Package 'RAM'

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Description Characterizing environmental microbiota diversity using amplicon-based next generation sequencing (NGS) data. Functions are developed to manipulate operational taxonomic unit (OTU) table, perform descriptive and inferential statistics, and generate publicatio quality plots.	n-
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Copyright Government of Canada	
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R topics documented:	
RAM-package	
1	

assist.ordination	
col.splitup	9
combine.OTU	11
core.OTU	12
core.OTU.rank	13
core.Taxa	14
correlation	15
data.clust	
data.revamp	
data.subset	
dissim	
dissim.heatmap	
dissim.plot	
diversity.indices	
envis.NB	
factor.abundance	
filter.META	
filter.OTU	
filter.Taxa	
fread.meta	
fread.OTU	
get.rank	
group.abund.Taxa	
group.abundance	
group.abundance.meta	
group.diversity	
group.heatmap	
group.heatmap.simple	
group.indicators	
group.OTU	
group.rich	
group.spatial	
group.spec	
group.Taxa.bar	
group.Taxa.box	54
group.temporal	56
group.venn	
TS1/ITS2	
LCA.OTU	
ocation.formatting	
match.data	
meta	
META.clust	
network_data	
OTU.diversity	
OTU.ord	
OTU.rarefy	
•	
OTU.recap	09

	pcoa.plot	7 0
	percent.classified	73
	phylog_taxonomy	74
	phylo_taxonomy	75
	RAM.dates	77
	RAM.factors	77
	RAM.input.formatting	78
	RAM.pal	<mark>79</mark>
	RAM.plotting	<mark>79</mark>
	RAM.rank.formatting	81
	read.meta	81
	read.OTU	82
	reset.META	83
	sample.locations	84
	sample.map	85
	sample.sites	87
	seq_var	88
	shared.OTU	90
		91
		92
		94
	tax.split	95
		96
	-661	99
	top.groups.prot	99
	transpose.LCA	01
	transpose.OTU	
	valid.OTU	
	valid.taxonomy	
	write.data	04
[]	1	06
Index	1	06
RAM-p	ackage Analysis of Amplicon-Based Metagenomic Data	

Description

The RAM package provides a series of functions to make amplicon based metagenomic analysis more accessible. The package is designed especially for those who have little or no experience with R. This package calls heavily upon other packages (such as vegan and ggplot2), but the functions in this package either extend their functionality, or increase the ease-of-use.

4 RAM-package

Details

Package: RAM
Type: Package
Version: 1.2.0
Date: 2014-12-10

License: MIT License, Copyright (c) 2014 Government of Canada

Load data from .csv-formatted OTU files with read.OTU or fread.OTU, then process the data with other commands. Type the command library(help = RAM) for a full index of all help topics, or ls("package:RAM") to get a list of all functions in the package.

Type data(ITS1, ITS2, meta) to load sample data sets of RAM, which include the following data of 16 samples: 1) ITS1: OTU table of fungal internal transcribed spacer region 3) ITS2: OTU table of fungal internal transcribed spacer region 2 3) meta: associated metadata 4) alignment for seq_var

Type citation("RAM") for how to cite this package.

This pacakge contains information licensed under the Open Government Licence - Canada. See group.spatial for further details.

Author(s)

Wen Chen and Joshua Simpson.

Maintainer: Wen Chen <wen.chen@agr.gc.ca>

See Also

```
vegan, ggplot2
```

```
## Not run:
# load data from your own files...
otu1 <- fread.OTU("path/to/OTU/table")
otu2 <- read.OTU("path/to/OTU/table")
meta1 <- fread.meta("path/to/meta/table")
meta2 <- read.meta("path/to/meta/table")

# ...or use the included sample data
data(ITS1, ITS2, meta)
data <- list(ITS1=ITS1, ITS2=ITS2)
dissim.heatmap(ITS1, meta, row.factor=c(City="City"))
dissim.alleig.plot(data)
data(alignment)

# type library(help = RAM) to get a full listing of help documents
## End(Not run)</pre>
```

alignment 5

alignment

Sample Alignment

Description

This is an alignment for seq_var package.

Usage

```
data(alignment)
```

Format

An alignment with sequence ID being formatted as follows: genus_name:accession:genus:species:strain_info/seqBegin-seqEnd. The location of each party can be rearranged, and the separator can be other speciall characters, such as "I".

Source

Wen Chen

Examples

```
data(alignment)
str(alignment)
alignment
```

assist.ado

Perform ADONIS Analysis for OTU Tables Or Taxonomic Abundance Matrix

Description

This function simplifies ADONIS analysis by abstracting away some of the complexity and returning a list of useful measures.

Usage

6 assist.ado

Arguments

data	an OTU table or a taxonomy abundance matrix.
is.OTU	logical. If the data is an OTU table, set is . OTU TRUE; otherwise, set it as FALSE.
meta	the metadata table to be used (must have same samples as data.
ranks	optional. If ranks is not provided, will test for OTUs, otherwise, will test on taxa at defined ranks. If data is a taxonomic abundance matrix, ranks can be NULL
data.trans	optional. Transform the data using method from the function decostand
dist	optional. the name of any method used in $vegdist$ to calculate pairwise distances. See also adonis and $vegdist$.
meta.strata	optional. A metadata variable within which to constrain permutations. See also ${\tt adonis}$
perm	a numeric number of replicate permutations used for the hypothesis test used in adonis. $ \\$
top	optional. Select the top taxa or OTUs. See also data.revamp
mode	a character vector, one of "percent" or "number". If number, then top groups will be selected based on total sequence count. If percent, then top groups will be selected based on relative abundance. See also data.revamp

Value

This function returns a list containing outputs from adonis test.

- If is.OTU is TRUE and ranks is not given: the output is a length one list named LCA_OTU.
- If is.OTU is TRUE and ranks is given: the output is a list with a length same as the number of taxonomic ranks provided. Each member of the list is named after the rank it processed at.
- If is.OTU is FALSE, the output is a length one list named Taxa.

Author(s)

Wen Chen.

See Also

adonis

assist.NB 7

assist.NB

Negative Binomial Test For OTUID or Taxon

Description

This function does negative binomial test for a given otuID or taxon

Usage

Arguments

data	an ecology data set to be analyzed.
meta	the metadata table to be analyzed.
is.OTU	logical. If an OTU table was provided, is . OTU should be set as TRUE; otherwise, it should be set as FALSE.
rank	optional. If no rank was provided, the data will be used as it is, if rank is provided, if data is an OTU table, it will be converted to taxonomic abundance matrix at the given rank, no change will be made for a data that has already been a taxonomic abundance matrix. See also tax.abund and data.revamp
meta.factors	optional. If provided, will only test the model on selected metadata variables; otherwise, will test all variables in the metadata table.
anov.fac	optional. Whether or not to do anova test on a metadata variable.
taxon	a length one charactor. Can either be an otuID or a taxon name.

Value

This function return a list of outputs of the negative bionomial modeling for a selected otuID or taxa. Members of this output list are: "NB.model", "tax.met", "taxon", "factors", "anova".

NB.model is the negative bionomial model
tax.met is a dataframe with combined the taxon and metadata

8 assist.ordination

taxon is either a taxon name or in LCA_otuID format, see also LCA.OTU

factors shows which metadata variable had significant impact

anova shows anova test of a metadata variable, this will not be available if anov. fac

is NULL

Author(s)

Wen Chen

Examples

assist.ordination

Perform CCA and RDA Analysis for OTU Tables

Description

This function simplifies CCA and RDA analysis by abstracting away some of the complexity and returning a list of useful measures.

Usage

Arguments

otu1	the first OTU table to be used.
otu2	the second OTU table to be used.
meta	the metadata table to be used (must have same samples as otu1/otu2).
full	logical. Should a full model be considered? (If not, a restricted model is used).
exclude	A vector, either numeric or logical, specifying the columns to be removed from meta. If a character vector, columns with those names will be removed; if a numeric vector, columns with those indices will be removed.
rank	a character vector representing a rank. Must be in one of three specific formats

(see ?RAM.rank.formatting for help).

col.splitup 9

na.action choice of one of the following: "na.fail", "na.omit" or "na.exclude", see na.action in cca for detail.

Value

If both otu1 and otu2 are given, a list of length 2 will be returned with the following items (if only otu1 is given, a list of length 1 will be returned with these items):

\$GOF the goodness of fit scores for the model.

\$VIF the VIF scores for the model.

\$percent_variation

the percent variation explained by each axis

\$CCA_eig Eigenvalues for CCA axes. \$CA_eig Eigenvalues for CA axes.

\$anova the ANOVA results for the model.

Author(s)

Wen Chen and Joshua Simpson.

See Also

```
cca, anova.cca
```

Examples

```
data(ITS1, meta)
cca.help <- assist.cca(ITS1, meta=meta, rank="p")
cca.help$anova</pre>
```

col.splitup

Split Column Of Data Frame

Description

This function output consumes a data frame and split one by defined separator.

Usage

```
col.splitup(df, col="", sep="", max=NULL, names=NULL, drop=TRUE)
```

10 col.splitup

Arguments

df	a data frame.
col	name of a column in df.
sep	the separator to split the column. It can be regular expression.
max	optional. The number of columns to be split to.
names	optional. The names for the new columns.
drop	logical. Whether or not to keep the original column to be split in the output.

Value

The value returned by this function is a data frame. The selected column is split each separator and appended to the original data frame. The original column may may not to be kept in the output as defined by option drop.

The number of columns to be split to depends on three factors, 1) the maximum columns that the original column can be split to by each separator; 2) the user definde max; and 3) the length of the column names defined by names. This function will split the column to the maximum number of the 3, empty columns will be filled with empty strings.

Author(s)

Wen Chen.

combine.OTU 11

combine.OTU

Combine Non Overlapped OTU tables From The Same Community

Description

This function combines out tables from the same community but based on independent sequencing runs. Such combined out table gives a more complete profile of the microbial community than each individual out table does. This function should NOT be used to combine ITS1 and ITS2 out tables if they were extracted from long NGS sequences.

Usage

```
combine.OTU(data, meta)
```

Arguments

data a list of otu tables to be combined.

meta the metadata that should have the same number and order of the samples as the

otu tables do.

Value

combine.OTU returns a data frame of combined otu tables which have the same samples. Samples in the output will match those in the metadata provided.

Author(s)

Wen Chen

See Also

```
match.data
```

12 core.OTU

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Summary Of Core OTUs

Description

This function returns a list showing otus that present in a pre-defined percent of samples in each level of a given metadata category.

Usage

```
core.OTU(data, meta, meta.factor="", percent=1)
```

Arguments

data a list of OTU tables to be analyzed. See RAM.input.formatting.

meta the metadata table to be analyzed.
meta.factor the metadata qualitative variable

percent the percent of samples in each level of the given metadata variable

Value

core.OTU returns a list containing of otus that present in a pre-defined percent of samples in each level of a given metadata category. The outputs describe the following information for each level of a given metadata variable: 1) core of otuID; 2) taxa the core of otus assigned to; and 3) percent of core of otus sequences vs. total sequences in each levels of the given metadata variable. The last item in the list show the same information of otus that in all levels.

Note

The OTUs are determined to be absent/present using the "pa" method from the function decostand.

Author(s)

Wen Chen

See Also

decostand

core.OTU.rank

core.OTU.rank Summary Of Core OTUs

Description

This function returns a list showing otus that present in a pre-defined percent of samples in each level of a given metadata category.

Usage

Arguments

data a list of OTU tables to be analyzed. See also RAM.input.formatting.

rank the taxonomic rank(s) of otu classification (see ?RAM.rank.formatting for for-

matting details).

drop.unclassified

logical, whether or not exclude unclassified groups.

meta the metadata table to be analyzed.
meta.factor the metadata qualitative variable

percent the percent of samples in each level of the given metadata variable

Value

core.OTU.rank returns a list containing otus that present in a pre-defined percent of samples in each level of a given metadata category. The outputs describe the following information for each level of a given metadata variable: 1) core otuID; 2) taxa the core otus assigned to; and 3) percent of core otus sequences vs. total sequences in each group. The last item in the list show the same information of otus that in all levels.

Note

The taxon groups are determined to be absent/present using the "pa" method from the function decostand.

Author(s)

Wen Chen

See Also

decostand

14 core.Taxa

Examples

core.Taxa

Show Summary of Core Taxa

Description

This function returns a list showing taxa at the given taxonomic rank that present in a pre-defined percent of samples in each level of a given metadata category.

Usage

Arguments

data a list of OTU tables or taxonomy abundance matrices to be analyzed.

is.OTU logical. If TRUE, data is an OTU table; otherwise a taxonomy abundance matrix

should be provided.

rank the taxonomic rank of classification (see ?RAM.rank.formatting for formatting

details).

drop.unclassified

logical, whether or not exclude unclassified groups. See also tax.abund

meta the metadata table to be analyzed.
meta.factor the metadata qualitative variable

percent the percent of samples in each level of the given metadata variable

Value

core. Taxa returns a list containing taxa at a given rank that present in a pre-defined percent of samples in each level of a given metadata category. The outputs describe the following information for each level of a given metadata variable: 1) core taxa; 2) percent of core taxa sequences vs. total sequences in each levels of the given metadata variable. The last item in the list show the same information of taxa that in all levels.

Note

The taxa are determined to be absent/present using the "pa" method from the function decostand.

correlation 15

Author(s)

Wen Chen

See Also

decostand

Examples

correlation

Plot Of Correlation Coefficient

Description

This function plot correlation relationship among taxa at a give rank and / or numeric variables of metadata.

Usage

Arguments

a data frame that either an OTU table or taxonomy abundance matrix, can be missing but if metadata is also missing, an error message will be raised.

is.OTU logical. Whether or not the data is an OTU table.

meta the metadata table to be used.

16 correlation

rank	the taxonomic rank to use (see ?RAM.rank.formatting for formatting details).
sel	optional. It is a character vector of selected otuIDs or taxa names at a given taxonomic rank. If provided, sel.OTU should be set to decribe the type of IDs, i.e. TRUE means otuIDs, FALSE means taxa names. If provide, only the selected taxa will be ploted; otherwise, all taxa will be ploted.
sel.OTU	logical. Whether or not the selected items from data are otuIDs. If FALSE, sel should be a string vector of taxa names at a given rank.
data.trans	a character string of one of the following, "total", "log", "hellinger" etc, see ?vegan::decostand for details and other data transformation methods.
method	a character string, can be one of the following, "pearson", "kendall", "spearman" for the calculation of correlation coefficient (or covariance) is to be computed (see ?stats::cor for details)
main	a character string. The title of the plot.
file	the file path where the image should be created (see ?RAM.plotting).
ext	filename extension, the type of image to be saved to. (see ?RAM.plotting).
height	the height of the image to be created (in inches).
width	the width of the image to be created (in inches).

Details

This function uses stats::cor to calculate correlation coefficient (or covariance), and uses lattice::levelplot to generate the graph. (see References)

Option sel is optional, however, it raises an error if the total number of variables to be plotted was too big, and no plot will be generated.

Value

This function generates a graph showing correlation relationship among OTUs or taxa at a given rank, and numeric variables of metadata

Author(s)

Wen Chen.

References

Sarkar, Deepayan (2008) _Lattice: Multivariate Data Visualization with R_, Springer. <URL: http://lmdvr.r-forge.r-project.org/>

Becker, R. A., Chambers, J. M. and Wilks, A. R. (1988) _The New S Language_. Wadsworth & Brooks/Cole.

See Also

cor levelplot

data.clust 17

Examples

data.clust

Plot Hierarchical Cluster Of Samples Based on OTU Table or Taxonomic Abundance Matrix

Description

This function plot hierarchical cluster Of ecology data set.

Usage

Arguments

data	an ecology data set to be analyzed.
is.OTU	logical. If an OTU table was provided, is . OTU should be set as TRUE; otherwise, it should be set as FALSE.
meta	the metadata table associated with ecology data set.
rank	optional. If no rank was provided, the data will be used as it is, if rank is provided, if data is an OTU table, it will be converted to taxonomic abundance matrix at the given rank, no change will be made for a data that has already been a taxonomic abundance matrix. See also tax.abund and data.revamp
top	the top otuIDs or taxa to be considered for the clustering analysis. See also data.revamp
mode	either be "number" or "percent". See also data.revamp
group	an integer or a metadata variable. If an integar, will cut tree into corresponding groups and color them accordingly; if a metadata variable was provided, tree leaves (sampleIDs) will be colored by each level.

18 data.revamp

data.trans	optional. If was provided, numeric data will be transformed. See also decostand
dist	optional. If was provided, distance matrix will be calculated using the given method; otherwise use vegdist default Bray-Curtis method. See also vegdist and gowdis.
clust	optional. If was not provided, will use the default agglomeration method used by hclust, i.e. "complete". Otherwise, will used user defined method for clustering. See also hclust.
type	optional. Can be one of the following: "triangle", "rectangle", "phylogram", "cladogram", "fan", "unrooted", "radial".
main	The title of the plot.
file	optional. Filename that the plot to be saved to.
ext	optional. File type that the plot to be saved to.
width	an integer, width of the plot.
height	an integer, height of the plot.

Value

This function returns a tree plot of the hierarchical cluster of the samples based on ecological data.

Author(s)

Wen Chen

Examples

data.revamp

Transform OTU Table

Description

This function consumes and transforms either an OTU table or a taxonomy abundance matrix. If an OTU table was provided, it will be either transposed without the "taxonomy" column, but each otuID will be renames with it's LCA classification appended; or being transformed to be taxonomic abundance matrix at the ranks set by ranks. If a taxonomic abundance matrix is provided, it will be kept the same with proper data transformation as defined by stand.method option.

Usage

data.revamp 19

Arguments

data an OTU table or a taxonomic abundance matrix. is.OTU logical. If an OTU table was provided, is.OTU should be set as TRUE; otherwise, it should be set as FALSE. ranks optional. If no ranks was provided, the OTU table will be processed by LCA.OTU and then transposed with sampleIDs being row names and otuIDs being column names. If ranks was provided, the OTU table will be processed by tax. abund at each given taxonomic ranks. See also RAM. rank. formatting. The unclassified taxon groups are removed. stand.method optional. Transform the output using method from the function decostand optional. Select the top taxa or OTUs. top a character vector, one of "percent" or "number". If number, then top many mode groups will be selected. If percent, then all groups with relative abundance in

Value

The value returned by this function is a list, so for convenience, any nested lists have been given descriptive items names to make accessing its elements simple (see Examples).

at least one sample above top will be selected.

- If is .OTU is TRUE and ranks is not given: the output is a length one list named LCA_OTU.
- If is.OTU is TRUE and ranks is given: the output is a list with a length same as the number of taxonomic ranks provided. Each member of the list is named after the rank it processed at.
- If is.OTU is FALSE, the output is a length one list named Taxa.

Author(s)

Wen Chen

20 data.subset

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data	CII	ncai	-

Subset OTU And Metadata

Description

This function subset OTUs and metadata based on user defined values of metadata variables.

Usage

```
data.subset(data, meta, factors="", values="", and=TRUE)
```

Arguments

data a list of otu tables to be processed. See also RAM.input.formatting.

meta the metadata for subset.

factors a vector containing metadata variables.

values a vector containing values of interest in metadata variables.

and logical. Determine whether all conditions needs to be met or not.

Value

The value returned by this function is a list containing otu tables matching the filtering requirement. The last item in the output list is the associated new metadata table fit the requirement.

Author(s)

Wen Chen

```
data(ITS1, ITS2, meta)
names(meta)
factors <- c("City", "Harvestmethod")</pre>
values <- c("City1", "Method1")</pre>
# match all requirements, and=TRUE
sub <- data.subset(data=list(ITS1=ITS1, ITS2=ITS2), meta=meta,</pre>
                     factors=factors, values=values, and=TRUE)
# match either of the requirements, and=FALSE
sub <- data.subset(data=list(ITS1=ITS1, ITS2=ITS2), meta=meta,</pre>
                     factors=factors, values=values, and=FALSE)
## Not run:
names(sub)
ITS1.sub <- sub[["ITS1"]]</pre>
ITS2.sub <- sub[["ITS2"]]</pre>
meta.sub <- sub[["meta"]]</pre>
## End(Not run)
```

dissim 21

dissim

Calculate Dissimilarity Matrix Data

Description

These functions calculate different measures related to dissimilarity matrices. All of these functions allow you to specify one of many dissimilarity indices to be used.

Usage

Arguments

elem	an ecology data set that can be an OTU table or a taxonomy abundance table. See RAM. input. formatting for details.
is.OTU	logical, whether the ecology data sets are OTU tables or taxonomy abundance matrices. See RAM.input.formatting for details.
stand.method	optional, if is.null, the standardization method for data transforamtion; must be one of the following: "total", "max", "frequency", "normalize", "range", "standardize", "pa", "chi.square", "hellinger", "log". See also decostand.
dist.method	the dissimilarity index to be used; one of "manhattan", "euclidean", "canberra", "bray", "kulczynski", "jaccard", "gower", "altGower", "morisita", "horn", "mountford", "raup", "binomial", "chao", or "cao". See also vegdist.
k	the number of dimensions desired. If NULL, the maximum value will be calculated and used.
clust.method	the method used for clustering the data. Must be one of "ward", "single", "complete", "average", "mcquitty", "median", or "centroid". See also hclust.

Value

dissim.clust returns a hierarchical clustering of the dissimilarity matrix.

dist.eigenval returns the eigenvalues of the dissimilarity matrix.

22 dissim.heatmap

dissim.ord	returns a list: the first item is the ordination distances, the second is the dissimilarity matrix distances.
dissim.GOF	returns the goodness of fit values of the dissimilarity matrix, for various numbers of dimensions used.
dissim.tree	returns a list: the first item is the tree distances, the second is the dissimilarity matrix distances.
dissim.pvar	returns a numeric vector containing the percent variation explained by each axis (where each sample corresponds to an axis).

Author(s)

Wen Chen and Joshua Simpson

See Also

```
decostand, vegdist, hclust, dissim.plot
```

Examples

```
data(ITS1)

# calculate clustering, using default method
dissim.clust(ITS1)

# calculate tree distances, specifying a distance method
# (but use default clustering method)
dissim.tree(ITS1, dist.method="euclidean")

# calculate ordination distances, specifying both distance
# and ordination methods
dissim.ord(ITS1, dist.method="bray", k=3)
```

dissim.heatmap

Plot Distance Matrix Heatmap for OTU Samples

Description

This function consumes an OTU table, along with some optional parameters, and creates a heatmap showing the distance matrix for the samples of the given OTU table.

Usage

dissim.heatmap 23

Arguments

data	the OTU table to be used.
is.OTU	logical. Whether or not the data is an OTU table.
meta	the metadata table to be used.
row.factor	a factor from the metadata to show along the rows of the heatmap (see Details below).
col.factor	a factor from the metadata to show along the columns of the heatmap (see Details below).
stand.method	a method used to standardize the OTU table. One of "total", "max", "freq", "normalize", "range", "standardize", "pa", "chi.square", "hellinger" or "log" (see ?decostand).
dissim.method	the dissimilarity index to be used; one of "manhattan", "euclidean", "canberra", "bray", "kulczynski", "jaccard", "gower", "altGower", "morisita", "horn", "mountford", "raup", "binomial", "chao", or "cao" (see ?vegdist).
file	the file path where the image should be created (see ?RAM.plotting).
ext	the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".
height	the height of the image to be created (in inches).
width	the width of the image to be created (in inches).
leg.x	how far the legend should be inset, on the x axis.
leg.y	how far the legend should be inset, on the y axis.

Details

Both row. factor and col. factor should be named character vectors specifying the names of the rows to be used from meta (see RAM. factors). They should also be factors; if they are not, a warning is raised and they are coerced to factors (see factor). A warning is also raised when a factor has more than 8 levels, as that is the most colours the current palettes support. The factor must also correspond to the OTU table; i.e. they should have the same samples. If not, an error is raised.

Note

This function creates the heatmap using the heatmap.2 function from the gplots package. That function calls layout to set up the plotting environment, which currently prevents plotting two data sets side by side, or to automatically place the (metadata) legend. Unfortunately, this means that the leg.x and leg.y parameters must be used to adjust the legend by trial and error. It is possible to move the legend outside of the plotting area; if not legend appears, try with small leg.x and leg.y values.

Author(s)

Wen Chen and Joshua Simpson.

See Also

decostand, vegdist, factor, heatmap. 2, RAM. factors

24 dissim.plot

Examples

dissim.plot

Plot Dissimilarity Matrix Data for Different Methods

Description

These functions all produce a plot of some measure related to dissimilarity matrices. All of these functions allow you to specify a vector of methods to be used when creating the plot.

Usage

```
dissim.clust.plot(data, is.OTU=TRUE, stand.method=NULL,
                  dist.methods=NULL,
                  clust.methods=NULL, file=NULL)
dissim.eig.plot(data, is.OTU=TRUE, stand.method=NULL,
                dist.methods=NULL, file=NULL)
dissim.alleig.plot(data, is.OTU=TRUE, stand.method=NULL,
                   dist.methods=NULL, file=NULL)
dissim.ord.plot(data, is.OTU=TRUE, stand.method=NULL,
                dist.methods=NULL, k=NULL, file=NULL)
dissim.GOF.plot(data, is.OTU=TRUE, stand.method=NULL,
                dist.methods=NULL, file=NULL)
dissim.tree.plot(data, is.OTU=TRUE, stand.method=NULL,
                 dist.methods=NULL,
                 clust.methods=NULL, file=NULL)
dissim.pvar.plot(data, is.OTU=TRUE, stand.method=NULL,
                 dist.methods=NULL, file=NULL)
```

Arguments

data a list of ecology data. See also RAM.input.formatting

is.OTU logical, whether the ecology data sets are OTU tables or taxonomy abundance

matrices.

dissim.plot 25

stand.method optional, if is.null, the standardization method for data transforamtion; must be one of the following: "total", "max", "frequency", "normalize", "range", "standardize", "pa", "chi.square", "hellinger", "log". See also decostand. dist.methods a character vector representing the dissimilarity indices to be used; each element must be one of one of "manhattan", "euclidean", "canberra", "bray", "kulczynski", "jaccard", "gower", "altGower", "morisita", "horn", "mountford", "raup", "binomial", "chao", or "cao". clust.methods a character vector representing the methods used for clustering the data. Each element must be one of "ward", "single", "complete", "average", "mcquitty", "median", or "centroid". k the number of dimensions desired. If NULL, the maximum value will be calculated and used. file the file path for the plot. If not provided (defaults to NULL), then the plot is displayed to the screen. If file is provided, that is where the .tiff file will be created.

Details

All of these functions (other than dissim.alleig.plot) call dissim. X counterparts and plot the data. If file is given, a .tiff file will be created at file; otherwise the plot is displayed to the screen.

Value

All functions create a plot and return the plotted data invisibly.

dissim.clust.plot

plots a hierarchical clustering of the dissimilarity matrix.

dissim.eig.plot

plots a bar plot of the eigenvalues of the dissimilarity matrix.

dissim.alleig.plot

plots a line plot showing the relative importance of all eigenvalues for a variety of methods.

dissim.ord.plot

plots a scatter plot comparing the "euclidean" distances among all samples in ordination space to the dissimilarity matrix distances.

dissim.GOF.plot

plots a scatter plot of the goodness of fit values of the dissimilarity matrix, for various numbers of dimensions used.

dissim.tree.plot

plots a scatter plot comparing the tree distances to the dissimilarity matrix distances.

dissim.pvar.plot

plots a bar plot showing the percent variation explained by each axis (where each sample corresponds to an axis).

26 dissim.plot

Note

If file does not end in ".tiff", then ".tiff" will be appended to the end of file.

Function dissim.alleig.plot uses the ggplot2 package for creating the plot, and returns the plot object. This means that you can store this plot and add other features manually, if desired (see Examples).

Author(s)

Wen Chen and Joshua Simpson

See Also

```
vegdist, hclust, dissim, ggplot
```

```
data(ITS1, ITS2)
data <- list(ITS1=ITS1, ITS2=ITS2)</pre>
# show percent variation for only ITS1 with default methods
dissim.pvar.plot(data=list(ITS1=ITS1))
## Not run:
# show clustering for ITS1 and ITS2 for set methods
dissim.clust.plot(data=data, is.OTU=TRUE, stand.method=NULL,
                   dist.methods=c("morisita", "bray"),
clust.methods=c("average", "centroid"))
dissim.ord.plot(data=data, is.OTU=TRUE, stand.method="total",
                 dist.method="bray")
# dissim.alleig.plot returns a ggplot2 object:
my.eig.plot <- dissim.alleig.plot(data)</pre>
class(my.eig.plot) # returns "gg" "ggplot"
my.eig.plot # view the plot
# update the title, then view the updated plot
my.eig.plot <- my.eig.plot + ggtitle("My New Title")</pre>
# update ggplot theme
require("grid")
new_theme <-RAM.color()</pre>
my.eig.plot <- my.eig.plot + new_theme</pre>
my.eig.plot
# save an image (named file.pdf) with GOF values for ITS1 and ITS2,
using # default methods
dissim.GOF.plot(data=data, file="~/Documents/my/file")
## End(Not run)
```

diversity.indices 27

diversity.indices

Calculate True Diversity and Evenness

Description

These functions calculate true diversity and evenness for all samples.

Usage

```
true.diversity(data, index = "simpson")
evenness(data, index = "simpson")
```

Arguments

data a list of otu tables to be processed. See RAM. input. formatting.

index the index to use for calculations; partial match to "simpson" or "shannon".

Details

For the following sections, S represents the number of species, λ represents the Simpson index, and H' represents the Shannon index.

The formulas for the true diversity of the indices are as follows:

• Simpson: $D_2 = frac1\lambda$

• Shannon: $D_1 = \exp H'$

The formulas for the evenness of the indices are as follows:

• Simpson: $\frac{\frac{1}{\lambda}}{S}$

• Shannon: $\frac{H'}{\ln S}$

Value

Both functions return a numeric data frame, where the rows are the given OTUs, and the columns are the samples.

Note

Credit goes to package vegan for the partial argument matching (see References).

Author(s)

Wen Chen and Joshua Simpson.

28 envis.NB

References

Jari Oksanen, F. Guillaume Blanchet, Roeland Kindt, Pierre Legendre, Peter R. Minchin, R. B. O'Hara, Gavin L. Simpson, Peter Solymos, M. Henry H. Stevens and Helene Wagner (2013). vegan: Community Ecology Package. R package version 2.0-10. http://CRAN.R-project.org/package=vegan

Diversity index. (2014, May 7). In Wikipedia, The Free Encyclopedia. Retrieved 14:57, May 28, 2014, from http://en.wikipedia.org/w/index.php?title=Diversity_index&oldid=607510424

Blackwood, C. B., Hudleston, D., Zak, D. R., & Buyer, J. S. (2007). Interpreting ecological diversity indices applied to terminal restriction fragment length polymorphism data: insights from simulated microbial communities. Applied and Environmental Microbiology, 73(16), 5276-5283.

Examples

```
data(ITS1, ITS2)

# true diversity, using default index (Simpson)
true.diversity(data=list(ITS1=ITS1))

# true diversity for ITS1 and ITS2, using Shannon
true.diversity(data=list(ITS1=ITS1, ITS2=ITS2), index="shannon")

# default evenness (Simpson) for ITS1/ITS2
evenness(data=list(ITS1=ITS1, ITS2=ITS2))

# Shannon evenness
evenness(data=list(ITS1=ITS1), index="shannon")
```

envis.NB

Visualize The Negative Binomial Model OF A Given Taxon OR OTUID

Description

This function plot the negative binomial model for a given otuID or taxon

Usage

Arguments

NB.model	the negative binomial model. Can be obtained by using assist.NB
tax.meta	the combined taxon/otuID and metadata. Can be obtained by using ${\tt assist.NB}$
taxon	the taxon or otuID. Can be obtained by using assist.NB
X	a metadata variable name for x axis.

envis.NB 29

num.col	optional. A metadata numerical variable that will be used as predictor.
group	optional. A metadata factor variable that will be used as predictor.
group.order	optional. The desired order for the group.
xlab	optional. X axis label.
ylab	optional. Y axis label.
fill	optional. Color for fill different categories.
facet	optional. Metadata category variables as faceting variables.
file	optional. Filename that the plot to be saved to.
ext	optional. Filename extension, type of image to be saved.
width	an integer. Filter OTU table by counts.
height	an integer. Filter OTU table by counts.

Value

This function plot predicted taxon/otuID under the impact of metadata variables.

Author(s)

Wen Chen

```
data(ITS1, meta)
# filter otu table
its1 <- filter.OTU(data=list(ITS1=ITS1), percent=0.01)[[1]]</pre>
m \leftarrow meta[, c(2,3,5,7)]
## Not run:
# test the model
nb <- assist.NB(its1, meta=m, rank="g", anov.fac="Harvestmethod",</pre>
              taxon=rownames(its1)[1])
NB.model<-nb[[1]]
tax.meta<-nb[[2]]</pre>
taxon<-nb[[3]]
envis.NB(NB.model=NB.model, tax.meta=tax.meta, taxon=taxon,
                x="Ergosterol_ppm", num.col="Ergosterol_ppm",
                 group="Crop", group.order=NULL,
                xlab="Ergosterol (ppm)", ylab=NULL,
                 fill="Harvestmethod", facet=c("City","Crop"))
## End(Not run)
```

30 factor.abundance

factor.abundance	Plot the Abundance of OTUs by Classification at a Given Taxonomic Rank For Each Level of A Metadata Category Variable.
	Kank For Each Level of A Meladala Calegory variable.

Description

This function consumes an OTU, and a rank, as well as various optional parameters. It creates a stacked bar plot showing the abundance of all classifications at the given taxonomic rank for each level of a metadata category variable.

Usage

Arguments

data	a list of OTU tables with names.	
rank	the taxonomic rank to use. See RAM.rank.formatting.	
top	the number of groups to select, starting with the most abundant. If \ensuremath{NULL} , all are selected.	
count	logical. If TRUE, the numerical counts for each OTU will be shown; otherwise the relative abundance will be shown.	
meta	metadata.	
meta.factor	a category variable in metadata	
drop.unclassified		
	logical. Should unclassified samples be excluded from the data?	
file	the file path where the image should be created (see ?RAM.plotting).	
ext	the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".	
height	the height of the image to be created (in inches).	
width	the width of the image to be created (in inches).	
main	the title of the plot	

Author(s)

Wen Chen

filter.META 31

Examples

filter.META

Select METADATA Variables

Description

This function will remove metadata variables with only one level and /or remove variables with missing data or neither numeric nor factor/character (NNF).

Usage

```
filter.META(meta=meta, excl.na=TRUE, excl.NNF=TRUE, exclude=NULL)
```

Arguments

meta	the metadata table to be analyzed.
excl.na	logical. Whether or not remove variables that contain missing data.
excl.NNF	logical. Whether or not remove variables that neither are numeric nor factor/character.
exclude	optional. If is NULL, the function only removes variables with only one level or NNF. Otherwise, the variables in the exclude will also be removed from the metadata table.

Value

The value returned by this function is a data frame with the following metadata variables being removed: 1) with missing data; 2) NNF if excl.NNF is TRUE; and 3) in the exclude list.

Author(s)

Wen Chen

32 filter.OTU

Examples

filter.OTU

Filter OTU

Description

This function filter OTU table by counts or relative abundance. If filter by counts, otus having total counts more than a threshhold will be kept. If filter by relative abundance, otus with the maximum relative abundance greater than a threshhold in at least one subject will be kept.

Usage

```
filter.OTU(data, percent=NULL, number=NULL)
```

Arguments

data a list of otu tables to be processed. See also RAM.input.formatting

percent a floating point greater than 0 and less or equals to 1. Filter OTU table by relative

abundance.

number an integer. Filter OTU table by counts.

Value

The value returned by this function is a list of filtered otu tables provided by the user

Author(s)

Wen Chen

filter.Taxa 33

Examples

```
data(ITS1, ITS2, meta)
data<-list(ITS1=ITS1, ITS2=ITS2)
## Not run:
otu.001 <- filter.OTU(data=data, percent=0.01)
length(otu.001)
names(otu.001)
otu.50 <- filter.OTU(data=data, number=50)
## End(Not run)</pre>
```

filter.Taxa

Filter Taxonomic Abundance Matrix by Total Counts Or Maximum Relative Abundance

Description

This function filter taxa group by counts or relative abundance. If filter by counts, taxa having total counts more than a threshhold will be kept. If filter by relative abundance, taxa with the maximum relative abundance greater than a threshhold in at least one subject will be kept.

Usage

Arguments

taxa the taxonomy abundance matrix: sample x species data frame. See also tax.abund drop.unclassified

logical, whether or not remove unclassified groups. See also tax. abund

percent a floating point greater than 0 and less or equals to 1. Filter Taxa table by relative

abundance.

number an integer. FilterTaxa table by total sequence counts.

Value

The value returned by this function is a data frame with taxa met the filter requirement only.

Author(s)

Wen Chen

```
data(ITS1)
g1 <- tax.abund(ITS1, rank="g", drop.unclassified=TRUE)
taxa.fil <- filter.Taxa(g1, percent=0.01)</pre>
```

34 fread.OTU

fread.meta

Load Metadata Table

Description

This function is same as read.meta to read in data; except using fread for loading large data sets.

Usage

```
fread.meta(file, sep="auto")
```

Arguments

file a character vector specifying the file path to your file.

sep the character used to separate cells in the file.

Value

Returns a data frame with the information from the file. The first row and column are used for the names of the other rows and columns.

Author(s)

Wen Chen

See Also

```
read.meta, fread
```

Examples

```
## Not run:
    my.meta <- fread.meta("path/to/meta")
## End(Not run)</pre>
```

fread.OTU

Fast Load Large OTU Table

Description

This function is same as read.OTU except using fread for loading large data sets.

Usage

```
fread.OTU(file, sep="auto")
```

get.rank 35

Arguments

file a character vector specifying the file path to your file.
sep the character used to separate cells in the file.

Value

Returns a data frame with the information from the file. The first row and column are used for the names of the other rows and columns.

Note

The OTU table should only contain classifications for the seven major taxonomic ranks, additional ranks will break some functions in the package. To remove those other classifications, try running

```
sed -i.backup -e 's/s[a-z]__[^;]*; //g' -e 's/t__[^;]*; //g' $FILE
```

where \$FILE is your OTU table. The letter t can be replaced in the second expression for any other letter which appears as a prefix. For example, adding $-e 's/u_{[^;]*; //g'}$ before \$FILE would remove any entries formatted like $u_{test_classification}$.

Additionally, if your OTU table starts with the entry #0TU ID, that cell needs to be removed before the table can be read with fread.0TU.

Author(s)

Wen Chen.

See Also

```
read.OTU, fread
```

Examples

```
## Not run:
    my.OTU <- fread.OTU("path/to/otu")
## End(Not run)</pre>
```

get.rank

Get OTUs Classified at Taxonomic Rank(s)

Description

This function returns the OTUs of the given OTU table(s) which are classified at the given taxonomic rank.

Usage

```
get.rank(otu1, otu2 = NULL, rank = NULL)
```

36 get.rank

Arguments

otu1	the first OTU table to be used.
otu2	the second OTU table to be used.
rank	a character vector representing a rank. Must be in one of three specific formats (see RAM.rank.formatting for help). If omitted, the function will repeat for all seven major taxonomic ranks.

Value

The value returned by this function may become nested lists, so for convenience, any nested lists have been given descriptive items names to make accessing its elements simple (see Examples).

- If otu2 is given:
 - If rank is given: a list containing two data frames (otu1 and otu2 selected at the given rank).
 - If rank is not given: a list containing two lists. The first sublist represents otu1, the second otu2. The sublists contain seven data frames, which are the OTU tables selected at each taxonomic rank (see Examples).
- If otu2 is not given:
 - If rank is given: a single data frame (otu1 selected at the given rank).
 - If rank is not given: a list containing seven data frames (otu1 selected at each taxonomic rank).

Author(s)

Wen Chen and Joshua Simpson.

```
data(ITS1, ITS2)

# the following are equivalent:
ITS1.p <- get.rank(ITS1, rank="p")
# this list has get.rank(ITS1, rank="k"),
# get.rank(ITS1, rank="p"), ...
lst <- get.rank(ITS1)
stopifnot(identical(ITS1.p, lst$phylum))
# true

# get a list of length 2: the item holds all ITS1 data, the
# second holds ITS2 data
lst.all <- get.rank(ITS1, ITS2)
stopifnot(identical(ITS1.p, lst.all$otu1$phylum))</pre>
```

group.abund.Taxa 37

group.abund.Taxa	Rarplot (of Distribution (Of Taxa In Groups
gi oup. abana. Taxa	Darpioi	j Distribution (oj rana in Groups

Description

This function do a barplot to show the distribution of selected taxa in each level of a given metadata variable

Usage

Arguments

, amends	
data	a list of otu tables or taxonomic abundance matrices. See also RAM. input. formatting.
is.OTU	logical. If an OTU table was provided, is.OTU should be set as TRUE; otherwise, it should be set as FALSE.
rank	a single taxonomic rank. See also RAM.rank.formatting
taxa	a vector containing taxa names for plotting.
drop.unclassifi	ed
	logical. Whether or not drop the unclassified taxon groups.
bar.width	width of bars
meta	the metadata table to be used (must have same samples as data.
meta.factor	a character string. Must be one of the metadata variables.
RAM.theme	customized ggplot_theme in RAM. See also ?theme_ggplot.
col.pal	color palettes to be used.
main	a character string. The title of the plot, default is an empty string.
file	filename to save the plot.
ext	filename extension, the type of image to be saved to.
width	an integer, width of the plot.
height	an integer, height of the plot.

Value

This function returns a Barplot of the distribution of seleted taxa within each level of a given metadata variable.

Note

This funtion provide an alternative view of taxa distribution as group. Taxa. bar.

38 group.abundance

Author(s)

Wen Chen.

Examples

group.abundance

Plot the Abundance of OTUs by Classification at a Given Taxonomic Rank

Description

This function consumes an OTU, and a rank, as well as various optional parameters. It creates a stacked bar plot showing the abundance of all classifications at the given taxonomic rank for each sample.

Usage

Arguments

data	a list of OTU tables.
rank	the taxonomic rank to use. See RAM.rank.formatting.
top	the number of groups to select, starting with the most abundant. If NULL, all are selected.
count	logical. If TRUE, the numerical counts for each OTU will be shown; otherwise
drop.unclassifi	the relative abundance will be shown.
	logical. Should unclassified samples be excluded from the data?
cex.x	optional. The size of x axis names.
main	the title of the plot
file	the file path where the image should be created (see ?RAM.plotting).
ext	the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".
height	the height of the image to be created (in inches).
width	the width of the image to be created (in inches).
bw	logical. Should the image be created in black and white?
ggplot2	logical. Should the ggplot2 package be used to produce the plot, or should the base graphics be used? (see ?RAM.plotting).

group.abundance.meta 39

Author(s)

Wen Chen and Joshua Simpson

Examples

Description

This function is an updated version of group. abundance, which groups samples by metadata category variables if provided.

Usage

Arguments

data	a list of OTU tables.
rank	the taxonomic rank to use. See RAM. rank. formatting.
top	the number of groups to select, starting with the most abundant. If \ensuremath{NULL} , all are selected.
count	logical. If TRUE, the numerical counts for each OTU will be shown; otherwise the relative abundance will be shown.
drop.unclassifi	ed
	lanical Charleton along if advantable and add from the data?

logical. Should unclassified samples be excluded from the data?

40 group.diversity

optional. The size of x axis names. cex.x the title of the plot main file the file path where the image should be created (see ?RAM.plotting). ext the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg". height the height of the image to be created (in inches). width the width of the image to be created (in inches). bw logical. Should the image be created in black and white? optional, associated metadata of the otu tables, sampleIDs and their orders in meta otu tables should be identical to those in metadata meta.factor optional, category variables in meta

Author(s)

Wen Chen

Examples

group.diversity

Boxplot To Compare Diversity Indices Among Groups

Description

This function first use OTU. diversity to calculate the diversity indices for each sample and then do a boxplot to compare the selected indices among different groups.

Usage

group.diversity 41

Arguments

data	a list, containing otu tables. See also RAM. input. formatting
meta	the metadata table to be used (must have same samples as data.
factors	a character vector. Must be variables in the metadata
indices	a character vector. Must be one or more of the following: "spec", "sim", "invsim", "shan", "sim_even", "shan_even", "sim_trudiv", "shan_trudiv", "chao", "ACE". See also OTU.diversity, true.diversity, evenness, and diversity.
diversity.info	logical. Whether the diversity indices have calculated and included in the metadata table. The diversity indices should be processed by OTU. diversity for the same otu tables and metadata table.
x.axis	optional. If NULL, will use the first variable in factors; otherwise, must be one factor in the metadata or 'SampleID'
compare	optional. If NULL, will use the first variable in factors; otherwise, must be one factor in the metadata ${\sf NULL}$
facet	optional. If provided, must be one factor in the metadata or 'SampleID'
facet.y	logical, whether the facet being used as strip text of y axis or x axis.
facet.x.cex	optional, an integer, the font size of the stip.text.x in ggplot
facet.y.cex	optional, an integer, the font size of the strip.text.y in ggplot.
scale.free	optional. Whether use free scale for y axis.
xlab	optional. If not provided, the $x.axis$ will be used as the title of the x axis, otherwise, will use the provided string.
ylab	optional. If not provided, "value" will be used as the title of the y axis, otherwise, will use the provided string.
legend.title	optional. If not provided, compare will be used as the title of the legend, otherwise, will use the provided string.
legend.labels	optional. If not provided, will use the levels of compare for the legends, otherwise, will use the provided vector of strings. The length of the provided vector of strings must equals to the levels of compare.
file	the filename to save the plot.
ext	the extention (file type) of the plot to saved.

ext the extention (file type) of the plot to saved.

width the width of the plot to be saved.
height the height of the plot to be saved.

Value

This function returns a boxplot to compared selected diversity indices among different groups.

Author(s)

Wen Chen.

See Also

 ${\tt OTU.diversity, true.diversity, evenness} \ {\tt and} \ {\tt diversity}$

42 group.heatmap

Examples

group.heatmap

Plot OTU Abundance at a Given Rank with Metadata Annotation

Description

This function plots the abundance for all taxon groups at a given rank. Additionally, it can display metadata for the samples as annotations along the rows of the heatmap.

Usage

Arguments

data the OTU table to be used.

is.OTU logical. Whether or not the data is an OTU table.

meta the metadata table to be used.

rank the taxonomic rank to use (see RAM. rank. formatting for formatting details).

factors a named character vector indicating the columns of the metadata table to be used

(see RAM. factors).

top the number of groups to select, starting with the most abundant. If NULL, all are

selected.

remove.unclassified

logical. Define whether OTUs labelled "unclassified" or missing classification

at the given taxonomic rank should be excluded.

group.heatmap 43

stand.method optional. Data standardization methods specified in decostand.

dist.method distance matrix calculation methods specified vegdist.

hclust.method the agglomeration methods specified in hclust. This should be unambiguous

abbreviation of one of the following: 'ward.D', 'ward.D2', 'single', 'complete',

'average', 'mcquitty', 'median' or 'centroid'.

dendro.row.status

whether or not to use the dendrogram to re-order the rows. It should be one of the following: 'yes' that use the dendrogram to re-order the rows/columns and display the dendrogram; 'hidden' means re-rorder, but do not display; 'no' means do not use the dendrogram at all. See also annHeatmap2

dendro.col.status

whether or not to use the dendrogram to re-order the columns. It should be one of the following: 'yes' that use the dendrogram to re-order the rows/columns and display the dendrogram; 'hidden' means re-rorder, but do not display; 'no' means do not use the dendrogram at all. See also annHeatmap2

row.labels logical. Whether or not to plot the row labels.
row.cex the size of row labels if row.labels is TRUE

cut the height at which to cut the sample tree, this will create distinct coloured

groups. Currently this will allow for at most nine groups (see Details).

file the file path where the image should be created (see ?RAM.plotting).

ext the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".

height the height of the image to be created (in inches). width the width of the image to be created (in inches).

Details

A warning from brewer.pal indicating "n too large, allowed maximum for palette Pastel1 is 9" means that the cut height is too low to allow for that many groups. This should be fixed in a future release.

Author(s)

Wen Chen and Joshua Simpson.

See Also

decostand, annHeatmap2

```
hclust.method="mcquitty", cut=0.5)
## End(Not run)
```

group.heatmap.simple Plot a Heatmap Showing OTU Abundance by Taxonomic Classification

Description

This function consumes an OTU table and a rank, as well as some optional parameters, and creates a heatmap showing the abundance of the OTUs at the given taxonomic rank for each sample.

Usage

Arguments

data	the OTU table to be used.
is.OTU	logical. Whether or not the data is an OTU table.
meta	the metadata table to be used.
rank	the taxonomic rank to use (see ?RAM.rank.formatting for formatting details).
row.factor	a factor from the metadata to show along the rows of the heatmap. (see Details below).
dendro	a character vector specifying on which axes (if any) a dendrogram should be plotted. Must be one of "none", "both", "column", or "row".
top	the number of groups to select, starting with the most abundant. If \ensuremath{NULL} , all are selected.
count	logical. Should the actual count of each OTU be shown, or should the relative abundances be shown?
drop.unclassifi	ed
	logical. Should OTUs labelled "unclassified" or missing classification at the given taxonomic rank be excluded?
file	the file path where the image should be created (see ?RAM.plotting).
ext	the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".
height	the height of the image to be created (in inches).
width	the width of the image to be created (in inches).
leg.x	how far the legend should be inset, on the x axis.
leg.y	how far the legend should be inset, on the y axis.

Details

row.factor should be a named character vector specifying the name of the row to be used from meta (see RAM.factors). It should also be a factor; if it is not, a warning is raised and it is coerced to a factor (see factor). A warning is also raised when a factor has more than 8 levels, as that is the most colours the current palettes support. The factor must also correspond to the OTU table; i.e. they should have the same samples. If not, an error is raised.

Note

This function creates the heatmap using the heatmap.2 function from the gplots package. That function calls layout to set up the plotting environment, which currently prevents plotting two data sets side by side, or to automatically place the (metadata) legend. Unfortunately, this means that the leg.x and leg.y parameters must be used to adjust the legend by trial and error. It is possible to move the legend outside of the plotting area; if no legend appears, try with small leg.x and leg.y values.

Author(s)

Wen Chen and Joshua Simpson.

See Also

```
factor, heatmap. 2, RAM. factors
```

46 group.indicators

group.indicators Plot Indicator Taxon Groups for Metadata Trends

Description

This function conumes one or two OTU tables, along with a metadata facotr, and creates a barplot showing the relative abundance of all groups which are statistical indicators of that factor.

Usage

Arguments

data	a list of OTUs or taxonomic abundance matrices. see also RAM.input.formatting
is.OTU	logical. Whether the input data are OTU tables.
meta	the metadata table to be used.
factor	a named character vector (of length 1) giving the name of the column in meta to be used when performing the analysis (see RAM. factors).
rank	the taxonomic rank to use (see ?RAM.rank.formatting for formatting details). if rank is NULL, will use otus as indicators which are annotated by the lca assigned to the otus, otherwise will use taxon names as indicators at the given taxonomic rank.
thresholds	a character vector of length 4 specifying the thresholds for the A, B, stat, and p values (see Details).
all.indicators	logical. Whether or not plot all identified indicators. If set as TRUE, will plot all indicators, otherwise, if the total indicators are less than 12, will plot them all; if the total indicators are more than 12, will plot the first 12 of them.
cex.x	optional. The size of x axis names.
file	the file path where the image should be created (see ?RAM.plotting).
ext	the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".
height	the height of the image to be created (in inches).
width	the width of the image to be created (in inches).

group.indicators 47

Details

This function uses indicspecies::multipatt to determine the indicators. After this analysis is performed, there will likely be some species determined to be 'significant,' but to varying degrees. To control how many groups are selected, you can adjust the thresholds argument. It consists of four components: (quotations taken from vignette("indicspeciesTutorial"), see References):

A the *specificity* of the indicator; "the probability that the surveyed site belongs to the target site group given the fact that the species has been found."

B the *fidelity* of the indicator; "the probability of finding the species in sites belonging to the site group."

stat the association strength for the combinations.

p.value "the degree of statistical significance of the association."

Only the species with A, B, and stat values above, and p. value below those given in thresholds will be kept.

Value

This function returns a stacked barplot and a vector of identified indicators, including the ones in the plot and the ones being excluded from the plot.

Author(s)

Wen Chen and Joshua Simpson.

References

De Caceres, M., Legendre, P. (2009). Associations between species and groups of sites: indices and statistical inference. Ecology, URL http://sites.google.com/site/miqueldecaceres/

See Also

```
multipatt
```

48 group.OTU

group.OTU

Plot Distribution of OTUs

Description

This function plot the distribution of otus in each level of a given metadata variable. The plot can be boxplot of barplot. The boxplot shows the range of relative abundance of a given otuID in each level of metadata category. The barplot shows the relative abundance of the total counts of a given otuID in each level of metadata category.

Usage

Arguments

otu the OTU table to be analyzed.

rank optional. if NULL, the lca of the OTUs will be retrieved; otherwise, the user

should provide a valid taxonomic rank that an otu being classified to (see ?RAM.rank.formatting

for formatting details).

otuIDs an vector of otuIDs in the OTU table
meta the metadata table to be analyzed.
meta.factor the metadata qualitative variable

boxplot logical. If TRUE, generate boxplot; otherwise generate barplot.

main title of the plot.

file the file path where the image should be created (see ?RAM.plotting).

ext the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".

height the height of the image to be created (in inches).
width the width of the image to be created (in inches).

Value

group. OTU returns boxplot or barplot for the distribution of a list of otuIDs.

group.rich 49

Note

The OTUs are determined to be absent/present using the "pa" method from the function decostand.

Author(s)

Wen Chen

See Also

ggplot

Examples

group.rich

Barplot Of Richness For Each Level Of A Given Metadata Variable

Description

This function first use specpool to estimate the extrapolated species richness in a species pool (levels of metadata variable), and the number of unobserved species, then do a barplot.

Usage

```
group.rich(otu, meta, factor, file=NULL, ext=NULL, width=8, height=8)
```

50 group.spatial

Arguments

otu	an OTU table.
meta	the metadata table to be used (must have same samples as data.
factor	a character string. Must be one of the metadata variables.
file	optional. Filename that the plot to be saved to.
ext	optional. Filename extension, type of image to be saved.
width	an integer. Filter OTU table by counts.
height	an integer. Filter OTU table by counts.

Value

This function returns a barplot of species richness for a given metadata variable.

Author(s)

Wen Chen.

See Also

```
specpool, specpool
```

Examples

```
data(ITS1, meta)
group.rich(ITS1, meta, "Crop")
```

group.spatial

Plot Spatial Collection Trends for Taxon Groups

Description

This function consumes an OTU table and its associated metadata table, and uses that information to produce a choropleth (essentially a heatmap, but superimposed imposed on an actual, cartographic map) to show how many counts of each taxon group were detected in each Canadian province/territory.

Usage

group.spatial 51

Arguments

data	the OTU table to be used.
meta	the metadata table to be used.
date.col	a character vector specifying which column in metadata contains the date information (see ${\sf RAM.dates}).$
province.col	a character vector specifying which column in metadata contains the province information (see Details).
rank	a character vector specifying the rank of the desired taxon groups. Note that all groups should come the same rank. (see RAM.rank.formatting).
group	a character vector giving the names of the groups to be plotted.
breaks	how many time segments should be plotted; see Details.
file	the file path where the image should be created. (see ?RAM.plotting).
ext	the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".
height	the height of the image to be created (in inches).
width	the width of the image to be created (in inches).

Details

This function currently only supports Canadian data. The entries in meta\$province.col should be specified as provinces, using the standard postal abbreviations (e.g. "Ontario" would be "ON").

The breaks argument is slightly buggy at the moment, possibly due to how R tries to split Date objects. breaks can be either an integer, in which case it will attempt to create that many levels (i.e. setting breaks=3 should split the data into three date 'blocks'.) breaks can also be a character vectors, such as "quarter" or "year" which attempts to split the date information accordingly. See cut.Date for more details and a complete specification of what is allowed for breaks.

Author(s)

Wen Chen and Joshua Simpson.

References

The file used to create the map of Canada is from GeoBase and is licensed under the Open Government License - Canada.

52 group.spec

group.spec	Boxplot Of Richness For Each Level Of A Given Metadata Variab	le
•		

Description

This function first use specpool to estimate the extrapolated species richness in a species pool (levels of metadata variable), and the number of unobserved species, then do a boxplot for percent of observed richness.

Usage

```
group.spec(otu, meta, factor, file=NULL, ext=NULL, width=8,height=8)
```

Arguments

otu	an OTU table.
meta	the metadata table to be used (must have same samples as data.
factor	a character string. Must be one of the metadata variables.
file	optional. Filename that the plot to be saved to.
ext	optional. Filename extension, type of image to be saved.
width	an integer. Filter OTU table by counts.
height	an integer. Filter OTU table by counts.

Value

This function returns a boxplot of species richness for a given metadata variable.

Author(s)

Wen Chen.

See Also

```
specpool, specpool
```

```
data(ITS1, meta)
group.spec(ITS1, meta, "Crop")
```

group.Taxa.bar 53

group.Taxa.bar	Barplot Of Taxa Distribution In Groups	

Description

This function do a barplot to show the distribution of selected taxa in each level of a given metadata variable

Usage

Arguments

data	a list of otu tables or taxonomic abundance matrices. See also RAM. input. formatting.
is.OTU	logical. If an OTU table was provided, is.OTU should be set as TRUE; otherwise, it should be set as FALSE.
rank	a single taxonomic rank. See also RAM.rank.formatting
taxa	a vector containing taxa names for plotting.
meta	the metadata table to be used (must have same samples as data.
meta.factor	a character string. Must be one of the metadata variables.
func	function to be used to aggregate count data, e.g. "sum" or "mean", default is "sum"
cex.y	size of y axis tick labels.
cex.x	size of x axis tick labels.
cex.main	size of title.
bar.width	width of bars
RAM.theme	customized ggplot_theme in RAM. See also ?theme_ggplot.
col.pal	color palettes to be used.
main	a character string. The title of the plot, default is an empty string.
file	filename to save the plot.
ext	filename extension, the type of image to be saved to.
width	an integer, width of the plot.
height	an integer, height of the plot.

Details

To use customized color palettes, must pass a vector of color names or hexadecimals. See examples for detail.

54 group.Taxa.box

Value

This function returns a barplot.

Author(s)

Wen Chen

Examples

```
data(ITS1, ITS2, meta)
taxa <- c("Fusarium", "Alternaria", "Cladosporium")</pre>
group.Taxa.bar(data=list(ITS1=ITS1, ITS2=ITS2), is.OTU=TRUE,
               rank="g", taxa=taxa, meta=meta, meta.factor="City",
               cex.y=5, cex.x=5, bar.width=1, RAM.theme=RAM.color())
## Not run:
# change default color schemes
col <- c("dodgerblue1", "darkcyan")</pre>
taxa.1 <- c("Fusarium", "Alternaria", "Cladosporium", "Verticillium",</pre>
           "Kondoa")
group.Taxa.bar(data=list(ITS1=ITS1, ITS2=ITS2), is.OTU=TRUE,
               rank="g", taxa=taxa.1, meta=meta, meta.factor="City",
               cex.y=5, cex.x=5, bar.width=1, RAM.theme=NULL,
               col.pal=col)
group.Taxa.bar(data=list(ITS1=ITS1, ITS2=ITS2), is.OTU=TRUE,
               rank="g", taxa=taxa.1, meta=meta, meta.factor="City",
               cex.y=5, cex.x=5, bar.width=1, RAM.theme=NULL,
               col.pal=col, func="mean")
# change ggplot theme
group.Taxa.bar(data=list(ITS1=ITS1, ITS2=ITS2), is.OTU=TRUE,
               rank="g", taxa=taxa.1, meta=meta, meta.factor="City",
               cex.y=5, cex.x=5, bar.width=1,
               col.pal=col, RAM.theme=RAM.border())
# save the plot
group.Taxa.bar(data=list(ITS1=ITS1, ITS2=ITS2), is.OTU=TRUE,
               rank="g", taxa=taxa.1, meta=meta, meta.factor="City",
               cex.y=5, cex.x=5, bar.width=1, RAM.theme=NULL,
               col.pal=col,main="", file="path/to/filename.pdf",
               ext="pdf", height=8, width=16)
## End(Not run)
```

group.Taxa.box

Boxplot Of Distribution Of Taxa In Each Level of A Metadata Variable

Description

This function do a boxplot to show the distribution of selected taxa in each level of a given metadata variable

group.Taxa.box 55

Usage

Arguments

data	a list of otu tables or taxonomic abundance matrices. See also RAM. input. formatting.
is.OTU	logical. If an OTU table was provided, is.OTU should be set as TRUE; otherwise, it should be set as FALSE.
rank	a single taxonomic rank. See also RAM.rank.formatting
taxa	a vector containing taxa names for plotting.
meta	the metadata table to be used (must have same samples as data.
meta.factor	a character string. Must be one of the metadata variables.
cex.y	size of y axis tick labels.
cex.x	size of x axis tick labels.
cex.main	size of title.
RAM.theme	customized ggplot_theme in RAM. See also ?theme_ggplot.
col.pal	color palettes to be used.
main	a character string. The title of the plot, default is an empty string.
file	filename to save the plot.
ext	filename extension, the type of image to be saved to.
width	an integer, width of the plot.
height	an integer, height of the plot.

Value

This function returns a boxplot of the distribution of seleted taxa within each level of a given metadata variable.

Author(s)

Wen Chen.

56 group.temporal

group.temporal

Plot Temporal Trends for Metadata and Taxon Groups

Description

This function consumes an OTU table and its associated metadata, and creates a plot showing how the collections of taxonomic groups, as well as metadata factors, evolve over time.

Usage

Arguments

the OTU table to be used.
the metadata table to be used.
a character vector specifying which column of the metadata has date information (see RAM. dates).
a named character vector specifying the names of the metadata columns to be plotted with the taxon group data. (see RAM.factors). NOTE: these factors must be <i>numeric</i> variables.
a character vector specifying the rank of the desired taxon groups. Note that all groups should come the same rank. (see RAM.rank.formatting).
a character vector giving the names of the groups to be plotted.
the file path where the image should be created (see ?RAM.plotting).
the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".
the height of the image to be created (in inches).
the width of the image to be created (in inches).

Details

The image created will contain several plots. It will always contain a large panel showing the counts collected for the specified taxon groups over time, and above that panel (on a common x-axis) will be a line graph for each metadata factor specified.

Note

If your data has collections being taken roughly annually, you may have a large amount of "empty space" in the middle of your plot. Consider subsetting the data by year, and plotting each year individually using this function.

group.venn 57

Author(s)

Wen Chen and Joshua Simpson

Examples

group.venn

Plot Venn Diagram For Two To Five Sets With Item Labels

Description

This function use draw.pairwise.venn to creates a venn diagram for two vectors

Usage

Arguments

vectors	a list of vectors with names. See also RAM.input.formatting.
cat.cex	size of the category names. (see venn. diagram for details).
cex	size of the label of the circles. (see venn.diagram for details).
cat.pos	optional. Location of the category names along the circles. (see venn.diagram for details).
cat.dist	optional. Distance of the category names to the circles. (see venn.diagram for details).
label	logical. If TRUE, will plot the item labels for 2 data sets. For more than 2 datasets or this is set as FALSE, the labels will be numbers for each circle. (see venn.diagram for details).
lab.cex	size of the labels.
lab.col	color of the labels.
fill	optional, color of the circles. (see venn.diagram for details).
file	the file path where the image should be created (see ?RAM.plotting).
ext	filename extension, the type of image to be saved to.
height	the height of the image to be created (in inches).
width	the width of the image to be created (in inches).

58 ITS1/ITS2

Value

group.venn returns a venn diagram for 2 to 5 sets. The user can choose to place item labels for 2 sets of data, however, the label locations can be wrong if the the smaller data set is part of the bigger data set, in this case, set label as FALSE. If the input datasets is more than 2, label will be ignored.

Author(s)

Wen Chen

See Also

```
see venn.diagram
```

Examples

ITS1/ITS2

Sample ITS1 and ITS2 Data

Description

Sample ITS1 and ITS2 OTU tables.

Usage

```
data(ITS1)
data(ITS2)
```

LCA.OTU 59

Format

A data frame with 4704 observations on the following 17 variables.

```
P1001.1M1, P1001.1M2, P1001.1M3, P1001.1M4, P1001.1M5, P1001.1M6, P1001.1M7, P1001.1M8, P1001.1M9, Collection samples.
```

taxonomy the taxonomic classification of the OTU.

Source

Wen Chen, AAFC-AAC

Examples

```
data(ITS1, ITS2)
str(ITS1)
str(ITS2)
```

LCA.OTU

Lowest Common Ancestor (LCA) OF EACH OTU

Description

This function consumes an OTU table and extract the LCA (lowest common ancestor) that each otu assigned to. See also tax.split.

Usage

```
LCA.OTU(otu, strip.format=FALSE, drop=TRUE)
```

Arguments

otu the OTU table to be used.

strip. format logical. Whether or not to remove the prefix of the taxonomy assignment at each

rank. see

drop logical. Whether or not drop taxonomic columns other than LCA.

Value

This function return a data frame same as the input OTU table, except the last column is the LCA of each otu, not the lineage. The taxonomy column can be kept, by using drop.

Note

tax.split returns the same OTU table with classification at a given taxonomic rank, which can be missing if an otu was not classified a that that taxonomic level. LCA.OTU, guaranteed that all OTUs will be preserved in the returned data table and provide the LCA for each OTU, although only higher taxonomic ranks were available.

60 location.formatting

Author(s)

Wen Chen

See Also

```
tax.split
```

Examples

```
data(ITS1)
## Not run:
# compare the following 2 commands:
# keep the rank prefix of the LCA column
ITS1.LCA <- LCA.OTU(ITS1, strip.format=TRUE, drop=TRUE)
# remove the rank prefix of the LCA column
ITS1.LCA <- LCA.OTU(ITS1, strip.format=FALSE, drop=TRUE)
## End(Not run)</pre>
```

location.formatting

Location Formatting

Description

Some functions in RAM expect to find a column with provincial/territorial data in the metadata. This data should use the standard Canadian provincial/territorial abbreviations:

- Alberta AB
- British Columbia BC
- Manitoba MB
- New Brunswick NB
- · Newfoundland and Labrador NL
- Novia Scotia NS
- Northwest Territories NT
- Nunavut NU
- Ontario ON
- Prince Edward Island PE
- Quebec QC
- Saskatchewan SK
- Yukon YT

Support for other locations is not available at this time.

Author(s)

Wen Chen and Joshua Simpson.

match.data 61

match.data

Match Samples In Ecology Data Sets and Metadata

Description

This function will match samples in ecology data sets, either OTU tables or taxonomy abundance matrices, and those in metadata. It makes sure that datasets contains same samples in the same order.

Usage

```
match.data(data, is.OTU=TRUE, meta)
```

Arguments

data a list of ecology data sets. if is.OTU is TRUE, they should be OTU tables, other-

wise should be taxonomy abundance matrices. See also RAM.input.formatting.

is.OTU logical, whether or not the ecology data sets are OTU tables.

meta metadata associated with input ecology data sets.

Author(s)

Wen Chen

See Also

```
RAM.input.formatting
```

62 meta

meta

Sample Metadata for ITS1/ITS2

Description

This data frame provides sample metadata for the ITS1/ITS2 data included in this package.

Usage

```
data(meta)
```

Format

A data frame with 16 observations on the following 10 variables.

Sample a factor with levels Sample1 Sample2 Sample3 Sample4 Sample5 Sample6 Sample7 Sample8

City a factor with levels City1 City2

Crop a factor with levels Crop1 Crop2

Plots a factor with levels 1 2

Harvestmethod a factor with levels Method1 Method2

Harvestdate a Date

Ergosterol_ppm a numeric vector

Province a character vector

Latitude a numeric vector

Longitude a numeric vector

Source

Wen Chen and Joshua Simpson.

Examples

```
data(meta)
```

str(meta)

META.clust 63

META.clust	Plot Hierarchical Cluster Of Metadata	

Description

This function plot hierarchical cluster Of metadata.

Usage

Arguments

meta	the metadata table to be clustered.
group	an integer or a metadata variable. If an integar, will cut tree into corresponding groups and color them accordingly; if a metadata variable was provided, tree leaves (sampleIDs) will be colored by each level.
data.trans	optional. If was provided, numeric data will be transformed. See also decostand
dist	optional. If was provided, distance matrix will be calculated using the give method for numeric variables; otherwise use vegdist default Bray-Curtis method. If metadata include qualitative variables, distance matrix will be calculated by gowdis.
clust	optional. If was not provided, will use the default agglomeration method used by hclust, i.e. "complete". Otherwise, will used user defined method for clustering. See also hclust.
type	optional. Can be one of the following: "triangle", "rectangle", "phylogram", "cladogram", "fan", "unrooted", "radial".
main	The title of the plot.
file	optional. Filename that the plot to be saved to.
ext	optional. File type that the plot to be saved to.
width	an integer, width of the plot.
height	an integer, height of the plot.

Value

This function return a plot of the hierarchical cluster analysis on a set of metadata.

Author(s)

Wen Chen

See Also

vegdist and gowdis.

network_data

Examples

```
data(meta)

META.clust(meta=meta, type="fan")

META.clust(meta=meta, type="fan", group="City")
```

network_data

Creates Nodes and Edge-List For An OTU Table

Description

This function creates nodes and edge-list for an otu table, which can be used for network plotting

Usage

```
network_data(data, is.OTU=TRUE, metadata)
```

Arguments

data an otu table of a species abundance matrix

is.OTU whether or not data is an otu table

metadata associated metadta of the otu table, should have same sampleIDs

Value

Return a list of network nodes and edges based on an otu table and the associated metadata

Author(s)

Wen Chen

```
data(ITS1, meta)
## Not run:
ITS1.01<-filter.OTU(data=list(ITS1=ITS1), percent=0.01)[[1]]

# create nodes and edges lists
b<-network_data(ITS1.01, is.OTU=TRUE, meta)
b_node<-b[[1]]
b_edge<-b[[2]]
head(b_node)
head(b_node)
head(b_edge)
library(igraph)
b1<-graph.data.frame(b_edge, directed=F)
lev <- levels(factor(E(b1)$Crop))</pre>
```

OTU.diversity 65

```
# vertices size
V(b1)$size<-degree(b1)
# vertices color
Crop1<-rownames(meta)[meta$Crop=="Crop1"]</pre>
Crop2<-rownames(meta)[meta$Crop=="Crop2"]</pre>
## vertices representing samples from crop1 will be in red,
## vertices representing samples from crop2 will be in blue;
## vertices representing taxa will be in pink
V(b1)$color<-ifelse((V(b1)$name
              ifelse((V(b1)$name
# edges color
col<-c("red", "blue")</pre>
for (i in 1:length(lev) ) { E(b1)$color[E(b1)$Crop==lev[i]] <- col[i]}
# plot
plot(b1, vertex.label.font=2,
         vertex.label.cex=0.5,
         layout=layout.kamada.kawai)
## End(Not run)
```

OTU.diversity

Summarize Diversity Indices for OTU Tables

Description

These functions calculate diversity indices for all samples and append outputs as new columns to metadata table.

Usage

```
OTU.diversity(data, meta)
```

Arguments

data a list of OTU tables.

meta the metadata to append the outputs.

Details

This function summarize the following diversity indices: specnumber, shannon, simpson, invsimpson, true diversity, evenness, chao and ACE indices, for a given otu table. See diversity.indices)

Value

This function return vectors of diversity indices for each sample, which are appended to a given metadata table.

66 OTU.ord

Note

Credit goes to package vegan for the partial argument matching (see References), and for the calculation of all diversity indices except for true diversity and evenness.

Author(s)

Wen Chen.

References

Jari Oksanen, F. Guillaume Blanchet, Roeland Kindt, Pierre Legendre, Peter R. Minchin, R. B. O'Hara, Gavin L. Simpson, Peter Solymos, M. Henry H. Stevens and Helene Wagner (2013). vegan: Community Ecology Package. R package version 2.0-10. http://CRAN.R-project.org/package=vegan

Diversity index. (2014, May 7). In Wikipedia, The Free Encyclopedia. Retrieved 14:57, May 28, 2014, from http://en.wikipedia.org/w/index.php?title=Diversity_index&oldid=607510424

Blackwood, C. B., Hudleston, D., Zak, D. R., & Buyer, J. S. (2007). Interpreting ecological OTU.diversity applied to terminal restriction fragment length polymorphism data: insights from simulated microbial communities. Applied and Environmental Microbiology, 73(16), 5276-5283.

Examples

```
data(ITS1, ITS2, meta)
data=list(ITS1=ITS1, ITS2=ITS2)
## Not run:
meta.diversity<-OTU.diversity(data=data, meta=meta)
## End(Not run)</pre>
```

OTU.ord

Ordination Plot For OTUs Using CCA or RDA Analysis

Description

This function consumes an OTU table, metadata factors, and graphing options, then produces a plot showing the cca or rda analysis of the OTU table.

Usage

OTU.ord 67

Arguments

otu the OTU table to be used.
meta the metadata table to be used.

factors a named character vector of length 1 or 2 specifying metadata factors for the

samples in the OTU table (see Details).

group a named character vector of length 1 or 2 specifying metadata factors for the

samples in the OTU table (see Details).

na.action choice of one of the following: "na.fail", "na.omit" or "na.exclude", see na.action

in cca for detail.

rank the rank to select the taxon groups at.

taxa an integer or a character vector of taxa names at the given rank. If it's an integer,

will display the top most abundant taxa; if it's a vector of taxa names, will plot

the selected taxa.

data.trans a method used to standardize the OTU table. One of "total", "max", "freq",

"normalize", "range", "standardize", "pa", "chi.square", "hellinger" or

"log" (see ?decostand).

plot.species whether plot sites or taxa, should be reflex to plot.scaling

plot.scaling one of the following: 1, 2, 3, or -1. See scaling in plot.cca for detail. See

also ordiplot

biplot.scale a numeric number, length of the biplot arrows

biplot.sig significance cutoff for biplot to be displayed. Currently disabled because in the

function, calculated ordination model cannot be passed to anova test.

biplot.label whether or not to plot biplot

mode one of the following: "cca" or "rda".

choice the chosen axes
main title of the plot
cex.point size of points

cex.leg size of lengend name cex.bp size of biplot labels

file the file path where the image should be created (see ?RAM.plotting).

ext the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".

width the width of the image to be created (in inches).

height the height of the image to be created (in inches).

Details

group should be a named character vector specifying the names of the columns to be used from meta (see RAM. factors). The values on the axes denote what fraction of the sum of all eigenvalues (i.e. from all axes) is explained by that (single) axis.

68 OTU.rarefy

Value

return a list of following: 1) ggplot object; 2) ordination model; 3) commodity data and 4) metadata used for the ordination model.

Author(s)

Wen Chen.

See Also

```
decostand, Taxa.ord, pcoa.plot
```

Examples

```
data(ITS1, meta)
its1<- filter.OTU(data=list(ITS1=ITS1), percent=0.001)[[1]]</pre>
factors=c("City", "Crop", "Harvestmethod", "Ergosterol_ppm")
## Not run:
# plot sites
ord1 <- OTU.ord(its1, meta=meta, data.trans="total",</pre>
        factors=factors, mode="cca", biplot.sig=0.1,
        taxa=20, biplot.scale=1.5, cex.point=5,
        plot.species=FALSE, rank="f", plot.scaling=3,
        group=c(City="City", Crop="Crop"))
# plot taxa
ord2 <- OTU.ord(its1, meta=meta, data.trans="total", plot.scaling=-1,</pre>
        factors=factors, mode="cca", biplot.sig=0.1,
        taxa=20, biplot.scale=3, cex.point=3,
        plot.species=TRUE, rank="g")
## End(Not run)
```

OTU.rarefy

Create Rarefied OTU Tables

Description

This function output rarefied OTU tables using rrarefy. This function may take long time for large dataset, e.g. over 100k otus x 45 samples.

Usage

```
OTU.rarefy(data, sample=NULL)
```

Arguments

```
data a list of otu tables. See also RAM.input.formatting. sample an integer represent the sampling size.
```

OTU.recap 69

Value

This function returns a list of rarefied otu tables.

Note

```
See also rrarefy
```

Author(s)

Wen Chen

See Also

```
RAM.input.formatting.
```

Examples

OTU.recap

Summarize OTU

Description

This function summarize OTU table at each given taxonomic ranks.

Usage

```
OTU.recap(data, ranks=c("p", "c", "o", "f", "g"),
brewer.pal="Pastel1", file=NULL, ext="pdf",
width=12, height=8)
```

Arguments

data	a list of otu tables. See also RAM.input.formatting.
ranks	a vector of taxonomic ranks. See also RAM.rank.formatting
brewer.pal	one of the color patterns available in RColorBrewer. See brewer.pal for available selections.
file	filename to save the plot.
ext	extention of the filename to save the plot.
width	width of the plot
height	heigth of the plot

70 pcoa.plot

Value

This function returns either a data frame or a list of data frames. If a single otu was provided, it returns the a dataframe with information of how many otuIDs and sequences being classified at selected taxonomic ranks. If more than 1 otu tables being provided, it returns a list, with the first a few are data frames of classification summary of each otu table, the last is a list showing taxa found only in one of the otu data set.

This function also generates a barplot for the percent classified otus and sequences at each given rank.

Note

warning is raised when run strsplit() and can be ignored.

Author(s)

Wen Chen

See Also

```
RAM.rank.formatting and RAM.input.formatting.
```

Examples

```
data(ITS1, ITS2)

ranks <- c("p", "c", "o", "f", "g")

df <- OTU.recap(data=list(ITS1=ITS1, ITS2=ITS2), ranks=ranks)
class(df)</pre>
```

pcoa.plot

Create a PCoA plot for an OTU Table

Description

This function consumes an OTU table, metadata factors, and graphing options, then produces a plot showing the PCoA analysis of the OTU table.

Usage

pcoa.plot 71

Arguments

data an OTU table or taxonomic abundance matrix to be used.
is.OTU logical. Whether or not the input data an OTU table.

meta the metadata table to be used.

factors a named character vector of length 1 or 2 specifying metadata factors for the

samples in the OTU table (see Details).

rank the rank to select the taxon groups at. For an OTU table, if rank is set NULL,

distance matrix will be calcuated using all OTUs, otherwise, the OTU table will be transformed to taxonomic abundance matrix before the calculation of the distance matrix. If a taxonomic abundance matrix is provided, i.e. is.OTU is set

TRUE, then the rank will be ignored.

stand.method a method used to standardize the OTU table. One of "total", "max", "freq",

"normalize", "range", "standardize", "pa", "chi.square", "hellinger" or

"log" (see ?decostand).

dist.method the dissimilarity index to be used; one of "manhattan", "euclidean", "canberra",

"bray", "kulczynski", "jaccard", "gower", "altGower", "morisita", "horn",

"mountford", "raup", "binomial", "chao", or "cao" (see vegdist).

sample.labels logical. Should the labels for the samples be displayed?

top how many taxon groups should be displayed, starting from the most abundant.

ellipse which of the metadata factors (if any) should have ellipses plotted around them.

Must be one of 1, 2, or FALSE (see Details).

main The title of the plot.

file the file path where the image should be created (see ?RAM.plotting).

ext the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".

height the height of the image to be created (in inches).
width the width of the image to be created (in inches).

ggplot2 logical. Should the ggplot2 package be used to produce the plot, or should the

base graphics be used? (see ?RAM.plotting).

bw logical. Should the image be created in black and white?

Details

This function uses pco in the labdsv package for the Principal coordinates analysis (PCoA). The distance matrix was square rooted before being passed to pco to avoid negative eigenvalues. factors should be a named character vector specifying the names of the columns to be used from meta (see RAM. factors). Those columns should be factors; if they are not, a warning is raised and they are coerced to factors (see factor). A warning is also raised when a factor has more than 9 levels, as that is the most colours the current palettes support.

The values on the axes denote what fraction of the sum of all eigenvalues (i.e. from all axes) is explained by that (single) axis.

When ellipse = FALSE, no ellipses will be plotted. When ellipse is a number, that 'number' metadata factor will have ellipses plotted. For example, if factors = c(Crop="Crop", City="City") and ellipse = 1, ellipses will be plotted for the different crops, but NOT the cities. Setting

72 pcoa.plot

factors = c(City="City") and ellipse = 2 is invalid, since there is no second metadata factor given. Ellipses can only be plotted for one factor currently. Furthermore, there need to be at least 3 samples for every level in every item in factors, otherwise ellipses cannot be plotted.

Value

When ggplot2 = TRUE, a ggplot object is returned; otherwise nothing ism returned (but the plot is shown on screen).

Note

The labels for the sample points are placed above, below, or next to the point itself at random. If labels are outside of the plotting area, or overlapping with each other, run your command again (without changing any arguments!) and the labels should move to new positions. Repeat until they are placed appropriately. This is done to ensure even tightly-grouped samples, or samples near the edge of the plot, have their labels shown. If the labels are too distracting, remember that they can be turned off by setting sample.labels = FALSE.

Author(s)

Wen Chen and Joshua Simpson.

See Also

vegdist

```
data(ITS1, meta)
# The argument for factors is a vector of length two; the first
# item is # Crop, which is a column from meta, and the second item
# is City, another # column from meta.
pcoa.plot(ITS1, meta=meta, rank="c",
          factors=c(Crop="Crop", City="City"))
## Not run:
# If you want to customize legend labels and plot the top 20 taxon
# groups at genus:
pcoa.plot(ITS1, meta=meta, rank="g", main="PCoA plot",
          factors=c(Place="City", Harvest_Method="Harvestmethod"))
# In black & white, using base graphics:
pcoa.plot(ITS1, meta=meta, rank="c", factors=c(Plot="Plots"),
          ggplot=F, bw=T)
pcoa.plot(ITS1, meta=meta, rank="c", factors=c(Plot="Plots"),
          ggplot=F, bw=T, dist.method="euc", stand.method="hell")
# Focus on the samples: hide all groups and plot ellipses for Crop:
pcoa.plot(ITS1, meta=meta, rank="g",
```

percent.classified 73

percent.classified

Calculate Percent of OTUs Classified at a Given Taxonomic Rank

Description

This function consumes an OTU table, and a vector containing taxnomic ranks, then returns what percent of OTUs in the given table are classified at each taxonomic rank.

Usage

```
percent.classified(data, ranks=c("f","g"))
```

Arguments

data a list of OTU tables to be processed. See also RAM.input.formatting

ranks a vector containing the taxonomic ranks you are interested in (see ?RAM.rank.formatting

for formatting details).

Value

A list of numeric vectors, containing the result for each taxonomic rank.

Author(s)

Wen Chen and Joshua Simpson.

```
data(ITS1, ITS2)
data <- list(ITS1=ITS1, ITS2=ITS2)
# find what percent of OTUs classified at family and genus levels
percent.classified(data=data, ranks=c("f","g"))</pre>
```

74 phylog_taxonomy

phylog_taxonomy	Plot Hierarchical Taxonomic Tree
priyiog_taxoriomy	I tot IIterarenteat Iaxonomic Iree

Description

This function plots hierarchical taxonomic tree, the leaves are taxa at a give rank, nonsplitting nodes are not collapsed as ape::plot.phylo does.

Usage

Arguments

guments	
otu	an otu table.
rank	optional. If no rank was provided, the data will be used as it is, if rank is provided, if data is an OTU table, it will be converted to taxonomic abundance matrix at the given rank, no change will be made for a data that has already been a taxonomic abundance matrix. See also tax.abund and data.revamp
rank.sep	the delimiter that separate the taxonomic ranks in the otu table; default is "; " (semi colon and a white space)
meta	the metadata table associated with otu table, samples must be in the same order in both otu table and metadata.
factors	the metadata variables, must be categories
sel.taxon	optional, a selected taxon at higher taxonomic rank (i.e. at phylum level). If given, only descendents of this taxa will be plotted.
sel.rank	the rank of sel.taxon
root	optional, the name of the root of the tree. If not given, will use "root"
cleaves	see ?ade4::plot.phylog
cnodes	see ?ade4::plot.phylog
clabel.leaves	see ?ade4::plot.phylog
clabel.nodes	see ?ade4::plot.phylog
f.phylog	see ?ade4::plot.phylog
sub	see ?ade4::plot.phylog
csub	see ?ade4::plot.phylog
possub	see ?ade4::plot.phylog
draw.box	see ?ade4::plot.phylog

phylo_taxonomy 75

Author(s)

Wen Chen

Examples

```
data(ITS1, meta)
## Not run:
ITS1.1<-filter.OTU(data=list(ITS1=ITS1), percent=0.01)[[1]]</pre>
factors=c("Crop", "City")
phylog_taxonomy(otu=ITS1.1, rank="family", rank.sep="; ",
                meta=meta, factors=NULL, sel.taxon=NULL,
                sel.rank=NULL, cleaves=1, cnodes=.5, root="k__Fungi",
                clabel.leaves = 0.5, clabel.nodes = 0.5,
                f.phylog = 0.8, sub = TRUE, csub = 1.25,
                possub = "bottomleft", draw.box = TRUE)
phylog_taxonomy(otu=ITS1.1, rank="family", rank.sep="; ";
                meta=meta, factors=c("Crop", "City", "Plots"),
                sel.taxon=NULL, sel.rank=NULL, cleaves=1,
                cnodes=.5, clabel.leaves = 0.5, clabel.nodes = 0.5,
                f.phylog = 0.8, sub = TRUE, csub = 1.25,
                possub = "bottomleft", draw.box = TRUE)
## End(Not run)
```

phylo_taxonomy

Plot Hierarchical Taxonomic Tree with Relative Abundance

Description

This function plots hierarchical taxonomic tree with relative abundance of all taxa at a give rank if category variables are provided. Nonsplitting nodes are collapsed as ape::plot.phylo does.

Usage

Arguments

```
otu an otu table.
rank taxonomic ranks, see ?RAM.rank.formatting
```

76 phylo_taxonomy

rank.sep the delimiter that separate the taxonomic ranks in the otu table; default is "; "

(semi colon and a white space)

meta the metadata table associated with otu table, samples must be in the same order

in both otu table and metadata.

factors the metadata variables, must be categories

plot.type tree type, default is phylogram. see ?plot.phylo

edge.width see ?ape::plot.phylo
cex size of tip labels.
font font of tip labels

x.lim a numeric vector of length one or two giving the limit(s) of the x-axis. see

?ape::plot.phylo

tip.offset the distance between tips of the phylogeny and their corresponding labels, see

?plot.phylo

tip.cex size of the pies

thermo add pies or thermometers to the tips
thermo.horiz orientiation of the thermometers
thermo.width width of the thermometers

thermo.height height of the thermometers node.frame type of frame around the nodes

node.bg background color of text frames of nodes

node.col color of the nodes

node.width width of the text frames of nodes node.height height of the text frames of nodes

node.cex size of text of nodes node.font font of text of nodes

Author(s)

Wen Chen

RAM.dates 77

```
display.pal(res[[1]][[i]])
}
par()
## End(Not run)
```

RAM.dates

Date Formatting for RAM

Description

This help page details the expected format for dates in RAM.

Details

For all functions expecting some type of date data, you will need to specify which column of the metadata table contains that information. The date information will likely be encoded as a character vector from read.meta, so these functions will try to coerce it to a Date object (see Date and as.Date), with a warning. These functions are expecting the date information to be in YYYY-MM-DD format.

RAM. factors

Factor Formatting for RAM

Description

This help page details how to pass metadata arguments in RAM.

Details

Many functions will expect arguments such as meta and factors (possibly row. factor or col. factor). These functions are expecting the full metadata table for meta (which you probably read into R using read.meta). The other argument, factor, should be a named character vector. The values of this vector should be the columns to be used from meta, while the names of the vector should be the labels you wish to have displayed in the plots. There are several ways to name a character vector:

Notice that myvec has *names* "This", "a", "character", but has *values* "is", "named", "vector". It is the names that will be used to label graphs in RAM, but the values that will be used to extract the actual data. This is useful if you have more complicated names; say we want the data from the column named "Precip_14d_before_harvest", but we want a nicer label for the plot. We can do the following:

```
> my.vec <- "Precip_14d_before_harvest"
> names(my.vec) <- "Precipitation (14 d. prior to Harvest, mm)"
```

Now we will be able to plot the value of the "Precip_14d_before_harvest" column, but we will have the (much nicer!) label "Precipitation (14 d. prior to Harvest, mm)" appear in our plots.

RAM.input.formatting Data Input Formatting

Description

When use some RAM functions for the comparison of multiple OTU tables or taxonomic abundance matrices, the user needs to privde all input data sets as list with names being provided.

```
one data set: data=list(data=otu)
multiple data sets: data=list(data1=otu1, data2=otu2, data3=otu3)
an OTU table: a data frame of otuIDs x sampleIDs with the last column named "taxonomy"
a taxonomy abundance matrix: a data frame of sampleIDs x taxa (e.g. species)
is.OTU: logical, many functions in RAM require the user to set is.OTU to be TRUE for OTU tables
```

is.OTU: logical, many functions in RAM require the user to set is.OTU to be TRUE for OTU tables or FALSE for a taxonomy abundance matrices.

Author(s)

Wen Chen.

RAM.pal 79

RAM.pal

Creat Color Palette

Description

This function creates color palette, especially if the number of colors required is more than 12.

Usage

```
RAM.pal(cols.needed=20)
```

Arguments

```
cols.needed an integer.
```

Author(s)

Wen Chen

Examples

```
col <- RAM.pal(40)</pre>
```

RAM.plotting

Creating Plots with RAM

Description

This help page details the standards for RAM plotting functions.

Overview

The RAM package contains many functions to produce plots and visualizations for metagenomic data. Currently, the plotting functions are grouped into 'casual' and 'publication' categories. The 'casual' plotting functions only accept a file argument and produce a .tiff file automatically. They are meant to quickly highlight certain aspects of the data, but are not meant to be published. The 'publication' quality plots accept many more graphing parameters, and should be of suitable quality for future publication. All 'publication' plots should accept the following parameters, in addition to those required to produce the plot:

- "file" the file name for the plot.
- "ext" the file type for the plot (see below).
- "height" the height of the plot, in inches.
- "width" the width of the plot, in inches.

80 RAM.plotting

Additionally, the following parameters are accepted by some functions:

• "bw" should the plot be in black and white?

For 'casual' plots, if file is not provided, the plot is displayed to the default graphics device (usually a new window), otherwise a .tiff file is created.

For 'publication' plots, if neither file nor ext are provided, the plot is displayed to the default graphics device (usually a new window). If both file and ext are provided, a file of type ext is created at file. If only one of file or ext is given, an error is raised.

In either case, the file extension will automatically be appended to the end of file, if file does not already end in the appropriate extension. For example, if file = ~/my/path.tiff and ext = png, the file will be called ~/my/path.tiff.png; but if file = ~/my/path.png, the file will just be called ~/my/path.png. Finally, if file = ~/my/path, the file will be called ~/my/path.png.

ggplot2

Furthermore, some of the 'publication' quality plots allow for a ggplot2 argument. If ggplot2 is TRUE, then the plot will be produced using the ggplot2 package, and a ggplot object will be returned. This allows for additional, personal customization of the plot for those who have used the package before. If ggplot2 is FALSE, then the plot will be created using the base plotting functions.

File Types

For 'publication' quality plots, the following file types are supported (use any of the following values for the ext argument): "pdf", "png", "tiff", "svg", "bmp", "jpeg".

Note

If file is given without a directory (e.g. file = my_fancy_file), then that file will be created in the current working directory (see ?getwd and ?setwd for more information).

The current default resolution is 1000 dpi for all plots.

See Also

ggplot

Author(s)

Wen Chen and Joshua Simpson.

RAM.rank.formatting 81

RAM.rank.formatting

Taxonomic Rank Formatting

Description

In all RAM functions requiring the user to input a taxonomic rank, three different formats for this taxon are accepted. All of these formats are simple character vectors (strings), and are provided for readability and convenience. The user only needs to specify any single element from any of the formats below. The formats are as follows:

```
Format 1: "kingdom", "phylum", "class", "order", "family", "genus", "species"
```

Format 2: "k", "p", "c", "o", "f", "g", "s"

Author(s)

Wen Chen and Joshua Simpson.

See Also

```
get.rank, tax.abund
```

read.meta

Open Metadata Table

Description

Opens the given file and return a data frame representing the metadata. This function use read.table to read in data; for large data sets, we recommend read.meta.

Usage

```
read.meta(file, sep=",")
```

Arguments

file a character vector specifying the file path to your file.

sep the character used to separate cells in the file.

Value

Returns a data frame with the information from the file. The first row and column are used for the names of the other rows and columns.

82 read.OTU

Author(s)

Wen Chen and Joshua Simpson

See Also

```
read.meta, read.table
```

Examples

```
## Not run:
    my.meta <- read.meta("path/to/meta")
## End(Not run)</pre>
```

read.OTU

Open OTU Table

Description

Opens the given file and returns a data frame representing the OTU table. This function use read.table function so is quite slow for large data sets, for which we recommend to use fread.OTU instead.

Usage

```
read.OTU(file, sep=",")
```

Arguments

file a character vector specifying the file path to your file.

sep the character used to separate cells in the file.

Value

Returns a data frame with the information from the file. The first row and column are used for the names of the other rows and columns.

Note

The OTU table should only contain classifications for the seven major taxonomic ranks, additional ranks will break some functions in the package. To remove those other classifications, try running

```
sed -i.backup -e 's/s[a-z]__[^;]*; //g' -e 's/t__[^;]*; //g' $FILE
```

where FILE is your OTU table. The letter t can be replaced in the second expression for any other letter which appears as a prefix. For example, adding -e 's/u__[^;]*; //g' before FILE would remove any entries formatted like u__test_classification; .

Additionally, if your OTU table starts with the entry #0TU ID, that cell needs to be removed before the table can be read with read.0TU.

reset.META 83

Author(s)

Wen Chen and Joshua Simpson.

See Also

```
getwd, setwd, read.table
```

Examples

```
## Not run:
my.OTU <- read.OTU("path/to/otu", sep=",")
## End(Not run)</pre>
```

reset.META

Reset OTU

Description

This function reset data type of metadata variables.

Usage

```
reset.META(meta, factor=NULL, numeric=NULL, date=NULL)
```

Arguments

meta	data frame. The metadata table to reset variable data type.
factor	a string or charactor vector, containing the column names of metadata variables to be set as factor. reset.META
numeric	a string or charactor vector, containing the column names of metadata variables to be set as numeric.
date	a string or charactor vector containing the column names of metadata variables

to be set as date.

Value

This function returns the same metadata with variables being reset to desired data type. Warnings or errors may be raise if the format of the original data cannot be recognized by R.

Author(s)

Wen Chen

84 sample.locations

Examples

sample.locations

Plot the Geographic Location of Samples

Description

This function consumes an OTU table, along with its associated metadata, and plots all the samples from that data as points on a map. The size of a point indicates the number of counts collected from that sample, while the colour of the point (optionally) shows a metadata factor for that sample.

Usage

Arguments

otu1	the OTU table to be used.
otu2	the (optional) second OTU table to be used.
meta	the metadata table to be used.
factor	(optional) a named character vector of length one specifying a column from the metadata table to be used to colour the points.
zoom	a positive integer in the range 3-21 (if source == "google") or 3-18 (if source == "osm") specifying the zoom for the map (low number means zoomed out).
source	the source to be used for the map; either "google" or "osm".
labels	a character vector giving one label per OTU.
lat.col	the name of the column in meta containing the latitude information.
long.col	the name of the column in meta containing the longitude information.
file	the file path where the image should be created (see ?RAM.plotting).
ext	the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".
height	the height of the image to be created (in inches).
width	the width of the image to be created (in inches).

sample.map 85

Details

Please note that this function is getting map information using either the Google Maps API or the OpenStreetMap API, and your usage is subject to the terms of those APIs.

Note

If you are getting a 403/503 error, that likely means that the current map provider is currently unavailable. This can be for a variety of reasons: if source == "google", you have likely maxed out your API call limit (this can be due to multiple users sharing an IP address; contact your system administrator for further details). If source == "osm", the server is likely under heavy load and unable to process your request. You can check the server load online. In either case, the issue will likely resolve itself. Try calling the function again in a few hours.

If you get a warning message of the form "Removed X rows containing missing values (geom_point).", this means that the current zoom level is too high to display some or all of the points. Try using a lower value forzoom.

Author(s)

Wen Chen and Joshua Simpson.

See Also

RAM. factors

Examples

```
data(ITS1, meta)
## Not run:
sample.locations(otu1=ITS1, otu2=ITS2, meta=meta, factor=c(Crop="Crop"))
## End(Not run)
```

sample.map

Plot The Geographic Location of Samples

Description

This function plot the number of samples collected from different locations that are DISTANT from each other, e.g. samples that collected from distant cities. This function is similar but not the same as sample.locations and sample.sites. The plot will also show the sample size of each location.

Usage

86 sample.map

Arguments

meta	the OTU table to be used.
siteID	IDs of sampling sites for each unique pair of longitude and latitude.
maptype	maptype to use, see also get_map.
shape	shape to be used, default is 16 (solid round dot).
colour	smooth gradient between two colours, default is c("red", "blue")
lat	latitude of each sampling location
lon	longitude of each sampling location
zoom	map zoom, an integer from 3 (continent) to 21 (building). see also get_map.
file	the file path where the image should be created (see ?RAM.plotting).
ext	the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".
height	the height of the image to be created (in inches).
width	the width of the image to be created (in inches).

Details

Please note that this function is getting map information using either the Google Maps API or the OpenStreetMap API, and your usage is subject to the terms of those APIs.

Note

If you are getting a 403/503 error, that likely means that the current map provider is currently unavailable. Try calling the function again in a few hours. If you get a warning message of the form "Removed X rows containing missing values (geom_point).", this means that the current zoom level is too high to display some or all of the points. Try using a lower value forzoom.

Author(s)

Wen Chen.

See Also

```
sample.locations, sample.sites
```

```
data(meta)
## Not run:
sample.map(meta=meta, zoom=8)
## End(Not run)
```

sample.sites 87

sample.sites	Plot The Geographic Location of Samples	

Description

This function plot the number of samples collected from different locations that are close to each other. This function is similar but not the same as sample.locations and sample.map.

Usage

Arguments

meta	the OTU table to be used.
siteID	IDs of sampling sites for each unique pair of longitude and latitude.
marker.size	maptype to use, see also get_map.
lat	latitude of each sampling location
lon	longitude of each sampling location
maptype	maptype to use, see also get_map.
zoom	map zoom, an integer from 3 (continent) to 21 (building). see also get_map.
file	the file path where the image should be created (see ?RAM.plotting).
ext	the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".
height	the height of the image to be created (in inches).
width	the width of the image to be created (in inches).

Note

This function is more suitable for plot sampling sites that are close to each other. If you are getting a 403/503 error, that likely means that the current map provider is currently unavailable. Try calling the function again in a few hours. If you get a warning message of the form "Removed X rows containing missing values (geom_point).", this means that the current zoom level is too high to display some or all of the points. Try using a lower value forzoom.

Author(s)

Wen Chen.

See Also

```
sample.locations, sample.map
```

seq_var

Examples

```
data(meta)
## Not run:
sample.sites(meta=meta, zoom=8)
## End(Not run)
```

seq_var

Plot Intra And Inter Specific Variation For An Alignment

Description

This function calculates and plots inter/intra specific variation for an alignment.

Usage

```
seq_var(taxon=NULL, region="ITS", align=NULL, file.align=NULL,
    file.align.format="fasta", outgroup.name=NULL,
    taxa.sep=":", col.total=5, col.genus=1, col.genus.syn=3,
    col.species=4, col.strain=5,
    box.cex.axis.text.x=6, box.cex.axis.text.y=6,
    box.cex.xlab=8, box.cex.ylab=8,
    intra.fill="purple", inter.fill="orange",
    den.cex.axis.text.x=6, den.cex.axis.text.y=6,
    den.cex.xlab=6, den.cex.ylab=6, frame.col="blue",
    table.cex=8, main=FALSE, main.cex=14,
    file=NULL, width=8, height=8)
```

Arguments

the name of the genus to be analyzed.
the DNA region of the alignment
optional, an alignment object in R environment
optional, a file of an alignment
nat
alignment file format, default is in FASTA format
the genus/genera names
the separator used to split alignment ID, default is ":". For example, as in "genus_name:seq_identifier:genus_synonym:species:strain"
using the taxa.sep, how many columns the alignment ID can be split in to.
genus name location
generic synonyms location
species name location
strain information location

seq_var 89

box.cex.axis.text.x

size of labels on x-axis of the boxplots

box.cex.axis.text.y

size of labels on y-axis of the boxplot

box.cex.xlab size of the xlab of boxplots

box.cex.ylab size of ylab of boxplots,

intra.fill color to fill the intra specific variation boxplot and density plot. Default is "pur-

ple".

inter.fill color to fill the intra specific variation boxplot and density plot. Default is "or-

ange",

den.cex.axis.text.x

size of labels on x-axis of the density plots

den.cex.axis.text.y

size of labels on y-axis of the density plots

den.cex.xlab size of the xlab of density plots

den.cex.ylab size of the ylab of boxplots

frame.col color of the frames separating inter and intra -specific variation plots and tables

table.cex the font size of the summary table

main whether or not to give a title to the plot, can be "FALSE", "TRUE", or a string

main.cex font size of the title of the plot

file whether or not save the plot to a PDF file.

width width of the plot

height height of the plot

Details

The sequence ID of the alignment to be analyzed should be in a similar format as follows: "Alternaria:gb_AF229477:Bipolaris:tetramera:strain_BMP_51_31_01", the separator, ":", and the location of genus / species / strain names can be changes or rearranged, as long as you provide such information to the function properly.

Value

This function calculate and plot inter/intra specific variation for an alignment.

Author(s)

Wen Chen & C. Andre Levesque

90 shared.OTU

Examples

```
data(alignment)
## Not run:
seq_var(taxon="Claviceps", region="ITS", align=alignment,
        file.align=NULL, outgroup.name=c("Talaromyces"),
       box.cex.axis.text.x=6, box.cex.axis.text.y=6, box.cex.xlab=8,
       box.cex.ylab=8, intra.fill="purple", inter.fill="orange",
       den.cex.axis.text.x=6, den.cex.axis.text.y=6, den.cex.xlab=6,
       den.cex.ylab=6, table.cex=8, frame.col="blue", main=TRUE,
        main.cex=14)
seq_var(taxon="Claviceps", region="ITS", align=NULL,
        file.align="/path/to/alignment_file", outgroup.name=c("Talaromyces"),
       box.cex.axis.text.x=6, box.cex.axis.text.y=6, box.cex.xlab=8,
       box.cex.ylab=8, intra.fill="purple", inter.fill="orange",
       den.cex.axis.text.x=6, den.cex.axis.text.y=6, den.cex.xlab=6,
       den.cex.ylab=6, table.cex=8, frame.col="blue", main=TRUE,
       main.cex=14)
## End(Not run)
```

shared.OTU

Summary of Shared OTUs Across ALL Subjects

Description

This function consumes OTU tables and returns a list summarizing information about the presence of the OTUs in samples.

Usage

```
shared.OTU(data)
```

Arguments

data

a list of OTU tables to be analyzed.

Value

shared.OTU returns a list containing the information calculated. The names associated with the list describe what that number represents; i.e. "#_of_OTUs_in_all_samples" shows how many OTUs in the given table were found to be present in all samples. The last item in the list is a character vector, containing the OTU number and taxonomic information of each OTU which was present in all samples. All entries in that column are of the form "OTU-taxonomic_classification".

Note

The OTUs are determined to be absent/present using the "pa" method from the function decostand.

shared.Taxa 91

Author(s)

Wen Chen and Joshua Simpson.

See Also

decostand

Examples

```
data(ITS1)
## Not run:
shared <- shared.OTU(data=list(ITS1=ITS1))
shared <- shared.OTU(data=list(ITS1=ITS1, ITS2=ITS2))
## End(Not run)</pre>
```

shared.Taxa

Summary of Shared Taxa Across ALL Subjects

Description

This function consumes OTU tables or a taxonomy matrices and returns a list summarizing information about the presence of the taxa in that table at a given taxonomic rank.

Usage

```
shared.Taxa(data, is.OTU=TRUE, rank="g")
```

Arguments

data a list of OTU tables or taxonomy abundance matrices to be analyzed.

is.OTU whether or not the input data are otu tables rank the taxonomic rank to be investigated

Value

shared. Taxa returns a list containing the information calculated. The names associated with the list describe what that number represents; i.e. "#_of_families_in_all_samples" shows how many taxa at the family level were found to be present in all samples. The last item in the list is a character vector, containing the taxon names of which were present in all samples.

Note

The taxa are determined to be absent/present using the "pa" method from the function decostand.

Author(s)

Wen Chen.

92 tax.abund

See Also

decostand

Examples

```
data(ITS1)
shared.Taxa(data=list(ITS1=ITS1))
## Not run:
g1 <- tax.abund(ITS1, rank="g", drop.unclassified=TRUE)
shared.Taxa(data=list(genus_ITS1=g1), rank="g", is.OTU=FALSE)
## End(Not run)</pre>
```

tax.abund

Aggregate OTU Data Based on Taxonomy

Description

This function consumes OTU table(s) and (optionally) a taxonomic rank, then extracts the classification of each OTU at the given taxonomic rank, groups by classification at the given rank, removes all groups with only 0 counts, optionally removes all unclassified groups, sorts groups based on abundance, and then returns the transpose.

Usage

Arguments

top

count

otu1	the first OTU table to be used.	
otu2	the second OTU table to be used.	
rank	a character vector representing a rank. Must be in one of three specific formats (see ?RAM.rank.formatting for help). If omitted, the function will repeat for all seven major taxonomic ranks.	
drop.unclassified		
	logical. Determine whether or not the OTUs labelled "unclassified" or missing classification at the given taxonomic rank should be excluded.	

the number of groups to select, starting with the most abundant. If NULL, all are

the number of groups to select, starting with the most abundant. If NULL, all are selected.

logical. Should the actual count of each OTU be shown, or should the relative

abundances be shown?

mode a character vector, one of "percent" or "number". If number, then top many

groups will be selected. If percent, then all groups with relative abundance in

at least one sample above top will be selected.

tax.abund 93

Value

The value returned by this function may become nested lists, so for convenience, any nested lists have been given descriptive items names to make accessing its elements simple (see Examples).

- If otu2 is given:
 - If rank is given: a list containing two data frames (otu1 and otu2 aggregated at the given rank).
 - If rank is not given: a list containing two lists. The first sublist represents otu1, the second otu2. The sublists contain seven data frames, the aggregation of the data at each taxonomic rank (see Examples).
- If otu2 is not given:
 - If rank is given: a single data frame (otu1 aggregated at the given rank).
 - If rank is not given: a list containing seven data frames (otu1 aggregated at each taxonomic rank).

Author(s)

Wen Chen and Joshua Simpson.

See Also

```
RAM.rank.formatting
```

```
data(ITS1, ITS2)
# aggregate based on phylum
ITS1.p <- tax.abund(ITS1, rank="p")

# aggregate based on all ranks; ignoring all unclassified OTUs
ITS1.taxa <- tax.abund(ITS1, drop.unclassified=FALSE)

# aggregate for one rank for both ITS1 and ITS2
lst <- tax.abund(ITS1, ITS2, rank="class")

# aggregate for all ranks for both ITS1 and ITS2
lst.all <- tax.abund(ITS1, ITS2)

stopifnot(identical(lst.all$otu1$phylum, ITS1.p))

# get the counts for all genera with relative abundance > 25%
tax.abund(ITS1, rank="g", top=25, mode="percent", count=TRUE)
```

94 tax.fill

tax.fill

Fill Missing Taxonomic Information

Description

This function consumes an OTU table and returns a new OTU table where the taxonomic classifications which are unidentified, unclassified, incertae sedis, or simply missing, are replaced with a more descriptive entry.

Usage

```
tax.fill(data, downstream = TRUE)
```

Arguments

data the OTU table to be used.

downstream logical. Should the replacement occur downstream, or upstream? (see Details)

Details

If downstream == TRUE, the function will start at the kingdom level and work its way down. Whenever an invalid group is encountered (i.e. one of "unclassified", "unidentified", "incertae_sedis", or simply missing, ignoring capitalization), the last known 'good' group will be substituted in the form "p__belongs_to_k_Fungi." If downstream == FALSE, the function will begin at the species level and work up.

This example should help clarify: given the taxonomy "k_Fungi; p_unidentified; c_Tremellomycetes", the new taxonomy is as follows (recall that an OTU table is required as input, and will be returned as output; this example simply shows the effect on the taxonomy):

- Downstream (Kingdom -> Species): "k__Fungi; p__belongs_to_k_Fungi; c__Tremellomycetes;
 o__belongs_to_c_Tremellomycetes; f__belongs_to_c_Tremellomycetes;
 s__belongs_to_c_Tremellomycetes"
- Upstream (Species -> Kingdom): "k__Fungi; p__belongs_to_c_Tremellomycetes; c__Tremellomycetes; o__belongs_to_no_taxonomy; f__belongs_to_no_taxonomy; g__belongs_to_no_taxonomy; s__belongs_to_no_taxonomy"

Usually, downstream = TRUE will provide a more useful output, however if the species is often known for your data, but other ranks are unknown, downstream = FALSE will provide a more descriptive taxonomy.

Value

Returns an OTU table as a data frame with the exact same numerical counts as data, but an updated taxonomy column.

Author(s)

Wen Chen and Joshua Simpson.

tax.split 95

See Also

```
RAM.rank.formatting
```

Examples

```
data(ITS1)
#\code{filter.OTU} returns a list
otu <- filter.OTU(data=list(ITS1=ITS1), percent=0.001)[[1]]
tax.fill(otu)</pre>
```

tax.split

Split OTU Tables By Taxonomic Rank

Description

This function consumes an OTU table and splits its taxonomy columns into the seven major taxonomic ranks. It returns a data frame preserving all numerical data, but changing the 'taxonomy' column to the name of the appropriate rank, and preserving only the classifications at that rank.

Usage

```
tax.split(otu1, otu2 = NULL, rank = NULL)
```

Arguments

otu1 the first OTU table to be used.

otu2 the second OTU table to be used.

rank the (optional) rank to split at and return (see ?RAM.rank.formatting for formatting details).

Value

The value returned by this function may become nested lists, so for convenience, any nested lists have been given descriptive items names to make accessing its elements simple (see Examples).

- If otu2 is given:
 - If rank is given: a list containing two data frames (otu1 and otu2 split at the given rank).
 - If rank is not given: a list containing two lists. The first sublist represents otu1, the second otu2. The sublists contain seven data frames, which are the data split at each taxonomic rank (see Examples).
- If otu2 is not given:
 - If rank is given: a single data frame (otu1 split at the given rank).
 - If rank is not given: a list containing seven data frames (otu1 split at each taxonomic rank).

96 Taxa.ord

Note

This function may seem similar to get.rank, but they are distinct. get.rank only returns the entries classified at that taxonomic rank, and so some OTUs might be omitted in the returned data frame. With tax.split, it is guaranteed that all OTUs will be preserved in the returned data table (although they may be missing taxonomic classification at that rank).

If no OTUs are classified at the given rank, the taxonomy column for that rank will be filled with empty strings.

Author(s)

Wen Chen and Joshua Simpson.

See Also

```
get.rank
```

Examples

```
data(ITS1, ITS2)
# split only ITS1 data at a single rank
ITS1.tax.p <- tax.split(ITS1, rank="phylum")
# split only ITS1 data at all ranks
ITS1.tax.all <- tax.split(ITS1)
# split ITS1 and IST2 data at a given rank
lst <- tax.split(ITS1, ITS2, rank="c")
# split ITS1 and ITS2 at every rank
lst.all <- tax.split(ITS1, ITS2)
stopifnot(identical(lst.all$otu1$phylum, ITS1.tax.p))</pre>
```

Taxa.ord

Ordination Plot For Taxa Groups Using CCA or RDA Analysis

Description

This function consumes an ecology data set, metadata factors, and graphing options, then produces a plot showing the vegan::ca or vegan::rda analysis.

Usage

Taxa.ord 97

```
mode=c("rda", "cca"), choice=c(1,2), main="",
cex.point=3, cex.label=1, cex.leg=12, cex.bp=3, cex.text=3,
file=NULL, ext=NULL, width=10, height=10)
```

Arguments

data an ecology data set, either an otu table or a taxonomy abundance matrix.

is.OTU whether or not the data an otu table

meta the metadata table to be used.

factors a named character vector of length 1 or 2 specifying metadata factors for the

samples in the OTU table (see Details).

group a named character vector of length 1 or 2 specifying metadata factors for the

samples in the OTU table (see Details).

rank the rank to select the taxon groups at.

taxa an integer or a character vector of taxa names at the given rank. if integer, plot

the top most abundant taxa, otherwise plot the taxa in the vector.

data.trans a method used to standardize the OTU table. One of "total", "max", "freq",

"normalize", "range", "standardize", "pa", "chi.square", "hellinger" or

"log" (see ?decostand).

plot. species whether plot sites or taxa, should be reflex to plot. scaling

plot.scaling one of the following: 1, 2, 3, or -1. See scaling in plot.cca for detail. See

also ordiplot

biplot.scale a numeric number, length of the biplot arrows

biplot.sig significance cutoff for biplot to be displayed. Currently disabled because in the

function, ordination model calcuated cannot be passed to anova test.

biplot.label whether or not to plot biplot

mode one of the following: "cca" or "rda".

choice the chosen axes

main title of the plot

cex.point size of points

cex.label size of taxa lables

cex.leg size of lengend name

cex.text size of taxon names if plot.species is set TRUE

cex.bp size of biplot labels

file the file path where the image should be created (see ?RAM.plotting).

ext the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".

width the width of the image to be created (in inches).

height the height of the image to be created (in inches).

98 Taxa.ord

Details

group should be a named character vector specifying the names of the columns to be used from meta (see RAM. factors). The values on the axes denote what fraction of the sum of all eigenvalues (i.e. from all axes) is explained by that (single) axis.

Value

return a list of following: 1) ggplot object; 2) ordination model; 3) commodity data and 4) metadata used for the ordination model.

Note

The labels for the taxa points are placed above, below, or next to the point itself at random. If labels are outside of the plotting area, or overlapping with each other, run your command again (without changing any arguments!) and the labels should move to new positions. Repeat until they are placed appropriately. This is done to ensure even tightly-grouped samples, or samples near the edge of the plot, have their labels shown. If the labels are too distracting, remember that they can be turned off by setting plot.species = FALSE.

Author(s)

Wen Chen.

See Also

decostand, OTU. ord, pcoa.plot

theme_ggplot 99

theme_ggplot

Customized Themes For GGPLOT

Description

RAM provides some customized ggplot themes to spice up your plots for presentations, but some of these additional features might be distractive and not be ideal for publications

Author(s)

Wen Chen

See Also

ggplot

Examples

```
## Not run:
data(ITS1, ITS2, meta)
data <- list(ITS1=ITS1, ITS2=ITS2)
# dissim.alleig.plot returns a ggplot2 object:
my.eig.plot <- dissim.alleig.plot(data)
my.eig.plot # view the plot

# update ggplot theme
require("grid")
new_theme <-RAM.color()
my.eig.plot <- my.eig.plot + new_theme
my.eig.plot
## End(Not run)</pre>
```

top.groups.plot

Plot the Top Taxon Groups

Description

These functions consume two OTU tables, along with (optionally) a file name and a parameter top. They create a box plot of the top number of OTUs (for plot.top.number), or all OTUs with relative abundance above top percent (for plot.top.percent) at the taxonomic ranks phylum, class, order, family, and genus.

100 top.groups.plot

Usage

Arguments

data a list of OTU tables.

top the number of OTUs to select (for top.number), or the minimum relative abun-

dance threshold to use for selecting OTUs (for top.percent).

ranks a vector of the taxonomic ranks. See also RAM.rank.formatting

drop.unclassified

logical. Should OTUs labelled "unclassified" or missing classification at the

given taxonomic rank be excluded?

cex.x optional. The size of the x axis names.

main the title of the plot

file the file path where the image should be created (see ?RAM.plotting).

ext the file type to be used; one of "pdf", "png", "tiff", "bmp", "jpg", or "svg".

height the height of the image to be created (in inches).
width the width of the image to be created (in inches).

bw logical. Should the image be created in black and white?

ggplot2 logical. Should the ggplot2 package be used to produce the plot, or should the

base graphics be used? (see ?RAM.plotting).

Note

Please be aware that the 'whiskers' in the plot may differ depending on the setting of ggplot2. Please see geom_boxplot boxplot, and boxplot.stats for more information on how the whiskers are calculated.

Author(s)

Wen Chen and Joshua Simpson.

See Also

RAM.plotting

transpose.LCA 101

Examples

transpose.LCA

Transpose OTU Tables With LCA Annotation For Each OTU

Description

Similar to transpose.OTU, but each OTU is annotated by the lowest common ancestor it was assigned to.

Usage

```
transpose.LCA(data)
```

Arguments

data

The OTU tables to be transposed. See also RAM. input. formatting.

Value

Returns a transposed OTU table, but the colname is formatted as: LCA_otuID.

Author(s)

Wen Chen.

```
data(ITS1, ITS2)
## Not run:
lca.t <- transpose.LCA(data=list(ITS1=ITS1, ITS2=ITS2))
## End(Not run)</pre>
```

102 valid.OTU

transpose.OTU

Take the Transpose of an OTU Table

Description

Returns the transpose of the given OTU table, excluding the last column (which should contain taxonomic information).

Usage

```
transpose.OTU(data)
```

Arguments

data

The OTU table to be transposed.

Value

Returns a data frame with rows equal to the columns of the original OTU, and columns equal to the rows of the original OTU. (Excluding the taxonomy column).

Author(s)

Wen Chen and Joshua Simpson.

Examples

```
data(ITS1)

ITS1.t <- transpose.OTU(ITS1)</pre>
```

valid.OTU

Validate an OTU Table

Description

This function consumes one or two OTU tables and checks if they are formatted properly and contain valid data.

Usage

```
valid.OTU(otu1, otu2 = NULL)
```

Arguments

otu1 the first OTU table to check.
otu2 the second OTU table to check.

valid.taxonomy 103

Value

If the table is not valid, an error will be raised with a description explaining the problem. If the table is valid, NULL will be returned invisibly.

Author(s)

Dr. Chen Wen and Joshua Simpson.

Examples

```
data(ITS1, ITS2)
valid.OTU(ITS1)
valid.OTU(ITS1, ITS2)
```

valid.taxonomy

Validate And Reformat The OTU Taxonomy Column

Description

A properly formatted taxonomy column of an otu table is critical for RAM functions to run properly. The taxonomy column of an otu table is composed of taxonomic lineages for otuIDs. RAM accept 7 ranks, including kingdom, phylum, class, order, family, genus and species, sub ranks are not supported. Taxa names at each rank should have prefix as "k_", "p_", "c_", "o_", _", "g_", and "s ", each rank should be separated by "; ", i.e. a semi colon and a white space, NOT just ";".

This function will check the format of the taxonomy column of the input of table and give suggetions that whether or not it needs to be reformatted using reformat.taxonomy of RAM.

However, RAM does accept missing ranks in lineages, as long as each rank is separated by "; " with proper prefix.

Usage

```
valid.taxonomy(data)
reformat.taxonomy(data, input.ranks, sep="; ")
```

Arguments

data a list of otu tables, see RAM.input.formatting.

input.ranks the ranks of the taxonomic lineages in the input otu tables.

sep the separator between each taxonomic rank in the lineage.

Author(s)

Wen Chen.

104 write.data

See Also

```
get.rank, tax.abund
```

Examples

write.data

Write Data To CSV File

Description

Creates a .csv-formatted file with the data. The file will be created in your current working directory (see ?getwd and ?setwd), unless specified otherwise by file. Note that if the file field does not end in .csv, ".csv" will be appended to the end of file.

Usage

```
write.data(data, file)
```

Arguments

data a data frame or matrix etc.

file The name of the .csv file to be created.

Author(s)

Wen Chen and Joshua Simpson.

See Also

```
write.csv, getwd, setwd
```

write.data 105

```
data(ITS1)
## Not run:
write.data(ITS1, "my_file_name.csv")
## End(Not run)
```

Index

*Topic IO	group.abund.Taxa,37
fread.meta, 34	group.abundance, 38
fread.OTU, 34	group.abundance.meta, 39
match.data,61	group.diversity,40
read.meta, 81	group.heatmap, 42
read.OTU, 82	group.heatmap.simple, 44
write.data, 104	group.indicators, 46
*Topic array	group.OTU,48
transpose.OTU, 102	group.rich,49
*Topic datagen	group.spatial,50
combine.OTU, 11	group.spec,52
core.OTU, 12	group.Taxa.bar,53
core.OTU.rank, 13	group.Taxa.box,54
core.Taxa, 14	group.temporal, 56
data.revamp, 18	group.venn,57
data.subset, 20	META.clust, 63
dissim, 21	OTU.ord, 66
filter.OTU,32	pcoa.plot, 70
filter.Taxa,33	RAM.pal, 79
shared.OTU, 90	sample.locations, 84
*Topic datasets	sample.map, 85
alignment, 5	sample.sites, 87
ITS1/ITS2, 58	Taxa.ord, 96
meta, 62	theme_ggplot, 99
*Topic error	top.groups.plot,99
valid.OTU, 102	*Topic manip
*Topic file	col.splitup,9
fread.meta, 34	diversity.indices, 27
fread.OTU, 34	filter.META, 31
match.data,61	get.rank,35
read.meta, 81	LCA.OTU, 59
read.OTU, 82	OTU.diversity,65
write.data, 104	percent.classified,73
*Topic hplot	reset.META,83
correlation, 15	tax.abund, 92
dissim.heatmap, 22	tax.fill, 94
dissim.plot, 24	tax.split,95
envis.NB, 28	transpose.LCA, 101
factor.abundance, 30	*Topic math

INDEX 107

OTU.rarefy, 68	dissim.eig.plot(dissim.plot), 24
OTU. recap, 69	dissim.GOF.plot(dissim.plot), 24
shared.Taxa, 91	dissim.heatmap, 22
*Topic models	dissim.ord.plot (dissim.plot), 24
assist.ado,5	dissim.plot, 22, 24
assist.NB, 7	dissim.pvar.plot(dissim.plot), 24
assist.ordination, 8	dissim.tree.plot(dissim.plot), 24
data.clust, 17	diversity, 41
network_data, 64	diversity.indices, 27, 65
phylo_taxonomy, 75	draw.pairwise.venn,57
phylog_taxonomy, 74	
seq_var, 88	envis.NB, 28
*Topic package	evenness, 41
RAM-package, 3	evenness (diversity.indices), 27
	factor, 23, 45, 71
adonis, 6	factor.abundance, 30
alignment, 5	filter.META, 31
annHeatmap2, 43	filter.OTU, 32
anova.cca, 9	filter.Taxa, 33
as.Date, 77	fread, <i>34</i> , <i>35</i>
assist.ado,5	fread.meta, 34
assist.cca (assist.ordination), 8	fread.OTU, 34, 82
assist.NB, 7, 28	
assist.ordination, 8	<pre>geom_boxplot, 100</pre>
assist.rda(assist.ordination), 8	get.rank, 35, 81, 96, 104
	get_map, <i>86</i> , <i>87</i>
boxplot, 100	getwd, <i>83</i> , <i>104</i>
boxplot.stats, 100	ggplot, 26, 49, 80, 99
brewer.pal,69	ggplot2,4
0.66.65	gowdis, <i>18</i> , <i>63</i>
cca, 9, 66, 67	group.abund.Taxa,37
col.splitup,9	group.abundance, 38, 39
combine.OTU, 11	group.abundance.meta,39
cor, 16	${\sf group.diversity,40}$
core.OTU, 12	group.heatmap, 42
core.OTU.rank, 13	group.heatmap.simple,44
core.Taxa, 14	group.indicators,46
correlation, 15	group.OTU,48
cut.Date, <i>51</i>	group.rich,49
1 . 17	group.spatial, $4,50$
data.clust, 17	group.spec,52
data.revamp, 6, 7, 17, 18, 74	group.Taxa.bar, <i>37</i> , <i>5</i> 3
data.subset, 20	group.Taxa.box,54
Date, 77	group.temporal, 56
decostand, 6, 12, 13, 15, 18, 19, 21–23, 25,	<pre>group.top.number(top.groups.plot), 99</pre>
43, 49, 63, 68, 90–92, 98	<pre>group.top.percent(top.groups.plot), 99</pre>
dissim, 21, 26	group.venn,57
dissim.alleig.plot(dissim.plot), 24	1 1 10 21 22 26 12 62
dissim.clust.plot(dissim.plot), 24	hclust, 18, 21, 22, 26, 43, 63

INDEX

heatmap.2, 23, 45	reformat.taxonomy, 103
	reformat.taxonomy (valid.taxonomy), 103
ITS1 (ITS1/ITS2), 58	reset.META,83
ITS1/ITS2, 58	rrarefy, 68, 69
ITS2 (ITS1/ITS2), 58	•
	sample.locations, 84, 85-87
LCA.OTU, 8, 19, 59, 59	sample.map, 85, 87
levelplot, <i>16</i>	sample.sites, 85, 86, 87
location.formatting, 60	seq_var, 4, 5, 88
	setwd, <i>83</i> , <i>104</i>
match.data, <i>11</i> , 61	shared.OTU, 90
meta, 62	shared. Taxa, 91
META.clust, 63	specpool, 49, 50, 52
multipatt, 47	Specpoo1, 49, 50, 52
· · · · · · · · · · · · · · · · · · ·	tax.abund, 7, 14, 17, 19, 33, 74, 81, 92, 104
network_data, 64	tax.fill, 94
ordiplot, <i>67</i> , <i>97</i>	tax.split, 59, 60, 95
OTU.cca (OTU.ord), 66	Taxa.cca (Taxa.ord), 96
OTU. diversity, 40, 41, 65	Taxa.ord, 68, 96
OTU. ord, 66, 98	Taxa.rda (Taxa.ord), 96
OTU.rarefy, 68	theme_ggplot,99
OTU.rda (OTU.ord), 66	top.groups.plot,99
OTU. recap, 69	transpose.LCA, 101
010.1 есар, 07	transpose.OTU, <i>101</i> , 102
pco, 71	true.diversity,41
pcoa.plot, 68, 70, 98	true.diversity(diversity.indices), 27
percent.classified, 73	
phylo_taxonomy, 75	valid.OTU, <u>102</u>
	valid.taxonomy, 103
phylog_taxonomy, 74	vegan, 4
plot.cca, 67, 97	vegdist, 6, 18, 21–23, 26, 43, 63, 71, 72
DAM (DAM-nackage) 2	venn.diagram, <i>57</i> , <i>58</i>
RAM (RAM-package), 3	5 , ,
RAM-package, 3	write.csv, <i>104</i>
RAM.border (theme_ggplot), 99	write.data, 104
RAM.color (theme_ggplot), 99	,
RAM. dates, <i>51</i> , <i>56</i> , 77	
RAM. factors, 23, 42, 45, 46, 56, 67, 71, 77,	
85, 98	
RAM.input.formatting, 12, 13, 20, 21, 24,	
27, 32, 37, 41, 46, 53, 55, 57, 61,	
68–70, 73, 78, 101	
RAM.pal, 79	
RAM.plotting, 79, 100	
RAM.rank.formatting, 19, 30, 36–39, 42, 51,	
53, 55, 56, 69, 70, 81, 93, 95, 100	
rda, <i>66</i>	
read.meta, 34, 77, 81, 81, 82	
read.OTU, <i>34</i> , <i>35</i> , 82	
read.table. <i>81–83</i>	