

Package ‘zitools’

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Title Analysis of zero-inflated count data

Version 1.3.0

Description zitools allows for zero inflated count data analysis by either using down-weighting of excess zeros or by replacing an appropriate proportion of excess zeros with NA. Through overloading frequently used statistical functions (such as mean, median, standard deviation), plotting functions (such as boxplots or heatmap) or differential abundance tests, it allows a wide range of downstream analyses for zero-inflated data in a less biased manner. This becomes applicable in the context of microbiome analyses, where the data is often overdispersed and zero-inflated, therefore making data analysis extremely challenging.

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Contents

zitoools-package	3
*	3
+	4
/	5
assays	5
boxplot	6
colData	7
colMeans2	7
colMedians	8
cor	9
cov	10
deinflatedcounts	10
heatmap	11
inputcounts	12
inputdata	12
log1p	13
log2p	14
mean	14
median	15
MissingValueHeatmap	16
model	16
mtx	17
otu_table	17
plot	18
quantile	19
resample_deinflatedcounts	19
rowData	20
rowQuantiles	21
rowSds	22
rowVars	23
rowWeightedMeans	23
rowWeightedSds	24
sample_data	25
sd	26
show	27
subset_feature	27
subset_sample	28
t	29
tax_table	30
var	30
weighted.mean	31
weightedSd	32
weights	33
Zi-class	33

zitoools-package3

zi2deseq2 34

zi2phyloseq 35

ziMain 35

Index38

zitoools-package

zitoools: Analysis of zero-inflated count data

Description

zitoools allows for zero inflated count data analysis by either using down-weighting of excess zeros or by replacing an appropriate proportion of excess zeros with NA. Through overloading frequently used statistical functions (such as mean, median, standard deviation), plotting functions (such as boxplots or heatmap) or differential abundance tests, it allows a wide range of downstream analyses for zero-inflated data in a less biased manner. This becomes applicable in the context of microbiome analyses, where the data is often overdispersed and zero-inflated, therefore making data analysis extremely challenging.

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See Also

- Useful links:
- <https://github.com/kreutz-lab/zitoools>
 - Report bugs at <https://github.com/kreutz-lab/zitoools/issues>

*

Arithmetic Operators

Description

Arithmetic operators for a Zi-class object

Usage

```
## S4 method for signature 'Zi,ANY'  
e1 * e2
```

Arguments

- e1

Zi-class object, matrix or number
- e2

Zi-class object, matrix or number

Value

a Zi-class object after a specific arithmetic operation is performed

Examples

```
data(mtx)
Zi <- ziMain(mtx)
Zi*Zi
Zi*2
```

+	<i>Arithmetic Operators</i>
---	-----------------------------

Description

Arithmetic operators for a Zi-class object

Arithmetic operators for a Zi-class object

Usage

```
## S4 method for signature 'Zi,ANY'
e1 + e2
```

```
## S4 method for signature 'Zi,ANY'
e1 - e2
```

Arguments

e1 [Zi-class object](#), matrix or number

e2 [Zi-class object](#), matrix or number

Value

a [Zi-class object](#) after a specific arithmetic operation is performed

a [Zi-class object](#) after a specific arithmetic operation is performed

Examples

```
data(mtx)
Zi <- ziMain(mtx)
Zi+Zi
Zi+2
data(mtx)
Zi <- ziMain(mtx)
Zi+Zi
Zi+2
```

/ *Arithmetic Operators*

Description

Arithmetic operators for a Zi-class object

Usage

```
## S4 method for signature 'Zi,ANY'
e1 / e2
```

Arguments

e1 [Zi](#)-class object, matrix or number
e2 [Zi](#)-class object, matrix or number

Value

a [Zi](#)-class object after a specific arithmetic operation is performed

Examples

```
data(mtx)
Zi <- ziMain(mtx)
Zi/Zi
Zi/2
```

assays *Access assays*

Description

access [assays](#) of an [Zi](#)-class object if the inputdata is an object of the class SummarizedExperiment

Usage

```
## S4 method for signature 'Zi'
assays(x, withDimnames = TRUE, ...)
```

Arguments

x [Zi](#)-class object
withDimnames A logical, indicating whether the dimnames of the SummarizedExperiment object should be applied (i.e. copied) to the extracted assays. see [assays](#)
... see [assays](#)

Value

list

Examples

```
data(mtx)
colData <- data.frame(SampleID = c('Sample1', 'Sample2', 'Sample3',
  'Sample4', 'Sample5', 'Sample6', 'Sample7', 'Sample8', 'Sample9',
  'Sample10'),
  Group = factor(x = c(1,1,1,1,1,2,2,2,2,2)))
rowData <- data.frame(Kingdom = c(rep('Bacteria', times = 100)),
  Phylum = c(rep('Bacteroidetes', times = 50),
  rep('Firmicutes', times = 50)))
se <- SummarizedExperiment::SummarizedExperiment(assays = list(counts = mtx),
  colData = colData, rowData = rowData)
Zi <- ziMain(se)
assays(Zi)
```

boxplot

*Create boxplots of a 'Zi'-class object***Description**

Create boxplots of a 'Zi'-class object.

Usage

```
## S3 method for class 'Zi'
boxplot(x, ...)
```

Arguments

x	'Zi'-class object
...	see boxplot.default

ValueA List with all information to create a boxplot see [boxplot.default](#)**See Also**[boxplot.default](#)**Examples**

```
data(mtx)
Zi <- ziMain(mtx)
boxplot(Zi)
boxplot(log1p(Zi))
```

colData	<i>Access the col Data</i>
---------	----------------------------

Description

access the `colData` of an `Zi`-class object if the inputdata is an object of the class `SummarizedExperiment`

Usage

```
## S4 method for signature 'Zi'
colData(x, ...)
```

Arguments

<code>x</code>	<code>Zi</code> -class object
<code>...</code>	<code>colData</code>

Value

DFrame

See Also

`colData`

Examples

```
data(mtx)
colData <- data.frame(SampleID = c('Sample1', 'Sample2', 'Sample3', 'Sample4',
  'Sample5', 'Sample6', 'Sample7', 'Sample8', 'Sample9', 'Sample10'),
  Group = factor(x = c(1,1,1,1,1,2,2,2,2,2)))
rowData <- data.frame(Kingdom = c(rep('Bacteria', times = 100)),
  Phylum = c(rep('Bacteroidetes', times = 50),
    rep('Firmicutes', times = 50)))
se <- SummarizedExperiment::SummarizedExperiment(assays = list(counts = mtx),
  colData = colData, rowData = rowData)
Zi <- ziMain(se)
colData(Zi)
```

colMeans2	<i>Calculate the row or column means of zero-inflated count data</i>
-----------	--

Description

Calculate row and column means of zero-inflated count data taking weights for structural zeros into account.

Usage

```
## S4 method for signature 'Zi'
colMeans2(x, rows = NULL, cols = NULL, na.rm = FALSE, useNames = TRUE)

## S4 method for signature 'Zi'
rowMeans2(x, rows = NULL, cols = NULL, na.rm = FALSE, useNames = TRUE)
```

Arguments

x	A Zi -class object
rows, cols	A vector indicating the subset of rows and/or columns to operate over. If NULL (default), no subsetting is done
na.rm	logical If TRUE NAs are excluded, otherwise not. default = FALSE
useNames	logical If TRUE (default), names attributes of result are set. Else if FALSE , no naming support is done.

Value

a numeric [vector](#) of row/column length

Examples

```
data(mtx)
Zi <- ziMain(mtx)
colMeans2(Zi)
rowMeans2(Zi)
```

colMedians	<i>Calculate the row or column median of zero-deinflated count data</i>
------------	---

Description

Calculate the row or column median of zero-deinflated data of a [Zi](#)-class object. To calculate the median, the `deinflatedcounts` matrix will be extracted.

Usage

```
## S4 method for signature 'Zi'
colMedians(x, rows = NULL, cols = NULL, na.rm = TRUE, ..., useNames = TRUE)

## S4 method for signature 'Zi'
rowMedians(x, rows = NULL, cols = NULL, na.rm = TRUE, ..., useNames = TRUE)
```

Arguments

x	Zi -class object
rows, cols	A vector indicating the subset of rows (and/or columns) to operate over. If NULL , no subsetting is done
na.rm	logical If TRUE NAs are excluded, otherwise not. default = TRUE
...	see colMedians
useNames	logical . If TRUE (default), names attributes of result are set. Else if FALSE , no naming support is done.

Value

returns a numeric vector of row/column length

Examples

```
data(mtx)
Zi <- ziMain(mtx)
colMedians(Zi, useNames = TRUE)
rowMedians(Zi, useNames = TRUE)
```

cor

Calculate weighted Pearson Correlation coefficients

Description

calculate the weighted pearson correlation coefficients of a count matrix of an Zi object taking weights for zero counts into account

Usage

```
## S4 method for signature 'Zi,ANY'
cor(x, y = NULL, use = "everything", method = "pearson")
```

Arguments

x	'Zi'-class object
y	'Zi'-class object
use	'everything' see cor
method	default = 'pearson', weighted correlation only implemented for person correlation

Value

correlation matrix

Examples

```
data(mtx)
Zi <- ziMain(mtx)
cor(Zi)
```

cov	<i>Calculate weighted Covariance</i>
-----	--------------------------------------

Description

calculate the weighted covariance of the columns of the count matrix of an Zi object taking weights for possible structural zero counts into account

Usage

```
## S4 method for signature 'Zi,ANY'
cov(x, y = NULL, use = "everything")
```

Arguments

x	'Zi'-class object
y	'Zi'-class object
use	'everything'

Value

covariance matrix

Examples

```
data(mtx)
Zi <- ziMain(mtx)
cov(Zi)
```

deinflatedcounts	<i>Access the model</i>
------------------	-------------------------

Description

access the deinflatedcounts of an Zi-class object

Usage

```
deinflatedcounts(x)

## S4 method for signature 'Zi'
deinflatedcounts(x)

deinflatedcounts(x) <- value

## S4 replacement method for signature 'Zi'
deinflatedcounts(x) <- value
```

Arguments

x [Zi](#)-class object
 value deinflatedcounts object

Value

deinflatedcounts

Examples

```
data(mtx)
Zi <- ziMain(mtx)
deinflatedcounts(Zi)
```

heatmap

Draw a Heat Map

Description

draw a heatmap of a given 'Zi'-class object, heatmap.Zi uses the deinflatedcounts matrix (drawn structural zeros) to produce a heatmap. NA values are white

Usage

```
## S3 method for class 'Zi'
heatmap(x, ...)
```

Arguments

x 'Zi'-class object
 ... see [heatmap](#)

Value

heatmap

Examples

```
data(mtx)
Zi <- ziMain(mtx)
#heatmap(Zi) # Error, clustering not possible
heatmap(Zi, Rowv=NA) # no clustering of rows
heatmap(Zi, Rowv=NA, Colv=NA) # no clustering of rows and cols
```

inputcounts	<i>Access the inputcounts</i>
-------------	-------------------------------

Description

access the inputcounts of an [Zi](#)-class object

Usage

```
inputcounts(x)

## S4 method for signature 'Zi'
inputcounts(x)

inputcounts(x) <- value

## S4 replacement method for signature 'Zi'
inputcounts(x) <- value
```

Arguments

x	Zi -class object
value	inputcounts object

Value

inputcounts

Examples

```
data(mtx)
Zi <- ziMain(mtx)
inputcounts(Zi)
```

inputdata	<i>Access and Set the inputdata</i>
-----------	-------------------------------------

Description

access the inputdata of an [Zi](#)-class object

Usage

```
inputdata(x)

## S4 method for signature 'Zi'
inputdata(x)

inputdata(x) <- value

## S4 replacement method for signature 'Zi'
inputdata(x) <- value
```

Arguments

x	Zi-class object
value	inputdata object

Value

inputdata

Examples

```
data(mtx)
Zi <- ziMain(mtx)
inputdata(Zi)
```

log1p	$\log(1+x)$
-------	-------------

Description

Calculate $\log(1+x)$ of all 'matrix' objects of a 'Zi'-class object, log calculates by default natural logarithms

Usage

```
## S4 method for signature 'Zi'
log1p(x)
```

Arguments

x	Zi-class object
---	---------------------------------

Value

a [Zi-class object](#) where the $\log(1+x)$ values of inputcounts, deinflatedcounts and weights are calculated.

See Also

[log1p](#), [log2p](#)

Examples

```
data(mtx)
Zi <- ziMain(mtx)
log1p(Zi)
```

log2p	$\log_2 p(x+1)$
-------	-----------------

Description

Calculate $\log_2(x+1)$ of all 'matrix' objects of a 'Zi'-class object

Usage

```
log2p(x)
```

Arguments

x [Zi](#)-class object

Value

a [Zi](#)-class object where the $\log_2(1+x)$ values of inputcounts, deinflatedcounts and weights are calculated.

Examples

```
data(mtx)
Zi <- ziMain(mtx)
log2p(Zi)
```

mean	<i>Arithmetic Mean</i>
------	------------------------

Description

Calculate the arithmetic mean of zero inflated data taking weights for structural zeros into account

Usage

```
## S3 method for class 'Zi'
mean(x, ...)
```

Arguments

x A [Zi](#)-class object
 ... [mean.default](#)

Value

mean value

See Also

[weighted.mean](#), [colMeans2](#), [rowMeans2](#)

Examples

```
data(mtx)
Zi <- ziMain(mtx)
mean(Zi)
```

median	<i>Calculate the median of zero-deinflated count data</i>
--------	---

Description

Caluclate the median of zero-deinflated data of a 'Zi'-class object. To calculate the median, the deinflatedcounts matrix will be extracted

Usage

```
## S3 method for class 'Zi'
median(x, na.rm = TRUE, ...)
```

Arguments

x	Zi -class object
na.rm	logical If TRUE NAs are excluded, otherwise not. default = TRUE
...	see median.default

Value

median value

See Also

[median](#), [colMedians](#), [rowMedians](#)

Examples

```
data(mtx)
Zi <- ziMain(mtx)
median(Zi)
```

MissingValueHeatmap	<i>Missing Value Heatmap</i>
---------------------	------------------------------

Description

Missing Value Heatmap

Usage

```
MissingValueHeatmap(ZiObject, title = "", xlab = "", ylab = "")
```

Arguments

ZiObject	ZiObject, result of the ziMain function
title	Title of the plot .
xlab	Title of the x axis.
ylab	Title of the y axis.

Value

heatmap

Examples

```
data(mtx)
```

model	<i>Access the model</i>
-------	-------------------------

Description

access the model of an [Zi](#)-class object

Usage

```
model(x)

## S4 method for signature 'Zi'
model(x)

model(x) <- value

## S4 replacement method for signature 'Zi'
model(x) <- value
```

Arguments

x	Zi -class object
value	model object

Value

model

Examples

```
data(mtx)
Zi <- ziMain(mtx)
model(Zi)
```

mtx	<i>Matrix Data</i>
-----	--------------------

Description

Zero-inflated matrix data

Usage

mtx

Format

'mtx':

A matrix with 100 rows and 10 columns

Value

a data matrix

otu_table	<i>Access the otu table</i>
-----------	-----------------------------

Description

access the [otu_table](#) of an [Zi](#)-class object if the inputdata slot is a phyloseq object

Usage

```
## S4 method for signature 'Zi'
otu_table(object)
```

Arguments

object [Zi](#)-class object

Value

otu_table

Examples

```
data(mtx)
OTU <- otu_table(mtx, taxa_are_rows = TRUE)
sample_data <- data.frame(SampleID = c('Sample1', 'Sample2', 'Sample3',
    'Sample4', 'Sample5', 'Sample6', 'Sample7', 'Sample8', 'Sample9',
    'Sample10'),
    Group = factor(x = c(1,1,1,1,1,1,2,2,2,2,2)))
SAM <- sample_data(sample_data)
tax_table <- data.frame(Kingdom = c(rep('Bacteria', times = 100)),
    Phylum = c(rep('Bacteroidetes', times = 50),
    rep('Firmicutes', times = 50)))
TAX <- tax_table(tax_table)
ps <- phyloseq::phyloseq(OTU, TAX, SAM)
Zi <- ziMain(ps)
otu_table(Zi)
```

plot	<i>Plotting</i>
------	-----------------

Description

plot

Usage

```
## S4 method for signature 'Zi,ANY'
plot(x, y, ...)
```

Arguments

- x [Zi-class](#) object
- y the y coordinates of points in the plot, optional if x is an appropriate structure
- ... Arguments to be passed to plot

Value

returns plot object

Examples

```
data(mtx)
Zi <- ziMain(mtx)
plot(Zi)
```

quantile	<i>Calculate the quantiles of zero-deinflated count data</i>
----------	--

Description

Calculate the quantiles of zero-deinflated data of a [Zi](#)-class object. To calculate the quantiles, the `deinflatedcounts` matrix will be extracted.

Usage

```
## S3 method for class 'Zi'
quantile(x, probs = seq(0, 1, 0.25), na.rm = TRUE, ...)
```

Arguments

<code>x</code>	A Zi -class object
<code>probs</code>	A numeric vector of J probabilities in [0,1]
<code>na.rm</code>	logical If TRUE NAs are excluded, otherwise not. default = TRUE
<code>...</code>	quantile

Value

quantile value

See Also

[quantile](#), [rowQuantiles](#), [colQuantiles](#)

Examples

```
data(mtx)
Zi <- ziMain(mtx)
quantile(Zi)
```

<code>resample_deinflatedcounts</code>	<i>Resample a Zi-class object</i>
--	---

Description

Resample the `deinflatedcounts` matrix of an [Zi](#)-class object. Resampling is done by drawing from a binomial distribution with a given probability that a count value (zero and non-zero) is a structural zero.

Usage

```
resample_deinflatedcounts(x)
```

Arguments

x [Zi-class object](#)

Value

a [Zi-class object](#) where the deinflatedcounts are resampled

Examples

```
data(mtx)
Zi <- ziMain(mtx)
resample_deinflatedcounts(Zi)
```

rowData	<i>Access the row data</i>
---------	----------------------------

Description

access the [rowData](#) of an [Zi-class object](#) if the inputdata is an object of the class SummarizedExperiment

Usage

```
## S4 method for signature 'Zi'
rowData(x, useNames = TRUE, ...)
```

Arguments

x [Zi-class object](#)

useNames returns a rowData dataframe with rownames

... [rowData](#)

Value

DFrame

Examples

```
data(mtx)
colData <- data.frame(SampleID = c('Sample1', 'Sample2', 'Sample3', 'Sample4',
  'Sample5', 'Sample6', 'Sample7', 'Sample8', 'Sample9', 'Sample10'),
  Group = factor(x = c(1,1,1,1,1,2,2,2,2,2)))
rowData <- data.frame(Kingdom = c(rep('Bacteria', times = 100)),
  Phylum = c(rep('Bacteroidetes', times = 50),
    rep('Firmicutes', times = 50)))
se <- SummarizedExperiment::SummarizedExperiment(assays = list(counts = mtx),
  colData = colData, rowData = rowData)
Zi <- ziMain(se)
rowData(Zi)
```

rowQuantiles

*Calculate the row or column quantiles of zero-deinflated count data***Description**

Calculate the row or column quantiles of zero-deinflated data of a [Zi](#)-class object. To calculate the quantiles, the deinflatedcounts matrix will be extracted

Usage

```
## S4 method for signature 'Zi'
rowQuantiles(
  x,
  rows = NULL,
  cols = NULL,
  probs = seq(from = 0, to = 1, by = 0.25),
  na.rm = TRUE,
  type = 7L,
  ...,
  useNames = TRUE,
  drop = TRUE
)

## S4 method for signature 'Zi'
colQuantiles(
  x,
  rows = NULL,
  cols = NULL,
  probs = seq(from = 0, to = 1, by = 0.25),
  na.rm = TRUE,
  type = 7L,
  ...,
  useNames = TRUE,
  drop = TRUE
)
```

Arguments

x	A Zi -class object
rows, cols	A vector indicating the subset of rows and/or columns to operate over. If NULL (default), no subsetting is done.
probs	A numeric vector of J probabilities in [0,1]
na.rm	logical If TRUE NAs are excluded, otherwise not. default = TRUE
type	An integer specifying the type of estimator
...	Additional arguments passed to specific methods rowQuantiles
useNames	logical If TRUE (default), names attributes of result are set. Else if FALSE , no naming support is done.
drop	If TRUE a vector is returned if J == 1.

Value

a numeric [vector](#) of row/column length

Examples

```
data(mtx)
Zi <- ziMain(mtx)
rowQuantiles(Zi, useNames = TRUE)
colQuantiles(Zi, useNames = TRUE)
```

rowSds

Row and Column Standard Deviations of zero inflated count data

Description

Calculate row and column standard deviations of zero inflated count data taking weights for structural zeros into account

Usage

```
## S4 method for signature 'Zi'
rowSds(x, rows = NULL, cols = NULL, na.rm = FALSE, useNames = TRUE)

## S4 method for signature 'Zi'
colSds(x, rows = NULL, cols = NULL, na.rm = FALSE, useNames = TRUE)
```

Arguments

x	A Zi -class object
rows, cols	A vector indicating the subset of rows and/or columns to operate over. If NULL (default), no subsetting is done
na.rm	logical If TRUE NAs are excluded, otherwise not. default = FALSE
useNames	logical If TRUE (default), names attributes of result are set. Else if FALSE , no naming support is done.

Value

a [vector](#) of row/column length

Examples

```
data(mtx)
Zi <- ziMain(mtx)
rowSds(Zi)
colSds(Zi)
```

rowVars

*Row and Column Variances of zero inflated count data***Description**

Calculate row and column variances of zero inflated count data taking weights for structural zeros into account.

Usage

```
## S4 method for signature 'Zi'
rowVars(x, rows = NULL, cols = NULL, na.rm = FALSE, useNames = TRUE)

## S4 method for signature 'Zi'
colVars(x, rows = NULL, cols = NULL, na.rm = FALSE, useNames = TRUE)
```

Arguments

x	A Zi -class object
rows, cols	A vector indicating the subset of rows and/or columns to operate over. If NULL (default), no subsetting is done
na.rm	logical If TRUE NAs are excluded, otherwise not. default = FALSE
useNames	logical If TRUE (default), names attributes of result are set. Else if FALSE , no naming support is done.

Value

a vector of row/col length

Examples

```
data(mtx)
Zi <- ziMain(mtx)
rowVars(Zi)
colVars(Zi)
```

rowWeightedMeans

*Row and Column weighted means of zero inflated count data***Description**

Calculate row and column weighted means of zero inflated count data, additionally taking weights for structural zeros into account.

Usage

```
## S4 method for signature 'Zi'
rowWeightedMeans(
  x,
  w,
  rows = NULL,
  cols = NULL,
  na.rm = FALSE,
  useNames = TRUE
)

## S4 method for signature 'Zi'
colWeightedMeans(
  x,
  w,
  rows = NULL,
  cols = NULL,
  na.rm = FALSE,
  useNames = TRUE
)
```

Arguments

x	A Zi -class object
w	a numerical vector of weights either of length = rows or length = cols giving the weights to use for elements of x
rows, cols	A vector indicating the subset of rows and/or columns to operate over. If NULL (default), no subsetting is done
na.rm	logical If TRUE NAs are excluded, otherwise not. default = FALSE
useNames	logical If TRUE (default), names attributes of result are set. Else if FALSE , no naming support is done.

Value

a numeric vector of length N(K)

Examples

```
data(mtx)
Zi <- ziMain(mtx)
rowWeightedMeans(Zi, w = runif(ncol(inputcounts(Zi)), 0.1,1))
colWeightedMeans(Zi, w = runif(nrow(inputcounts(Zi)), 0.1,1))
```

rowWeightedSds	<i>Row and column weighted standard deviations or variances of zero inflated count data</i>
----------------	---

Description

Calculate row and column standard deviations or variances of zero inflated count data, additionally taking weights for structural zeros into account.

Usage

```
## S4 method for signature 'Zi'
rowWeightedSds(x, w, rows = NULL, cols = NULL, na.rm = FALSE, useNames = TRUE)

## S4 method for signature 'Zi'
colWeightedSds(x, w, rows = NULL, cols = NULL, na.rm = FALSE, useNames = TRUE)

## S4 method for signature 'Zi'
rowWeightedVars(x, w, rows = NULL, cols = NULL, na.rm = FALSE, useNames = TRUE)

## S4 method for signature 'Zi'
colWeightedVars(x, w, rows = NULL, cols = NULL, na.rm = FALSE, useNames = TRUE)
```

Arguments

x	A Zi -class object
w	a numerical vector of weights either of length = rows or length = cols giving the weights to use for elements of x
rows, cols	A vector indicating the subset of rows and/or columns to operate over. If NULL (default), no subsetting is done
na.rm	logical If TRUE NAs are excluded, otherwise not. default = FALSE
useNames	logical If TRUE (default), names attributes of result are set. Else if FALSE , no naming support is done.

Value

a numeric vector of length N(K)

Examples

```
data(mtx)
Zi <- ziMain(mtx)
rowWeightedSds(Zi, w = runif(ncol(inputcounts(Zi)), 0.1,1))
colWeightedSds(Zi, w = runif(nrow(inputcounts(Zi)), 0.1,1))
rowWeightedVars(Zi, w = runif(ncol(inputcounts(Zi)), 0.1,1))
colWeightedVars(Zi, w = runif(nrow(inputcounts(Zi)), 0.1,1))
```

sample_data

Access the sample data

Description

access the [sample_data](#) of an [Zi](#)-class object if the inputdata slot is a phyloseq object

Usage

```
## S4 method for signature 'Zi'
sample_data(object)
```

Arguments

object [Zi](#)-class object

Value

sample_data

Examples

```
data(mtx)
OTU <- otu_table(mtx, taxa_are_rows = TRUE)
sample_data <- data.frame(SampleID = c('Sample1', 'Sample2', 'Sample3',
    'Sample4', 'Sample5', 'Sample6', 'Sample7', 'Sample8', 'Sample9',
    'Sample10'),
    Group = factor(x = c(1,1,1,1,1,1,2,2,2,2,2)))
SAM <- sample_data(sample_data)
tax_table <- data.frame(Kingdom = c(rep('Bacteria', times = 100)),
    Phylum = c(rep('Bacteroidetes', times = 50),
    rep('Firmicutes', times = 50)))
TAX <- tax_table(tax_table)
ps <- phyloseq::phyloseq(OTU, TAX, SAM)
Zi <- ziMain(ps)
sample_data(Zi)
```

sd	<i>Standard Deviation of zero inflated count data</i>
----	---

Description

Calculate the standard deviation of zero inflated count data taking weights for structural zeros into account.

Usage

```
## S4 method for signature 'Zi'
sd(x, na.rm = FALSE)
```

Arguments

x A [Zi](#)-class object
na.rm [logical](#) If [TRUE](#) NAs are excluded, otherwise not. default = [FALSE](#)

Value

standard deviation value

See Also

[weightedSd](#), [rowSds](#), [colSds](#)

Examples

```
data(mtx)
Zi <- ziMain(mtx)
sd(Zi)
```

show	<i>Show summary of Zi object</i>
------	----------------------------------

Description

Message printed at command line

Usage

```
## S4 method for signature 'Zi'
show(object)
```

Arguments

object [Zi](#)-class object

Value

returns a numeric vector of row/column length

Examples

```
data(mtx)
Zi <- ziMain(mtx)
Zi
show(Zi)
```

subset_feature	<i>Subset a Zi-class object based on feature data</i>
----------------	---

Description

Subset a [Zi](#)-class object based on tax_table of a phyloseq object or on rowData of a Summarized-Experiment object

Usage

```
subset_feature(Zi, ...)
```

Arguments

Zi [Zi](#)-class object

... The subsetting expression that should be applied, see [subset](#) for more details

Value

a [Zi](#)-class object after subsetting is done

Examples

```
data(mtx)
OTU <- otu_table(mtx, taxa_are_rows = TRUE)
sample_data <- data.frame(SampleID = c('Sample1', 'Sample2', 'Sample3',
  'Sample4', 'Sample5', 'Sample6', 'Sample7', 'Sample8', 'Sample9',
  'Sample10'),
  Group = factor(x = c(1,1,1,1,1,2,2,2,2,2)))
SAM <- sample_data(sample_data)
tax_table <- data.frame(Kingdom = c(rep('Bacteria', times = 100)),
  Phylum = c(rep('Bacteroidetes', times = 50),
  rep('Firmicutes', times = 50)))
TAX <- tax_table(tax_table)
ps <- phyloseq::phyloseq(OTU, TAX, SAM)
Zi <- ziMain(ps)
subset_Zi_phylo <- subset_feature(Zi, ta2 == 'Bacteroidetes')
```

subset_sample	<i>Subset a Zi-class object based on sample data</i>
---------------	--

Description

Subset a [Zi](#)-class object based on sample_data of an phyloseq object or on colData based on a SummarizedExperiment object

Usage

```
subset_sample(Zi, ...)
```

Arguments

Zi [Zi](#)-class object

... The subsetting expression that should be applied, see [subset](#) for more details

Value

a [Zi](#)-class object after subsetting is done

Examples

```
data(mtx)
OTU <- otu_table(mtx, taxa_are_rows = TRUE)
sample_data <- data.frame(SampleID = c('Sample1', 'Sample2', 'Sample3',
    'Sample4', 'Sample5', 'Sample6', 'Sample7', 'Sample8', 'Sample9',
    'Sample10'),
    Group = factor(x = c(1,1,1,1,1,1,2,2,2,2,2)))
SAM <- sample_data(sample_data)
tax_table <- data.frame(Kingdom = c(rep('Bacteria', times = 100)),
    Phylum = c(rep('Bacteroidetes', times = 50),
    rep('Firmicutes', times = 50)))
TAX <- tax_table(tax_table)
ps <- phyloseq::phyloseq(OTU, TAX, SAM)
Zi <- ziMain(ps)
subset_Zi <- subset_sample(Zi, SampleID %in% c('Sample1','Sample2'))
```

t

Transpose a [Zi](#)-class object

Description

transpose all matrices of a [Zi](#)-class object

Usage

```
## S4 method for signature 'Zi'
t(x)
```

Arguments

x [Zi](#)-class object

Value

[Zi](#)-class object

Examples

```
data(mtx)
Zi <- ziMain(mtx)
t(Zi)
```

tax_table	<i>Access the taxonomy table</i>
-----------	----------------------------------

Description

access the taxonomy table ([tax_table](#)) of an [Zi](#)-class object if the inputdata slot is a phyloseq object

Usage

```
## S4 method for signature 'Zi'
tax_table(object)
```

Arguments

object [Zi](#)-class object

Value

tax_table

Examples

```
data(mtx)
OTU <- otu_table(mtx, taxa_are_rows = TRUE)
sample_data <- data.frame(SampleID = c('Sample1', 'Sample2', 'Sample3',
  'Sample4', 'Sample5', 'Sample6', 'Sample7', 'Sample8', 'Sample9',
  'Sample10'),
  Group = factor(x = c(1,1,1,1,1,1,2,2,2,2,2)))
SAM <- sample_data(sample_data)
tax_table <- data.frame(Kingdom = c(rep('Bacteria', times = 100)),
  Phylum = c(rep('Bacteroidetes', times = 50),
  rep('Firmicutes', times = 50)))
TAX <- tax_table(tax_table)
ps <- phyloseq::phyloseq(OTU, TAX, SAM)
Zi <- ziMain(ps)
tax_table(Zi)
```

var	<i>Variance of zero inflated count data</i>
-----	---

Description

Calculate the variance of zero inflated count data taking weights for structural zeros into account.

Usage

```
## S4 method for signature 'Zi,ANY'
var(x, na.rm = FALSE)
```

Arguments

`x` A [Zi](#)-class object
`na.rm` [logical](#) If [TRUE](#) NAs are excluded, otherwise not. default = [FALSE](#)

Value

variance value

See Also

[weightedVar](#), [rowVars](#), [colVars](#)

Examples

```
data(mtx)
Zi <- ziMain(mtx)
var(Zi)
```

`weighted.mean`

Weighted Arithmetic Mean of zero inflated count data

Description

Calculate a weighted mean of zero inflated count data, additionally taking weights for structural zeros into account

Usage

```
## S4 method for signature 'Zi'
weighted.mean(x, w, ...)
```

Arguments

`x` A [Zi](#)-class object
`w` a numerical [vector](#) of weight the same length as `x` giving the weights to use for elements of `x`
`...` [weighted.mean](#)

Value

weighted mean value

See Also

[weighted.mean](#), [rowWeightedMeans](#), [colWeightedMeans](#)

Examples

```
data(mtx)
Zi <- ziMain(mtx)
weight <- runif(length(inputcounts(Zi)), 0.1, 1)
weighted.mean(Zi, w= weight)
```

weightedSd

*Weighted Variance and weighted Standard Deviation***Description**

Calculate a weighted variance and standard deviation of zero inflated count data, additionally taking weights for structural zeros into account

Usage

```
weightedSd(x, w = NULL, idxs = NULL, na.rm = FALSE, center = NULL, ...)
```

```
weightedVar(x, w = NULL, idxs = NULL, na.rm = FALSE, center = NULL, ...)
```

Arguments

x	A Zi -class object
w	a numerical vector of weight the same length as x giving the weights to use for elements of x
idxs	A vector indicating subset of elements to operate over. If NULL , no subsetting is done.
na.rm	logical If TRUE NAs are excluded, otherwise not. default = FALSE
center	numeric scalar specifying the center location of the data. If NULL , it is estimated from data.
...	weightedVar

Value

a [numeric](#) scalar

See Also

[weightedVar](#), [rowWeightedVars](#), [colWeightedVars](#)

Examples

```
data(mtx)
Zi <- ziMain(mtx)
weight <- runif(length(inputcounts(Zi)), 0.1, 1)
weightedVar(Zi, w= weight)
weightedSd(Zi, w = weight)
```

weights	<i>Access the weights</i>
---------	---------------------------

Description

access the weights of an [Zi](#)-class object

Usage

```
weights(x)

## S4 method for signature 'Zi'
weights(x)

weights(x) <- value

## S4 replacement method for signature 'Zi'
weights(x) <- value
```

Arguments

x	Zi -class object
value	weights object

Value

weights

Examples

```
data(mtx)
Zi <- ziMain(mtx)
weights(Zi)
```

Zi-class	<i>Class Zi</i>
----------	-----------------

Description

Objects of this class store all the results of the ZiMain function to continue zero inflated data analysis

Value

[Zi](#)-class object

Slots

`inputdata` a matrix, phyloseq or SummarizedExperiment object.

`inputcounts` matrix. The count matrix, features as rows, samples as columns

`model` list. The result of fitting a zero inflated model using [zeroinfl](#)

`deinflatedcounts` matrix. The matrix where predicted structural zeros are omitted and stored as NA values

`weights` matrix. A matrix containing weights for zero counts

zi2deseq2	<i>Convert a Zi-class object to a DESeq2 dds object</i>
-----------	---

Description

A [Zi](#)-class object is converted to a DESeqDataSet object, which can be used for DESeq2 analysis. Both, weight and count matrices will be stored in assays of the DESeqDataSet.

Usage

```
zi2deseq2(ZiObject, design, colData, ...)
```

Arguments

<code>ZiObject</code>	Zi -class object
<code>design</code>	A formula which specifies the design of the experiment, taking the form formula(~ x + y + z). That is, a formula with right-hand side only. By default, the functions in this package and DESeq2 will use the last variable in the formula (e.g. z) for presenting results (fold changes, etc.) and plotting. When considering your specification of experimental design, you will want to re-order the levels so that the NULL set is first.
<code>colData</code>	if the inputdata of the Zi -class object is a matrix: a DataFrame or data.frame with at least a single column. Rows of colData correspond to columns of count-Data
<code>...</code>	phyloseq::phyloseq_to_deseq2 if the inputdata of the 'Zi'-object is a phyloseq object DESeq2::DESeqDataSet if the inputdata the 'Zi'-object is a SummarizedExperiment object

Value

a dds class object

Examples

```
data(mtx)
Zi <- ziMain(mtx)
colData <- data.frame(group = factor(x = c(1,1,1,1,1,2,2,2,2,2)))
zi2deseq2(Zi, ~group, colData)
```

zi2phyloseq	<i>Replace the otu table of a phyloseq object</i>
-------------	---

Description

Replace the OTU table of a phyloseq object with the OTU table of zero de-inflated count data

Usage

```
zi2phyloseq(ZiObject)
```

Arguments

ZiObject [Zi](#)-class object with a phyloseq object as input

Value

a 'phyloseq'-class object

Examples

```
data(mtx)
OTU <- otu_table(mtx, taxa_are_rows = TRUE)
sample_data <- data.frame(SampleID = c('Sample1', 'Sample2', 'Sample3',
    'Sample4', 'Sample5', 'Sample6', 'Sample7', 'Sample8', 'Sample9',
    'Sample10'),
    Group = factor(x = c(1,1,1,1,1,2,2,2,2,2)))
SAM <- sample_data(sample_data)
tax_table <- data.frame(Kingdom = c(rep('Bacteria', times = 100)),
    Phylum = c(rep('Bacteroidetes', times = 50),
    rep('Firmicutes', times = 50)))
TAX <- tax_table(tax_table)
ps <- phyloseq::phyloseq(OTU, TAX, SAM)
Zi <- ziMain(ps)
new_ps <- zi2phyloseq(Zi)
new_ps
```

ziMain	<i>ziMain - main function to fit a zero inflation model and calculate weights for structural zeros</i>
--------	--

Description

This function fits a zero-inflated mixture model (either Poisson or negative binomial distribution) to count data and calculates weights for all zeros indicating whether a zero is a real count (weight close to 1) or whether it is a structural zero (weight close to 0). The default model is a zero inflated negative binomial model.

The input inputdata of the ziMain function is either a phyloseq object, SummarizedExperiment object or count matrix.

In order to reduce calculation times, the count matrix is divided into blocks of around 5000 count values. Then, a zero inflation model (either Poisson or negative binomial distribution) is fitted to the data. The response variable count is estimated using the predictor variables `sample(columns)` and `feature(rows)`. Using the fitted zero inflated model, probabilities given that a zero in the count matrix is a structural zero are predicted. Those probabilities are used in two ways: 1) A zero-deinflated count matrix is generated where a appropriate proportion of zeros are randomly replaced by NA. This count matrix can be used for analysis methods which cannot deal with weights. 2) Weights

$$w = \frac{(1 - \pi) f_{\text{NB}}(y; \mu, \theta)}{f_{\text{ZINB}}(y; \mu, \theta, \pi)}.$$

(see Van den Berge, K., Perraudeau, F., Soneson, C. et al.) are calculated in order to down-weight structural zeros in analyses which can account for weighting of individual data points.

all zero counts are calculated given the following formula:

The result of the `ziMain` function can be used to analyze zero inflated count data.

Usage

```
ziMain(
  inputdata,
  feature = "feature",
  formula = count ~ sample + feature,
  dist = "negbin",
  link = "logit",
  zeroRows.rm = FALSE,
  ...
)
```

Arguments

<code>inputdata</code>	phyloseq object, SummarizedExperiment object, or matrix (rows =features, columns=samples)
<code>feature</code>	'feature', 'gene', 'OTU', 'phylum', etc. By default, rownames are labelled as feature1, feature2, ...
<code>formula</code>	formula to fit the zero inflated model $y \sim x_1 + x_2 + \dots$, default = <code>count ~ sample + feature</code> . A different set of regressors can be specified using $y \sim x_1 + x_2 + \dots z_1 + z_2$... where the first part describes the count data model and the second part describes the zero inflation model
<code>dist</code>	= distribution, either poisson ('poisson'), negative binomial ('negbin')
<code>link</code>	= link function, either 'logit', 'probit', 'cloglog', 'cauchit'
<code>zeroRows.rm</code>	= logical, indicating whether rows that only contain zeros should be removed (TRUE) or not (FALSE) (they are removed to fit a zero inflated model and will be added afterwards count matrix per default = 0 and weights = 1)
...	additional parameters to describe the model, see zeroinfl

Value

[Zi](#)-class object

Slots

`inputdata` a matrix, phyloseq or SummarizedExperiment object.
`inputcounts` matrix. The count matrix, features as rows, samples as columns
`model` list. The result of fitting a zero inflated model using [zeroinfl](#)
`deinflatedcounts` matrix. A matrix where zero counts are randomly replaced according to the estimated probability of being a structural zero
`weights` matrix. A matrix containing weights for zero counts

References

Van den Berge, K., Perraudeau, F., Soneson, C. et al. Observation weights unlock bulk RNA-seq tools for zero inflation and single-cell applications. *Genome Biol* 19, 24 (2018). <https://doi.org/10.1186/s13059-018-1406-4>

See Also

[zeroinfl](#)

Examples

```
# zero-inflated count matrix
data(mtx)
# calling ziMain function:
Zi <- ziMain(mtx)
#Example Data Sets from other R packages
#data(enterotype)
#data(GlobalPatterns)
#data(esophagus)
#ziMain(esophagus)
#data(soilrep)
```

Index

- * **datasets**
 - mtx, [17](#)
- * **internal**
 - zitoools-package, [3](#)
- [*, 3](#)
- [*, Zi, ANY-method \(*\), 3](#)
- [+, 4](#)
- [+, Zi, ANY-method \(+\), 4](#)
- [-, Zi, ANY-method \(-\), 4](#)
- [/, 5](#)
- [/, Zi, ANY-method \(/\), 5](#)
- [assays, 5, 5](#)
- [assays, Zi-method \(assays\), 5](#)
- [boxplot, 6](#)
- [boxplot.default, 6](#)
- [colData, 7, 7](#)
- [colData, Zi-method \(colData\), 7](#)
- [colMeans2, 7, 14](#)
- [colMeans2, Zi-method \(colMeans2\), 7](#)
- [colMedians, 8, 8, 15](#)
- [colMedians, Zi-method \(colMedians\), 8](#)
- [colQuantiles, 19](#)
- [colQuantiles \(rowQuantiles\), 21](#)
- [colQuantiles, Zi-method \(rowQuantiles\), 21](#)
- [colSds, 27](#)
- [colSds \(rowSds\), 22](#)
- [colSds, Zi-method \(rowSds\), 22](#)
- [colVars, 31](#)
- [colVars \(rowVars\), 23](#)
- [colVars, Zi-method \(rowVars\), 23](#)
- [colWeightedMeans, 31](#)
- [colWeightedMeans \(rowWeightedMeans\), 23](#)
- [colWeightedMeans, Zi-method \(rowWeightedMeans\), 23](#)
- [colWeightedSds \(rowWeightedSds\), 24](#)
- [colWeightedSds, Zi-method \(rowWeightedSds\), 24](#)
- [colWeightedVars, 32](#)
- [colWeightedVars \(rowWeightedSds\), 24](#)
- [colWeightedVars, Zi-method \(rowWeightedSds\), 24](#)
- [cor, 9, 9](#)
- [cor, Zi, ANY-method \(cor\), 9](#)
- [cov, 10](#)
- [cov, Zi, ANY-method \(cov\), 10](#)
- [deinflatedcounts, 10](#)
- [deinflatedcounts, Zi-method \(deinflatedcounts\), 10](#)
- [deinflatedcounts<- \(deinflatedcounts\), 10](#)
- [deinflatedcounts<-, Zi-method \(deinflatedcounts\), 10](#)
- [DESeq2: :DESeqDataSet, 34](#)
- [FALSE, 8, 21–26, 31, 32, 36](#)
- [heatmap, 11, 11](#)
- [inputcounts, 12](#)
- [inputcounts, Zi-method \(inputcounts\), 12](#)
- [inputcounts<- \(inputcounts\), 12](#)
- [inputcounts<-, Zi-method \(inputcounts\), 12](#)
- [inputdata, 12](#)
- [inputdata, Zi-method \(inputdata\), 12](#)
- [inputdata<- \(inputdata\), 12](#)
- [inputdata<-, Zi-method \(inputdata\), 12](#)
- [log1p, 13, 13](#)
- [log1p, Zi-method \(log1p\), 13](#)
- [log2p, 13, 14](#)
- [log2p, Zi-method \(log2p\), 14](#)
- [logical, 8, 15, 19, 21–26, 31, 32](#)
- [mean, 14](#)
- [mean.default, 14](#)
- [median, 15, 15](#)
- [median.default, 15](#)
- [MissingValueHeatmap, 16](#)
- [model, 16](#)
- [model, Zi-method \(model\), 16](#)
- [model<- \(model\), 16](#)
- [model<-, Zi-method \(model\), 16](#)

- mtx, 17
- NA, 8, 21–26, 31, 32
- NULL, 8, 21–25, 32
- numeric, 32
- otu_table, 17, 17
- otu_table, Zi-method (otu_table), 17
- phyloseq::phyloseq_to_deseq2, 34
- plot, 18
- plot, Zi, ANY-method (plot), 18
- quantile, 19, 19
- resample_deinflatedcounts, 19
- rowData, 20, 20
- rowData, Zi-method (rowData), 20
- rowMeans2, 14
- rowMeans2 (colMeans2), 7
- rowMeans2, Zi-method (colMeans2), 7
- rowMedians, 15
- rowMedians (colMedians), 8
- rowMedians, Zi-method (colMedians), 8
- rowQuantiles, 19, 21, 21
- rowQuantiles, Zi-method (rowQuantiles), 21
- rowSds, 22, 27
- rowSds, Zi-method (rowSds), 22
- rowVars, 23, 31
- rowVars, Zi-method (rowVars), 23
- rowWeightedMeans, 23, 31
- rowWeightedMeans, Zi-method (rowWeightedMeans), 23
- rowWeightedSds, 24
- rowWeightedSds, Zi-method (rowWeightedSds), 24
- rowWeightedVars, 32
- rowWeightedVars (rowWeightedSds), 24
- rowWeightedVars, Zi-method (rowWeightedSds), 24
- sample_data, 25, 25
- sample_data, Zi-method (sample_data), 25
- sd, 26
- sd, Zi-method (sd), 26
- show, 27
- show, Zi-method (show), 27
- subset, 28
- subset_feature, 27
- subset_sample, 28
- t, 29
- t, Zi-method (t), 29
- tax_table, 30, 30
- tax_table, Zi-method (tax_table), 30
- TRUE, 8, 15, 19, 21–26, 31, 32, 36
- var, 30
- var, Zi, ANY-method (var), 30
- vector, 8, 19, 21–25, 31, 32
- weighted.mean, 14, 31, 31
- weighted.mean, Zi-method (weighted.mean), 31
- weightedSd, 27, 32
- weightedSd, Zi-method (weightedSd), 32
- weightedVar, 31, 32
- weightedVar (weightedSd), 32
- weightedVar, Zi-method (weightedSd), 32
- weights, 33
- weights, Zi-method (weights), 33
- weights<- (weights), 33
- weights<- , Zi-method (weights), 33
- zeroinfl, 34, 36, 37
- Zi, 3–5, 7, 8, 10–36
- Zi-class, 33
- zi2deseq2, 34
- zi2phyloseq, 35
- ziMain, 35
- ziMain, phyloseq-method (ziMain), 35
- ziMain, SummarizedExperiment-method (ziMain), 35
- zitoools (zitoools-package), 3
- zitoools-package, 3
- ~` (+), 4