

Package ‘msgbsR’

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Type Package

Title msgbsR: methylation sensitive genotyping by sequencing (MS-GBS)
R functions

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GenomicFeatures, GenomeInfoDb, ggbio, ggplot2, IRanges,
parallel, plyr, Rsamtools, R.utils, stats,
SummarizedExperiment, S4Vectors, utils

Suggests roxygen2, BSgenome.Rnorvegicus.UCSC.rn6

biocViews ImmunoOncology, DifferentialMethylation, DataImport,
Epigenetics, MethylationSeq

Description Pipeline for the analysis of a MS-GBS experiment.

License GPL-2

LazyLoad yes

Collate 'msgbsR.R' 'rawCounts.R' 'checkCuts.R' 'plotCounts.R'
'diffMeth.R' 'plotCircos.R'

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Contents

| | |
|---------------------|---|
| checkCuts | 2 |
| cuts | 3 |
| diffMeth | 3 |

| | |
|----------------------|---|
| msgbsR | 4 |
| plotCircos | 4 |
| plotCounts | 5 |
| ratdata | 5 |
| ratdata2 | 6 |
| rawCounts | 7 |

| | |
|--------------|----------|
| Index | 8 |
|--------------|----------|

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|-----------|------------------|
| checkCuts | <i>checkCuts</i> |
|-----------|------------------|

Description

Determines the sequence around a cut site using a fasta file or BSgenome

Usage

```
checkCuts(cutSites, genome, fasta = FALSE, seq)
```

Arguments

| | |
|----------|---|
| cutSites | A GRanges object containing the locations of the cut sites to be checked for sequence match. The names of the correct cut sites will be returned as a GRanges object. |
| genome | The path to a fasta file or a BSgenome object to check for genomic sequences. |
| fasta | TRUE if a fasta file has been supplied. Default = FALSE |
| seq | The desired recognition sequence that the enzyme should have cut. |

Value

A GRanges object containing the names of the sites that had the correct sequence.

Author(s)

Benjamin Mayne

Examples

```
library(GenomicRanges)
library(SummarizedExperiment)
library(BSgenome.Rnorvegicus.UCSC.rn6)
# Load the positions of possible MspI cut sites
data(ratdata)
# Extract the cut sites
cutSites <- rowRanges(ratdata)
# Adjust the cut sites to overlap recognition site on each strand
start(cutSites) <- ifelse(test = strand(cutSites) == '+',
  yes = start(cutSites) - 1, no = start(cutSites) - 2)
end(cutSites) <- ifelse(test = strand(cutSites) == '+',
  yes = end(cutSites) + 2, no = end(cutSites) + 1)
correctCuts <- checkCuts(cutSites = cutSites, genome = "rn6", seq = "CCGG")
```

| | |
|------|---|
| cuts | <i>A GRanges object of differentially methylated MspI cut sites on chromosome 20 in Rat from a MS-GBS experiment.</i> |
|------|---|

Description

The GRanges object was created from a list of differentially methylated cut sites from a MS-GBS experiment between two groups of rats that were fed either a control diet or a high fat diet.

Usage

```
data(cuts)
```

Format

A GRanges object of length 10.

Details

- Positions of MspI cut sites differentially methylated in the prostate on chromosome 20 in Rats.

The data set contains 10 differentially methylated sites in the prostate between rats fed a control or high fat diet.

Value

A GRanges object of length 10.

| | |
|----------|-----------------|
| diffMeth | <i>diffMeth</i> |
|----------|-----------------|

Description

Determines differential methylated sites from a RangedSummarizedExperiment

Usage

```
diffMeth(se, category, condition1, condition2,
         block = NULL, cpmThreshold, thresholdSamples)
```

Arguments

| | |
|------------------|--|
| se | A RangedSummarizedExperiment containing meta data of the samples. |
| category | The heading name in the sample data to be tested for differential methylation. |
| condition1 | The reference group within the category. |
| condition2 | The experimental group within the category. |
| block | The heading name in the sample data if differential methylation is to be tested with a blocking factor. Default is NULL. |
| cpmThreshold | Counts per million threshold of read counts to be filtered out of the analysis. |
| thresholdSamples | Minimum number of samples to contain the counts per million threshold. |

Value

A data frame containing which cut sites that are differentially methylated.

Author(s)

Benjamin Mayne

Examples

```
# Load data
data(ratdata2)
top <- diffMeth(se = ratdata2, cateogory = "Group",
               condition1 = "Control", condition2 = "Experimental",
               cpmThreshold = 1, thresholdSamples = 1)
```

| | |
|--------|---------------|
| msgbsR | <i>msgbsR</i> |
|--------|---------------|

Description

msgbsR

| | |
|------------|-------------------|
| plotCircos | <i>plotCircos</i> |
|------------|-------------------|

Description

Plot a circos representing the cut site locations

Usage

```
plotCircos(cutSites, seqlengths, cutSite.colour, seqlengths.colour)
```

Arguments

| | |
|-------------------|---|
| cutSites | A GRanges object containing the locations of the cut sites to be plotted. |
| seqlengths | An integer with the lengths of the chromosomes. |
| cutSite.colour | The colour of the cut sites. |
| seqlengths.colour | The colour of the chromosomes |

Value

A circos plot showing the locations of the cut sites.

Author(s)

Benjamin Mayne

Examples

```
# load example cut site positions
data(cuts)
# Obtain the length of chromosome 20 in rn6
library(BSgenome.Rnorvegicus.UCSC.rn6)
chr20 <- seqlengths(BSgenome.Rnorvegicus.UCSC.rn6)["chr20"]
plotCircos(cutSites = cuts, seqlengths = chr20,
           cutSite.colour = "red", seqlengths.colour = "blue")
```

plotCounts

*plotCounts***Description**

Plots the total number of reads vs total number of cut sites per sample

Usage

```
plotCounts(se, category)
```

Arguments

| | |
|----------|---|
| se | A RangedSummarizedExperiment containing meta data of the samples. |
| category | The heading name in the sample data to distinguish groups. |

Value

Produces a plot showing the total number reads vs total number of cut sites per sample.

Author(s)

Benjamin Mayne

Examples

```
data(ratdata2)
plotCounts(se = ratdata2, category = "Group")
```

ratdata

Read counts of potential MspI cut sites from a MS-GBS experiment of prostates from rats

Description

A RangedSummarizedExperiment containing read counts generated from a MS-GBS experiment using the restriction enzyme MspI, focusing on chromosome 20 of Rat.

Usage

```
data(ratdata)
```

Format

RangedSummarizedExperiment

Details

- ratdata A RangedSummarizedExperiment with 16047 potential MspI cut sites on chromosome 20 in Rat and six samples (3 Control and 3 Experimental).

This dataset contains six prostate samples from rats: 3 control and 3 experimental high fat diet.

Value

RangedSummarizedExperiment

| | |
|----------|--|
| ratdata2 | <i>Read counts of correct MspI cut sites from a MS-GBS experiment of prostates from rats</i> |
|----------|--|

Description

A RangedSummarizedExperiment containing read counts generated from a MS-GBS experiment using the restriction enzyme MspI, focusing on chromosome 20 of Rat. The sites have been checked for the correct recognition site.

Usage

```
data(ratdata2)
```

Format

RangedSummarizedExperiment

Details

- ratdata2 A RangedSummarizedExperiment containing data for 13983 MspI cut sites on chromosome 20 in Rat and six samples (3 Control and 3 Experimental).

This dataset contains six prostate samples from rats: 3 control and 3 experimental high fat diet. The data can be used for differential methylation analyses.

Value

RangedSummarizedExperiment

`rawCounts`*rawCounts*

Description

Imports the raw read counts from sorted and indexed bam file(s)

Usage

```
rawCounts(bamFilepath, threads = 1)
```

Arguments

| | |
|--------------------------|--|
| <code>bamFilepath</code> | The path to the location of the bam file(s). |
| <code>threads</code> | The total number of usable threads to be used. Default is 1. |

Value

Produces a `RangedSummarizedExperiment`. Columns are samples and the rows are cut sites. The cut site IDs are in the format `chr:position-position:strand`.

Author(s)

Benjamin Mayne, Sam Buckberry

Examples

```
my_path <- system.file("extdata", package = "msgbsR")
my_data <- rawCounts(bamFilepath = my_path)
```

Index

* datasets

cuts, [3](#)

ratdata, [5](#)

ratdata2, [6](#)

checkCuts, [2](#)

cuts, [3](#)

diffMeth, [3](#)

msgbsR, [4](#)

msgbsR-package (msgbsR), [4](#)

plotCircos, [4](#)

plotCounts, [5](#)

ratdata, [5](#)

ratdata2, [6](#)

rawCounts, [7](#)