Package ‘miRtest’

October 13, 2022

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Title Combined miRNA- And mRNA-Testing
Type Package
LazyLoad yes
Author Stephan Artmann, Klaus Jung, Tim Beissbarth
Description Package for combined miRNA- and mRNA-testing.
Version 2.0
Date 2022-05-23
Depends corpcor, MASS
Imports globaltest, GlobalAncova, limma
Collate ‘miRtest.R’
NeedsCompilation no
Repository CRAN
Date/Publication 2022-05-23 20:50:02 UTC

R topics documented:

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contingency.table

Description

Part of expression data from Nielsen et al.

Author(s)

Stephan Artmann taken from Nielsen et al.

contingency.table

Description


Usage

contingency.table(gene.set, p.val, sign=0.05)

Arguments

gene.set Vector of gene sets.
p.val Vector with p-values.
sign Significance threshold.

Author(s)

Stephan Artmann
Description
Fisher method of p value combination.

Usage
fisher.combination(p1, p2, check.range=FALSE)

Arguments
p1, p2  one-sided p-values that shall be combined.
check.range  If set to "TRUE" values above 1 will be set to 1.

Value
Combined p-value.

Author(s)
Stephan Artmann

generate.A  Turn a data...

Description
Turn a data.frame indicating gene sets into the allocation matrix.

Usage
generate.A(df, X, Y, verbose=TRUE)

Arguments
df  data.frame with mRNAs in its first and miRNAs in its second column.
X  Expression matrix of miRNAs whose row names will be used to generate the list of miRNAs.
Y  Expression matrix of mRNAs whose row names will be used to generate the list of mRNAs.
verbose  Logical. Shall progress be printed?
**Value**

Allocation matrix A necessary for "miR.test" function.

**Author(s)**

Stephan Artmann

**Examples**

```
# Generate random expression data of 3 miRNAs
# with 8 replicates
set.seed(1)
X = rnorm(24);
dim(X) = c(3,8);
rownames(X) = 1:3;
# Generate random mRNA expression data with 20 mRNAs
# and 10 replicates
Y = rnorm(200);
dim(Y) = c(20,10);
rownames(Y) = 1:20;
# Let's assume that we want to compare 2 miRNA groups, each of 4 replicates:
# ... and that the corresponding mRNA experiments had 5 replicates in each group
group.miRNA = factor(c(1,1,1,1,2,2,2,2));
group.mRNA = factor(c(1,1,1,1,1,2,2,2,2,2));

library(miRtest)
# Let miRNA 1 attack mRNAs 1 to 9 and miRNA 2 attack mRNAs 10 to 17.
# mRNAs 18 to 20 are not attacked. miRNA 3 has no gene set.
miR = c(rep(1,9),c(rep(2,8)));
mRNAs = 1:17;
A = data.frame(mRNAs,miR); # Note that the miRNAs MUST be in the second column!
set.seed(1)
P = miR.test(X,Y,A,group.miRNA,group.mRNA)
P
```

```
## For a faster result: use other gene set tests ##
# Wilcoxon two-sample test is recommended for fast results
# Note that results may vary depending on how much genes correlate
P.gsWilcox = miR.test(X,Y,A,group.miRNA,group.mRNA,gene.set.tests="W")
P.gsWilcox

### We can use an allocation matrix as A ###
```
Internal function for gene set testing.

Description

Internal function for gene set testing.

Usage

gs.test(A, X, Y, group, tests, permutation=FALSE, nrot=1000, design, allocation.matrix=FALSE, verbose=FALSE)

Arguments

A Allocation matrix as in "miR.test" function.
X miRNA expression matrix as in 'miR.test' function. Only necessary when allocation.matrix=TRUE.
Y mRNA expression matrix as in "miR.test" function.
group group as in 'miR.test' function
tests Test applied, see gene.set.tests
permutation Shall permutation procedure for global tests be applied? Put 'FALSE' to use approximate results or give a number for the number of permutations.
nrot Number of rotations of rotation tests. Defaults to 1000 to be able to show p-values as low as $10^{-3}$.

design If specified, group will be ignored. Design matrix as used in ‘limma’ package. Cannot be used with global tests.

allocation.matrix Logical, is A an allocation matrix with mRNAs in its columns and miRNAs in its rows, or is it an allocation data.frame?

verbose Defaults to FALSE. If TRUE, progress is printed.

Value List of the following, for up- and for down-regulation: Matrix with testing results for every gene set in its rows and the applied gene set test in its columns.

Author(s) Stephan Artmann

References


inverse.normal.combination

Inverse-normal method for p value combination.

Description
Inverse-normal method for p value combination.

Usage
inverse.normal.combination(p1, p2)

Arguments
p1, p2 one-sided p-values that shall be combined.

Value
Two-sided combined p-value.

Author(s)
Stephan Artmann

limma.one.sided Internal algorithm: Make limma test one-sided...

Description
Internal algorithm: Make limma test one-sided

Usage
limma.one.sided(fit, lower=FALSE)

Arguments
fit Result of "lmFit" and "eBayes" functions in "limma" package.
lower Shall one-sided p-value indicated down-regulation?
**Description**

internal algorithm for author’s convenience. Create a linear model with the limma package.

**Usage**

```r
limma.test(X, group, design)
```

**Arguments**

- `X`  
  Expression matrix.
- `group`  
  Group membership of replicates.
- `design`  
  Design as specified in limma (design matrix, see model.matrix).

**Author(s)**

Stephan Artmann

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**m.combine**

*Internal function for author’s convenience and more legible code.*

**Description**

Internal function for author’s convenience and more legible code. Applies a function to every column vector of a matrix and a vector.

**Usage**

```r
m.combine(M, v, FUN, ...)
```

**Arguments**

- `M`  
  The matrix for whose column vectors mapply shall be used.
- `v`  
  The vector.
- `FUN`  
  The function.
- `...`  
  Further arguments to be given to FUN.

**Author(s)**

Stephan Artmann
miR.test

Main Function of miRtest package.

Description

Main Function of miRtest package.

Usage

miR.test(X, Y, A, group.miRNA, group.mRNA, gene.set.tests="romer", design.miRNA, design.mRNA, adjust="none", permutation=FALSE, nrot=1000, allocation.matrix=FALSE, verbose=FALSE, errors=TRUE)

Arguments

X
miRNA expression matrix with genes in rows and replicates in columns

Y
mRNA expression matrix with genes in rows and replicates in columns

A
Allocation data.frame or Allocation matrix. An allocation data.frame contains the mRNAs in its first column and the miRNAs in its second column. See vignette ‘miRtest’ for information on Allocation matrices.

group.miRNA
Vector of miRNA group membership, being either numeric or a factor (**this makes a difference**). E. g. if you have four replicates in a control group and three replicates in a treated group, you may choose c(1,1,1,2,2,2)

design.miRNA
If specified, group.miRNA will be ignored. Here you can specify a design matrix as it is returned from the model.matrix ‘limma’ function.

design.mRNA
If specified, group.mRNA will be ignored. Here you can specify a design matrix as it is returned from the model.matrix ‘limma’ function.

group.mRNA
Vector of mRNA group membership, being either numeric or a factor (**this makes a difference**). E. g. if you have four replicates in a control group and three replicates in a treated group, you may choose c(1,1,1,2,2,2)

gene.set.tests
Test to be applied for gene set testing. Can be one or more of the following: ‘globaltest’, ‘GA’, ‘RHD’, ‘KS’, ‘W’, ‘Fisher’, ‘roast’, ‘romer’, or ‘all’ if you want to do all tests.

adjust
Multiple hypothesis testing adjustment. Same options as in "p.adjust" function.

permutation
Number of permutations for ‘globaltest’ or ‘GlobalAncova’ gene set tests. Put to ‘FALSE’ to use the approximate p-values instead of permutation ones.

nrot
Number of rotations for rotation tests ‘ROAST’ and ‘romer’

allocation.matrix
Logical, is A an allocation matrix with mRNAs in its columns and miRNAs in its rows, or is it an allocation data.frame?

verbose
Defaults to FALSE. If TRUE, output on progress is printed.

errors
Defaults to TRUE. If set to FALSE, some errors checking correct sizes of matrices are turned into warning messages.
Value
Matrix with testing results for every miRNA in its rows and the applied gene set test in its columns. Note that result will depend on whether multiple hypothesis testing correction was applied or not.

Author(s)
Stephan Artmann

References


Examples

# Generate random expression data

# Generate random miRNA expression data of 3 miRNAs
# with 8 replicates
set.seed(1)
X = rnorm(24);
dim(X) = c(3,8);
rownames(X) = 1:3;

# Generate random mRNA expression data with 20 mRNAs
# and 10 replicates
Y = rnorm(200);
dim(Y) = c(20,10);
rownames(Y) = 1:20;
# Let's assume that we want to compare 2 miRNA groups, each of 4 replicates:
group.miRNA = factor(c(1,1,1,1,2,2,2,2));
# ... and that the corresponding mRNA experiments had 5 replicates in each group
group.mRNA = factor(c(1,1,1,1,1,2,2,2,2,2));

library(miRtest)
#Let miRNA 1 attack mRNAs 1 to 9 and miRNA 2 attack mRNAs 10 to 17.
# mRNAs 18 to 20 are not attacked. miRNA 3 has no gene set.
miR = c(rep(1,9),c(rep(2,8)));
mRNAs = 1:17;
A = data.frame(mRNAs,miR);  # Note that the miRNAs MUST be in the second column!
A
set.seed(1)
P = miR.test(X,Y,A,group.miRNA,group.mRNA)
P

### For a faster result: use other gene set tests ###
# Wilcoxon two-sample test is recommended for fast results
# Note that results may vary depending on how much genes correlate
P.gsWilcox = miR.test(X,Y,A,group.miRNA,group.mRNA,gene.set.tests="W")
P.gsWilcox

### Other Designs ###
# Some more complicated designs are implemented, check the vignette "miRtest" for details.
group.miRNA = 1:8
group.mRNA = 1:10
covariable.miRNA = factor(c(1,2,3,4,1,2,3,4))  ### A covariable in miRNAs.
covariable.mRNA = factor(c(1,2,3,4,5,1,2,3,4,5))  ### A covariable in mRNAs.
library(limma)
design.miRNA = model.matrix(~group.miRNA + covariable.miRNA)
design.mRNA = model.matrix(~group.mRNA + covariable.mRNA)
\[ P = \text{miR.test}(X,Y,A,\text{design.miRNA}=\text{design.miRNA},\text{design.mRNA}=\text{design.mRNA},\text{allocation.matrix}=\text{TRUE}) \]

<table>
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<th>miRtest</th>
<th>Package Description: Two-group combined miRNA- and mRNA- expression testing.</th>
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</thead>
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**Description**

Looking for differential expression in miRNA-data can have low power. Taking their respective mRNA-gene sets on the other hand can lead to too liberal results. In Artmann et al. we proposed a method to combine both information sources and generate p-values that can detect either miRNA- and target gene set expression differences.

**Details**

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</table>

For a detailed help check vignette("miRtest")

You can start the test with the "miR.test" function, which needs the expression matrix X of miRNAs, the expression matrix Y of mRNAs and the allocation matrix.

**Author(s)**

Stephan Artmann <stephanartmann@gmx.net>, Klaus Jung, Tim Beissbarth

Maintainer: Stephan Artmann <stephanartmann@gmx.net>

**References**


See Also

Function "generate.A" as well as main function "miR.test"

Examples

```
#######################################
### Generate random expression data ###
#######################################
# Generate random miRNA expression data of 3 miRNAs
# with 8 replicates
set.seed(1)
X = rnorm(24);
dim(X) = c(3,8);
rownames(X) = 1:3;
# Generate random mRNA expression data with 20 mRNAs
# and 10 replicates
Y = rnorm(200);
dim(Y) = c(20,10);
rownames(Y) = 1:20;
# Let's assume that we want to compare 2 miRNA groups, each of 4 replicates:
group.miRNA = factor(c(1,1,1,1,2,2,2,2));
# and that the corresponding mRNA experiments had 5 replicates in each group
group.mRNA = factor(c(1,1,1,1,1,2,2,2,2,2));

library(miRtest)
# Let miRNA 1 attack mRNAs 1 to 9 and miRNA 2 attack mRNAs 10 to 17.
# mRNAs 18 to 20 are not attacked. miRNA 3 has no gene set.
miR = c(rep(1,9),c(rep(2,8)));
mRNAs = 1:17;
A = data.frame(mRNAs,miR); # Note that the mRNAs MUST be in the second column!
A
```
```r
set.seed(1)
P = miR.test(X,Y,A,group.miRNA,group.mRNA)
P

#####################################################
### For a faster result: use other gene set tests ###
#####################################################
# Wilcoxon two-sample test is recommended for fast results
# Note that results may vary depending on how much genes correlate
P.gsWilcox = miR.test(X,Y,A,group.miRNA,group.mRNA,gene.set.tests="W")
P.gsWilcox

############################################
### We can use an allocation matrix as A ###
############################################
A = generate.A(A,X=X,Y=Y,verbose=FALSE);
A
# Now we can test as before
set.seed(1)
P = miR.test(X,Y,A,group.miRNA,group.mRNA,allocation.matrix=TRUE)
P

###############################
### Other Designs ###
###############################
# Some more complicated designs are implemented, check the vignette "miRtest" for details.
group.miRNA = 1:8
group.mRNA = 1:10
covariable.miRNA = factor(c(1,2,3,4,1,2,3,4)) ### A covariable in miRNAs.
covariable.mRNA = factor(c(1,2,3,4,5,1,2,3,4,5)) ### A covariable in mRNAs.
library(limma)
design.miRNA = model.matrix(~group.miRNA + covariable.miRNA)
design.mRNA = model.matrix(~group.mRNA + covariable.mRNA)
P = miR.test(X,Y,A,design.miRNA=design.miRNA,design.mRNA=design.mRNA,allocation.matrix=TRUE)
P

X X

Description
Part of expression data from Nielsen et al.

Author(s)
Stephan Artmann taken from Nielsen et al.
**Description**

Part of expression data from Nielsen et al.

**Author(s)**

Stephan Artmann taken from Nielsen et al.
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