

Package ‘funtooNorm’

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Type Package

Title Normalization Procedure for Infinium HumanMethylation450
BeadChip Kit

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Description Provides a function to normalize Illumina Infinium Human
Methylation 450 BeadChip (Illumina 450K), correcting for tissue and/or cell
type.

License GPL-3

Imports pls, matrixStats, minfi, methods,
IlluminaHumanMethylation450kmanifest,
IlluminaHumanMethylation450kanno.ilmn12.hg19, GenomeInfoDb,
grDevices, graphics, stats

Suggests prettydoc, minfiData, knitr, rmarkdown

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funtooNorm-package	<i>funtooNorm</i>
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Description

The funtooNorm Package provides a normalization method for data arising from the Illumina Infinium Human Methylation 450 BeadChip (Illumina 450K), including explicit considerations of differences between tissues or cell types. This method should only be used when the data set contains samples fom multiple different tissues or cell types.

Details

Package: funtooNorm
Type: Package
License: GPL-3

Author(s)

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agreement	<i>Function to measure intra-replicate agreement for methylation data.</i>
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Description

Function to measure intra-replicate agreement for methylation data.

Usage

```
agreement(Beta, individualID)
```

Arguments

Beta : Matrix with beta-values, rows corresponding to probes, columns corresponding to samples.

individualID : a vector where 2 replicates have the exact same value for two technical replicates. Order of samples should nmatch the samples (columns) in Beta

Details

We expect that the values returned by the agreement function after normalization by funtooNorm to be smaller than before.

Value

The average value of the square distance between replicates: a measure of agreement between replicates in methylation data.

Examples

```
agreement(cbind(rnorm(n = 10), rnorm(n = 10), rnorm(n = 10)), c(1, 1, 1))
```

fromGenStudFiles	<i>Creates a S4 object of class 'SampleSet' from GenomeStudio files</i>
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Description

Creates a S4 object of class 'SampleSet' from GenomeStudio files

Usage

```
fromGenStudFiles(controlProbeFile, signalFile, cell_type)
```

Arguments

controlProbeFile	The control probe file exported from GenomeStudio
signalFile	The signals exported from GenomeStudio samples must be in same order as the control probe File
cell_type	A vector of cell types, names must match control probes and signal files.

Value

An object of class 'SampleSet'.

fromRGChannelSet	<i>Creates an object of class SampleSet from a RGChannelSet minfi</i>
------------------	---

Description

Creates a object of class SampleSet from the raw unprocessed data in RGChannelSet

Usage

```
fromRGChannelSet(myRGChannelSet)
```

Arguments

myRGChannelSet : RGChannelSet, from minfi package, should contain a cell_type vector in pData

Value

An object of class 'SampleSet'

Examples

```
require(minfiData)
pData(RGsetEx)$cell_type <- rep(c("type1", "type2"), 3)
mySampleSet=fromRGChannelSet(RGsetEx)
```

funtooNorm	<i>The funtooNorm normalization function</i>
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Description

funtooNorm Returns the normalized signals to the SampleSet object

Usage

```
funtooNorm(object, type.fits = "PCR", ncmp = 4, force = FALSE,
           sex = NULL)
```

```
## S4 method for signature 'SampleSet'
funtooNorm(object, type.fits = "PCR", ncmp = 4,
           force = FALSE, sex = NULL)
```

Arguments

object	Object of class SampleSet
type.fits	Choice between "PCR" or "PLS" (default="PCR")
ncmp	Number of components included in the analysis (default=4)
force	If set to TRUE, forces the normalization procedure to re-compute
sex	Boolean vector if male. if NULL Beta values from ChrY are used for classification.

Details

This is a generic function which applies to autosomes and the X chromosome. Chromosome Y requires separate analysis as there are few probes on Y. We use a straightforward quantile normalization applied to males only.

Value

a S4 object of class SampleSet containing the normalized signal

Methods (by class)

- SampleSet: The funtooNorm normalization function

Examples

```
require(minfiData)
pData(RGsetEx)$cell_type <- rep(c("type1", "type2"), 3)
mySampleSet=fromRGChannelSet(RGsetEx)
mySampleSet=funtooNorm(mySampleSet)
```

getGRanges	<i>Build GRRange object of methylation probes</i>
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Description

Build GRRange object of methylation probes

Usage

```
getGRanges(object)

## S4 method for signature 'SampleSet'
getGRanges(object)
```

Arguments

object Object of class SampleSet.

Value

A GRRange object of the positions of each cpg.

Methods (by class)

- SampleSet: Build GRRange object of methylation probes

Examples

```
require(minfiData)
pData(RGsetEx)$cell_type <- rep(c("type1", "type2"), 3)
mySampleSet=fromRGChannelSet(RGsetEx)
gr=getGRanges(mySampleSet)
```

getNormBeta	<i>Computes Beta values from normalized signals</i>
-------------	---

Description

Computes Beta values from normalized signals

Usage

```
getNormBeta(object, offset = 100)

## S4 method for signature 'SampleSet'
getNormBeta(object, offset = 100)
```

Arguments

object of type SampleSet
 offset default is 100 as Illumina standard

Value

a matrix containing beta after normalization value for each CpG position and each samples

Methods (by class)

- SampleSet: Computes Beta values from normalized signals

Examples

```
require(minfiData)
pData(RGsetEx)$cell_type <- rep(c("type1", "type2"), 3)
mySampleSet=fromRGChannelSet(RGsetEx)
b=getNormBeta(funtooNorm(mySampleSet))
```

getNormM

Computes M values, log₂(Meth/Unmeth), from normalized signals

Description

Computes M values, log₂(Meth/Unmeth), from normalized signals

Usage

```
getNormM(object)

## S4 method for signature 'SampleSet'
getNormM(object)
```

Arguments

object An object of class SampleSet

Value

a matrix containing M values, log₂(Meth/Unmeth), after normalization

Methods (by class)

- SampleSet: Computes M values, log₂(Meth/Unmeth), from normalized signals

Examples

```
require(minfiData)
pData(RGsetEx)$cell_type <- rep(c("type1", "type2"), 3)
mySampleSet=fromRGChannelSet(RGsetEx)
m=getNormM(funtooNorm(mySampleSet))
```

getRawBeta

Computes Beta value from raw signals

Description

Computes Beta value from raw signals

Usage

```
getRawBeta(object, offset = 100)

## S4 method for signature 'SampleSet'
getRawBeta(object, offset = 100)
```

Arguments

object	object of class SampleSet
offset	default is 100 as Illumina standard

Value

a matrix containing the raw beta value for each position and each samples

Methods (by class)

- SampleSet: Computes Beta value from raw signals

Examples

```
require(minfiData)
pData(RGsetEx)$cell_type <- rep(c("type1", "type2"), 3)
mySampleSet=fromRGChannelSet(RGsetEx)
r=getRawBeta(mySampleSet)
```

getSnpM

*Computes M values after normalization of SNP data.***Description**

Computes M values after normalization of SNP data.

Usage

```
getSnpM(object)

## S4 method for signature 'SampleSet'
getSnpM(object)
```

Arguments

object of class SampleSet

Value

a matrix containing M values, $\log_2(\text{Meth}/\text{Unmeth})$, after normalization for SNP data

Methods (by class)

- SampleSet: Computes M values, $\log_2(\text{Meth}/\text{Unmeth})$, for normalized SNP data

Examples

```
require(minfiData)
pData(RGsetEx)$cell_type <- rep(c("type1", "type2"), 3)
mySampleSet=fromRGChannelSet(RGsetEx)
snp=getSnpM(funtooNorm(mySampleSet))
```

plotValidationGraph

*plot of Validation Graph for determing number of components***Description**

Plots a series of graphs for each signal type, to determine the number of components to include in the normalization procedure.

Usage

```
plotValidationGraph(object, type.fits = "PCR", pdf.file = NULL)

## S4 method for signature 'SampleSet'
plotValidationGraph(object, type.fits = "PCR",
  pdf.file = NULL)
```

Arguments

<code>object</code>	of class <code>SampleSet</code>
<code>type.fits</code>	can be "PCR" or "PLS" (default "PCR")
<code>pdf.file</code>	if no file name is provided print pdf file <code>plotValidationGraph.pdf</code> in working directory.

Value

No value is returned. The function prints the plots to a pdf file.

Methods (by class)

- `SampleSet`: Plots a series of graphs for each signal type, to determine the number of components to include in the normalization procedure.

Examples

```
require(minfiData)
pData(RGsetEx)$cell_type <- rep(c("type1", "type2"), 3)
mySampleSet=fromRGChannelSet(RGsetEx)
plotValidationGraph(mySampleSet)
```

<code>SampleSet-class</code>	<i>S4 class object <code>SampleSet</code></i>
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Description

`SampleSet` is an S4 class defined for the purpose of running the `funtooNorm` algorithm. They are lists containing signal data and different variables useful for `funtooNorm`. The data is separated into the 3 probes types, each having 2 channels (methylated and unmethylated ie : A and B) We then define then the 6 (2*3) labels: AIGrn BIGrn AIRed BIRed AII BII

Value

a S4 object of class `SampleSet`

Slots

type Character: is 'minfi' or 'GenomeStudio'
 sampleNames character vector: contain the list of sample names in order used
 sampleSize numeric: the number of samples
 nPos numeric: the number of positions in the ILLUMINA chip
 annotation character: the annotation object from minfi package
 cell_type factor: vector of the cell type for each sample as factors
 qntllist numeric: vector of ordered quantiles
 quantiles list: list of 6 quantiles tables for the 6 signal types
 ctl.covmat matrix: covariance matrix for the model fit
 signal list: list of the values for all 6 probe types.
 names list: list of probes for each type
 predmat list: list of the normalized values for all 6 probe types.

Examples

```
showClass("SampleSet")
```

show, SampleSet-method *Show Object SampleSet*

Description

Display informations about the SampleSet object

Usage

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

Arguments

object an object of class SampleSet
 ... optional arguments passed to or from other methods.

Value

No value is returned. The function prints the summary of object of class SampleSet to screen

Examples

```

require(minfiData)
pData(RGsetEx)$cell_type <- rep(c("type1", "type2"), 3)
mySampleSet=fromRGChannelSet(RGsetEx)
mySampleSet
```

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