# Package 'msgbsR'

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**Type** Package

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

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**Author** Benjamin Mayne

**Maintainer** Benjamin Mayne <benjamin.mayne@adelaide.edu.au>

**Depends** R (>= 3.4), GenomicRanges, methods

**Imports** BSgenome, easyRNASeq, edgeR, GenomicAlignments, 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, MethylSeq

**Description** Pipeline for the anaysis of a MS-GBS experiment.

**License** GPL-2

**LazyLoad** yes

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

**RoxygenNote** 5.0.1

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`checkCuts` *checkCuts*

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## 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 *A GRanges object of differentially methylated MspI cut sites on chromosome 20 in Rat from a MS-GBS experiment.*

---

## 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 *diffMeth*

## Description

Determines differential methylated sites from a RangedSummarizedExperiment

## Usage

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

## Arguments

| | |
|---|---|
| se | A RangedSummarizedExperiment containing meta data of the samples. |
| cateogory | The heading name in the sample data to be tested for differential methylation. |
| condition1 | The reference group within the cateogory. |
| condition2 | The experimental group within the cateogory. |
| 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 differenitally 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 *msgbsR*

## Description

msgbsR

plotCircos *plotCircos*

## 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, cateogory)
```

## Arguments

| | |
|---|---|
| se | A RangedSummarizedExperiment containing meta data of the samples. |
| cateogory | 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, cateogory = "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` *Read counts of correct 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. 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

| | |
|---|---|
| bamFilepath | The path to the location of the bam file(s). |
| threads | 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)
```

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