

Package ‘reconsi’

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Type Package

Title Resampling Collapsed Null Distributions for Simultaneous Inference

Version 1.23.0

Description Improves simultaneous inference under dependence of tests by estimating a collapsed null distribution through resampling. Accounting for the dependence between tests increases the power while reducing the variability of the false discovery proportion. This dependence is common in genomics applications, e.g. when combining flow cytometry measurements with microbiome sequence counts.

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VignetteBuilder knitr

biocViews Metagenomics, Microbiome, MultipleComparison, FlowCytometry

BugReports <https://github.com/CenterForStatistics-UGent/reconsi/issues>

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| | |
|----------|---|
| binStats | <i>Bin the test statistic into equally sized bins</i> |
|----------|---|

Description

Bin the test statistic into equally sized bins

Usage

```
binStats(z, nBins = 82L, binEdges = c(-4.1, 4.1))
```

Arguments

| | |
|----------|---|
| z | the matrix of permuted test statistics |
| nBins | an integer, the number of bins |
| binEdges | A vector of length 2 with the outer bin edges |

Value

Matrix of binned test statistics

| | |
|-------------|--|
| calcWeights | <i>Obtain weights as posterior probabilities to calculate the consensus null</i> |
|-------------|--|

Description

Obtain weights as posterior probabilities to calculate the consensus null

Usage

```
calcWeights(logDensPerm, fdr)
```

Arguments

| | |
|-------------|---|
| logDensPerm | A matrix with B rows of logged density estimates of the B permutation distributions, and p columns for the p observed test statistics |
| fdr | A vector of local false discovery rates for the observed tests statistics of length p |

Value

A vector of weights of length B

| | |
|-----------|---|
| estNormal | <i>Fast estimation of mean and standard deviation of a normal distribution, optionally with weights</i> |
|-----------|---|

Description

Fast estimation of mean and standard deviation of a normal distribution, optionally with weights

Usage

```
estNormal(y, w = NULL, p = length(y))
```

Arguments

| | |
|---|------------------------|
| y | vector of observations |
| w | optional weight vector |
| p | The number of features |

Value

A vector of length 2 with mean and standard deviation

| | |
|-------|---|
| estP0 | <i>Estimate the fraction of true null hypotheses.</i> |
|-------|---|

Description

Estimate the fraction of true null hypotheses.

Usage

```
estP0(statObs, fitAll, z0quantRange, smooth.df, evalVal, assumeNormal)
```

Arguments

| | |
|--------------|---|
| statObs | A vector of observed z-values |
| fitAll | the estimated normal null |
| z0quantRange | a number of quantiles between 0 and 0.5 |
| smooth.df | degrees of freedom for the spline smoother |
| evalVal | the value of q at which to evaluate the spline |
| assumeNormal | A boolean, should normality be assumed for the null distribution? |

Details

A natural spline is used over a range of intervals. Based on the `qvalue::qvalue()` function and Storey and Tibshirani, 2003

Value

The estimated null fraction, the value of the spline evaluated at the first element of `z0quantRange`

| | |
|----------------|--|
| getApproxCovar | <i>Obtain a null covariance matrix of binned test statistics</i> |
|----------------|--|

Description

Obtain a null covariance matrix of binned test statistics

Usage

```
getApproxCovar(statsPerm, ...)
```

Arguments

| | |
|-----------|--|
| statsPerm | The $p \times B$ matrix of permutation z-values in the columns |
| ... | passed on to <code>binStats</code> |

Value

The covariance matrix of binned z-values

Note

This is not the covariance matrix of the p test statistic, nor of the data! It is an approximate covariance matrix of binned test statistics for visualization and diagnostic purposes.

Examples

```
p = 200; n = 50; B = 5e1
x = rep(c(0,1), each = n/2)
mat = cbind(
  matrix(rnorm(n*p/10, mean = 5+x),n,p/10), #DA
  matrix(rnorm(n*p*9/10, mean = 5),n,p*9/10) #Non DA
)
mat = mat = mat + rnorm(n, sd = 0.3) #Introduce some dependence
fdrRes = reconsi(mat, x, B = B)
corMat = getApproxCovar(fdrRes$statsPerm)
```

| | |
|-----------|--|
| getC1prop | <i>Find the dependence pat C1 of the approximate covariance matrix, and extract the ratio of the first eigenvalue to the sum of all positive eigenvalues</i> |
|-----------|--|

Description

Find the dependence pat C1 of the approximate covariance matrix, and extract the ratio of the first eigenvalue to the sum of all positive eigenvalues

Usage

```
getC1prop(statsPerm, numEig = 1, ...)
```

Arguments

| | |
|-----------|---|
| statsPerm | Matrix of permuted test statistics |
| numEig | An integer, number of first eigenvalues |
| ... | passed onto binStats |

Value

A proportion indicating the ratio of the first eigenvalues to the sum of all eigenvalues

Examples

```

p = 200; n = 50; B = 5e1
x = rep(c(0,1), each = n/2)
mat = cbind(
  matrix(rnorm(n*p/10, mean = 5+x),n,p/10), #DA
  matrix(rnorm(n*p*9/10, mean = 5),n,p*9/10) #Non DA
)
mat = mat + rnorm(n, sd = 0.3) #Introduce some dependence
fdrRes = reconsi(mat, x, B = B)
getC1prop(fdrRes$statsPerm)

```

| | |
|--------|--|
| getFdr | <i>Calculate tail-area (Fdr) and local (fdr) false discovery rates, based on a certain null distribution</i> |
|--------|--|

Description

Calculate tail-area (Fdr) and local (fdr) false discovery rates, based on a certain null distribution

Usage

```

getFdr(
  statObs,
  fitAll,
  fdr,
  p,
  p0,
  zValsDensObs,
  smoothObs,
  assumeNormal,
  fitObs,
  ...
)

```

Arguments

| | |
|--------------|---|
| statObs | Vector of observed z-values |
| fitAll | The parameters of the estimated random null |
| fdr | local false discovery rate, already estimated |
| p | the number of hypotheses |
| p0 | The estimated fraction of null hypotheses |
| zValsDensObs | estimated densities of observed test statistics |
| smoothObs | A boolean, should estimated observed densities of the test statistics be used in estimating the Fdr |
| assumeNormal | A boolean, should normality be assumed for the null distribution? |
| fitObs | The kernel density estimate object of all test statistics |
| ... | more arguments, ignored |

Value

A list with components

Fdr Tail are false discovery rate
fdr Local false discovery rate

getG0 *Obtain the consensus null*

Description

Obtain the consensus null

Usage

```
getG0(
  statObs,
  statsPerm,
  z0Quant,
  gridsize,
  maxIter,
  tol,
  estP0args,
  testPargs,
  B,
  p,
  pi0,
  assumeNormal,
  resamAssumeNormal
)
```

Arguments

| | |
|-----------|---|
| statObs | A vector of length p with observed test statistics |
| statsPerm | A pxB matrix with permutation z-values |
| z0Quant | a vector of length of quantiles defining the central part R of the distribution. If a single number is supplied, then (z0quant, 1-z0quant) will be used |
| gridsize | An integer, the gridsize for the density estimation |
| maxIter | An integer, the maximum number of iterations in determining R |
| tol | The convergence tolerance. |
| estP0args | A list of arguments passed on to the estP0args() function |
| testPargs | A list of arguments passed on to quantileFun |
| B | an integer, the number of permutations |
| p | an integer, the number of hypotheses |

| | |
|-------------------|---|
| pi0 | A known fraction of true null hypotheses |
| assumeNormal | A boolean, should normality be assumed for the null distribution? |
| resamAssumeNormal | A boolean, should normality be assumed for resampling dists |

Value

A list with following entries

| | |
|--------------|--|
| PermDensFits | The permutation density fits |
| zSeq | The support of the kernel for density estimation |
| zValsDensObs | The estimated densities of the observed z-values |
| convergence | A boolean, has the algorithm converged? |
| weights | Vector of length B with weights for the permutation distributions |
| fdr | Estimated local false discovery rate along the support of the kernel |
| p0 | The estimated fraction of true null hypotheses |
| iter | The number of iterations |
| fitAll | The consensus null fit |

| | |
|--------------|---|
| getTestStats | <i>A function to calculate observed and permutation z-statistics on a n-by-p matrix of observations</i> |
|--------------|---|

Description

A function to calculate observed and permutation z-statistics on a n-by-p matrix of observations

Usage

```
getTestStats(
  Y,
  center,
  test = "wilcox.test",
  x,
  B,
  argList,
  tieBreakRan,
  replace,
  scale
)
```

Arguments

| | |
|-------------|--|
| Y | The nxp data matrix |
| center | a boolean, should data be centered prior to permutation |
| test | A function name, possibly user defined. See details. |
| x | A vector defining the groups. Will be coerced to factor. |
| B | an integer, the number of permutations |
| argList | A list of further arguments passed on to the test function |
| tieBreakRan | A boolean, should ties of permutation test statistics be broken randomly? If not, midranks are used |
| replace | A boolean. If FALSE, samples are permuted (resampled without replacement), if TRUE the samples are bootstrapped (resampled with replacement) |
| scale | a boolean, should data be scaled prior to resampling |

Details

For test "wilcox.test" and "t.test", fast custom implementations are used. Other functions can be supplied but must accept a y outcome variable, a x as grouping variable, and possibly a list of other arguments. It must return all arguments needed to evaluate its quantile function if z-values are to be used.

Value

A list with components

| | |
|-------------|--|
| statObs | A vector of length p of observed test statistics |
| statsPerm | A p-by-B matrix of permutation test statistics |
| resamDesign | The resampling design |

| | |
|----------|---|
| getTstat | <i>A function to obtain a t-test statistic efficiently. For internal use only</i> |
|----------|---|

Description

A function to obtain a t-test statistic efficiently. For internal use only

Usage

```
getTstat(y1, y2, mm, nn)
```

Arguments

| | |
|--------|--|
| y1, y2 | vectors of observed values in the two groups |
| mm, nn | number of observations in the corresponding groups |

Value

A list with items

tstat The t-test statistic
df The degrees of freedom (Welch approximation)

plotApproxCovar *Plot an approximation of the correlation structure of the test statistics*

Description

Plot an approximation of the correlation structure of the test statistics

Usage

```
plotApproxCovar(
  reconsiFit,
  col = colorRampPalette(c("yellow", "blue"))(12),
  x = seq(-4.2, 4.2, 0.1),
  y = seq(-4.2, 4.2, 0.1),
  xlab = "Z-values",
  ylab = "Z-values",
  nBins = 82L,
  binEdges = c(-4.1, 4.1),
  ...
)
```

Arguments

reconsiFit The reconsi fit
col, x, y, xlab, ylab, ...
 A list of arguments for the image() function.
nBins, binEdges passed on to the getApproxCovar function

Details

By default, yellow indicates negative correlaton between bin counts, blue positive correlation

Value

invisible()

Note

This is not the covariance matrix of the p test statistic, nor of the data! It is an approximate covari-
ance matrix of binned test statistics for visualization purposes. See [plotCovar](#) for the full covariance
matrix.

See Also

[plotCovar](#), [getApproxCovar](#)

Examples

```
p = 200; n = 50; B = 5e1
x = rep(c(0,1), each = n/2)
mat = cbind(
  matrix(rnorm(n*p/10, mean = 5+x),n,p/10), #DA
  matrix(rnorm(n*p*9/10, mean = 5),n,p*9/10) #Non DA
)
mat = mat + rnorm(n, sd = 0.3) #Introduce some dependence
fdrRes = reconsi(mat, x, B = B)
plotApproxCovar(fdrRes)
```

| | |
|-----------|--|
| plotCovar | <i>Plot an the covariance matrix of the test statistics estimated through permutations</i> |
|-----------|--|

Description

Plot an the covariance matrix of the test statistics estimated through permutations

Usage

```
plotCovar(
  reconsiFit,
  col = colorRampPalette(c("yellow", "blue"))(12),
  xlab = "Test statistic index",
  ylab = xlab,
  ...
)
```

Arguments

reconsiFit The reconsi fit
col, xlab, ylab, ...
 A list of arguments for the image() function.

Details

By default, yellow indicates negative correlaton between test statistics, blue positive correlation

Value

invisible()

Note

Note the difference with the plotApproxCovar function, where the covariances between binned test statistics are shown to get an idea between covariances between tail and center values of the univariate null distribution. Here the covariance matrix between all test statistics is shown

See Also

[plotApproxCovar](#)

Examples

```
p = 200; n = 50; B = 5e1
x = rep(c(0,1), each = n/2)
mat = cbind(
  matrix(rnorm(n*p/10, mean = 5+x),n,p/10), #DA
  matrix(rnorm(n*p*9/10, mean = 5),n,p*9/10) #Non DA
)
mat = mat = mat + rnorm(n, sd = 0.3) #Introduce some dependence
fdrRes = reconsi(mat, x, B = B)
plotCovar(fdrRes)
```

| | |
|----------|---|
| plotNull | <i>Plot the obtained null distribution along with a histogram of observed test statistics</i> |
|----------|---|

Description

Plot the obtained null distribution along with a histogram of observed test statistics

Usage

```
plotNull(
  fit,
  lowColor = "yellow",
  highColor = "blue",
  idNull = NULL,
  nResampleCurves = length(fit$Weights),
  hSize = 0.5
)
```

Arguments

| | |
|---------------------|---|
| fit | an object returned by the reconsi() (or testDAA()) function |
| lowColor, highColor | The low and high ends of the colour scale |
| idNull | indices of known null taxa |
| nResampleCurves | The number of resampling null distributions to plot |
| hSize | A double, the size of the line of the collapsed null estimate |

Value

a ggplot2 plot object

Examples

```
p = 180; n = 50; B = 1e2
#Low number of resamples keeps computation time down
x = rep(c(0,1), each = n/2)
mat = cbind(
  matrix(rnorm(n*p/10, mean = 5+x),n,p/10), #DA
  matrix(rnorm(n*p*9/10, mean = 5),n,p*9/10) #Non DA
)
#Provide just the matrix and grouping factor, and test using the random null
fdrRes = reconsi(mat, x, B = B)
plotNull(fdrRes)
```

| | |
|--------|--|
| ptEdit | <i>A custom function to calculate the distribution function of the t-test statistic. For internal use only</i> |
|--------|--|

Description

A custom function to calculate the distribution function of the t-test statistic. For internal use only

Usage

```
ptEdit(q)
```

Arguments

q a vector with t-statistic and degrees of freedom

Value

A value between 0 and 1, the evaluation of the cdf

| | |
|--------|--|
| qtEdit | <i>A custom function to calculate the quantile function of the t-test statistic. For internal use only</i> |
|--------|--|

Description

A custom function to calculate the quantile function of the t-test statistic. For internal use only

Usage

```
qtEdit(p)
```

Arguments

`p` a vector with quantile and degrees of freedom

Value

the corresponding quantile

| | |
|---------------------------|--|
| <code>quantCorrect</code> | <i>Correct quantiles by not returning 0 or 1</i> |
|---------------------------|--|

Description

Correct quantiles by not returning 0 or 1

Usage

```
quantCorrect(quants)
```

Arguments

`quants` A vector of quantiles

Value

The same vector of quantiles but without 0 or 1 values

| | |
|----------------------|---|
| <code>reconsi</code> | <i>Perform simultaneous inference through collapsed resampling null distributions</i> |
|----------------------|---|

Description

Perform simultaneous inference through collapsed resampling null distributions

Usage

```
reconsi(
  Y,
  x = NULL,
  B = 1000L,
  test = "wilcox.test",
  argList = list(),
  distFun = "pnorm",
  zValues = TRUE,
  testPargs = list(),
  z0Quant = 0.25,
```

```

    gridsize = 801L,
    maxIter = 100L,
    tol = 1e-06,
    zVals = NULL,
    center = FALSE,
    replace = is.null(x),
    assumeNormal = TRUE,
    estP0args = list(z0quantRange = seq(0.05, 0.45, 0.0125), smooth.df = 3, evalVal = 0.05),
    resamZvals = FALSE,
    smoothObs = TRUE,
    scale = FALSE,
    tieBreakRan = FALSE,
    pi0 = NULL,
    resamAssumeNormal = TRUE
  )

```

Arguments

| | |
|-----------|---|
| Y | the matrix of sequencing counts |
| x | a grouping factor. If provided, this grouping factor is permuted. Otherwise a bootstrap procedure is performed |
| B | the number of resampling instances |
| test | Character string, giving the name of the function to test for differential absolute abundance. Must accept the formula interface. Features with tests resulting in observed NA test statistics will be discarded |
| argList | A list of arguments, passed on to the testing function |
| distFun | the distribution function of the test statistic, or its name. Must at least accept an argument named 'q', 'p' and 'x' respectively. |
| zValues | A boolean, should test statistics be converted to z-values. See details |
| testPargs | A list of arguments passed on to distFun |
| z0Quant | A vector of length 2 of quantiles of the null distribution, in between which only null values are expected |
| gridsize | The number of bins for the kernel density estimates |
| maxIter | An integer, the maximum number of iterations in the estimation of the null distribution |
| tol | The tolerance for the infinity norm of the central borders in the iterative procedure |
| zVals | An optional list of observed (statObs) and resampling (statsPerm) z-values. If supplied, the calculation of the observed and resampling test statistics is skipped and the algorithm proceeds with calculation of the consensus null distribution |
| center | A boolean, should observations be centered in each group prior to permutations? See details. |
| replace | A boolean. Should resampling occur with replacement (bootstrap) or without replacement (permutation) |

| | |
|-------------------|--|
| assumeNormal | A boolean, should normality be assumed for the null distribution? |
| estP0args | A list of arguments passed on to the estP0 function |
| resamZvals | A boolean, should resampling rather than theoretical null distributions be used? |
| smoothObs | A boolean, should the fitted rather than estimated observed distribution be used in the Fdr calculation? |
| scale | a boolean, should data be scaled prior to resampling |
| tieBreakRan | A boolean, should ties of resampling test statistics be broken randomly? If not, midranks are used |
| pi0 | A known fraction of true null hypotheses. If provided, the fraction of true null hypotheses will not be estimated. Mainly for oracle purposes. |
| resamAssumeNormal | A boolean, should normality be assumed for resampling dists |

Details

Efron (2007) centers the observations in each group prior to permutation. As permutations will remove any genuine group differences anyway, we skip this step by default. If `zValues = FALSE`, the density is fitted on the original test statistics rather than converted to z-values. This unlocks the procedure for test statistics with unknown distributions, but may be numerically less stable.

Value

A list with entries

| | |
|--------------|--|
| statsPerm | Resampling Z-values |
| statObs | Observed Z-values |
| distFun | Density, distribution and quantile function as given |
| testPargs | Same as given |
| zValues | A boolean, were z-values used? |
| resamZvals | A boolean, were the resampling null distribution used? |
| cdfValObs | Cumulative distribution function evaluation of observed test statistics |
| p0estimated | A boolean, was the fraction of true null hypotheses estimated from the data? |
| Fdr, fdr | Estimates of tail-area and local false discovery rates |
| p0 | Estimated or supplied fraction of true null hypotheses |
| iter | Number of iterations executed |
| fitAll | Mean and standard deviation estimated collapsed null |
| PermDensFits | Mean and standard deviations of resamples |
| convergence | A boolean, did the iterative algorithm converge? |
| zSeq | Basis for the evaluation of the densities |
| weights | weights of the resampling distributions |
| zValsDensObs | Estimated overall densities, evaluated in zSeq |

Note

Ideally, it would be better to only use unique resamples, to avoid unnecessary replicated calculations of the same test statistics. Yet this issue is almost always ignored in practice; as the sample size grows it also becomes irrelevant. Notice also that this would require to place weights in case of the bootstrap, as some bootstrap samples are more likely than others.

Examples

```
#Important notice: low number of resamples B necessary to keep
# computation time down, but not recommended. Pray set B at 200 or higher.
p = 60; n = 20; B = 5e1
x = rep(c(0,1), each = n/2)
mat = cbind(
matrix(rnorm(n*p/10, mean = 5+x), n, p/10), #DA
matrix(rnorm(n*p*9/10, mean = 5), n, p*9/10) #Non DA
)
fdrRes = reconsi(mat, x, B = B)
fdrRes$p0
#Indeed close to 0.9
estFdr = fdrRes$Fdr
#The estimated tail-area false discovery rates.

#With another type of test. Need to supply quantile function in this case
fdrResLm = reconsi(mat, x, B = B,
test = function(x, y){
fit = lm(y~x)
c(summary(fit)$coef["x","t value"], fit$df.residual)},
distFun = function(q){pt(q = q[1], df = q[2])})

#With a test statistic without known null distribution(for small samples)
fdrResKruskal = reconsi(mat, x, B = B,
test = function(x, y){
kruskal.test(y~x)$statistic}, zValues = FALSE)

#Provide an additional covariate through the 'argList' argument
z = rpois(n , lambda = 2)
fdrResLmZ = reconsi(mat, x, B = B,
test = function(x, y, z){
fit = lm(y~x+z)
c(summary(fit)$coef["x","t value"], fit$df.residual)},
distFun = function(q){pt(q = q[1], df = q[2])},
argList = list(z = z))

#When nog grouping variable is provided, a bootstrap is performed
matBoot = cbind(
matrix(rnorm(n*p/10, mean = 1), n, p/10), #DA
matrix(rnorm(n*p*9/10, mean = 0), n, p*9/10) #Non DA
)
fdrResBoot = reconsi(matBoot, B = B,
test = function(y, x){testRes = t.test(y, mu = 0, var.equal = TRUE);
c(testRes$statistic, testRes$parameter)},
distFun = function(q){pt(q = q[1], df = q[2])},
```

center = TRUE, replace = TRUE)

| | |
|-------------|---|
| rowMultiply | <i>A function to efficiently row multiply a a-by-b matrix by a vector of length b. More memory intensive but that does not matter with given matrix sizes</i> |
|-------------|---|

Description

A function to efficiently row multiply a a-by-b matrix by a vector of length b. More memory intensive but that does not matter with given matrix sizes

Usage

```
rowMultiply(matrix, vector)
```

Arguments

| | |
|--------|--------------------------------------|
| matrix | a numeric matrix of dimension a-by-b |
| vector | a numeric vector of length b |

Details

t(t(matrix)*vector) but then faster

Value

a matrix, row multiplied by the vector

| | |
|---------|---|
| stabExp | <i>A function to numerically stabilize an exponentiation. For internal use only</i> |
|---------|---|

Description

A function to numerically stabilize an exponentiation. For internal use only

Usage

```
stabExp(exps)
```

Arguments

| | |
|------|--------------------------------|
| exps | the vector to be exponentiated |
|------|--------------------------------|

Value

the vector with the maximum subtracted

| | |
|---------|--|
| testDAA | <i>A function to test for differential absolute abundance on a phyloseq object</i> |
|---------|--|

Description

A function to test for differential absolute abundance on a phyloseq object

Usage

```
testDAA(Y, ...)

## S4 method for signature 'phyloseq'
testDAA(Y, groupName, FCname, ...)

## S4 method for signature 'matrix'
testDAA(Y, FC, x, S = rowSums(Y), tieBreakRan = TRUE, assumeNormal = TRUE, ...)
```

Arguments

| | |
|--------------|--|
| Y | A phyloseq object, or a data matrix with samples in the rows and OTUs in the columns |
| ... | passed on to the reconsi() function |
| groupName | A character string, the name of a variable in physeq indicating the grouping factor |
| FCname | A character string, the name of a variable in physeq containing the total cell count |
| FC | a vector of length n with total flow cytometry cell counts |
| x | a grouping factor of length n |
| S | a vector of library sizes. Will be calculated if not provided |
| tieBreakRan | A boolean, should ties be broken at random. |
| assumeNormal | A boolean, should normality be assumed for the null distribution? |

Value

See the reconsi() function

Examples

```
#Test for phyloseq object
library(phyloseq)
data("VandeputteData")
VandeputtePruned = prune_samples(Vandeputte,
samples = sample_names(Vandeputte)[20:40])
testVanDePutte = testDAA(VandeputtePruned, "Health.status", "absCountFrozen",
```

```
B = 15)
#Test for matrix
testMat = testDAA(as(otu_table(VandeputtePruned), "matrix"),
  get_variable(VandeputtePruned, "Health.status"),
  get_variable(VandeputtePruned,"absCountFrozen"), B = 15)
```

Vandeputte

Microbiomes of Crohn's disease patients and healthy controls

Description

Microbiome sequencing data of Crohn's disease patients, and healthy controls, together with other baseline covariates. Both sequencing and flow cytometry data are available.

Usage

Vandeputte

Format

A phyloseq object with an OTU-table and sample data

otu_table Count data matrix of 234 taxa in 135 samples

sample_data Data frame of patient covariates

Source

<https://www.ncbi.nlm.nih.gov/pubmed/29143816>

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