset default 1; offset for the node size.

annotation\_shape default 22; shape used in the annotation legend.

annotation\_shape\_size default 5; size used in the annotation legend.

Returns: ggplot.

Examples:

```
\donttest{
t1$plot_diff_cladogram(use_taxa_num = 100, use_feature_num = 30, select_show_labels = NULL)
}
```

**Method clone():** The objects of this class are cloneable with this method.

Usage:

```
trans_diff$clone(deep = FALSE)
```

Arguments:

deep Whether to make a deep clone.

## Examples

```
## -----
## Method `trans_diff$new`
## -----

data(dataset)
t1 <- trans_diff$new(dataset = dataset, method = "lfc", group = "Group")
t1 <- trans_diff$new(dataset = dataset, method = "rf", group = "Group")
t1 <- trans_diff$new(dataset = dataset, method = "metastat", group = "Group", taxa_level = "Genus")
t1 <- trans_diff$new(dataset = dataset, method = "wilcox", group = "Group")
t1 <- trans_diff$new(dataset = dataset, method = "KW_dunn", group = "Group", taxa_level = "Phylum")

## -----
## Method `trans_diff$plot_diff_abund`
## -----

t1 <- trans_diff$new(dataset = dataset, method = "anova", group = "Group", taxa_level = "Genus")
t1$plot_diff_abund(use_number = 1:10)
t1$plot_diff_abund(use_number = 1:10, add_sig = TRUE)
t1 <- trans_diff$new(dataset = dataset, method = "wilcox", group = "Group")
t1$plot_diff_abund(use_number = 1:20)
t1$plot_diff_abund(use_number = 1:20, add_sig = TRUE)
t1 <- trans_diff$new(dataset = dataset, method = "lfc", group = "Group")
t1$plot_diff_abund(use_number = 1:20)
t1$plot_diff_abund(use_number = 1:20, add_sig = TRUE)
```

```

## -----
## Method `trans_diff$plot_diff_bar`
## -----

t1$plot_diff_bar(use_number = 1:20)

## -----
## Method `trans_diff$plot_diff_cladogram`
## -----

t1$plot_diff_cladogram(use_taxa_num = 100, use_feature_num = 30, select_show_labels = NULL)

```

---

trans_env	<i>Create trans_env object for the analysis of the effects of environmental factors on communities.</i>
-----------	---

---

## Description

This class is a wrapper for a series of operations associated with environmental measurements, including redundancy analysis, mantel test, correlation analysis and linear fitting based on An et al. (2019) <doi:10.1016/j.geoderma.2018.09.035>.

## Methods

### Public methods:

- `trans_env$new()`
- `trans_env$scal_diff()`
- `trans_env$scal_autocor()`
- `trans_env$scal_ordination()`
- `trans_env$scal_ordination_envsquare()`
- `trans_env$trans_ordination()`
- `trans_env$plot_ordination()`
- `trans_env$scal_mantel()`
- `trans_env$scal_cor()`
- `trans_env$plot_cor()`
- `trans_env$plot_scatterfit()`
- `trans_env$print()`
- `trans_env$clone()`

### Method `new()`:

*Usage:*

```
trans_env$new(
  dataset = NULL,
  env_cols = NULL,
  add_data = NULL,
  character2numeric = TRUE,
  complete_na = FALSE
)
```

*Arguments:*

`dataset` the object of `microtable` Class.

`env_cols` default NULL; either numeric vector or character vector to select columns in `sample_table` of your `microtable` object. This parameter should be used in the case that all the required environmental data is in `sample_table` of your `microtable` object. Otherwise, please use `add_data` parameter.

`add_data` default NULL; data.frame format; provide the environmental data in the format data.frame; rownames should be sample names. This parameter should be used when the `sample_table` in your `microtable` object has no environmental data. Under this circumstance, the `env_cols` parameter can not be used because no data can be selected.

`character2numeric` default TRUE; whether transform the characters or factors to numeric attributes.

`complete_na` default FALSE; Whether fill the NA (missing value) in the environmental data; If TRUE, the function can run the interpolation with the `mice` package; to use this parameter, please first install `mice` package.

*Returns:* `data_env` in `trans_env` object.

*Examples:*

```
data(dataset)
data(env_data_16S)
t1 <- trans_env$new(dataset = dataset, add_data = env_data_16S[, 4:11])
```

**Method** `cal_diff()`: Test the difference of environmental variable across groups.

*Usage:*

```
trans_env$cal_diff(
  group = NULL,
  method = c("KW", "KW_dunn", "wilcox", "t.test", "anova")[1],
  measure = NULL,
  p_adjust_method = "fdr",
  anova_set = NULL,
  ...
)
```

*Arguments:*

`group` default NULL; a colname of `sample_table` used to compare values across groups.

`method` default "KW"; see the following available options:

**'KW'** KW: Kruskal-Wallis Rank Sum Test for all groups ( $\geq 2$ )

**'KW\_dunn'** Dunn's Kruskal-Wallis Multiple Comparisons, see `dunnTest` function in `FSA` package

**'wilcox'** Wilcoxon Rank Sum and Signed Rank Tests for all paired groups

**'t.test'** Student's t-Test for all paired groups

**'anova'** Duncan's multiple range test for anova

measure default NULL; a vector; if null, all variables will be calculated.

p\_adjust\_method default "fdr"; p.adjust method; see method parameter of p.adjust function for available options.

anova\_set default NULL; specified group set for anova, such as 'block + N\*P\*K', see [aov](#).

... parameters passed to `kruskal.test` or `wilcox.test` function (method = "KW") or `dunnTest` function of FSA package (method = "KW\_dunn") or `agricolae::duncan.test` (method = "anova").

*Returns:* `res_diff` in object. A data.frame generally. A list for anova when `anova_set` is assigned. In the data frame, 'Group' column means that the group has the maximum median or mean value across the test groups; For non-parametric methods, maximum median value; For `t.test`, maximum mean value.

*Examples:*

```
\donttest{
t1$cal_diff(group = "Group", method = "KW")
t1$cal_diff(group = "Group", method = "KW_dunn")
t1$cal_diff(group = "Group", method = "anova")
}
```

**Method** `cal_autocor()`: Calculate the autocorrelations among environmental variables and plot the result.

*Usage:*

```
trans_env$cal_autocor(
  group = NULL,
  color_values = RColorBrewer::brewer.pal(8, "Dark2"),
  alpha = 0.8,
  ...
)
```

*Arguments:*

group default NULL; a colname of `sample_table`; used to perform calculations for different groups.

color\_values default `RColorBrewer::brewer.pal(8, "Dark2")`; colors palette.

alpha default 0.8; the alpha value to add transparency in colors; useful when group is not NULL.

... default parameters passed to `GGally::ggpairs`.

*Returns:* `ggmatrix`.

*Examples:*

```
\donttest{
t1$cal_autocor(method = "GGally")
}
```

**Method** `cal_ordination()`: Redundancy analysis (RDA) and Correspondence Analysis (CCA) based on the `vegan` package.

*Usage:*

```
trans_env$cal_ordination(
  method = c("RDA", "dbRDA", "CCA")[1],
  feature_sel = FALSE,
  taxa_level = NULL,
  taxa_filter_thres = NULL,
  use_measure = NULL,
  add_matrix = NULL,
  ...
)
```

*Arguments:*

`method` default `c("RDA", "dbRDA", "CCA")[1]`; the ordination method.

`feature_sel` default `FALSE`; whether perform the feature selection based on forward selection method.

`taxa_level` default `NULL`; If use `RDA` or `CCA`, provide the taxonomic rank, such as "Phylum" or "Genus"; If use `otu_table`; please input "OTU".

`taxa_filter_thres` default `NULL`; If want to filter taxa, provide the relative abundance threshold.

`use_measure` default `NULL`; a name of beta diversity matrix; only useful when parameter `method = "dbRDA"`; If not provided, use the first beta diversity matrix automatically.

`add_matrix` default `NULL`; additional distance matrix provided, when the user does not want to use the beta diversity matrix within the dataset; only available when `method = "dbRDA"`.

... parameters pass to `dbrda` or `rda` or `cca` function according to the input of `method`.

*Returns:* `res_ordination`, `res_ordination_R2`, `res_ordination_terms` and `res_ordination_axis` in object.

*Examples:*

```
\donttest{
t1$cal_ordination(method = "dbRDA", use_measure = "bray")
t1$cal_ordination(method = "RDA", taxa_level = "Genus")
t1$cal_ordination(method = "CCA", taxa_level = "Genus")
}
```

**Method** `cal_ordination_envsquare()`: Fits each environmental vector onto the ordination to obtain the contribution of each variable.

*Usage:*

```
trans_env$cal_ordination_envsquare(...)
```

*Arguments:*

... the parameters passing to `vegan::envfit` function.

*Returns:* `res_ordination_envsquare` in object.

*Examples:*

```
\donttest{
t1$cal_ordination_envsquare()
}
```

**Method** `trans_ordination()`: transform ordination result for the following plotting.

*Usage:*

```
trans_env$trans_ordination(
  show_taxa = 10,
  adjust_arrow_length = FALSE,
  min_perc_env = 0.1,
  max_perc_env = 0.8,
  min_perc_tax = 0.1,
  max_perc_tax = 0.8
)
```

*Arguments:*

show\_taxa default 10; taxa number shown in the plot.

adjust\_arrow\_length default FALSE; whether adjust the arrow length to be clearer.

min\_perc\_env default 0.1; used for scaling up the minimum of env arrow; multiply by the maximum distance between samples and origin.

max\_perc\_env default 0.8; used for scaling up the maximum of env arrow; multiply by the maximum distance between samples and origin.

min\_perc\_tax default 0.1; used for scaling up the minimum of tax arrow; multiply by the maximum distance between samples and origin.

max\_perc\_tax default 0.8; used for scaling up the maximum of tax arrow; multiply by the maximum distance between samples and origin.

*Returns:* res\_ordination\_trans in object.

*Examples:*

```
\donttest{
t1$trans_ordination(adjust_arrow_length = TRUE, min_perc_env = 0.1, max_perc_env = 1)
}
```

**Method** plot\_ordination(): plot ordination result.

*Usage:*

```
trans_env$plot_ordination(
  plot_color = NULL,
  plot_shape = NULL,
  color_values = RColorBrewer::brewer.pal(8, "Dark2"),
  shape_values = c(16, 17, 7, 8, 15, 18, 11, 10, 12, 13, 9, 3, 4, 0, 1, 2, 14),
  env_text_color = "black",
  env_arrow_color = "grey30",
  taxa_text_color = "firebrick1",
  taxa_arrow_color = "firebrick1",
  env_text_size = 3.7,
  taxa_text_size = 3,
  taxa_text_italic = TRUE,
  plot_type = "point",
  point_size = 3,
  point_alpha = 0.8,
  centroid_segment_alpha = 0.6,
  centroid_segment_size = 1,
  centroid_segment_linetype = 3,
```

```

ellipse_chull_fill = TRUE,
ellipse_chull_alpha = 0.1,
ellipse_level = 0.9,
ellipse_type = "t",
add_sample_label = NULL,
env_nudge_x = NULL,
env_nudge_y = NULL,
taxa_nudge_x = NULL,
taxa_nudge_y = NULL,
...
)

```

*Arguments:*

`plot_color` default NULL; a colname of `sample_table` to assign colors to different groups in plot.

`plot_shape` default NULL; a colname of `sample_table` to assign shapes to different groups in plot.

`color_values` default `RColorBrewer::brewer.pal(8, "Dark2")`; color pallete for different groups.

`shape_values` default `c(16, 17, 7, 8, 15, 18, 11, 10, 12, 13, 9, 3, 4, 0, 1, 2, 14)`; a vector for point shape types of groups, see `ggplot2` tutorial.

`env_text_color` default "black"; environmental variable text color.

`env_arrow_color` default "grey30"; environmental variable arrow color.

`taxa_text_color` default "firebrick1"; taxa text color.

`taxa_arrow_color` default "firebrick1"; taxa arrow color.

`env_text_size` default 3.7; environmental variable text size.

`taxa_text_size` default 3; taxa text size.

`taxa_text_italic` default TRUE; "italic"; whether use "italic" style for the taxa text in the plot.

`plot_type` default "point"; one or more elements of "point", "ellipse", "chull" and "centroid".

- 'point'** add point
- 'ellipse'** add confidence ellipse for points of each group
- 'chull'** add convex hull for points of each group
- 'centroid'** add centroid line of each group

`point_size` default 3; point size in plot when "point" is in `plot_type`.

`point_alpha` default .8; point transparency in plot when "point" is in `plot_type`.

`centroid_segment_alpha` default 0.6; segment transparency in plot when "centroid" is in `plot_type`.

`centroid_segment_size` default 1; segment size in plot when "centroid" is in `plot_type`.

`centroid_segment_linetype` default 3; an integer; the line type related with centroid in plot when "centroid" is in `plot_type`.

`ellipse_chull_fill` default TRUE; whether fill colors to the area of ellipse or chull.

`ellipse_chull_alpha` default 0.1; color transparency in the ellipse or convex hull depending on whether "ellipse" or "centroid" is in `plot_type`.

`ellipse_level` default .9; confidence level of ellipse when "ellipse" is in `plot_type`.

`ellipse_type` default "t"; ellipse type when "ellipse" is in `plot_type`; see type in [stat\\_ellipse](#).



`add_sample_label` default NULL; the column name in sample table, if provided, show the point name in plot.

`env_nudge_x` default NULL; numeric vector to adjust the env text x axis position; passed to `nudge_x` parameter of `geom_text_repel` function of `ggrepel` package; default NULL represents automatic adjustment; the length must be same with the row number of `object$res_ordination_trans$df_arrows`. For example, if there are 5 env variables, `env_nudge_x` should be something like `c(0.1, 0, -0.2, 0, 0)`. Note that this parameter and `env_nudge_y` is generally used when the automatic text adjustment is not very well.

`env_nudge_y` default NULL; numeric vector to adjust the env text y axis position; passed to `nudge_y` parameter of `ggrepel::geom_text_repel` function; default NULL represents automatic adjustment; the length must be same with the row number of `object$res_ordination_trans$df_arrows`. For example, if there are 5 env variables, `env_nudge_y` should be something like `c(0.1, 0, -0.2, 0, 0)`.

`taxa_nudge_x` default NULL; numeric vector to adjust the taxa text x axis position; passed to `nudge_x` parameter of `ggrepel::geom_text_repel` function; default NULL represents automatic adjustment; the length must be same with the row number of `object$res_ordination_trans$df_arrows_spe`. For example, if 3 taxa are shown, `taxa_nudge_x` should be something like `c(0.3, -0.2, 0)`.

`taxa_nudge_y` default NULL; numeric vector to adjust the taxa text y axis position; passed to `nudge_y` parameter of `ggrepel::geom_text_repel` function; default NULL represents automatic adjustment; the length must be same with the row number of `object$res_ordination_trans$df_arrows_spe`. For example, if 3 taxa are shown, `taxa_nudge_y` should be something like `c(-0.2, 0, 0.4)`.

... parameters pass to `geom_point` for controlling sample points.

*Returns:* ggplot object.

*Examples:*

```
\donttest{
t1$cal_ordination(method = "RDA")
t1$trans_ordination(adjust_arrow_length = TRUE, max_perc_env = 1.5)
t1$plot_ordination(plot_color = "Group")
t1$plot_ordination(plot_color = "Group", plot_shape = "Group", plot_type = c("point", "ellipse"))
t1$plot_ordination(plot_color = "Group", plot_type = c("point", "chull"))
t1$plot_ordination(plot_color = "Group", plot_type = c("point", "centroid"),
  centroid_segment_linetype = 1)
t1$plot_ordination(plot_color = "Group", env_nudge_x = c(0.4, 0, 0, 0, 0, -0.2, 0, 0),
  env_nudge_y = c(0.6, 0, 0.2, 0.5, 0, 0.1, 0, 0.2))
}
```

**Method** `cal_mantel()`: Mantel test between beta diversity matrix and environmental data.

*Usage:*

```
trans_env$cal_mantel(
  select_env_data = NULL,
  partial_mantel = FALSE,
  add_matrix = NULL,
  use_measure = NULL,
  method = "pearson",
  p_adjust_method = "fdr",
  ...
)
```

*Arguments:*

select\_env\_data default NULL; numeric or character vector to select columns in data\_env; if not provided, automatically select the columns with numeric attributes.

partial\_mantel default FALSE; whether use partial mantel test; If TRUE, use other measurements as the zdis.

add\_matrix default NULL; additional distance matrix provided, if you donot want to use the beta diversity matrix in the dataset.

use\_measure default NULL; name of beta diversity matrix. If necessary and not provided, use the first beta diversity matrix.

method default "pearson"; one of "pearson", "spearman" and "kendall"; correlation method; see method parameter in mantel function of vegan package.

p\_adjust\_method default "fdr"; p.adjust method; see method parameter of p.adjust function for available options.

... parameters pass to [mantel](#) of vegan package.

*Returns:* res\_mantel in object.

*Examples:*

```
\donttest{
t1$cal_mantel(use_measure = "bray")
t1$cal_mantel(partial_mantel = TRUE, use_measure = "bray")
}
```

**Method** cal\_cor(): Calculating the correlations between taxa abundance and environmental variables. Actually, it can also be used for calculating other correlation between any two variables from two tables.

*Usage:*

```
trans_env$cal_cor(
  use_data = c("Genus", "all", "other")[1],
  select_env_data = NULL,
  cor_method = c("pearson", "spearman", "kendall")[1],
  p_adjust_method = "fdr",
  p_adjust_type = c("Type", "Taxa", "Env")[3],
  add_abund_table = NULL,
  by_group = NULL,
  use_taxa_num = NULL,
  other_taxa = NULL,
  group_use = NULL,
  group_select = NULL,
  taxa_name_full = TRUE
)
```

*Arguments:*

use\_data default "Genus"; "Genus", "all" or "other"; "Genus" or other taxonomic name: use genus or other taxonomic abundance table in taxa\_abund; "all": use all merged taxa abundance table; "other": provide additional taxa name with other\_taxa parameter which is necessary.

select\_env\_data default NULL; numeric or character vector to select columns in data\_env; if not provided, automatically select the columns with numeric attributes.

cor\_method default "pearson"; "pearson", "spearman" or "kendall"; correlation method.

p\_adjust\_method default "fdr"; p.adjust method; see method parameter of p.adjust function for available options.

p\_adjust\_type default "Env"; "Type", "Taxa" or "Env"; p.adjust type; Env: environmental data; Taxa: taxa data; Type: group used.

add\_abund\_table default NULL; additional data table to be used. Samples must be rows.

by\_group default NULL; one column name or number in sample\_table; calculate correlations for different groups separately.

use\_taxa\_num default NULL; integer; a number used to select high abundant taxa; only useful when use\_data parameter is a taxonomic level, e.g., "Genus".

other\_taxa default NULL; character vector containing a series of taxa names; used when use\_data = "other"; the provided names should be standard full names used to select taxa from all the tables in taxa\_abund list of the microtable object; please see the example.

group\_use default NULL; numeric or character vector to select one column in sample\_table for selecting samples; together with group\_select.

group\_select default NULL; the group name used; remain samples within the group.

taxa\_name\_full default TRUE; Whether use the complete taxonomic name of taxa.

*Returns:* res\_cor in object.

*Examples:*

```
\donttest{
t2 <- trans_diff$new(dataset = dataset, method = "rf", group = "Group", rf_taxa_level = "Genus")
t1 <- trans_env$new(dataset = dataset, add_data = env_data_16S[, 4:11])
t1$cal_cor(use_data = "other", p_adjust_method = "fdr", other_taxa = t2$res_diff$Taxa[1:40])
}
```

**Method** plot\_cor(): Plot correlation heatmap.

*Usage:*

```
trans_env$plot_cor(
  color_vector = c("#053061", "white", "#A50026"),
  color_palette = NULL,
  pheatmap = FALSE,
  filter_feature = NULL,
  ylab_type_italic = FALSE,
  keep_full_name = FALSE,
  keep_prefix = TRUE,
  text_y_order = NULL,
  text_x_order = NULL,
  font_family = NULL,
  cluster_ggplot = "none",
  cluster_height_rows = 0.2,
  cluster_height_cols = 0.2,
  text_y_position = "right",
  mylabels_x = NULL,
  ...
)
```

*Arguments:*

color\_vector default c("#053061", "white", "#A50026"); colors with only three values representing low, middle and high value.  
 color\_palette default NULL; a customized palette with more color values; if provided, use it instead of color\_vector.  
 pheatmap default FALSE; whether use pheatmap package to plot the heatmap.  
 filter\_feature default NULL; character vector; used to filter features that only have significance labels in the filter\_feature vector. For example, filter\_feature = "" can be used to filter features that only have "", no any "\*".  
 ylab\_type\_italic default FALSE; whether use italic type for y lab text.  
 keep\_full\_name default FALSE; whether use the complete taxonomic name.  
 keep\_prefix default TRUE; whether retain the taxonomic prefix.  
 text\_y\_order default NULL; character vector; provide customized text order for y axis; shown in the plot from the top down.  
 text\_x\_order default NULL; character vector; provide customized text order for x axis.  
 font\_family default NULL; font family used in ggplot2; only available when pheatmap = FALSE.  
 cluster\_ggplot default "none"; add clustering dendrogram for ggplot2 based heatmap; available options: "none", "row", "col" or "both". "none": no any clustering used; "row": add clustering for rows; "col": add clustering for columns; "both": add clustering for both rows and columns. Only available when pheatmap = FALSE.  
 cluster\_height\_rows default 0.2, the dendrogram plot height for rows; available when cluster\_ggplot != "none".  
 cluster\_height\_cols default 0.2, the dendrogram plot height for columns; available cluster\_ggplot != "none".  
 text\_y\_position default "right"; "left" or "right"; the y axis text position; ggplot2 based heatmap.  
 mylabels\_x default NULL; provide x axis text labels additionally; only available when pheatmap = TRUE.  
 ... parameters pass to ggplot2::geom\_tile or pheatmap, depending on the pheatmap = FALSE or TRUE.

*Returns:* plot.

*Examples:*

```

\donttest{
t1$plot_cor(pheatmap = FALSE)
}

```

**Method** plot\_scatterfit(): Scatter plot and add fitted line. The most important thing is to make sure that the input x and y have corresponding sample orders. If one of x and y is a matrix, the other will be also transformed to matrix with Euclidean distance. Then, both of them are transformed to be vectors. If x or y is a vector with a single value, x or y will be assigned according to the column selection of the data\_env inside.

*Usage:*

```

trans_env$plot_scatterfit(
  x = NULL,
  y = NULL,

```

```

group = NULL,
group_order = NULL,
color_values = RColorBrewer::brewer.pal(8, "Dark2"),
shape_values = NULL,
type = c("cor", "lm")[1],
cor_method = "pearson",
label_sep = ";",
label.x.npc = "left",
label.y.npc = "top",
label.x = NULL,
label.y = NULL,
x_axis_title = "",
y_axis_title = "",
point_size = 5,
point_alpha = 0.6,
line_size = 0.8,
line_alpha = 1,
line_color = "black",
line_se = TRUE,
line_se_color = "grey70",
pvalue_trim = 4,
cor_coef_trim = 3,
lm_fir_trim = 2,
lm_sec_trim = 2,
lm_squ_trim = 2,
...
)

```

*Arguments:*

- x default NULL; a single numeric or character value or a vector or a distance matrix used for the x axis. If x is a single value, it will be used to select the column of data\_env inside. If x is a distance matrix, it will be transformed to be a vector.
- y default NULL; a single numeric or character value or a vector or a distance matrix used for the y axis. If y is a single value, it will be used to select the column of data\_env inside. If y is a distance matrix, it will be transformed to be a vector.
- group default NULL; a character vector; if length is 1, must be a colname of dataset\$sample\_table; Otherwise, group should be a vector with same length of x/y (for vector) or ncol of x/y (for matrix).
- group\_order default NULL; a vector to order groups, i.e. reorder the legend and colors in plot when group is not NULL; If group\_order is NULL and group is provided, the function can first check whether the group column of dataset\$sample\_table is factor. If provided, overlook the levels in the group of dataset\$sample\_table.
- color\_values default RColorBrewer::brewer.pal(8, "Dark2"); color pallete for different groups.
- shape\_values default NULL; a numeric vector for point shape types of groups when group is not NULL, see ggplot2 tutorial.
- type default c("cor", "lm")[1]; "cor": correlation; "lm" for regression.
- cor\_method default "pearson"; one of "pearson", "kendall" and "spearman"; correlation method.
- label\_sep default ";"; the separator string between different label parts.

label.x.npc default "left"; can be numeric or character vector of the same length as the number of groups and/or panels. If too short they will be recycled.

**numeric** value should be between 0 and 1. Coordinates to be used for positioning the label, expressed in "normalized parent coordinates"

**character** allowed values include: i) one of c('right', 'left', 'center', 'centre', 'middle') for x-axis; ii) and one of c('bottom', 'top', 'center', 'centre', 'middle') for y-axis.

label.y.npc default "top"; same usage with label.x.npc; see also label.y.npc parameter of stat\_cor of ggpubr package.

label.x default NULL; x axis absolute position for adding the statistic label.

label.y default NULL; y axis absolute position for adding the statistic label.

x\_axis\_title default ""; the title of x axis.

y\_axis\_title default ""; the title of y axis.

point\_size default 5; point size value.

point\_alpha default 0.6; alpha value for the point color transparency.

line\_size default 0.8; line size value.

line\_alpha default 1; alpha value for the line color transparency.

line\_color default "black"; fitted line color only useful when group = NULL.

line\_se default TRUE; Whether show the confidence interval for the fitting.

line\_se\_color default "grey70"; the color to fill the confidence interval when line\_se = TRUE.

pvalue\_trim default 4; trim the decimal places of p value.

cor\_coef\_trim default 3; trim the decimal places of correlation coefficient.

lm\_fir\_trim default 2; trim the decimal places of regression first coefficient.

lm\_sec\_trim default 2; trim the decimal places of regression second coefficient.

lm\_squ\_trim default 2; trim the decimal places of regression R square.

... other arguments to pass to geom\_text or geom\_label.

*Returns:* plot.

*Examples:*

```
\donttest{
t1$plot_scatterfit(x = 1, y = 2, type = "cor")
t1$plot_scatterfit(x = 1, y = 2, type = "lm", point_alpha = .3)
t1$plot_scatterfit(x = "pH", y = "TOC", type = "lm", group = "Group", line_se = FALSE)
t1$plot_scatterfit(x =
  dataset$beta_diversity$bray[rownames(t1$data_env), rownames(t1$data_env)], y = "pH")
}
```

**Method** print(): Print the trans\_env object.

*Usage:*

```
trans_env$print()
```

**Method** clone(): The objects of this class are cloneable with this method.

*Usage:*

```
trans_env$clone(deep = FALSE)
```

*Arguments:*

deep Whether to make a deep clone.

**Examples**

```

## -----
## Method `trans_env$new`
## -----

data(dataset)
data(env_data_16S)
t1 <- trans_env$new(dataset = dataset, add_data = env_data_16S[, 4:11])

## -----
## Method `trans_env$scal_diff`
## -----

t1$scal_diff(group = "Group", method = "KW")
t1$scal_diff(group = "Group", method = "KW_dunn")
t1$scal_diff(group = "Group", method = "anova")

## -----
## Method `trans_env$scal_autocor`
## -----

t1$scal_autocor(method = "GGally")

## -----
## Method `trans_env$scal_ordination`
## -----

t1$scal_ordination(method = "dbRDA", use_measure = "bray")
t1$scal_ordination(method = "RDA", taxa_level = "Genus")
t1$scal_ordination(method = "CCA", taxa_level = "Genus")

## -----
## Method `trans_env$scal_ordination_envsquare`
## -----

t1$scal_ordination_envsquare()

## -----
## Method `trans_env$trans_ordination`
## -----

t1$trans_ordination(adjust_arrow_length = TRUE, min_perc_env = 0.1, max_perc_env = 1)

```

```

## -----
## Method `trans_env$plot_ordination`
## -----

t1$cal_ordination(method = "RDA")
t1$trans_ordination(adjust_arrow_length = TRUE, max_perc_env = 1.5)
t1$plot_ordination(plot_color = "Group")
t1$plot_ordination(plot_color = "Group", plot_shape = "Group", plot_type = c("point", "ellipse"))
t1$plot_ordination(plot_color = "Group", plot_type = c("point", "chull"))
t1$plot_ordination(plot_color = "Group", plot_type = c("point", "centroid"),
  centroid_segment_linetype = 1)
t1$plot_ordination(plot_color = "Group", env_nudge_x = c(0.4, 0, 0, 0, 0, -0.2, 0, 0),
  env_nudge_y = c(0.6, 0, 0.2, 0.5, 0, 0.1, 0, 0.2))

## -----
## Method `trans_env$cal_mantel`
## -----

t1$cal_mantel(use_measure = "bray")
t1$cal_mantel(partial_mantel = TRUE, use_measure = "bray")

## -----
## Method `trans_env$cal_cor`
## -----

t2 <- trans_diff$new(dataset = dataset, method = "rf", group = "Group", rf_taxa_level = "Genus")
t1 <- trans_env$new(dataset = dataset, add_data = env_data_16S[, 4:11])
t1$cal_cor(use_data = "other", p_adjust_method = "fdr", other_taxa = t2$res_diff$Taxa[1:40])

## -----
## Method `trans_env$plot_cor`
## -----

t1$plot_cor(heatmap = FALSE)

## -----
## Method `trans_env$plot_scatterfit`
## -----

t1$plot_scatterfit(x = 1, y = 2, type = "cor")
t1$plot_scatterfit(x = 1, y = 2, type = "lm", point_alpha = .3)
t1$plot_scatterfit(x = "pH", y = "TOC", type = "lm", group = "Group", line_se = FALSE)

```



```
t1$plot_scatterfit(x =
  dataset$beta_diversity$bray[rownames(t1$data_env), rownames(t1$data_env)], y = "pH")
```

---

trans\_func

*Create trans\_func object for functional analysis.*


---

## Description

This class is a wrapper for a series of functional analysis on species and communities, including the prokaryotes function identification based on Louca et al. (2016) <doi:10.1126/science.aaf4507> and Lim et al. (2020) <10.1038/s41597-020-0516-5>, or fungi function identification based on Nguyen et al. (2016) <10.1016/j.funeco.2015.06.006> and Polme et al. (2020) <doi:10.1007/s13225-020-00466-2>; functional redundancy calculation and metabolic pathway abundance prediction Abhauer et al. (2015) <10.1093/bioinformatics/btv287>.

## Active bindings

func\_group\_list store and show the function group list

## Methods

### Public methods:

- `trans_func$new()`
- `trans_func$cal_spe_func()`
- `trans_func$cal_spe_func_perc()`
- `trans_func$show_prok_func()`
- `trans_func$plot_spe_func_perc()`
- `trans_func$cal_tax4fun()`
- `trans_func$cal_tax4fun2()`
- `trans_func$cal_tax4fun2_FRI()`
- `trans_func$print()`
- `trans_func$clone()`

**Method** `new()`: Create the `trans_func` object. This function can identify the data type for Prokaryotes or Fungi automatically.

*Usage:*

```
trans_func$new(dataset = NULL)
```

*Arguments:*

`dataset` the object of `microtable` Class.

*Returns:* `for_what` : "prok" or "fungi" or NA, "prok" represent prokaryotes. "fungi" represent fungi. NA stand for not identified according to the Kingdom information, at this time, if you want to use the functions to identify species traits, you need provide "prok" or "fungi" manually, e.g. `dataset$for_what <- "prok"`.

*Examples:*

```
data(dataset)
t1 <- trans_func$new(dataset = dataset)
```

**Method** `cal_spe_func()`: Confirm traits of each feature by matching the taxonomic assignments to the functional database.

*Usage:*

```
trans_func$cal_spe_func(
  prok_database = c("FAPROTAX", "NJC19")[1],
  fungi_database = c("FUNGuild", "FungalTraits")[1]
)
```

*Arguments:*

`prok_database` default "FAPROTAX"; "FAPROTAX" or "NJC19"; select a prokaryotic trait database; see the details:

**'FAPROTAX'** FAPROTAX v1.2.4 Reference: Louca et al. (2016). Decoupling function and taxonomy in the global ocean microbiome. *Science*, 353(6305), 1272. <doi:10.1126/science.aaf4507>

**'NJC19'** NJC19: Lim et al. (2020). Large-scale metabolic interaction network of the mouse and human gut microbiota. *Scientific Data*, 7(1). <10.1038/s41597-020-0516-5>

`fungi_database` default "FUNGuild"; "FUNGuild" or "FungalTraits"; select a fungal trait database; see the details:

**'FUNGuild'** Nguyen et al. (2016) FUNGuild: An open annotation tool for parsing fungal community datasets by ecological guild. *Fungal Ecology*, 20(1), 241-248, <doi:10.1016/j.funeco.2015.06.006>

**'FungalTraits'** Polme et al. FungalTraits: a user-friendly traits database of fungi and fungus-like stramenopiles. *Fungal Diversity* 105, 1-16 (2020). <doi:10.1007/s13225-020-00466-2>

*Returns:* `res_spe_func` stored in object.

*Examples:*

```
\donttest{
t1$cal_spe_func(prok_database = "FAPROTAX")
t1$cal_spe_func(fungi_database = "FungalTraits")
}
```

**Method** `cal_spe_func_perc()`: Calculating the percentages of species with specific trait in communities. The percentages of the taxa with specific trait can reflect the potential of the corresponding function in the community. So this method is a simple calculation of functional redundancy without the consideration of phylogenetic distance among taxa.

*Usage:*

```
trans_func$cal_spe_func_perc(abundance_weighted = FALSE)
```

*Arguments:*

`abundance_weighted` default FALSE; whether use abundance of taxa. If FALSE, calculate the functional population percentage. If TRUE, calculate the functional individual percentage.

*Returns:* `res_spe_func_perc` stored in the object.

*Examples:*

```
\donttest{
t1$cal_spe_func_perc(abundance_weighted = TRUE)
}
```

**Method** show\_prok\_func(): Show the annotation information for a function of prokaryotes from FAPROTAX database.

*Usage:*

```
trans_func$show_prok_func(use_func = NULL)
```

*Arguments:*

use\_func default NULL; the function name.

*Returns:* None.

*Examples:*

```
\donttest{
t1$show_prok_func(use_func = "methanotrophy")
}
```

**Method** plot\_spe\_func\_perc(): Plot the percentages of species with specific trait in communities or network modules.

*Usage:*

```
trans_func$plot_spe_func_perc(
  filter_func = NULL,
  use_group_list = TRUE,
  add_facet = TRUE,
  select_samples = NULL,
  color_gradient_low = "#00008B",
  color_gradient_high = "#9E0142"
)
```

*Arguments:*

filter\_func default NULL; a vector of function names used to show in the plot.

use\_group\_list default TRUE; If TRUE, use default group list; If user want to use personalized group list, please first set trans\_func\$func\_group\_list object with a list of group names and functions.

add\_facet default TRUE; whether use group names as the facets in the plot, see trans\_func\$func\_group\_list object.

select\_samples default NULL; character vector; select partial samples to show.

color\_gradient\_low default "#00008B"; the color used as the low end in the color gradient.

color\_gradient\_high default "#9E0142"; the color used as the high end in the color gradient.

*Returns:* ggplot2.

*Examples:*

```
\donttest{
t1$plot_spe_func_perc(use_group_list = TRUE)
}
```

**Method** `cal_tax4fun()`: Predict functional potential of communities using tax4fun. please cite: Tax4Fun: Predicting functional profiles from metagenomic 16S rRNA data. *Bioinformatics*, 31(17), 2882-2884, <doi:10.1093/bioinformatics/btv287>. Note that this function requires a standard prefix in taxonomic table with double underlines (e.g. `g__`).

*Usage:*

```
trans_func$cal_tax4fun(keep_tem = FALSE, folderReferenceData = NULL)
```

*Arguments:*

`keep_tem` default FALSE; whether keep the intermediate file, that is, the feature table in local place.

`folderReferenceData` default NULL; the folder, see <http://tax4fun.gobics.de/> and Tax4Fun function in Tax4Fun package.

*Returns:* tax4fun\_KO and tax4fun\_path in object.

**Method** `cal_tax4fun2()`: Predict functional potential of communities with Tax4Fun2 method. The function was adapted from the raw Tax4Fun2 package to make it compatible with the microtable object. Please cite: Tax4Fun2: prediction of habitat-specific functional profiles and functional redundancy based on 16S rRNA gene sequences. *Environmental Microbiome* 15, 11 (2020). <doi:10.1186/s40793-020-00358-7>

*Usage:*

```
trans_func$cal_tax4fun2(
  blast_tool_path = NULL,
  path_to_reference_data = "Tax4Fun2_ReferenceData_v2",
  path_to_temp_folder = NULL,
  database_mode = "Ref99NR",
  normalize_by_copy_number = T,
  min_identity_to_reference = 97,
  use_uproc = T,
  num_threads = 1,
  normalize_pathways = F
)
```

*Arguments:*

`blast_tool_path` default NULL; the folder path, e.g., ncbi-blast-2.5.0+/bin ; blast tools folder downloaded from "ftp://ftp.ncbi.nlm.nih.gov/blast/executables/blast+" ; e.g., ncbi-blast-2.5.0+-x64-win64.tar.gz for windows system; if `blast_tool_path` is NULL, search the tools in the environmental path variable.

`path_to_reference_data` default "Tax4Fun2\_ReferenceData\_v2"; the path that points to files used in the prediction; The directory must contain the Ref99NR or Ref100NR folder; download Ref99NR.zip from <https://cloudstor.aarnet.edu.au/plus/s/DkoZIyZpMNbrzSw/download> or Ref100NR.zip from <https://cloudstor.aarnet.edu.au/plus/s/jIByczak9ZAFUB4/download>.

`path_to_temp_folder` default NULL; The temporary folder to store the logfile, intermediate file and result files; if NULL, use the default temporary in the computer system.

`database_mode` default 'Ref99NR'; "Ref99NR" or "Ref100NR"; Ref99NR: 99% clustering reference database; Ref100NR: no clustering.

`normalize_by_copy_number` default TRUE; whether normalize the result by the 16S rRNA copy number in the genomes.

min\_identity\_to\_reference default 97; the identity threshold used for finding the nearest species.

use\_uproc default TRUE; whether use UProC to functionally annotate the genomes in the reference data.

num\_threads default 1; the threads used in the blastn.

normalize\_pathways default FALSE; Different to Tax4Fun, when converting from KEGG functions to KEGG pathways, Tax4Fun2 does not equally split KO gene abundances between pathways a functions is affiliated to. The full predicted abundance is affiliated to each pathway. Use TRUE to split the abundances (default is FALSE).

*Returns:* res\_tax4fun2\_KO and res\_tax4fun2\_pathway in object.

*Examples:*

```
\dontrun{
t1$cal_tax4fun2(blast_tool_path = "ncbi-blast-2.5.0+/bin",
  path_to_reference_data = "Tax4Fun2_ReferenceData_v2")
}
```

**Method** cal\_tax4fun2\_FRI(): Calculate (multi-) functional redundancy index (FRI) of prokaryotic community with Tax4Fun2 method. This function is used to calculating aFRI and rFRI use the intermediate files generated by the function cal\_tax4fun2(). please also cite: Tax4Fun2: prediction of habitat-specific functional profiles and functional redundancy based on 16S rRNA gene sequences. Environmental Microbiome 15, 11 (2020). <doi:10.1186/s40793-020-00358-7>

*Usage:*

```
trans_func$cal_tax4fun2_FRI()
```

*Returns:* res\_tax4fun2\_aFRI and res\_tax4fun2\_rFRI in object.

*Examples:*

```
\dontrun{
t1$cal_tax4fun2_FRI()
}
```

**Method** print(): Print the trans\_func object.

*Usage:*

```
trans_func$print()
```

**Method** clone(): The objects of this class are cloneable with this method.

*Usage:*

```
trans_func$clone(deep = FALSE)
```

*Arguments:*

deep Whether to make a deep clone.

## Examples

```
## -----
## Method `trans_func$new`
## -----
```

```

data(dataset)
t1 <- trans_func$new(dataset = dataset)

## -----
## Method `trans_func$cal_spe_func`
## -----

t1$cal_spe_func(prok_database = "FAPROTAX")
t1$cal_spe_func(fungi_database = "FungalTraits")

## -----
## Method `trans_func$cal_spe_func_perc`
## -----

t1$cal_spe_func_perc(abundance_weighted = TRUE)

## -----
## Method `trans_func$show_prok_func`
## -----

t1$show_prok_func(use_func = "methanotrophy")

## -----
## Method `trans_func$plot_spe_func_perc`
## -----

t1$plot_spe_func_perc(use_group_list = TRUE)

## -----
## Method `trans_func$cal_tax4fun2`
## -----

## Not run:
t1$cal_tax4fun2(blast_tool_path = "ncbi-blast-2.5.0+/bin",
  path_to_reference_data = "Tax4Fun2_ReferenceData_v2")

## End(Not run)

## -----
## Method `trans_func$cal_tax4fun2_FRI`
## -----

## Not run:
t1$cal_tax4fun2_FRI()

```

```
## End(Not run)
```

---

```
trans_network      Create trans_network object for co-occurrence network analysis.
```

---

## Description

This class is a wrapper for a series of network analysis methods, including the network construction approaches, network attributes analysis, eigengene analysis, network subsetting, node and edge properties extraction, network plotting, and other network operations.

## Methods

### Public methods:

- `trans_network$new()`
- `trans_network$cal_network()`
- `trans_network$cal_module()`
- `trans_network$save_network()`
- `trans_network$cal_network_attr()`
- `trans_network$cal_node_type()`
- `trans_network$get_node_table()`
- `trans_network$get_edge_table()`
- `trans_network$get_adjacency_matrix()`
- `trans_network$plot_network()`
- `trans_network$cal_eigen()`
- `trans_network$plot_taxa_roles()`
- `trans_network$subset_network()`
- `trans_network$cal_powerlaw()`
- `trans_network$trans_comm()`
- `trans_network$print()`
- `trans_network$clone()`

**Method** `new()`: This function is used to create the `trans_network` object, store the important intermediate data and calculate correlations if `cal_cor` parameter is selected.

### Usage:

```
trans_network$new(
  dataset = NULL,
  cor_method = c("pearson", "spearman", "kendall")[1],
  cal_cor = c("base", "WGCNA", "SparCC", NA)[1],
  taxa_level = "OTU",
  filter_thres = 0,
  nThreads = 1,
  SparCC_simu_num = 100,
```

```

    env_cols = NULL,
    add_data = NULL
  )

```

*Arguments:*

`dataset` the object of `microtable` Class.

`cor_method` default "pearson"; "pearson", "spearman" or "kendall"; correlation algorithm, only use for correlation-based network.

`cal_cor` default "base"; "base", "WGCNA", "SparCC" or NA; correlation method; NA represents no correlation calculation, used for non-correlation based network, such as SpiecEasi and FlashWeave methods.

`taxa_level` default "OTU"; taxonomic rank; 'OTU' represents using feature table directly; other available options should be one of the colnames of `microtable$tax_table`.

`filter_thres` default 0; the relative abundance threshold.

`nThreads` default 1; the thread number used for "WGCNA" and SparCC.

`SparCC_simu_num` default 100; SparCC simulation number for bootstrap.

`env_cols` default NULL; numeric or character vector to select the column names of environmental data in `dataset$sample_table`; the environmental data can be used in the correlation network (as the nodes) or FlashWeave network.

`add_data` default NULL; provide environmental table additionally instead of `env_cols` parameter; rownames must be sample names.

*Returns:* `res_cor_p` list; include the correlation matrix and p value matrix.

*Examples:*

```

\donttest{
data(dataset)
# for correlation network
t1 <- trans_network$new(dataset = dataset, cal_cor = "base",
taxa_level = "OTU", filter_thres = 0.0001)
# for other network
t1 <- trans_network$new(dataset = dataset, cal_cor = NA)
}

```

**Method** `cal_network()`: Calculate network based on the correlation method or SpiecEasi package or julia FlashWeave package or beamStatic package.

*Usage:*

```

trans_network$cal_network(
  network_method = c("COR", "SpiecEasi", "FlashWeave", "beamStatic")[1],
  COR_p_thres = 0.01,
  COR_p_adjust = "fdr",
  COR_weight = TRUE,
  COR_cut = 0.6,
  COR_optimization = FALSE,
  COR_optimization_low_high = c(0.4, 0.8),
  SpiecEasi_method = "mb",
  FlashWeave_tempdir = NULL,
  FlashWeave_meta_data = FALSE,

```



```
FlashWeave_other_para = "alpha=0.01,sensitive=true,heterogeneous=true",
beemStatic_t_strength = 0.001,
beemStatic_t_stab = 0.8,
add_taxa_name = "Phylum",
username_rawtaxa_when_taxalevel_notOTU = FALSE,
...
)
```

#### Arguments:

network\_method default "COR"; "COR", "SpiecEasi", "FlashWeave" or "beemStatic"; The option details:

**'COR'** correlation-based network; use the correlation and p value matrixes in object\$res\_cor\_p returned from trans\_network\$new; See Deng et al. (2012) <doi:10.1186/1471-2105-13-113> for other details

**'SpiecEasi'** SpiecEasi network; relies on algorithms for sparse neighborhood and inverse covariance selection; belong to the category of conditional dependence and graphical models; see <https://github.com/zdk123/SpiecEasi> for installing the R package; see Kurtz et al. (2015) <doi:10.1371/journal.pcbi.1004226> for the algorithm details

**'FlashWeave'** FlashWeave network; Local-to-global learning framework; belong to the category of conditional dependence and graphical models; good performance on heterogeneous datasets to find direct associations among taxa; see <https://github.com/meringlab/FlashWeave.jl> for installing julia language and FlashWeave package; julia must be in the computer system env path, otherwise the program can not find julia; see Tackmann et al. (2019) <doi:10.1016/j.cels.2019.08.002> for the algorithm details

**'beemStatic'** beemStatic network; extend generalized Lotka-Volterra model to cases of cross-sectional datasets to infer interaction among taxa based on expectation-maximization algorithm; see <https://github.com/CSB5/BEEM-static> for installing the R package; see Li et al. (2021) <doi:10.1371/journal.pcbi.1009343> for algorithm details

COR\_p\_thres default 0.01; the p value threshold for the correlation-based network.

COR\_p\_adjust default "fdr"; p value adjustment method, see method of p.adjust function for available options.

COR\_weight default TRUE; whether use correlation coefficient as the weight of edges; FALSE represents weight = 1 for all edges.

COR\_cut default 0.6; correlation coefficient threshold for the correlation network.

COR\_optimization default FALSE; whether use random matrix theory (RMT) based method to determine the correlation coefficient; see <https://doi.org/10.1186/1471-2105-13-113>

COR\_optimization\_low\_high default c(0.4, 0.8); the low and high value threshold used for the RMT optimization; only useful when COR\_optimization = TRUE.

SpiecEasi\_method default "mb"; either 'glasso' or 'mb'; see spiec.easi function in package SpiecEasi and <https://github.com/zdk123/SpiecEasi>.

FlashWeave\_tempdir default NULL; The temporary directory used to save the temporary files for running FlashWeave; If not assigned, use the system user temp.

FlashWeave\_meta\_data default FALSE; whether use env data for the optimization, If TRUE, the function automatically find the object\$env\_data in the object and generate a file for meta\_data\_path parameter of FlashWeave.

FlashWeave\_other\_para default "alpha=0.01,sensitive=true,heterogeneous=true"; the parameters used for FlashWeave; user can change the parameters or add more according to Flash-

Weave help document; An exception is meta\_data\_path parameter as it is generated based on the data inside the object, see FlashWeave\_meta\_data parameter for the description.

beemStatic\_t\_strength default 0.001; for network\_method = "beemStatic"; the threshold used to limit the number of interactions (strength); same with the t\_strength parameter in showInteraction function of beemStatic package.

beemStatic\_t\_stab default 0.8; for network\_method = "beemStatic"; the threshold used to limit the number of interactions (stability); same with the t\_stab parameter in showInteraction function of beemStatic package.

add\_taxa\_name default "Phylum"; one or more taxonomic rank name; used to add taxonomic rank name to network node properties.

username\_rawtaxa\_when\_taxalevel\_notOTU default FALSE; whether replace the name of nodes using the taxonomic information.

... parameters pass to spiec.easi function of SpiecEasi package when network\_method = "SpiecEasi" or func.EM function of beemStatic package when network\_method = "beemStatic".

*Returns:* res\_network stored in object.

*Examples:*

```
\donttest{
# for correlation network
t1 <- trans_network$new(dataset = dataset, cal_cor = "base",
  taxa_level = "OTU", filter_thres = 0.0001)
t1$cal_network(COR_p_thres = 0.01, COR_cut = 0.6)
t1 <- trans_network$new(dataset = dataset, cal_cor = NA, filter_thres = 0.0001)
t1$cal_network(network_method = "SpiecEasi")
t1 <- trans_network$new(dataset = dataset, cal_cor = NA, filter_thres = 0.0001)
t1$cal_network(network_method = "FlashWeave")
}
```

**Method** cal\_module(): Calculate network modules and add module names to the network node properties.

*Usage:*

```
trans_network$cal_module(
  method = "cluster_fast_greedy",
  module_name_prefix = "M"
)
```

*Arguments:*

method default "cluster\_fast\_greedy"; the method used to find the optimal community structure of a graph; the following are available functions (options) from igraph package: "cluster\_fast\_greedy", "cluster\_optimal", "cluster\_edge\_betweenness", "cluster\_infomap", "cluster\_label\_prop", "cluster\_leading\_eigen", "cluster\_louvain", "cluster\_spinglass", "cluster\_walktrap". For the details of these functions, see the help document, such as help(cluster\_fast\_greedy); Note that the default "cluster\_fast\_greedy" method can only be used for undirected network. If the user selects network\_method = "beemStatic" in cal\_network function or provides other directed network, please use cluster\_optimal or others for the modules identification.

module\_name\_prefix default "M"; the prefix of module names; module names are made of the module\_name\_prefix and numbers; numbers are assigned according to the sorting result of node numbers in modules with decreasing trend.

*Returns:* res\_network with modules, stored in object.

*Examples:*

```
\donttest{
t1 <- trans_network$new(dataset = dataset, cal_cor = "base",
taxa_level = "OTU", filter_thres = 0.0001)
t1$cal_network(COR_p_thres = 0.01, COR_cut = 0.6)
t1$cal_module(method = "cluster_fast_greedy")
}
```

**Method** save\_network(): Save network as gexf style, which can be opened by Gephi (<https://gephi.org/>).

*Usage:*

```
trans_network$save_network(filepath = "network.gexf")
```

*Arguments:*

filepath default "network.gexf"; file path to save the network.

*Returns:* None.

*Examples:*

```
\dontrun{
t1$save_network(filepath = "network.gexf")
}
```

**Method** cal\_network\_attr(): Calculate network properties.

*Usage:*

```
trans_network$cal_network_attr()
```

*Returns:* res\_network\_attr stored in object.

*Examples:*

```
\donttest{
t1$cal_network_attr()
}
```

**Method** cal\_node\_type(): Calculate node properties. This function will be deprecated in the next release! Please use get\_node\_table function!

*Usage:*

```
trans_network$cal_node_type()
```

*Returns:* see the Return part in function get\_node\_table.

**Method** get\_node\_table(): Get the node property table. The properties may include the node names, modules allocation, degree, betweenness, abundance, taxonomy, within-module connectivity and among-module connectivity <doi:10.1016/j.geoderma.2022.115866>.

Authors: Chi Liu, Umer Zeeshan Ijaz

*Usage:*

```
trans_network$get_node_table(node_roles = TRUE)
```

*Arguments:*

node\_roles default TRUE; whether calculate node roles, i.e. Module hubs, Network hubs, Connectors and Peripherals <doi:10.1016/j.geoderma.2022.115866>.

*Returns:* res\_node\_table in object; Abundance expressed as a percentage; z represents within-module connectivity; p represents among-module connectivity.

*Examples:*

```
\donttest{
t1$get_node_table(node_roles = TRUE)
}
```

**Method** get\_edge\_table(): Get the edge property table, including connected nodes, label and weight.

*Usage:*

```
trans_network$get_edge_table()
```

*Returns:* res\_edge\_table in object.

*Examples:*

```
\donttest{
t1$get_edge_table()
}
```

**Method** get\_adjacency\_matrix(): Get the adjacency matrix from the network graph.

*Usage:*

```
trans_network$get_adjacency_matrix(...)
```

*Arguments:*

... parameters passed to as\_adjacency\_matrix function of igraph package.

*Returns:* res\_adjacency\_matrix in object.

*Examples:*

```
\donttest{
t1$get_adjacency_matrix(attr = "weight")
}
```

**Method** plot\_network(): Plot the network based on a series of methods from other packages, such as igraph, ggraph and networkD3. The networkD3 package provides dynamic network. It is especially useful for a glimpse of the whole network structure and finding the interested nodes and edges in a large network. In contrast, the igraph and ggraph methods are suitable for relatively small network.

*Usage:*

```
trans_network$plot_network(
  method = c("igraph", "ggraph", "networkD3")[1],
  node_label = "name",
  node_color = NULL,
  ggraph_layout = "fr",
  ggraph_node_size = 2,
  ggraph_text_color = NULL,
  ggraph_text_size = 3,
```

```

networkD3_node_legend = TRUE,
networkD3_zoom = TRUE,
...
)

```

*Arguments:*

method default "igraph"; The available options:

**'igraph'** call plot.igraph function in igraph package for a static network; see plot.igraph for the parameters

**'ggraph'** call ggraph function in ggraph package for a static network

**'networkD3'** use forceNetwork function in networkD3 package for a dynamic network; see forceNetwork function for the parameters

node\_label default "name"; node label shown in the plot for method = "ggraph" or method = "networkD3"; Please see the column names of object\$res\_node\_table, which is the returned table of function object\$get\_node\_table; User can select other column names in res\_node\_table.

node\_color default NULL; node color assignment for method = "ggraph" or method = "networkD3"; Select a column name of object\$res\_node\_table, such as "module".

ggraph\_layout default "fr"; for method = "ggraph"; see layout parameter of create\_layout function in ggraph package.

ggraph\_node\_size default 2; for method = "ggraph"; the node size.

ggraph\_text\_color default NULL; for method = "ggraph"; a column name of object\$res\_node\_table; User can select other column names or change the content of object\$res\_node\_table.

ggraph\_text\_size default 3; for method = "ggraph"; the node label text size.

networkD3\_node\_legend default TRUE; used for method = "networkD3"; logical value to enable node colour legends; Please see the legend parameter in networkD3::forceNetwork function.

networkD3\_zoom default TRUE; used for method = "networkD3"; logical value to enable (TRUE) or disable (FALSE) zooming; Please see the zoom parameter in networkD3::forceNetwork function.

... parameters passed to plot.igraph function when method = "igraph" or forceNetwork function when method = "networkD3".

*Returns:* network plot.

*Examples:*

```

\donttest{
t1$plot_network(method = "igraph", layout = layout_with_kk)
t1$plot_network(method = "ggraph", node_color = "module")
t1$plot_network(method = "networkD3", node_color = "module")
}

```

**Method cal\_eigen():** Calculate eigengenes of modules, i.e. the first principal component based on PCA analysis, and the percentage of variance <doi:10.1186/1471-2105-13-113>.

*Usage:*

```
trans_network$cal_eigen()
```

*Returns:* res\_eigen and res\_eigen\_expla in object.

*Examples:*

```
\donttest{
t1$cal_eigen()
}
```

**Method** `plot_taxa_roles()`: Plot the classification and importance of nodes, see `object$res_node_table` for the variable names used in the parameters.

*Usage:*

```
trans_network$plot_taxa_roles(
  use_type = c(1, 2)[1],
  roles_color_background = FALSE,
  roles_color_values = NULL,
  plot_module = FALSE,
  x_lim = c(0, 1),
  use_level = "Phylum",
  show_value = c("z", "p"),
  show_number = 1:10,
  plot_color = "Phylum",
  plot_shape = "taxa_roles",
  plot_size = "Abundance",
  color_values = RColorBrewer::brewer.pal(12, "Paired"),
  shape_values = c(16, 17, 7, 8, 15, 18, 11, 10, 12, 13, 9, 3, 4, 0, 1, 2, 14),
  ...
)
```

*Arguments:*

`use_type` default 1; 1 or 2; 1 represents taxa roles area plot; 2 represents the layered plot with taxa as x axis.

`roles_color_background` default FALSE; for `use_type=1`; TRUE: use background colors for each area; FALSE: use classic point colors.

`roles_color_values` default NULL; for `use_type=1`; color palette for background or points.

`plot_module` default FALSE; for `use_type=1`; whether plot the modules information.

`x_lim` default `c(0, 1)`; for `use_type=1`; x axis range when `roles_color_background = FALSE`.

`use_level` default "Phylum"; for `use_type=2`; used taxonomic level in x axis.

`show_value` default `c("z", "p")`; for `use_type=2`; used variable in y axis.

`show_number` default 1:10; for `use_type=2`; showed number in x axis, sorting according to the nodes number.

`plot_color` default "Phylum"; for `use_type=2`; used variable for color.

`plot_shape` default "taxa\_roles"; for `use_type=2`; used variable for shape.

`plot_size` default "Abundance"; for `use_type=2`; used for point size; a fixed number (e.g. 5) is also available.

`color_values` default `RColorBrewer::brewer.pal(12, "Paired")`; for `use_type=2`; color vector

`shape_values` default `c(16, 17, 7, 8, 15, 18, 11, 10, 12, 13, 9, 3, 4, 0, 1, 2, 14)`; for `use_type=2`; shape vector, see `ggplot2` tutorial for the shape meaning.

... parameters pass to `geom_point`.

*Returns:* `ggplot`.

*Examples:*

```
\donttest{
t1$plot_taxa_roles(roles_color_background = FALSE)
}
```

**Method** subset\_network(): Subset of the network.

*Usage:*

```
trans_network$subset_network(node = NULL, edge = NULL, rm_single = TRUE)
```

*Arguments:*

node default NULL; provide the node names that you want to use in the sub-network.

edge default NULL; provide the edge name needed; must be one of "+" or "-".

rm\_single default TRUE; whether remove the nodes without any edge in the sub-network.

*Returns:* a new network

*Examples:*

```
\donttest{
t1$subset_network(node = t1$res_node_table %>% base::subset(module == "M1") %>%
  rownames, rm_single = TRUE)
# return a sub network that contains all nodes of module M1
}
```

**Method** cal\_powerlaw(): Fit degrees to a power law distribution. First, perform a bootstrapping hypothesis test to determine whether degrees follow a power law distribution. If the distribution follows power law, then fit degrees to power law distribution and return the parameters.

*Usage:*

```
trans_network$cal_powerlaw(...)
```

*Arguments:*

... parameters pass to fit\_power\_law function in igraph package.

*Returns:* res\_powerlaw\_p and res\_powerlaw\_fit; see bootstrap\_p function in powerLaw package for the bootstrapping p value details; see fit\_power\_law function in igraph package for the power law fit return details.

*Examples:*

```
\donttest{
t1$cal_powerlaw()
}
```

**Method** trans\_comm(): Transform classified features to community-like microtable object for further analysis, such as module-taxa table.

*Usage:*

```
trans_network$trans_comm(use_col = "module", abundance = TRUE)
```

*Arguments:*

use\_col default "module"; which column to use as the 'community'; must be one of the name of res\_node\_table from function get\_node\_table.

abundance default TRUE; whether sum abundance of taxa. TRUE: sum the abundance for a taxon across all samples; FALSE: sum the frequency for a taxon across all samples.

*Returns:* a new `microtable` class.

*Examples:*

```
\donttest{
t2 <- t1$trans_comm(use_col = "module")
}
```

**Method** `print()`: Print the `trans_network` object.

*Usage:*

```
trans_network$print()
```

**Method** `clone()`: The objects of this class are cloneable with this method.

*Usage:*

```
trans_network$clone(deep = FALSE)
```

*Arguments:*

`deep` Whether to make a deep clone.

## Examples

```
## -----
## Method `trans_network$new`
## -----

data(dataset)
# for correlation network
t1 <- trans_network$new(dataset = dataset, cal_cor = "base",
taxa_level = "OTU", filter_thres = 0.0001)
# for other network
t1 <- trans_network$new(dataset = dataset, cal_cor = NA)

## -----
## Method `trans_network$cal_network`
## -----

# for correlation network
t1 <- trans_network$new(dataset = dataset, cal_cor = "base",
taxa_level = "OTU", filter_thres = 0.0001)
t1$cal_network(COR_p_thres = 0.01, COR_cut = 0.6)
t1 <- trans_network$new(dataset = dataset, cal_cor = NA, filter_thres = 0.0001)
t1$cal_network(network_method = "SpiecEasi")
t1 <- trans_network$new(dataset = dataset, cal_cor = NA, filter_thres = 0.0001)
t1$cal_network(network_method = "FlashWeave")

## -----
## Method `trans_network$cal_module`
## -----
```



```
t1 <- trans_network$new(dataset = dataset, cal_cor = "base",
taxa_level = "OTU", filter_thres = 0.0001)
t1$cal_network(COR_p_thres = 0.01, COR_cut = 0.6)
t1$cal_module(method = "cluster_fast_greedy")

## -----
## Method `trans_network$save_network`
## -----

## Not run:
t1$save_network(filepath = "network.gexf")

## End(Not run)

## -----
## Method `trans_network$cal_network_attr`
## -----

t1$cal_network_attr()

## -----
## Method `trans_network$get_node_table`
## -----

t1$get_node_table(node_roles = TRUE)

## -----
## Method `trans_network$get_edge_table`
## -----

t1$get_edge_table()

## -----
## Method `trans_network$get_adjacency_matrix`
## -----

t1$get_adjacency_matrix(attr = "weight")

## -----
## Method `trans_network$plot_network`
## -----
```

```

t1$plot_network(method = "igraph", layout = layout_with_kk)
t1$plot_network(method = "ggraph", node_color = "module")
t1$plot_network(method = "networkD3", node_color = "module")

## -----
## Method `trans_network$scal_eigen`
## -----

t1$scal_eigen()

## -----
## Method `trans_network$plot_taxa_roles`
## -----

t1$plot_taxa_roles(roles_color_background = FALSE)

## -----
## Method `trans_network$subset_network`
## -----

t1$subset_network(node = t1$res_node_table %>% base::subset(module == "M1") %>%
  rownames, rm_single = TRUE)
# return a sub network that contains all nodes of module M1

## -----
## Method `trans_network$scal_powerlaw`
## -----

t1$scal_powerlaw()

## -----
## Method `trans_network$trans_comm`
## -----

t2 <- t1$trans_comm(use_col = "module")

```

---

trans\_nullmodel

---

*Create trans\_nullmodel object for phylogeny- and taxonomy-based null model analysis.*


---

## Description

This class is a wrapper for a series of null model related approaches, including the mantel correlogram analysis of phylogenetic signal, beta nearest taxon index (betaNTI), beta net relatedness index (betaNRI), NTI, NRI and RCbray calculations; See Stegen et al. (2013) <10.1038/ismej.2013.93> and Liu et al. (2017) <doi:10.1038/s41598-017-17736-w> for the algorithms and applications.

## Methods

### Public methods:

- `trans_nullmodel$new()`
- `trans_nullmodel$scal_mantel_corr()`
- `trans_nullmodel$plot_mantel_corr()`
- `trans_nullmodel$scal_betampd()`
- `trans_nullmodel$scal_betamntd()`
- `trans_nullmodel$scal_ses_betampd()`
- `trans_nullmodel$scal_ses_betamntd()`
- `trans_nullmodel$scal_rcbray()`
- `trans_nullmodel$scal_process()`
- `trans_nullmodel$scal_NRI()`
- `trans_nullmodel$scal_NTI()`
- `trans_nullmodel$scal_Cscore()`
- `trans_nullmodel$scal_tNST()`
- `trans_nullmodel$scal_tNST_test()`
- `trans_nullmodel$clone()`

### Method `new()`:

#### *Usage:*

```
trans_nullmodel$new(
  dataset = NULL,
  filter_thres = 0,
  taxa_number = NULL,
  group = NULL,
  select_group = NULL,
  env_cols = NULL,
  add_data = NULL,
  complete_na = FALSE
)
```

#### *Arguments:*

`dataset` the object of `microtable` Class.

`filter_thres` default 0; the relative abundance threshold.

`taxa_number` default NULL; how many taxa the user want to keep, if provided, `filter_thres` parameter will be forcible invalid.

`group` default NULL; which group column name in `sample_table` is selected.

`select_group` default NULL; the group name, used following the group to filter samples.

env\_cols default NULL; number or name vector to select the environmental data in dataset\$sample\_table.  
 add\_data default NULL; provide environmental data table additionally.  
 complete\_na default FALSE; whether fill the NA in environmental data based on the method  
 in mice package.

*Returns:* data\_comm and data\_tree in object.

*Examples:*

```
data(dataset)
data(env_data_16S)
t1 <- trans_nullmodel$new(dataset, filter_thres = 0.0005, add_data = env_data_16S)
```

**Method** cal\_mantel\_corr(): Calculate mantel correlogram.

*Usage:*

```
trans_nullmodel$cal_mantel_corr(
  use_env = NULL,
  break.pts = seq(0, 1, 0.02),
  cutoff = FALSE,
  ...
)
```

*Arguments:*

use\_env default NULL; numeric or character vector to select env\_data; if provide multiple variables or NULL, use PCA (principal component analysis) to reduce dimensionality.  
 break.pts default seq(0, 1, 0.02); see break.pts parameter in [mantel.correlog](#) of vegan package.  
 cutoff default FALSE; see cutoff parameter in [mantel.correlog](#).  
 ... parameters pass to [mantel.correlog](#)

*Returns:* res\_mantel\_corr in object.

*Examples:*

```
\donttest{
t1$cal_mantel_corr(use_env = "pH")
}
```

**Method** plot\_mantel\_corr(): Plot mantel correlogram.

*Usage:*

```
trans_nullmodel$plot_mantel_corr(point_shape = 22, point_size = 3)
```

*Arguments:*

point\_shape default 22; the number for selecting point shape type; see ggplot2 manual for the number meaning.  
 point\_size default 3; the point size.

*Returns:* ggplot.

*Examples:*

```
\donttest{
t1$plot_mantel_corr()
}
```

**Method** `cal_betampd()`: Calculate betaMPD (mean pairwise distance). Same with `comdist` in `picante` package, but faster.

*Usage:*

```
trans_nullmodel$cal_betampd(abundance.weighted = TRUE)
```

*Arguments:*

`abundance.weighted` default TRUE; whether use abundance-weighted method.

*Returns:* `res_betampd` in object.

*Examples:*

```
\donttest{
t1$cal_betampd(abundance.weighted = TRUE)
}
```

**Method** `cal_betamntd()`: Calculate betaMNTD (mean nearest taxon distance). Same with `comdistnt` in `picante` package, but faster.

*Usage:*

```
trans_nullmodel$cal_betamntd(
  abundance.weighted = TRUE,
  exclude.conspecifics = FALSE,
  use_iCAMP = FALSE,
  use_iCAMP_force = TRUE,
  iCAMP_tempdir = NULL,
  ...
)
```

*Arguments:*

`abundance.weighted` default TRUE; whether use abundance-weighted method.

`exclude.conspecifics` default FALSE; see `exclude.conspecifics` parameter in `comdistnt` function of `picante` package.

`use_iCAMP` default FALSE; whether use `bmntd.big` function of `iCAMP` package to calculate betaMNTD. This method can store the phylogenetic distance matrix on the disk to lower the memory spending and perform the calculation parallelly.

`use_iCAMP_force` default FALSE; whether use `bmntd.big` function of `iCAMP` package automatically when the feature number is large.

`iCAMP_tempdir` default NULL; the temporary directory used to place the large tree file; If NULL; use the system user tempdir.

... parameters pass to `iCAMP::pdist.big` function.

*Returns:* `res_betamntd` in object.

*Examples:*

```
\donttest{
t1$cal_betamntd(abundance.weighted = TRUE)
}
```

**Method** `cal_ses_betampd()`: Calculate standardized effect size of betaMPD, i.e. beta net relatedness index (betaNRI).

*Usage:*

```
trans_nullmodel$cal_ses_betampd(
  runs = 1000,
  null.model = c("taxa.labels", "richness", "frequency", "sample.pool",
    "phylogeny.pool", "independentswap", "trialswap")[1],
  abundance.weighted = TRUE,
  iterations = 1000
)
```

*Arguments:*

runs default 1000; simulation runs.

null.model default "taxa.labels"; The available options include "taxa.labels", "richness", "frequency", "sample.pool", "phylogeny.pool", "independentswap" and "trialswap"; see null.model parameter of ses.mntd function in picante package for the algorithm details.

abundance.weighted default TRUE; whether use weighted abundance.

iterations default 1000; iteration number for part null models to perform; see iterations parameter of picante::randomizeMatrix function.

*Returns:* res\_ses\_betampd in object.

*Examples:*

```
\donttest{
# run 50 times for the example; default 1000
t1$cal_ses_betampd(runs = 50, abundance.weighted = TRUE)
}
```

**Method** cal\_ses\_betamntd(): Calculate standardized effect size of betaMNTD, i.e. beta nearest taxon index (betaNTI).

*Usage:*

```
trans_nullmodel$cal_ses_betamntd(
  runs = 1000,
  null.model = c("taxa.labels", "richness", "frequency", "sample.pool",
    "phylogeny.pool", "independentswap", "trialswap")[1],
  abundance.weighted = TRUE,
  exclude.conspecifics = FALSE,
  use_iCAMP = FALSE,
  use_iCAMP_force = TRUE,
  iCAMP_tempdir = NULL,
  nworker = 2,
  iterations = 1000
)
```

*Arguments:*

runs default 1000; simulation number of null model.

null.model default "taxa.labels"; The available options include "taxa.labels", "richness", "frequency", "sample.pool", "phylogeny.pool", "independentswap" and "trialswap"; see null.model parameter of ses.mntd function in picante package for the algorithm details.

abundance.weighted default TRUE; whether use abundance-weighted method.

exclude.conspecifics default FALSE; see comdistnt in picante package.

use\_iCAMP default FALSE; whether use `bmntd.big` function of `iCAMP` package to calculate `betaMNTD`. This method can store the phylogenetic distance matrix on the disk to lower the memory spending and perform the calculation parallelly.

use\_iCAMP\_force default FALSE; whether to make `use_iCAMP` to be TRUE when the feature number is large.

iCAMP\_tempdir default NULL; the temporary directory used to place the large tree file; If NULL; use the system user tempdir.

nworker default 2; the CPU thread number.

iterations default 1000; iteration number for part null models to perform; see `iterations` parameter of `picante::randomizeMatrix` function.

*Returns:* `res_ses_betamntd` in object.

*Examples:*

```
\donttest{
# run 50 times for the example; default 1000
t1$cal_ses_betamntd(runs = 50, abundance.weighted = TRUE, exclude.conspecifics = FALSE)
}
```

**Method** `cal_rcbray()`: Calculate Bray–Curtis-based Raup–Crick (RCbray).

*Usage:*

```
trans_nullmodel$cal_rcbray(
  runs = 1000,
  verbose = TRUE,
  null.model = "independentswap"
)
```

*Arguments:*

`runs` default 1000; simulation runs.

`verbose` default TRUE; whether show the calculation process message.

`null.model` default "independentswap"; see more available options in `randomizeMatrix` function of `picante` package.

*Returns:* `res_rcbray` in object.

*Examples:*

```
\donttest{
# run 50 times for the example; default 1000
t1$cal_rcbray(runs = 50)
}
```

**Method** `cal_process()`: Infer the ecological processes according to `ses.betaMNTD` `ses.betaMPD` and `rcbray`.

*Usage:*

```
trans_nullmodel$cal_process(use_betamntd = TRUE)
```

*Arguments:*

`use_betamntd` default TRUE; whether use `ses.betaMNTD`; if false, use `ses.betaMPD`.

*Returns:* `res_rcbray` in object.

*Examples:*

```
\donttest{
t1$cal_process(use_betamtd = TRUE)
}
```

**Method** `cal_NRI()`: Calculates Nearest Relative Index (NRI), equivalent to -1 times the standardized effect size of MPD.

*Usage:*

```
trans_nullmodel$cal_NRI(
  null.model = "taxa.labels",
  abundance.weighted = FALSE,
  runs = 999,
  ...
)
```

*Arguments:*

`null.model` default "taxa.labels"; Null model to use; see `null.model` parameter in `ses.mpd` function of `picante` package for available options.

`abundance.weighted` default FALSE; Should mean nearest relative distances for each species be weighted by species abundance?

`runs` default 999; Number of randomizations.

... parameters pass to `ses.mpd` function in `picante` package.

*Returns:* `res_NRI` in object, equivalent to -1 times `ses.mpd`.

*Examples:*

```
\donttest{
t1$cal_NRI()
}
```

**Method** `cal_NTI()`: Calculates Nearest Taxon Index (NTI), equivalent to -1 times the standardized effect size of MNTD.

*Usage:*

```
trans_nullmodel$cal_NTI(
  null.model = "taxa.labels",
  abundance.weighted = FALSE,
  runs = 999,
  ...
)
```

*Arguments:*

`null.model` default "taxa.labels"; Null model to use; see `null.model` parameter in `ses.mntd` function of `picante` package for available options.

`abundance.weighted` default FALSE; Should mean nearest taxon distances for each species be weighted by species abundance?

`runs` default 999; Number of randomizations.

... parameters pass to `ses.mntd` function in `picante` package.

*Returns:* `res_NTI` in object, equivalent to -1 times `ses.mntd`.



*Examples:*

```
\donttest{
t1$cal_NTI(null.model = "taxa.labels", abundance.weighted = TRUE)
}
```

**Method** `cal_Cscore()`: Calculates the (normalised) mean number of checkerboard combinations (C-score) using C.score function in bipartite package.

*Usage:*

```
trans_nullmodel$cal_Cscore(by_group = NULL, ...)
```

*Arguments:*

`by_group` default NULL; one column name or number in `sample_table`; calculate C-score for different groups separately.

... parameters pass to C.score function in bipartite package.

*Returns:* results directly.

*Examples:*

```
\donttest{
t1$cal_Cscore()
}
```

**Method** `cal_tNST()`: Calculate normalized stochasticity ratio (NST) based on the tNST function of NST package.

*Usage:*

```
trans_nullmodel$cal_tNST(group, ...)
```

*Arguments:*

`group` a colname of `sample_table`; the function can select the data from `sample_table` to generate a one-column (n x 1) matrix and provide it to the group parameter of tNST function.

... parameters pass to tNST function of NST package; see the documents of tNST function for more details.

*Returns:* .

*Examples:*

```
\donttest{
t1$cal_tNST(group = "Group", dist.method = "bray", output.rand = TRUE, SES = TRUE)
}
```

**Method** `cal_tNST_test()`: Test the significance of NST difference between each pair of groups.

*Usage:*

```
trans_nullmodel$cal_tNST_test(method = "nst.boot", ...)
```

*Arguments:*

`method` default "nst.boot"; "nst.boot" or "nst.panova"; see NST::nst.boot function or NST::nst.panova function for the details.

... parameters pass to NST::nst.boot when `method = "nst.boot"` or NST::nst.panova when `method = "nst.panova"`

*Returns:* .

*Examples:*

```
\donttest{
t1$cal_tNST_test()
}
```

**Method** clone(): The objects of this class are cloneable with this method.

*Usage:*

```
trans_nullmodel$clone(deep = FALSE)
```

*Arguments:*

deep Whether to make a deep clone.

## Examples

```
## -----
## Method `trans_nullmodel$new`
## -----

data(dataset)
data(env_data_16S)
t1 <- trans_nullmodel$new(dataset, filter_thres = 0.0005, add_data = env_data_16S)

## -----
## Method `trans_nullmodel$cal_mantel_corr`
## -----

t1$cal_mantel_corr(use_env = "pH")

## -----
## Method `trans_nullmodel$plot_mantel_corr`
## -----

t1$plot_mantel_corr()

## -----
## Method `trans_nullmodel$cal_betampd`
## -----

t1$cal_betampd(abundance.weighted = TRUE)

## -----
## Method `trans_nullmodel$cal_betamntd`
## -----
```

```
t1$cal_betamntd(abundance.weighted = TRUE)

## -----
## Method `trans_nullmodel$cal_ses_betampd`
## -----

# run 50 times for the example; default 1000
t1$cal_ses_betampd(runs = 50, abundance.weighted = TRUE)

## -----
## Method `trans_nullmodel$cal_ses_betamntd`
## -----

# run 50 times for the example; default 1000
t1$cal_ses_betamntd(runs = 50, abundance.weighted = TRUE, exclude.conspecifics = FALSE)

## -----
## Method `trans_nullmodel$cal_rcbray`
## -----

# run 50 times for the example; default 1000
t1$cal_rcbray(runs = 50)

## -----
## Method `trans_nullmodel$cal_process`
## -----

t1$cal_process(use_betamntd = TRUE)

## -----
## Method `trans_nullmodel$cal_NRI`
## -----

t1$cal_NRI()

## -----
## Method `trans_nullmodel$cal_NTI`
## -----

t1$cal_NTI(null.model = "taxa.labels", abundance.weighted = TRUE)
```

```

## -----
## Method `trans_nullmodel$cal_Cscore`
## -----

t1$cal_Cscore()

## -----
## Method `trans_nullmodel$cal_tNST`
## -----

t1$cal_tNST(group = "Group", dist.method = "bray", output.rand = TRUE, SES = TRUE)

## -----
## Method `trans_nullmodel$cal_tNST_test`
## -----

t1$cal_tNST_test()

```

---

trans\_venn

*Create trans\_venn object.*


---

## Description

This class is a wrapper for a series of venn analysis related methods, including venn result, 2- to 5-way venn diagram, more than 5-way petal plot and venn result transformations based on David et al. (2012) <doi:10.1128/AEM.01459-12>.

## Methods

### Public methods:

- `trans_venn$new()`
- `trans_venn$plot_venn()`
- `trans_venn$trans_commm()`
- `trans_venn$print()`
- `trans_venn$clone()`

### Method `new()`:

*Usage:*

```
trans_venn$new(
  dataset = NULL,
  sample_names = NULL,
  ratio = NULL,
  add_abund_table = NULL
)
```

*Arguments:*

`dataset` the object of `microtable` Class.

`sample_names` default NULL; if provided, filter the samples.

`ratio` default NULL; NULL, "numratio" or "seqratio"; numratio: calculate number percentage; seqratio: calculate sequence percentage; NULL: no additional percentage.

`add_abund_table` default NULL; data.frame or matrix format; additional data provided instead of `dataset$otu_table`; Features must be rows.

*Returns:* `data_details` and `data_summary` stored in `trans_venn` object.

*Examples:*

```
\donttest{
data(dataset)
t1 <- dataset$merge_samples(use_group = "Group")
t1 <- trans_venn$new(dataset = t1, ratio = "numratio")
}
```

**Method** `plot_venn()`: Plot venn diagram.

*Usage:*

```
trans_venn$plot_venn(
  color_circle = RColorBrewer::brewer.pal(8, "Dark2"),
  fill_color = TRUE,
  text_size = 4.5,
  text_name_size = 6,
  text_name_position = NULL,
  alpha = 0.3,
  linesize = 1.1,
  petal_plot = FALSE,
  petal_color = "#BEAED4",
  petal_color_center = "#BEBADA",
  petal_a = 4,
  petal_r = 1,
  petal_use_lim = c(-12, 12),
  petal_center_size = 40,
  petal_move_xy = 4,
  petal_move_k = 2.3,
  petal_move_k_count = 1.3,
  petal_text_move = 40,
  other_text_show = NULL,
  other_text_position = c(2, 2),
  other_text_size = 5
)
```

*Arguments:*

color\_circle default RColorBrewer::brewer.pal(8, "Dark2"); color palette  
 fill\_color default TRUE; whether fill the area color  
 text\_size default 4.5; text size in plot  
 text\_name\_size default 6; name size in plot  
 text\_name\_position default NULL; name position in plot  
 alpha default .3; alpha for transparency  
 linesize default 1.1; cycle line size  
 petal\_plot default FALSE; whether use petal plot.  
 petal\_color default "#BEAED4"; color of the petals.  
 petal\_color\_center default "#BEBADA"; color of the center in the petal plot.  
 petal\_a default 4; the length of the ellipse  
 petal\_r default 1; scaling up the size of the ellipse  
 petal\_use\_lim default c(-12, 12); the width of the plot  
 petal\_center\_size default 40; petal center circle size  
 petal\_move\_xy default 4; the distance of text to circle  
 petal\_move\_k default 2.3; the distance of title to circle  
 petal\_move\_k\_count default 1.3; the distance of data text to circle  
 petal\_text\_move default 40; the distance between two data text  
 other\_text\_show default NULL; other characters used to show in the plot  
 other\_text\_position default c(1, 1); the text position for text in other\_text\_show  
 other\_text\_size default 5; the text size for text in other\_text\_show

*Returns:* ggplot.

*Examples:*

```
\donttest{
t1$plot_venn()
}
```

**Method** trans\_comm(): Transform venn result to community-like microtable object for further composition analysis.

*Usage:*

```
trans_venn$trans_comm(use_frequency = TRUE)
```

*Arguments:*

use\_frequency default TRUE; whether only use OTUs occurrence frequency, i.e. presence/absence data; if FALSE, use abundance data.

*Returns:* a new [microtable](#) class.

*Examples:*

```
\donttest{
t2 <- t1$trans_comm(use_frequency = TRUE)
}
```

**Method** print(): Print the trans\_venn object.

*Usage:*

```
trans_venn$print()
```

**Method** clone(): The objects of this class are cloneable with this method.

*Usage:*

```
trans_venn$clone(deep = FALSE)
```

*Arguments:*

deep Whether to make a deep clone.

## Examples

```
## -----  
## Method `trans_venn$new`  
## -----  
  
data(dataset)  
t1 <- dataset$merge_samples(use_group = "Group")  
t1 <- trans_venn$new(dataset = t1, ratio = "numratio")  
  
## -----  
## Method `trans_venn$plot_venn`  
## -----  
  
t1$plot_venn()  
  
## -----  
## Method `trans_venn$trans_comm`  
## -----  
  
t2 <- t1$trans_comm(use_frequency = TRUE)
```

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