# Package 'lumi'

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Type Package

**Title** BeadArray Specific Methods for Illumina Methylation and Expression Microarrays

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**Depends** R (>= 2.10), Biobase (>= 2.5.5)

Imports affy (>= 1.23.4), methylumi (>= 2.3.2), GenomicFeatures, GenomicRanges, annotate, lattice, mgcv (>= 1.4-0), nleqslv, KernSmooth, preprocessCore, RSQLite, DBI, AnnotationDbi, MASS, graphics, stats, stats4, methods

**Suggests** beadarray, limma, vsn, lumiBarnes, lumiHumanAll.db, lumiHumanIDMapping, genefilter, RColorBrewer

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Description The lumi package provides an integrated solution for the Illumina microarray data analysis. It includes functions of Illumina BeadStudio (GenomeStudio) data input, quality control, BeadArray-specific variance stabilization, normalization and gene annotation at the probe level. It also includes the functions of processing Illumina methylation microarrays, especially Illumina Infinium methylation microarrays.

**License** LGPL (>= 2)

LazyLoad yes

**biocViews** Microarray, OneChannel, Preprocessing, DNAMethylation, QualityControl, TwoChannel

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lumi-package

A package for preprocessing Illumina microarray data

#### **Description**

lumi R package is designed to preprocess the Illumina microarray (BeadArray) data. It includes functions of Illumina data input, quality control, variance stabilization, normalization and gene annotation.

#### **Details**

Package: lumi
Type: Package
Version: 1.1.0
Date: 2007-03-23

License: LGPL version 2 or newer

#### Author(s)

Pan Du, Simon Lin Maintainer: Pan Du <dupan@northwestern.edu>

#### References

- 1. Du, P., Kibbe, W.A. and Lin, S.M., (2008) 'lumi: a pipeline for processing Illumina microarray', Bioinformatics 24(13):1547-1548
- 2. Lin, S.M., Du, P., Kibbe, W.A., (2008) 'Model-based Variance-stabilizing Transformation for Illumina Microarray Data', Nucleic Acids Res. 36, e11
- 3. Du, P., Kibbe, W.A. and Lin, S.M., (2007) 'nuID: A universal naming schema of oligonucleotides for Illumina, Affymetrix, and other microarrays', Biology Direct, 2, 16

addAnnotationInfo

Add probe color channel and basic annotation information based on the annotation library of Illumina methylation microarray

### Description

Add probe color channel and basic annotation information based on the annotation library of Illumina methylation microarray

# Usage

addAnnotationInfo(methyLumiM, lib = 'FDb.InfiniumMethylation.hg19', annotationColumn=c('COLOR\_CHANNEI

addControlData2lumi 5

#### **Arguments**

methyLumiM a MethyLumiM object includes Illumina Infinium methylation data

1ib Annotation library of Illumina methylation microarray.

annotationColumn

only include 'COLOR\_CHANNEL', 'CHROMOSOME' and 'POSITION' in-

formation

#### **Details**

The "lib" parameter supports both FeatureDb based annotation libraries and old array-based annotation libraries. 'FDb.InfiniumMethylation.hg19' is the FeatureDb based annotation library, which includes both 450k and 27k data. "IlluminaHumanMethylation27k.db" (for 27k array) and "IlluminaHumanMethylation450k.db" (450k infinium array) are old types of annotation libraries.

#### Value

return the MethyLumiM object with COLOR\_CHANNEL, CHROMOSOME and chromome PO-SITION information added to the featureData.

#### Author(s)

Pan DU

#### See Also

lumiMethyR

# **Examples**

```
data(example.lumiMethy)
head(pData(featureData(example.lumiMethy)))
## removing color channel information
# testData = example.lumiMethy
# pData(featureData(testData))$COLOR_CHANNEL = NULL
# testData = addAnnotationInfo(testData, lib="IlluminaHumanMethylation27k.db")
## check whether the color channel information is added
# head(pData(featureData(testData)))
```

addControlData2lumi

Add the control probe data into the controlData slot of LumiBatch object

#### **Description**

Add the control probe profile data, outputted by BeadStudio, into the controlData slot of LumiBatch object.

#### Usage

```
addControlData2lumi(controlData, x.lumi)
```

#### **Arguments**

controlData the control data can be a data.frame or the control probe filename outputted by

BeadStudio

x.lumi a LumiBatch object, to which controlData will be added.

#### **Details**

The controlData slot in LumiBatch object is a data.frame with first two columns as "controlType" and "ProbeID". The rest columns are the expression amplitudes for individual samples.

### Value

Return the LumiBatch object with controlData slot filled.

### Author(s)

Pan Du

#### See Also

```
getControlData, plotControlData
```

# **Examples**

```
## Not runnable
# controlFile <- 'Control_Probe_Profile.txt'
# x.lumi <- addControlData2lumi(controlFile, x.lumi)</pre>
```

addControlData2methyLumiM

Add methylation control data to a MethyLumiM object

#### **Description**

Add methylation control data to a MethyLumiM object

### Usage

```
addControlData2methyLumiM(controlData, methyLumiM, checkConsistency = TRUE, ...)
```

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#### **Arguments**

controlData a methylation control data file (output by GenomeStudio), or a MethyLumiQC

object

methyLumiM a MethyLumiM object to add control data

checkConsistency

whether to check the sample names consistency between methyLumiM and con-

trolData

... other parameters for reading controlData

#### **Details**

This function aims to add the controlData (MethyLumiQC object) to the controlData slot of a methyLumiM object For control data, methylated data matrix in assayData slot corresponds to green channel, and unmethylated data matrix in assayData slot corresponds to red channel.

#### Value

Return the methyLumiM object with the controlData added

#### Author(s)

Pan DU

#### See Also

lumiMethyR

addNuID2lumi

Add the nuID information to the LumiBatch object

# **Description**

Replace the Illumina Id (Target ID or Probe Id) as nuID (nucleotide universal identifier) for indexing genes in the LumiBatch object

#### Usage

```
addNuID2lumi(x.lumi, annotationFile=NULL, sep = NULL, lib.mapping = NULL, annotationColName = c(sequer
```

#### **Arguments**

x.lumi a LumiBatch object

annotationFile a annotation file, which includes the Illumina ID (target or probe ids) and probe

sequence information

sep the separation used in the annotation file. Automatically detect the separator if

it is "," or "\t".

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lib.mapping a Illumina ID mapping package, e.g, lumiHumanIDMapping annotationColName

the annotation column name in the annotation file used for the probe sequence

and TargetID and ProbeID

verbose a boolean to decide whether to print out some messages

#### **Details**

Since the default Illumina IDs (TargetID (ILMN\\_Gene ID) and ProbeId (Probe\\_Id)) are not consistent between different arrays and batches, we invented a nuID, which is one-to-one matching with the probe sequence. This function is to replace the Illumina ID with the nuID. If the annotation library (the unzipped manifest file (.bgx)) is provided, the function will automatically check whether the Illumina ID is provided for the microarray data. We recommend output the data using ProbeID when using Illumina BeadStudio software, because the TargetID (ILMN\\_Gene ID) are not unique.

#### Value

a LumiBatch object with Illumina ID replaced by nuID.

#### Author(s)

Pan Du

### References

Du, P., Kibbe, W.A., Lin, S.M., "nuID: A universal naming schema of oligonucleotides for Illumina, Affymetrix, and other microarrays", submitted.

### See Also

```
IlluminaID2nuID, lumiR
```

```
## load example data
# data(example.lumi)

## specify the annotation file for the Illumina chip
# annotationFile <- 'Human_RefSeq-8.csv'

## Replace the Target ID with nuID
# lumi.nuID <- addNuID2lumi(example.lumi, annotationFile)

## An alternative way is to load the Annotation library and match the targetID (or Probe Id) with nuID
# lumi.nuId <- addNuID2lumi(example.lumi, lib.mapping='lumiHumanIDMapping')</pre>
```

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adjColorBias.quantile Color bias adjustment of Illumina Infinium methylaton microarrays using smooth quantile normalization

### Description

Color bias adjustment of Illumina Infinium methylaton microarrays using smooth quantile normalization smoothQuantileNormalization

### Usage

```
adjColorBias.quantile(methyLumiM, refChannel = c("green", "red"), logMode = TRUE, verbose = TRUE,...)
```

### Arguments

methyLumiM	a MethyLumiM object or any eSet object with "methylated" and "unmethylated" data matrix element in the assayData slot
refChannel	the reference color channel for color bias adjustment
logMode	whether perform the adjustment in log scale or not
verbose	whether print extra information during processing
	other parameters used by smoothQuantileNormalization

#### **Details**

Perform color bias adjustment of Illumina Infinium methylaton microarrays. It requires the input methyLumiM object includes the color channel information in the featureData. Basically, there should be a "COLOR\_CHANNEL" column in the data.frame returned by pData(featureData(methyLumiM)).

The basic idea of color bias adjustment is to treat it as the normalization between two color channels. It uses smooth quantile normalization smoothQuantileNormalization to normalize two color channels.

#### Value

Return an object (same class as input methyLumiM) with updated "methylated" and "unmethylated" data matrix after color bias adjustment.

# Author(s)

Pan DU

#### See Also

See Also lumiMethyC, smoothQuantileNormalization and adjColorBias.ssn

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#### **Examples**

```
data(example.lumiMethy)
# before adjustment
plotColorBias1D(example.lumiMethy)
lumiMethy.adj = adjColorBias.quantile(example.lumiMethy)
# after adjustment
plotColorBias1D(lumiMethy.adj)
```

adjColorBias.ssn

Color bias adjustment of Illumina Infinium methylaton microarrays

using simple shift and scaling normalization

### **Description**

Color bias adjustment of Illumina Infinium methylaton microarrays using simple shift and scaling normalization

# Usage

```
adjColorBias.ssn(methyLumiM, refChannel = c("green", "red", "mean"))
```

### Arguments

methyLumiM a MethyLumiM object or any eSet object with "methylated" and "unmethylated"

data matrix element in the assayData slot

refChannel the reference color channel for color bias adjustment

### Details

Perform color bias adjustment of Illumina Infinium methylaton microarrays. It requires the input methyLumiM object includes the color channel information in the featureData. Basically, there should be a "COLOR\_CHANNEL" column in the data.frame returned by pData(featureData(methyLumiM)).

The basic idea of color bias adjustment is to treat it as the normalization between two color channels. It uses simple scaling normalization to normalize two color channels. The background levels are estimated using function estimateMethylationBG.

### Value

Return an object (same class as input methyLumiM) with updated "methylated" and "unmethylated" data matrix after color bias adjustment.

#### Author(s)

Pan DU

### See Also

See Also lumiMethyC, estimateMethylationBG and adjColorBias.quantile

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### **Examples**

```
data(example.lumiMethy)
# before adjustment
plotColorBias1D(example.lumiMethy)
lumiMethy.adj = adjColorBias.ssn(example.lumiMethy)
# after adjustment
plotColorBias1D(lumiMethy.adj)
```

asBigMatrix-methods

convert the data matrix in the assayData of a ExpressionSet as Big-Matrix

#### **Description**

convert the data matrix in the assayData of a ExpressionSet as BigMatrix

#### Usage

```
## S4 method for signature 'ExpressionSet'
asBigMatrix(object, rowInd=NULL, colInd=NULL, nCol=NULL, dimNames=NULL, saveDir='.', savePrefix=NULL,
```

### **Arguments**

object	an object of ExpressionSet or its inherited class
rowInd	the subset of row index
colInd	the subset of column index
nCol	the number of columns of the data, which can be larger than the real data dimension. It is designed for adding future data.
dimNames	the dimension names, which is a list of two character vectors (rownames and colnames)
saveDir	the parent directory to save the BigMatrix data files
savePrefix	the folder name prefix of the directory to save the BigMatrix data files. The fold name will be like this: paste(savePrefix, '_bigmat', sep=")
	optional arguments to BigMatrix

### **Details**

This function does not work in Windows because the dependent package bigmemoryExtras does not support it. In order to make lumi package still compilation under Windows, I deliberately remove the dependency of bigmemoryExtras package. As a result, users need to manually load the bigmemoryExtras function before using this function.

The BigMatrix data files will be save in the directory file.path(saveDir, paste(savePrefix, '\_bigmat', sep="))

#### See Also

```
BigMatrix
```

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beta2m

Convert methylation Beta-value to M-value

### **Description**

Convert methylation Beta-value to M-value through a logistic transformation

# Usage

```
beta2m(beta)
```

### **Arguments**

beta

a matrix or vector of methylation Beta-value

#### **Details**

Convert methylation Beta-value to M-value through a logistic transformation

#### Value

return methylation M-value with the same size of input Beta-value

### Author(s)

Pan Du

### References

Du, P., Zhang, X, Huang, C.C., Jafari, N., Kibbe, W.A., Hou, L., and Lin, S.M., (2010) 'Comparison of Beta-value and M-value methods for quantifying methylation levels by microarray analysis', (under review)

### See Also

See Also as m2beta

bgAdjust 13

bgAdjust	Background adjustment for Illumina data

#### **Description**

The method adjusts the data by subtracting an offset, which is estimated based on the quantile of the control probes

# Usage

```
bgAdjust(lumiBatch, probs = 0.5, ...)
```

# Arguments

 lumiBatch
 A LumiBatch object with controlData slot include control probe information

 probs
 The quantile used to estimate the background

 ...
 other parameters used by quantile method

#### **Details**

The method adjusts the data by subtracting an offset, which is estimated based on the quantile of the control probes. The control probe information is kept in the controlData slot of the LumiBatch object. If no control data information, the method will do nothing.

### Value

It returns a LumiBatch object with background adjusted.

# Author(s)

Pan Du

### See Also

1umiB

```
data(example.lumi)
## Here will assume the minimum of the control probe as the background,
## because there is no negative control (blank beads) information for the Barnes data.
example.lumi.b <- bgAdjust(example.lumi, probs=0)</pre>
```

bgAdjustMethylation

### **Description**

Estimate and adjust the background levels of Illumina Infinium methylaton microarrays

#### Usage

bgAdjustMethylation(methyLumiM, separateColor = FALSE, targetBGLevel = 300, negPercTh = 0.25)

### **Arguments**

methyLumiM a MethyLumiM object or any eSet object with "methylated" and "unmethylated"

data matrix element in the assayData slot

separateColor determine whether separately process two color channels targetBGLevel adjust background level to a non-zero target background level

negPercTh the threshold of the percentage of negative values after subtract estimated back-

ground levels. A warning will be given if too many probes having intensities

lower than background levels.

#### **Details**

The estimation of background level of Infinium methylaton microarray is based on the assumption that the lots of CpG sites are unmethylated, which results in a density mode of the intensities measured by methylated probes. The position of this mode represents the background level.

#### Value

Return an object (same class as input methyLumiM) with updated "methylated" and "unmethylated" data matrix after background level adjustment. The estimated background level was kept in the attribute, "EstimatedBG", of the returned methyLumiM object.

### Author(s)

Pan DU

#### See Also

See Also lumiMethyB and estimateMethylationBG

```
data(example.lumiMethy)
lumiMethy.bgAdj = bgAdjustMethylation(example.lumiMethy)
attr(lumiMethy.bgAdj, "EstimatedBG")
```

boxplot, MethyLumiM-method

boxplot of a MethyLumiM object

# Description

Creating a hdr.boxplot of the M-value in a MethyLumiM object

# Usage

```
## S4 method for signature 'MethyLumiM'
boxplot(x, main, logMode = TRUE, ...)
```

### **Arguments**

x a MethyLumiM-class object

main title of the boxplot

logMode only works when the dataType of x is "Intensity"

... optional arguments to bwplot.

#### **Details**

Because the density plot of M-values usually includes two modes, using the traditional boxplot cannot accurately represent the distribution of the data. Here we use violin plot to show the density of M-values by samples

### See Also

MethyLumiM-class, panel.violin and boxplot,ExpressionSet-method

```
## load example data
data(example.lumiMethy)
boxplot(example.lumiMethy)
```

16 boxplot-methods

boxplot-methods	boxplot of a ExpressionSet object

# **Description**

Creating boxplot of sample intensities in a ExpressionSet object

### Usage

```
## S4 method for signature 'ExpressionSet'
boxplot(x, range = 0, main, logMode = TRUE, subset = NULL, xlab = "", ylab = "Amplitude", ...)
```

### **Arguments**

X	a ExpressionSet object
range	parameter of boxplot
main	title of the boxplot
logMode	whether plot the data in log2 scale or not
subset	subset of rows used to plot. It can be an index vector, or the length of a random subset
xlab	xlab of the plot
ylab	ylab of the plot
	optional arguments to boxplot.

### **Details**

The boxplot function has a "subset" parameter. By default, it is set as 5000, i.e., randomly selected 5000 probes to plot the boxplot. The purpose of this is to plot the picture faster, but it will also make the boxplot has slightly different each time. If the user wants to make sure the boxplot is the same each time, you can set the "subset" parameter as NULL.

# See Also

LumiBatch-class, boxplot and boxplot, MethyLumiM-method

```
## load example data
data(example.lumi)
boxplot(example.lumi)
```

boxplotColorBias 17

boxplotColorBias Plot the Illumina Infinium methylation color bias in terms of boxplot	boxplotColorBias	Plot the Illumina Infinium methylation color bias in terms of boxplot
--	------------------	---

#### **Description**

Plot the Illumina Infinium methylation color bias in terms of boxplot. boxplot of red and green color channel will be plotted side by side

### Usage

```
boxplotColorBias(methyLumiM, logMode = TRUE, channel = c("both", "unmethy", "methy", "sum"), grid = TRU
```

### **Arguments**

methyLumiM MethyLumiM-class object or eSet-class object, which include methylated and

unmethylated probe intensities

logMode whether plot the intensities in log-scale channel estimate the intensity in different methods

grid whether to add grid on the plot

main title of the plot mar margin of the plot

verbose whether print verbose information during plot

subset plot subset of randomly selected rows. All data will be plotted if it is NULL.

... other parameters of boxplot

# **Details**

Plot the Illumina Infinium methylation color bias in terms of boxplot. boxplot of red and green color channel will be plotted side by side

#### Value

Invisibly return TRUE if plot successfully.

# Author(s)

Pan DU

#### See Also

See Also as boxplot and plotColorBias1D

```
data(example.lumiMethy)
boxplotColorBias(example.lumiMethy)
```

18 density-methods

|--|--|--|

#### **Description**

A summary of colorBias information, which is a data.frame summarizing the intensities of individual samples

### Usage

```
colorBiasSummary(methyLumiM, logMode = TRUE, channel = c("both", "unmethy", "methy", "sum"))
```

### **Arguments**

methyLumiM MethyLumiM-class object or eSet-class object, which include methylated and

unmethylated probe intensities

logMode Whether plot the intensities in log-scale channel estimate the intensity in different methods

#### **Details**

A summary of colorBias information. There are four options using "channel" parameter to plot the density plot. "both": estimate the density by pooling together methylated and unmethylated probe intensities. "unmethy" and "methy": plot either unmethylated or methylated probe density. "sum" plot the density of the sum of methylated and unmethylated probe intensitys.

#### Value

A data frame summarizing the intensities of individual samples

# Author(s)

Pan DU

thous Density plot of a Expressionset object	density-methods	Density plot of a ExpressionSet object
--	-----------------	--

### Description

Creating density plot of sample intensities in a ExpressionSet object. It is equivalent to hist-methods.

density-methods 19

### Usage

```
## S4 method for signature 'ExpressionSet'
density(x, logMode=TRUE, xlab = NULL, ylab = "density", type = "l",
col=1:dim(x)[2], lty=1:dim(x)[2], lwd=1, xlim = NULL, index.highlight = NULL, color.highlight = 2,
symmetry = NULL, addLegend = TRUE, legendPos="topright", subset = NULL, main="", ...)
```

### **Arguments**

x	a ExpressionSet object
logMode	determine whether the density plot is based on a log2 scale
xlab	xlab of the density plot
ylab	ylab of the density plot
type	parameter of plot function
col	line colors of the density plot
lty	line types of the density plot
lwd	line width of plot function
xlim	parameter of the plot function
index.highligh	t
	the column index of the highlighted density curve
color.highligh	
	color of highlighted density curve
symmetry	the boundary position suppose to be symmetric distributed
addLegend	whether add legend to the plot or not
legendPos	the legend position. It can be a string specifying the position, or a length two vector specifying the x and y position. Please check legend for more details.
subset	subset of rows used to plot. It can be an index vector, or the length of a random subset
main	title for the plot
	additional parameters for density function

#### See Also

LumiBatch-class, hist-methods, density

```
## load example data
data(example.lumi)
density(example.lumi)
```

20 detectionCall

				_
det	ect	i∩n	Cal	ı

Estimate the detectable probe ratio

### Description

Estimate the detectable probe ratio of each probe, sample or just return an AP matrix

### Usage

```
detectionCall(x.lumi, Th = 0.01, type = c('probe', 'sample', 'matrix'))
```

### **Arguments**

x. lumi a LumiBatch or MethyLumiM object

Th the threshold. By default, when the detection p-value is less than 0.01, we sup-

pose it is detectable. For the old version of BeadStudio output (version 2 or earlier), the threshold will automatically transferred as 1 - Th, because in the old

format, value close to 1 is suppose to be detectable.

type determine to calculate the detection count by probe or by sample

#### Value

If the type is 'probe', then returns the presentCount of each probe. If the type is 'sample', then return the detectable probe ratio of each sample. If the type is 'matrix', then return the AP matrix, in which 'A' represents absent (the detect p-value less than threshold) and 'P' represents present.

### Author(s)

Pan Du

#### See Also

lumiQ

```
## load example data
data(example.lumi)
## load example data
data(example.lumi)

## estimate the detect call (percentage of expressed genes) of each sample
temp <- detectionCall(example.lumi, type='sample')
print(temp)

## estimate the present count of each gene (probe)
temp <- detectionCall(example.lumi, type='probe')
hist(temp)</pre>
```

detectOutlier 21

sample (or gene)	Detect the out	detectOutlier
------------------	----------------	---------------

#### **Description**

Detect the outlier sample (or gene) based on distance to the cluster center

# Usage

```
detectOutlier(x, metric = "euclidean", standardize = TRUE, Th = 2, ifPlot = FALSE)
```

### **Arguments**

x a LumiBatch object, ExpressionSet object or a matrix with each column corre-

sponding to a sample or other profile

metric the distance matric

standardize standardize the profile or not

Th the threshold of outlier,

ifPlot to plot the result (as a hierarchical tree) or not

#### **Details**

The current outlier detection is based on the distance from the sample to the center (average of all samples after removing 10 percent samples farthest away from the center). The assumption of the outlier detection is that there is only one single cluster and the distance from the sample to the center is Gaussian distributed.

The outlier is detected when its distance to the center is larger than a certain threshold. The threshold is calculated as Th \* median distances to the center.

The profile relations can be visualized as a hierarchical tree.

### Value

Plot the results or return the outlier (a logic vector) with the distance matrix and threshold as attributes.

#### Author(s)

Pan Du

#### See Also

lumiQ

22 estimateBeta

#### **Examples**

```
## load example data
data(example.lumi)

## detect the outlier (Further improvement needed.)
temp <- detectOutlier(example.lumi, ifPlot=TRUE)</pre>
```

estimateBeta

Estimate methylation Beta-value matrix

### **Description**

Estimate methylation Beta-value matrix from MethyLumiM-class object or eSet-class object, which include methylated and unmethylated probe intensities

#### Usage

```
estimateBeta(methyLumiM, returnType=c("ExpressionSet", "matrix"), offset = 100)
```

# Arguments

methyLumiM MethyLumiM-class object or eSet-class object, which include methylated and

unmethylated probe intensities

returnType determine whether return an ExpressionSet or matrix object

offset An offset value added to the denominator to avoid close to zero intensities

### Details

Beta-value is ratio between Illumina methylated probe intensity and total probe intensities (sum of methylated and unmethylated probe intensities, see <a href="mateIntensity">estimateIntensity</a>). An offset value added to the denominator to avoid close to zero intensities in the denominator. Beta-value is in the range of 0 and 1. If we assume the probe intensity follows Gamma distribution, then the Beta-value follows a Beta distribution.

#### Value

An ExpressionSet or matrix object of methylation Beta-value

### Author(s)

Pan DU

### See Also

See Also as estimateIntensity and estimateM

estimateIntensity 23

### **Examples**

```
data(example.lumiMethy)
methyLumiBeta = estimateBeta(example.lumiMethy)
density(methyLumiBeta)
```

estimateIntensity

Estimate intensity of Illumina methylation data

### **Description**

Estimate intensity of Illumina methylation data, which is the sum of Illumina methylated and unmethylated probe intensities

### Usage

```
estimateIntensity(methyLumiM, returnType=c("ExpressionSet", "matrix"))
```

#### **Arguments**

methyLumiM MethyLumiM-class object or eSet-class object, which include methylated and

unmethylated probe intensities

returnType determine whether return an ExpressionSet or matrix object

#### **Details**

The Intensity basically is the sum of Illumina methylated and unmethylated probe intensities.

#### Value

An ExpressionSet or matrix object of methylation Intensity-value

### Author(s)

Pan DU

# See Also

See Also as estimateBeta and estimateM

```
data(example.lumiMethy)
methyLumiIntensity = estimateIntensity(example.lumiMethy)
boxplot(methyLumiIntensity)
```

24 estimateLumiCV

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Estimate the coefficient of variance matrix of LumiBatch object

# Description

Estimate the coefficient of variance matrix of LumiBatch object for each measurement or probe.

### Usage

```
estimateLumiCV(x.lumi, type = c("measurement", "probe"), ifPlot = FALSE, ...)
```

# **Arguments**

x.lumi a LumiBatch object type estimate the coefficient of variance of each measurement or each probe

ifPlot determine whether to plot the density plot or not

... optional arguments to plot.

#### **Details**

By default, the coefficient of variance is the ratio of the mean and variance of the bead expression values. Basically, it is the ration of exprs and se.exprs element of LumiBatch object. If the type is "probe", it is the ratio of the mean and variance of probe expression profile.

#### Value

A matrix of coefficient of variance

# Author(s)

Pan Du

### See Also

lumiQ

```
## load example data
data(example.lumi)

## estimate the coefficient of variance and plot the density plot of it
cv <- estimateLumiCV(example.lumi, ifPlot = TRUE)</pre>
```

estimateM 25

estimateM	Estimate methylation M-value matrix	

### **Description**

Estimate methylation M-value matrix from MethyLumiM-class object or eSet-class object, which include methylated and unmethylated probe intensities

### Usage

```
estimateM(methyLumiM, returnType=c("ExpressionSet", "matrix"), offset=100)
```

### **Arguments**

offset

methyLumiM	MethyLumiM-class object or eSet-class object, which include methylated and unmethylated probe intensities
returnType	determine whether return an ExpressionSet (MethyLumiM in this case) or matrix object

offset added to the methylated and unmethylated probe intensities when estimating the Marsha

ing the M-value

# **Details**

M-value is the log2 ratio between Illumina methylated and unmethylated probe intensities. As variations of small intensities can cause big changes in the ratio estimation, so an offset is added to methylated and unmethylated probe intensities when estimating the M-value.

### Value

A MethyLumiM or matrix object of methylation M-value

# Author(s)

Pan DU

### References

Du, P., Zhang, X, Huang, C.C., Jafari, N., Kibbe, W.A., Hou, L., and Lin, S.M., (2010) 'Comparison of Beta-value and M-value methods for quantifying methylation levels by microarray analysis', (under review)

#### See Also

See Also as estimateBeta, estimateIntensity

### **Examples**

```
data(example.lumiMethy)
methyLumiM = estimateM(example.lumiMethy)
boxplot(methyLumiM)
```

### Description

Estimate the background levels of Illumina Infinium methylaton microarrays. It is called by function bgAdjustMethylation

### Usage

```
estimateMethylationBG(methyLumiM, separateColor = FALSE, nbin = 1000)
```

#### **Arguments**

methyLumiM a MethyLumiM object or any eSet object with "methylated" and "unmethylated"

data matrix element in the assayData slot

separateColor determine whether to separately process two color channels

nbin the number of bins in the histogram used to estimate the mode position of the

density

#### **Details**

When the controlData includes the negative control probe information, the background estimation will be the median of the negative control probes. Red and Green color channels will be estimated separately.

In the case the negative control data is not available, the background will be estimated based on the mode positions of unmethylated or methylated distribution (the smaller one). The assumption is that the lots of CpG sites are unmethylated, which results in a density mode of the intensities measured by methylated probes. The position of this mode represents the background level.

#### Value

a vector of estimated background levels for individual samples.

#### Author(s)

Pan DU

### See Also

See Also lumiMethyB and bgAdjustMethylation

example.lumi 27

### **Examples**

```
data(example.lumiMethy)
estimatedBG = estimateMethylationBG(example.lumiMethy)
estimatedBG
```

example.lumi

Example Illumina Expression data in LumiBatch class

#### **Description**

Example data as a LumiBatch object which is a subset of Barnes data (Barnes, 2005)

### Usage

```
data(example.lumi)
```

#### **Format**

A 'LumiBatch' object

#### **Details**

The data is from (Barnes, 2005). It used Sentrix HumanRef-8 Expression BeadChip. Two samples "100US" and "95US:5P" (each has two technique replicates) were selected. In order to save space, 8000 genes were randomly selected. As a result, the example data includes 8000 genes, each has 4 measurements. The full data set was included in the Bioconductor Experiment data package lumiBarnes.

The entire data set has been built as a lumiBarnes data object and can be downloaded from Bioconductor Experiment Data.

#### References

Barnes, M., Freudenberg, J., Thompson, S., Aronow, B. and Pavlidis, P. (2005) Ex-perimental comparison and cross-validation of the Affymetrix and Illumina gene expression analysis platforms, Nucleic Acids Res, 33, 5914-5923.

The detailed data information can be found at: http://www.bioinformatics.ubc.ca/pavlidis/lab/platformCompare/

```
## load the data
data(example.lumi)
## summary of the data
example.lumi
```

example.lumiMethy

Example Illumina Infinium Methylation data in MethyLumiM class

#### **Description**

An example Illumina Infinium Methylation27k dataset, which includes a control and treatment dataset with both technique and biological replicates

### Usage

```
data(example.lumiMethy)
```

#### **Details**

The example dataset includes four control and four treatment samples together with their technique replicates. The original samples and technique replicates were measured in two batches. Here are the names of sixteen samples: Treat1, Treat2, Treat3, Treat4, Ctrl1, Ctrl2, Ctrl3, Ctrl4, Treat1.rep, Treat2.rep, Treat3.rep, Treat4.rep, Ctrl1.rep, Ctrl2.rep, Ctrl3.rep, Ctrl4.rep.

To save storage space, we randomly subset 5000 CpG sites among about 27000 measured CpG sites.

# **Examples**

```
data(example.lumiMethy)
sampleNames(example.lumiMethy)
```

```
example.methyTitration
```

Example Illumina Infinium Methylation titration data in MethyLumiM class

### Description

An example Illumina Infinium Methylation27k dataset, which includes a titration dataset

#### Usage

```
data(example.methyTitration)
```

#### **Details**

The example dataset is a titration dataset. It includes 8 samples: "A1", "A2", "B1", "B2", "C1", "C2", "D" and "E". They are mixtures of Sample A (a B-lymphocyte sample) and Sample B (is (a colon cancer sample from a female donor) at five different titration ratios: 100:0 (A), 90:10 (C), 75:25 (D), 50:50 (E) and 0:100 (B).

To save storage space, we randomly subset 10000 CpG sites among about 27000 measured CpG sites.

gammaFitEM 29

### **Examples**

```
data(example.methyTitration)
sampleNames(example.methyTitration)
```

gammaFitEM	Estimate the methylation status by fitting a Gamma mixture model using EM algorithm

### **Description**

Estimate the methylation status by fitting a two component Gamma mixture model using EM algorithm based on the all M-values of a particular sample

### Usage

```
gammaFitEM(M, initialFit = NULL, fix.k = NULL, weighted = TRUE, maxIteration = 50, tol = 1e-04, plotMode
```

### **Arguments**

М	a vector of M-values covering the whole genome
initialFit	the initial estimation of the gamma parameters returned by .initial $Gamma\ Estimation$ function
fix.k	the k parameter of the gamma function which is fixed during estimation
weighted	determine whether to down-weight the long tails of two component densities beyond their modes
maxIteration	maximum iterations allowed before converging
tol	the difference threshold used to determine convergence
plotMode	determine whether plot the histogram and density plot estimation
truncate	determine whether to truncate the tails beyond the modes during parameter estimation
verbose	determine whether plot intermediate messages during iterations

#### **Details**

The assumption of this function is that the M-value distribution is composed of the mixture of two shifted gamma distributions, which are defined as: dgamma(x-s[1], shape=k[1], scale=theta[1]) and dgamma(s[2]-x, shape=k[2], scale=theta[2]). Here s represents the shift.

NOTE: the methylation status modeling algorithm was developed based on 27K methylation array. It has not been tested for 450K array. Considering 450K array covers both promoter and gene body, the two component Gamma mixture model assumption may not be valid any more.

30 getChipInfo

#### Value

The return is a list with "gammaFit" class attribute, which includes the following items:

logLikelihood the log-likelihood of the fitting model k parameter k of gamma distribution theta parameter theta of gamma distribution shift parameter shift of gamma distribution

proportion the proportion of two components (gamma distributions)

mode the mode positions of the gamma distributions

probability the estimated methylation status posterior probability of each CpG site

#### Author(s)

Pan Du

#### See Also

```
methylationCall and plotGammaFit
```

### **Examples**

```
data(example.lumiMethy)
M <- exprs(example.lumiMethy)
fittedGamma <- gammaFitEM(M[,1], initialFit=NULL, maxIteration=50, tol=0.0001, plotMode=TRUE, verbose=FALSE)</pre>
```

getChipInfo

Get Illumina Chip Information based on probe identifiers

### **Description**

Retrieve the matched Illumina chip information by searching the provided probe identifiers through the Illumina identifiers in all manifest files.

### Usage

```
getChipInfo(x, lib.mapping = NULL, species = c("Human", "Mouse", "Rat", "Unknown"), chipVersion = NULL,
```

### **Arguments**

x a vector of probe identifiers, ExpressionSet object or a matrix with probe iden-

tifiers as row names

the ID mapping library. If it is provided, the parameter "species" will be ignored. species species of the chip designed for. If users do not know it, it can be set as "Un-

known".

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chipVersion chipVersion information returned by function getChipInfo

idMapping determine whether return the idMapping information (between Illumina ID and

nuID)

returnAllMatches

determine whether return all matches or just the best match

verbose determine whether print some warning information

#### **Details**

The function searches the provided probe Identifiers (Illumina IDs or nuIDs) through all the manifest file ID information kept in the IDMapping libraries (lumiHumanIDMapping, lumiMouseI-DMapping, lumiRatIDMapping). The Illumina IDs kept in the library include "Search\\_key" ("Search\\_Key"), "Target" ("ILMN\\_Gene"), "Accession", "Symbol", "ProbeId" ("Probe\\_Id"). To determine the best match, the function calculate the number of matched probes. The higher "matchedProbeNumber" is claimed as better. When the "matchedProbeNumber" is the same, the manifest file with fewer probes is claimed as better. If x is NULL and chipVersion is provided, it will return the entire mapping table of the chip.

#### Value

The function returns a list with following items:

chipVersion the file name of the manifest file for the corresponding version and release

species the species of the chip designed for

IDType the type of probe identifier

chipProbeNumber

the number of probes in the manifest file

matchedProbeNumber

the number of input probes matching the manifest file

idMapping id mapping information between Illumina ID and nuID

When parameter "returnAllMatches" is TRUE, the items of "chipVersion", "IDType", "chipProbe-Number", "inputProbeNumber", "matchedProbeNumber" will be a vector corresponding to the matched manifest files, whose "matchedProbeNumber" is larger than zero, and the "idMapping" will be a matrix with each column corresponding to one matched manifest file. All of the items are sorted from the best match to worst (The higher "matchedProbeNumber" is claimed as better. When the "matchedProbeNumber" is the same, the manifest file with fewer probes is claimed as better.).

#### Author(s)

Pan Du

#### See Also

nuID2IlluminaID, IlluminaID2nuID

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#### **Examples**

```
## load example data
data(example.lumi)
if (require(lumiHumanIDMapping)) {
  chipInfo <- getChipInfo(example.lumi, species='Human')
  chipInfo
}</pre>
```

getChrInfo

get the chromosome location information of methylation probes

### **Description**

get the chromosome location information of methylation probes

# Usage

```
getChrInfo(methyData, lib = NULL, ...)
```

# Arguments

```
methyData a MethyLumiM object
```

lib Methylation annotation library

... optional arguments to addAnnotationInfo.

#### Value

a data.frame

#### Author(s)

Pan Du

getControlData

Get control probe information

# Description

Get control probe information from Bead Studio output or a LumiBatch object.

### Usage

```
getControlData(x, type = c('data.frame', 'LumiBatch'), ...)
```

getControlProbe 33

### **Arguments**

X	the control data can be a LumiBatch object or the Control Probe Profile file outputted by BeadStudio
type	determine the return data type
	other parameters used by lumiR function

#### Value

By default, it returns a data frame with first two columns as "controlType" and "ProbeID". The rest columns are the expression amplitudes for individual samples. When type is 'LumiBatch', it returns a LumiBatch object, which basically is the return of lumiR without combining duplicated TargetIDs. As the return is a LumiBatch object, it includes more information, like probe number, detection p-value and standard error of the measurement.

### Author(s)

Pan Du

#### See Also

addControlData2lumi

# Examples

```
controlFile <- system.file('doc', 'Control_Probe_Profile.txt', package='lumi')
if (file.exists(controlFile)) {
    ## return a data.frame
    controlData <- getControlData(controlFile)
    class(controlData)
    names(controlData)

## return a LumiBatch object
    controlData <- getControlData(controlFile, type='LumiBatch')
    summary(controlData)
}</pre>
```

getControlProbe

Get the control probe Ids

### Description

Get the control probe Ids corresponding to the control probe type provided. The control probe ids are kept in the second column of controlData data.frame.

### Usage

```
getControlProbe(controlData, type = NULL)
```

34 getControlType

# **Arguments**

controlData a LumiBatch object including control data or a control data data.frame

type the type of control probe (case insensitive), which can be get by using getControlType

function

#### Value

returns the corresponding probe Ids for the control type.

#### Author(s)

Pan Du

#### See Also

addControlData2lumi

# **Examples**

```
controlFile <- system.file('doc', 'Control_Probe_Profile.txt', package='lumi')
if (file.exists(controlFile)) {
    ## return a data.frame
    controlData <- getControlData(controlFile)
    getControlType(controlData)
    getControlProbe(controlData, type='housekeeping')
}</pre>
```

getControlType

Get the types of the control probes

# Description

Get the types of the control probes, which is in the first column of the controlData data.frame for LumiBatch objects. For methylation data, it is the return of controlTypes function

#### Usage

```
getControlType(controlData)
```

### **Arguments**

controlData

a LumiBatch object including control data, a control data data.frame, or a Methy-LumiQC object for methylation data

### Value

return the unique type of control probe type.

getNuIDMappingInfo 35

### Author(s)

Pan Du

#### See Also

addControlData2lumi, controlTypes for methylation data

### **Examples**

```
controlFile <- system.file('doc', 'Control_Probe_Profile.txt', package='lumi')
if (file.exists(controlFile)) {
    ## return a data.frame
    controlData <- getControlData(controlFile)
    getControlType(controlData)
}</pre>
```

getNuIDMappingInfo

get the mapping information from nuID to RefSeq ID

### **Description**

Get the mapping information (including mapping quality information) of nuIDs to the most recent RefSeq release. These information was kept in the IDMapping libraries.

### Usage

```
getNuIDMappingInfo(nuID = NULL, lib.mapping)
```

#### **Arguments**

nuID a vector of nuIDs. If it is NULL, all mappings will be returned.

lib.mapping the ID mapping library

#### **Details**

The function basically return the nuID mapping information kept in the "nuID\\_MappingInfo" table of IDMapping libraries (lumiHumanIDMapping, lumiMouseIDMapping, lumiRatIDMapping). For more details of nuID mapping, please refer to the help of corresponding IDMapping library.

### Value

It returns a data.frame with each row corresponding to an input nuID.

# Author(s)

Warren Kibbe, Pan Du, Simon Lin

36 hist-methods

### **Examples**

```
## load example data
data(example.lumi)
if (require(lumiHumanIDMapping)) {
  nuIDs <- featureNames(example.lumi)
  mappingInfo <- getNuIDMappingInfo(nuIDs, lib.mapping='lumiHumanIDMapping')
  head(mappingInfo)
}</pre>
```

hist-methods

Density plot of a ExpressionSet object

# Description

Creating density plot of sample intensities in a ExpressionSet object. It is equivalent to density-methods.

### Usage

```
## S4 method for signature 'ExpressionSet' hist(x, ...)
```

### **Arguments**

```
x a ExpressionSet object... other parameters for density-methods function
```

### See Also

LumiBatch-class, density-methods, hist

```
## load example data
data(example.lumi)
hist(example.lumi)
```

id2seq 37

id2seq

Transfer a nuID as a nucleotide sequence

## **Description**

The nuID (nucleotide universal identifier) is uniquely corresponding to probe sequence. The nuID is also self-identification and error checking

## Usage

```
id2seq(id)
```

## **Arguments**

id

a nuID (nucleotide universal identifier)

### **Details**

A reverse of seq2id. Please refer to reference for more details.

## Value

a string of nucleotide sequence

## Author(s)

Pan Du

## References

Du, P., Kibbe, W.A. and Lin, S.M., "nuID: A universal naming schema of oligonucleotides for Illumina, Affymetrix, and other microarrays", Biology Direct 2007, 2:16 (31May2007).

### See Also

```
seq2id
```

```
seq <- 'ACGTAAATTTCAGTTTAAAACCCCCCG'
id <- seq2id(seq)
id
id2seq(id)</pre>
```

38 IlluminaID2nuID

IlluminaID2nuID	Matching Illumina IDs to nuID based on Illumina ID mapping library

# Description

Matching Illumina IDs to nuID based on Illumina ID mapping libraries.

# Usage

```
IlluminaID2nuID(IlluminaID, lib.mapping=NULL, species = c("Human", "Mouse", "Rat", "Unknown"), chipVer
```

### **Arguments**

IlluminaID	a vector of Illumina IDs
lib.mapping	the ID mapping library. If it is provided, the parameter "species" will be ignored.
species	the species of the chip designed for. If users do not know it, it can be set as "Unknown".
chipVersion	chipVersion information returned by function getChipInfo
	other parameters of getChipInfo

# **Details**

When the parameter "chipVersion" is not provided, this function basically returned the "idMapping" item returned by function getChipInfo.

## Value

The mapping information from Illumina ID to nuID. It will be a matrix with each column corresponding to one matched manifest file when parameter "returnAllMatches" is TRUE. In this case, the columns are sorted from the best match to worst. If IlluminaID is NULL and chipVersion is provided, it will return all mapping information of the chip.

### Author(s)

Pan Du

### See Also

getChipInfo, nuID2IlluminaID

importMethyIDAT 39

importMethyIDAT	Import Illumina methylation .idat files as an MethyLumiM object
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## Description

Import Illumina methylation .idat files as an MethyLumiM object. An extension of lumIDAT function

### Usage

```
importMethyIDAT(sampleInfo, dataPath = getwd(), lib = NULL, bigMatrix=FALSE, dir.bigMatrix='.', savePr
```

### **Arguments**

sampleInfo A data.frame of sample information or a character vector of barcodes.

dataPath The path of .idat files
lib Annotation library

bigMatrix whether to save the data as BigMatrix (designed for very large dataset)

dir.bigMatrix the parent directory to save the BigMatrix data files

savePrefix.bigMatrix

the folder name prefix of the directory to save the BigMatrix data files. The fold

name will be like this: paste(savePrefix.bigMatrix, '\_bigmat', sep=")

... other parameters used by lumIDAT function

#### **Details**

This function is an extension of lumIDAT. It adds sample information and probe annotation information to the data. As Illumina organizes the output .idat files by barcodes, the function will automatically check the sub-folders in the names of barcodes for .idat files. The "sampleInfo" parameter can be either a barcode vector, e.g., "7310440039\_R04C02" "7310440039\_R05C02". Or a data.frame with required columns of 'Sentrix\_Barcode' and 'Sentrix\_Position'. If "sampleInfo" is a data.frame, it will be added as the pData of the output MethyLumiM object.

#### Value

A MethyLumiM object

#### Author(s)

Pan Du, Tim Triche

#### See Also

lumIDAT, lumiMethyR, addAnnotationInfo

40 inverse VST

inverseVST

Inverse VST transform

## Description

Inverse transform of VST (variance stabilizing transform), see vst.

### Usage

```
inverseVST(x, fun = c('asinh', 'log'), parameter)
```

### **Arguments**

x a VST transformed LumiBatch object or a numeric matrix or vector

fun function used in VST transform
parameter parameter of VST function

#### **Details**

Recover the raw data from VST transformed data returned by vst. This function can be directly applied to the VST transformed or VST + RSN normalized LumiBatch object to reverse transform the data to the original scale.

### Value

Return the raw data before VST transform

### Author(s)

Pan Du

#### References

Lin, S.M., Du, P., Kibbe, W.A., "Model-based Variance-stabilizing Transformation for Illumina Mi-croarray Data", submitted

## See Also

vst

```
## load example data
data(example.lumi)

## get the gene expression mean for one chip
u <- exprs(example.lumi)[,1]
## get the gene standard deviation for one chip</pre>
```

is.nuID 41

```
std <- se.exprs(example.lumi)[,1]
## do variance stabilizing transform
transformedU <- vst(u, std)
## do inverse transform and recover the raw data
parameter <- attr(transformedU, 'parameter')
transformFun <- attr(transformedU, 'transformFun')
recoveredU <- inverseVST(transformedU, fun=transformFun, parameter=parameter)
## compare with the raw data
print(u[1:5])
print(recoveredU[1:5])
## do inverse transform of the VST + RSN processed data
lumi.N <- lumiExpresso(example.lumi[,1:2])
## Inverse transform.
## Note: as the normalization is involved, the processed data will be different from the raw data.
lumi.N.raw <- inverseVST(lumi.N)</pre>
```

is.nuID

nuID self-identification

### **Description**

Self-identify nuID (nucleotide universal identifier) by verify the check code value and the checksum value

## Usage

is.nuID(id)

### **Arguments**

id

nuId or other string

#### Value

Return TRUE if id is a nuID, or else return FALSE.

### Author(s)

Pan Du

### References

Du, P., Kibbe, W.A. and Lin, S.M., "nuID: A universal naming schema of oligonucleotides for Illumina, Affymetrix, and other microarrays", Biology Direct 2007, 2:16 (31May2007).

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### See Also

```
seq2id, id2seq
```

### **Examples**

```
## check the function using a random sequence
id <- 'adfasdfafd'
is.nuID(id) # FALSE

## check the function using a read nuID
seq <- 'ACGTAAATTTCAGTTTAAAACCCCCCG'
id <- seq2id(seq)
is.nuID(id) # TRUE</pre>
```

lumiB

Background correction of Illumina Expression data

# Description

Background correction of Illumina Expression data

### Usage

```
lumiB(x.lumi, method = c('none', 'bgAdjust', 'forcePositive', 'bgAdjust.affy'), verbose = TRUE, ...)
```

# Arguments

x.lumi	an ExpressionSet inherited object or a data matrix with columns as samples and rows as genes. For 'bgAdjust' method, it should be a LumiBatch Object
method	the background correction method, it can be any function with a ExpressionSet Object or matrix as the first argument and return an processed object with the same class
verbose	a boolean to decide whether to print out some messages
	other parameters used by the user provided background correction method

### **Details**

We assume the BeadStudio output data is background corrected. So by default, it will do nothing. The 'bgAdjust' method will estimate the background based on the control probe information, which is kept in the controlData slot of LumiBatch object. The 'forcePositive' method will force all expression values to be positive by adding an offset (minus minimum value plus one), it does nothing if all expression values are positive. The purpose of this is to avoid NA when do logarithm transformation. 'none' does not but return the LumiBatch object. 'bgAdjust.affy' will call the bg.adjust function in affy package. User can also provide their own function with a LumiBatch Object as the first argument and return a LumiBatch Object with background corrected.

Thanks Kevin Coombes (M.D. Anderson Cancer Center) suggested adding this function.

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## Value

Return an object with background corrected. The class of the return object is the same as the input object x.lumi.

#### Author(s)

Pan Du, Kevin Coombes

### See Also

```
bgAdjust, lumiExpresso
```

# Examples

```
## load example data
data(example.lumi)

## Do the default background correction method
lumi.B <- lumiB(example.lumi, method='bgAdjust', probs=0)</pre>
```

LumiBatch-class

Class LumiBatch: contain and describe Illumina microarray data

# Description

This is a class representation for Illumina microarray data. It extends ExpressionSet.

### **Extends**

Directly extends class ExpressionSet.

# **Creating Objects**

```
new("LumiBatch", exprs = [matrix],se.exprs = [matrix],beadNum = [matrix],detection =
[matrix], phenoData = [AnnotatedDataFrame], history = [data.frame], ...)
```

LumiBatch instances are usually created through new("LumiBatch", ...). The arguments to new should include exprs and se.exprs, others can be missing, in which case they are assigned default values.

Objects can be created using the function lumiR.

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#### **Slots**

Slot specific to LumiBatch:

history: a data.frame recording the operation history of the LumiBatch object.

controlData: a data.frame with first two columns as "controlType" and "ProbeID". The rest columns are the control probe expression amplitudes for individual samples.

QC: a the quality control information of the LumiBatch object, returned by lumiQ function.

Slots inherited from ExpressionSet:

assayData contains equal dimensional matrices: exprs (contains gene expression level, which is the mean of its bead replicates.), se.exprs (contains gene expression standard error, which is the standard error of its bead replicates.), beadNum (records the number of beads for the probe.), detection (records the detection p-value of the probe. The number is from [0,1]. By default, < 0.01 indicates good detection.). For more details of assayData, please see ExpressionSet

phenoData: See eSet
experimentData: See eSet
annotation: See eSet

#### Methods

### **Class-specific methods:**

se.exprs(LumiBatch), se.exprs(LumiBatch, matrix)<-: Access and set elements named se.exprs in the AssayData-class slot.

beadNum(LumiBatch), beadNum(LumiBatch)<-: Access and set elements named beadNum in the AssayData-class slot. Use "beadNum(LumiBatch) <- NULL" to remove the beadNum element

detection(LumiBatch), detection(LumiBatch)<-: Access and set elements named detection in the AssayData-class slot. Use "detection(LumiBatch) <- NULL" to remove the detection element.

getHistory(LumiBatch): Access the operation history of LumiBatch object.

**Derived from** ExpressionSet (For the directly inherited methods, please see ExpressionSet and eSet):

combine(LumiBatch, missing): Combine two LumiBatch objects, including history slot. See eSet

exprs(LumiBatch), exprs(LumiBatch, matrix)<-: Access and set elements named exprs in the AssayData-class slot.

object[(i,j): Conduct subsetting of the data in a LumiBatch object

**Standard generic methods** (For the directly inherited methods, please see ExpressionSet and eSet):

initialize(LumiBatch): Object instantiation, used by new; not to be called directly by the user.
validObject(LumiBatch): Validity-checking method, ensuring that exprs and se.exprs is a
 member of assayData. Other validity check is the same as checkValidity(ExpressionSet).
show(LumiBatch) A summary of the LumiBatch object.

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### Author(s)

Pan Du, Simon Lin

#### See Also

```
lumiR, lumiT, lumiN, boxplot-methods, pairs-methods, MAplot-methods
```

### **Examples**

```
## load example data
data(example.lumi)

## show the summary of the data
# summary(example.lumi)
example.lumi

## get expression matrix
temp <- exprs(example.lumi)

## get a subset
temp <- example.lumi[,1] ## retrieve the first sample

## get the probe id
featureNames(example.lumi)[1:3]

## combine LumiBatch objects
temp <- combine(example.lumi[,1], example.lumi[,3])
temp</pre>
```

lumiExpresso

From raw Illumina probe intensities to expression values

## **Description**

Goes from raw Illumina probe intensities to expression values

# Usage

```
lumiExpresso(lumiBatch, bg.correct = TRUE, bgcorrect.param = list(method='bgAdjust'), variance.stabil:
varianceStabilize.param = list(), normalize = TRUE, normalize.param = list(), QC.evaluation = TRUE,
QC.param = list(), verbose = TRUE)
```

### **Arguments**

```
lumiBatch a LumiBatch object, which can be the return of lumiR
bg.correct a boolean to decide whether to do background correction or not
```

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```
bgcorrect.param
```

a list of parameters of lumiB

variance.stabilize

a boolean to decide whether to do variance stabilization or not

varianceStabilize.param

a list of parameters of lumiT

normalize a boolean to decide whether to do normalization or not

normalize.param

a list of parameters of lumiN

QC. evaluation a boolean to decide whether to do quality control estimation before and after

preprocessing

QC.param a list of parameters of lumiQ

verbose a boolean to decide whether to print out some messages

#### **Details**

The function is to encapsulate the major functions of Illumina preprocessing. It is organized in a similar way as the expresso function in affy package.

#### Value

return a processed LumiBatch object. The operation history can be track in the history slot of the object.

## Author(s)

Pan Du

### See Also

```
lumiB, lumiT, lumiN
```

```
## load example data
data(example.lumi)

## Do all the default preprocessing in one step
lumi.N <- lumiExpresso(example.lumi)

## Do customized preprocessing. No variance stabilizing or log transform, use Quantile normalization.
lumi.N <- lumiExpresso(example.lumi, variance.stabilize=FALSE, normalize.param = list(method='quantile'))</pre>
```

IumiMethyB 47

lumiMethyB	Adjust background level of Illumina Infinium methylation data

# Description

Adjust background level of Illumina Infinium methylation data, which is an object in MethyLumiM class.

## Usage

```
lumiMethyB(methyLumiM, method = c("bgAdjust2C", "forcePositive", "none"), separateColor = FALSE, verbo
```

### **Arguments**

methyLumiM a MethyLumiM object includes Illumina Infinium methylation data

method background adjustment methods or user provided function, whose input and

output should be a intensity matrix (pool of methylated and unmethylated probe

intensities)

separateColor determine whether to separately process two color channels

verbose a boolean to decide whether to print out some messages

overwriteBigMatrix

whether to overwrite the result to the BigMatrix data, only valid when the input

data is BigMatrix-based

... other parameters used by corresponding method

### Value

Return an object (same class as input methyLumiM) with updated "methylated" and "unmethylated" data matrix after background level adjustment.

### Author(s)

Pan DU

### See Also

See Also bgAdjustMethylation and estimateMethylationBG

```
data(example.lumiMethy)
lumiMethy.bgAdj = lumiB(example.lumiMethy)
attr(lumiMethy.bgAdj, "EstimatedBG")
```

48 lumiMethyC

lumiMethyC	Color bias adjust of Illumina Infinium methylation data

## **Description**

Color bias adjust of Illumina Infinium methylation data, which is an object in MethyLumiM class.

### Usage

```
lumiMethyC(methyLumiM, method = c("quantile", "ssn", "none"), verbose = TRUE, overwriteBigMatrix=FALSE
```

## **Arguments**

methyLumiM a MethyLumiM object includes Illumina Infinium methylation data

method color bias adjustment methods or user provided function, see "details" for more

information of user defined function.

verbose a boolean to decide whether to print out some messages

overwriteBigMatrix

whether to overwrite the result to the BigMatrix data, only valid when the input

data is BigMatrix-based

... other parameters used by corresponding method

## Details

The first two arguments of the user defined function should be two intensity matrix (pool of methylated and unmethylated probe intensities) of red and green channel respectively. The return of the user defined function should be a list including color adjusted matrix of red and green channel. For example: return(list(red=redData, green=grnData)). "redData" and "grnData" are two color adjusted matrix.

#### Value

Return an object (same class as input methyLumiM) with updated "methylated" and "unmethylated" data matrix after background level adjustment.

### Author(s)

Pan DU

#### See Also

See Also adjColorBias.quantile and adjColorBias.ssn

lumiMethyN 49

### **Examples**

```
data(example.lumiMethy)
# before adjustment
plotColorBias1D(example.lumiMethy)
# plot in 2D plot of one selected sample
plotColorBias2D(example.lumiMethy, selSample = 1)
lumiMethy.adj = lumiMethyC(example.lumiMethy)
# after adjustment
plotColorBias1D(lumiMethy.adj)
# plot in 2D plot of one selected sample
plotColorBias2D(lumiMethy.adj, selSample = 1)
```

lumiMethyN

Normalize the Illumina Infinium methylation data

### **Description**

Normalize the Illumina Infinium methylation data, which is an object in MethyLumiM class.

### Usage

```
lumiMethyN(methyLumiM, method = c("quantile", "ssn", "none"), separateColor = FALSE, verbose = TRUE, ov
```

# Arguments

methyLumiM a MethyLumiM object includes Illumina Infinium methylation data

method supported normalization methods or user provided function, whose input and

output should be a intensity matrix (pool of methylated and unmethylated probe

intensities)

separateColor determine whether to separately process two color channels

verbose a boolean to decide whether to print out some messages

overwriteBigMatrix

whether to overwrite the result to the BigMatrix data, only valid when the input

data is BigMatrix-based

... other parameters used by corresponding method

#### Value

Return an object (same class as input methyLumiM) with updated "methylated" and "unmethylated" data matrix after background level adjustment.

### Author(s)

Pan DU

50 lumiMethyR

### See Also

See Also normalizeMethylation.ssn and normalizeMethylation.quantile

### **Examples**

```
data(example.lumiMethy)
lumiMethy.norm = lumiN(example.lumiMethy)
```

lumiMethyR

Reading Illumina methylation microarray data

# Description

This function is a wrap of methylumiR function in methylumi package.

### Usage

```
lumiMethyR(filename, lib=NULL, controlData=NULL, qcfile=NULL, sampleDescriptions=NULL, sep = NULL)
```

### **Arguments**

filename file name output by GenomeStudio

1ib Annotation library of Illumina methylation microarray

controlData the controlData file name or a MethyLumiQC object to be added to the "con-

trolData" slot of the MethyLumiM object

qcfile parameter of methylumiR function

sampleDescriptions

parameter of methylumiR function

sep parameter of methylumiR function

### **Details**

This function is a wrap of methylumiR function in methylumi package. It will coerce the returned object as MethyLumiM class. The methylated and unmethylated probe intensity information is required for color-bias adjustment and normalization. If users have the Illumina IDAT files, we suggest use importMethyIDAT function to import the data. The importMethyIDAT function will automatically retrieve the required information and return a MethyLumiM object.

### Value

return a MethyLumiM object

### Author(s)

Pan Du

lumiMethyStatus 51

### See Also

See Also importMethyIDAT, methylumiR and addControlData2methyLumiM

lumiMethyStatus

Estimate the methylation status of individual methylation sites

### **Description**

Estimate the methylation status of individual methylation sites by fitting a two component Gamma mixture model for each sample

### Usage

```
lumiMethyStatus(methyLumiM, ...)
```

## **Arguments**

```
methyLumiM a MethyLumiM class object
... Other parameters used by methylationCall
```

### **Details**

This function calls methylationCall and returns the methylation status of individual methylation sites. The methylation status includes: "Unmethy" (unmethylation probability > unmethylation threshold), "Methy" (methylation probability > methylation threshold), or "Margin". The methylation probability is returned as an attribute of "probability".

### Value

return a methylation status matrix with "probability" attribute

### Author(s)

Pan Du

### See Also

See Also methylationCall and gammaFitEM

```
data(example.lumiMethy)
methyCall <- lumiMethyStatus(example.lumiMethy)
head(methyCall)</pre>
```

52 lumiN

lumiN	Between chip normalization of a LumiBatch object	

### **Description**

A main function of between chip normalization of a LumiBatch object. Currently, four methods ("rsn", "ssn", "quantile", "loess", "vsn") are supported.

### Usage

```
lumiN(x.lumi, method = c("quantile", "rsn", "ssn", "loess", "vsn", "rankinvariant"), verbose = TRUE, ...
```

# Arguments

x.lumi	an ExpressionSet inherited object or a data matrix with columns as samples and rows as genes
method	five different between chips normalization methods ("quantile", "rsn", "ssn", "loess", "vsn", "rankinvariant") are supported
verbose	a boolean to decide whether to print out some messages
	other parameters used by corresponding method

### **Details**

lumiN is an interface for different normalization methods. Currently it supports "RSN" (See rsn), "SSN" (See ssn), "loess" (See normalize.loess), "quantile" (See normalize.quantiles), "VSN" (See vsn) and "rankinvariant" (See rankinvariant). See details in individual functions. Note: the "VSN" normalization should be directly applied to the raw data instead of the lumiT processed data.

### Value

Return an object with expression values normalized. The class of the return object is the same as the input object x.lumi. If it is a LumiBatch object, it also includes the VST transform function and its parameters as attributes: "transformFun", "parameter". See inverseVST for details.

### Author(s)

Pan Du, Simon Lin

#### See Also

rsn, ssn, rankinvariant

lumiQ 53

### **Examples**

```
## load example data
data(example.lumi)

## Do lumi transform
lumi.T <- lumiT(example.lumi)

## Do lumi between chip normaliazation
lumi.N <- lumiN(lumi.T, method='rsn', ifPlot=TRUE)</pre>
```

lumiQ

Quality control evaluation of the LumiBatch object

### **Description**

Quality control evaluation of the LumiBatch object and returns a summary of the data

### Usage

```
lumiQ(x.lumi, logMode = TRUE, detectionTh = 0.01, verbose = TRUE)
```

### **Arguments**

x.lumi a LumiBatch object

logMode transform as log2 or not (the function can check whether it is already log trans-

formed.)

detectionTh the detection threshold used by detectionCall

verbose a boolean to decide whether to print out some messages

### **Details**

Quality control of a LumiBatch object includes estimating the mean and standard deviation of the chips, detectable probe ratio of each chip, sample (chip) relations, detecting outliers of samples (chips). The produced QC information is kept in the QC slot of LumiBatch class. The summary function will provide a summary of the QC information (See example).

#### Value

a LumiBatch object with QC slot keeping the QC information

#### Author(s)

Pan Du

### See Also

```
LumiBatch, plot, ExpressionSet-method
```

54 lumiR

### **Examples**

```
## load example data
data(example.lumi)

## Do quality control estimation
lumi.Q <- lumiQ(example.lumi)

## A summary of the QC
summary(lumi.Q, 'QC')

## Plot the results
## plot the pairwise sample correlation
plot(lumi.Q, what='pair')

## see more examples in "plot,ExpressionSet-method" help documents</pre>
```

lumiR

Read in Illumina expression data

# Description

Read in Illumina expression data. We assume the data was saved in a comma or tab separated text file.

### Usage

lumiR(fileName, sep = NULL, detectionTh = 0.01, na.rm = TRUE, convertNuID = TRUE, lib.mapping = NULL, det QC = TRUE, columnNameGrepPattern = list(exprs='AVG\_SIGNAL', se.exprs='BEAD\_STD', detection='DETECTION inputAnnotation=TRUE, annotationColumn=c('ACCESSION', 'SYMBOL', 'PROBE\_SEQUENCE', 'PROBE\_START', 'CHF

## **Arguments**

fileName of the data file

sep the separation character used in the text file.

detectionTh the p-value threshold of determining detectability of the expression. See more

details in lumiQ

na.rm determine whether to remove NA

convertNuID determine whether convert the probe identifier as nuID

lib.mapping a Illumina ID mapping package, e.g, lumiHumanIDMapping, used by addNuID2lumi

dec the character used in the file for decimal points.

parseColumnName

determine whether to parse the column names and retrieve the sample informa-

tion (Assume the sample information is separated by "\\_".)

checkDupId determine whether to check duplicated TargetIDs or ProbeIds. The duplicated

ones will be averaged.

lumiR 55

QC determine whether to do quality control assessment after read in the data. columnNameGrepPattern

the string grep patterns used to determine the slot corresponding columns.

inputAnnotation

determine whether input the annotation information outputted by BeadStudio if exists.

annotationColumn

the column names of the annotation information outputted by BeadStudio

verbose a boolean to decide whether to print out some messages

... other parameters used by read. table function

#### **Details**

The function can automatically determine the separation character if it is Tab or comma. Otherwise, the user should specify the separator manually. If the annotation library is provided, the Illumina Id will be replaced with nuID, which is used as the index Id for the lumi annotation packages. If the annotation library is not provided, it will try to directly convert the probe sequence (if provided in the BeadStudio output file) as nuIDs.

The parameter "columnNameGrepPattern" is designed for some advanced users. It defines the string grep patterns used to determine the slot corresponding columns. For example, for the "exprs" slot in LumiBatch object, it is composed of the columns whose name includes "AVG\\_SIGNAL". In some cases, the user may not want to read the "detection" and "beadNum" related columns to save memory. The user can set the "detection" and "beadNum" as NA in "columnNameGrepPattern". If the 'se.exprs' is set as NA or the corresponding columns are not available, then lumiR will create a ExpressionSet object instead of LumiBatch object.

The parameter "parseColumnName" is designed to parse the column names and retrieve the sample information. We assume the sample information is separated by "\\_" and the last element after "\\_" is the sample label (sample names of the LumiBatch object). If the parsed sample labels are not unique, then the entire string will be used as the sample label. For example: "1881436055\\_A\\_STA 27aR" is included in one of the column names of BeadStudio output file. Here, the program will first treat "STA 27aR" as the sample label. If it is not unique across the samples, "1881436055\\_A\\_STA 27aR" will be the sample label. If it is still not unique, the program will report warning messages. All the parsed information is kept in the phenoData slot. By default, "parseColumnName" is FALSE. We suggest the users use it only when they know what they are doing.

Current version of lumiR can adaptively read the output of BeadStudio Verson 1 and 3. The format Version 3 made quite a few changes comparing with previous versions. One change is the detection value. It was called detectable when the detection value is close to one for Version 1 format. However, the detection value became a p-value in the Version 3. As a result, the detectionTh is automatically changed based on the version. The detectionTh 0.01 for the Version 3 will be changed as the detectionTh 0.99 for Version 1. Another big change is that Version 3 separately output the control probe (gene) information and a "Samples Table". As a result, the controlData slot in LumiBatch class was added to keep the control probe (gene) information, and a QC slot to keep the quality control information, including the "Sample Table" output by BeadStudio version 3.

The recent version of BeadStudio can also output the annotation information together with the expression data. In the users also want to input the annotation information, they can set the parameter "inputAnnotation" as TRUE. At the same time, they can also specify which columns to be inputted

56 lumiR.batch

by setting parameter "annotationColumn". The BeadStudio annotation columns include: SPECIES, TRANSCRIPT, ILMN\\_GENE, UNIGENE\\_ID, GI, ACCESSION, SYMBOL, PROBE\\_ID, AR-RAY\\_ADDRESS\\_ID, PROBE\\_TYPE, PROBE\\_START, PROBE\\_SEQUENCE, CHROMOSOME, PROBE\\_CHR\\_ORIENTATION, PROBE\\_COORDINATES, DEFINITION, ONTOLOGY\\_COMPONENT, ONTOLOGY\\_PROCESS, ONTOLOGY\\_FUNCTION, SYNONYMS, OBSOLETE\\_PROBE\\_ID. As the annotation data is huge, by default, we only input: ACCESSION, SYMBOL, PROBE\\_START, CHROMOSOME, PROBE\\_CHR\\_ORIENTATION, PROBE\\_COORDINATES, DEFINITION. This annotation information is kept in the featureData slot of ExpressionSet, which can be retrieved using pData(featureData(x.lumi)), suppose x.lumi is the LumiBatch object. As some annotation information may be outdated. We recommend using Bioconductor annotation packages to retrieve the annotation information.

The BeadStudio may output either STDEV or STDERR (standard error of the mean) columns. As the variance stabilization (see vst function) requires the information of the standard deviation instead of the standard error of the mean, the value correction is required. The lumiR function will automatically check whether the BeadStudio output file includes STDEV or STDERR columns. If it is STDERR columns, it will correct STDERR as STDEV. The corrected value will be x \* sqrt(N), where x is the STDERR value (standard error of the mean), N is the number of beads corresponding to the probe. (Thanks Sebastian Balbach and Gordon Smyth kindly provided this information.). This correction was previous implemented in the lumiT function.

#### Value

return a LumiBatch object

### Author(s)

Simon Lin, Pan Du

### See Also

LumiBatch, addNuID2lumi

### **Examples**

```
## specify the file name
# fileName <- 'Barnes_gene_profile.txt' # Not Run
## load the data
# x.lumi <- lumiR(fileName)
## load the data with empty detection and beadNum slots
# x.lumi <- lumiR(fileName, columnNameGrepPattern=list(detection=NA, beadNum=NA))</pre>
```

lumiR.batch

Read BeadStudio output files in batch

# Description

Read BeadStudio output files in batch and combine them as a single LumiBatch object

lumiR.batch 57

#### Usage

lumiR.batch(fileList, convertNuID = TRUE, lib.mapping = NULL, detectionTh = 0.01, QC = TRUE, transform =

#### **Arguments**

a vector of file names or a directory keeping the data files in the format of .csv fileList determine whether convert the probe identifier as nuID convertNuID same as lumiR parameter lib.mapping (optional) lib.mapping the p-value threshold of determining detectability of the expression. See more detectionTh details in lumiQ QC determine whether to do quality control assessment after read in the data. transform determine whether to do transform after input each file sampleInfoFile a Tab-separated text file or a data frame keeping the sample information (opverbose a boolean to decide whether to print out some messages

... other parameters used by lumiR

#### **Details**

The function basically call lumiR for individual files and then combine the returns. The sampleInfoFile parameter is optional. It provides the sample information (for phenoData slot in LumiBatch object), it is a Tab-separated text file. ID column is required. It represents sample ID, which is defined based on the column names of BeadStudio output file. For example, sample ID of column "1881436070\\_A\\_STA.AVG\\_Signal" is "1881436070\\_A\\_STA". The sample ID column can also be found in the "Samples Table.txt" file output by BeadStudio. Another "Label" column (if provided) will be used as the sampleNames of LumiBatch object. All information of sampleInfoFile will be directly added in the phenoData slot in LumiBatch object.

To save memory space in the case of reading large data set, we can do transformation using lumiT function right after input the data, and the information like se.exprs, beadNum will be removed from the LumiBatch object after transformation.

#### Value

A LumiBatch object which combines the individual LumiBatch object corresponding to each file

### Author(s)

Pan Du

## See Also

lumiR

```
## fileList <- c('file1.csv', 'file2.cvs')
## x.lumi <- lumiR.batch(fileList, sampleInfoFile='sampleInfo.txt')</pre>
```

58 lumiT

lumiT Transfer the Illumina data to stabilize the variance	
--	--

# Description

Transfer the Illumina data to stabilize the variance.

### Usage

```
lumiT(x.lumi, method = c("vst", 'log2', 'cubicRoot'), ifPlot = FALSE, simpleOutput = TRUE, verbose = TRU
```

## **Arguments**

x.lumi	LumiBatch object
method	four methods are supported: "vst", "log2", "cubicRoot"
ifPlot	determine whether to plot the intermediate results
simpleOutput	determine whether to simplify the output LumiBatch object, which will set the se.exprs, detection and beadNum slots as NULL.
verbose	a boolean to decide whether to print out some messages
	other parameters used by vst

### **Details**

lumiT is an interface of difference variance stabilizing transformation. See vst for details of VST (Variance Stabilizing Transform) of Illumina data.

NOTE: This correction of STDERR as STDEV was moved to the lumiR function.

## Value

Return a LumiBatch object with transformed expression values. It also includes the VST transform function and its parameters as attributes: "transformFun", "parameter". See inverseVST for details.

## Author(s)

Pan Du, Simon Lin

### References

Lin, S.M., Du, P., Kibbe, W.A., (2008) 'Model-based Variance-stabilizing Transformation for Illumina Microarray Data', Nucleic Acids Res. 36, e11

### See Also

vst

m2beta 59

### **Examples**

```
## load example data
data(example.lumi)

## Do default VST variance stabilizing transform
lumi.T <- lumiT(example.lumi, ifPlot=TRUE)</pre>
```

m2beta

Convert methylation M-value to Beta-value

## **Description**

Convert methylation M-value to Beta-value

# Usage

m2beta(m)

## **Arguments**

m

a matrix or vector of methylation M-value

### **Details**

Convert methylation M-value to Beta-value

## Value

return methylation Beta-value with the same size of input M-value

# Author(s)

Pan Du

### References

Du, P., Zhang, X, Huang, C.C., Jafari, N., Kibbe, W.A., Hou, L., and Lin, S.M., (2010) 'Comparison of Beta-value and M-value methods for quantifying methylation levels by microarray analysis', (under review)

# See Also

See Also as beta2m

60 MAplot-methods

MAplot-methods	MAplot of a ExpressionSet object	

# Description

Creating pairwise MAplot of sample intensities in a ExpressionSet object

# Usage

```
## S4 method for signature 'ExpressionSet'
MAplot(object, ..., smoothScatter = FALSE, logMode = TRUE, subset = 5000, main = NULL)
```

### **Arguments**

object an ExpressionSet object
... optional arguments to MAplot.

smoothScatter whether use smoothScatter function to plot points

logMode whether plot the data in log2 scale or not

subset subset of rows used to plot. It can be an index vector, or the length of a random

subset

main title of the plot

### **Details**

To increase the plot efficiency, by default, we only plot RANDOMLY selected subset of points (based on parameter "subset"). If users want to plot all the points, they can set the parameter "subset = NULL". When smoothScatter is set as TRUE, the subsetting will be suppressed because smoothScatter function has good plot efficiency for large number of points.

### See Also

```
LumiBatch-class, MAplot
```

```
## load example data
data(example.lumi)

MAplot(example.lumi, smoothScatter=TRUE)
```

methylationCall 61

methvl	ationCall

Estimated methylation call

### **Description**

Estimated methylation call based on the fitting results of gammaFitEM

## Usage

```
methylationCall(x, threshold = 0.95, ...)
```

# **Arguments**

x a vector of M-values covering the whole genome or a "gammaF	it" class object
---	------------------

returned by gammaFitEM

threshold the probability threshold to make a methylation call. The threshold can be a

vector of two: unmethylation threshold and methylation threshold

... other parameters used by gammaFitEM

### **Details**

Retrieve the probability element returned by gammaFitEM, and convert it as three status calls based on probability threshold

NOTE: the methylation status modeling algorithm was developed based on 27K methylation array. It has not been tested for 450K array. Considering 450K array covers both promoter and gene body, the two component Gamma mixture model assumption may not be valid any more.

### Value

A vector of three methylation status: "Unmethy" (unmethylation posterior probability > unmethylation threshold), "Methy" (methylation posterior probability > methylation threshold), or "Margin". The sum of unmethylation posterior probability and methylation posterior probability equals one. The methylation probability is returned as an attribute of "probability".

### Author(s)

Pan DU

#### See Also

gammaFitEM

62 monoSmu

### **Examples**

```
data(example.lumiMethy)
M <- exprs(example.lumiMethy)
fittedGamma <- gammaFitEM(M[,1], initialFit=NULL, maxIteration=50, tol=0.0001, plotMode=TRUE, verbose=FALSE)
methyCall <- methylationCall(fittedGamma)
table(methyCall)</pre>
```

monoSmu

Monotonic smooth method

# **Description**

Fit the monotonic-constraint spline curve

## Usage

```
monoSmu(x, y, newX = NULL, nSupport = min(200, length(x)), nKnots = 6, rotate = FALSE, ifPlot = FALSE, xl
```

## **Arguments**

X	a vector represents x values
у	a vector represents y values
newX	the new values to be transformed. If not provided, "x" will be used.
nSupport	downsampled data points
nKnots	parameter used by monoSpline
rotate	determine whether to rotate the axis with 45 degrees in clockwise, i.e., fit the curve in the MA-plot.
ifPlot	determine whether to plot intermediate results
1 1	4 11 64 14

xlab the xlab of the plot ylab the ylab of the plot

... parameters used by supsmu and plot

### **Details**

function called by lumiN.rsn. The function first fits a monotonic spline between vector x and y, then transforms the vector newX based on the fitted spline. (After transformation the fitted spline is supposed to be a diagonal line, i.e., x=y)

#### Value

Return the transformed "newX" based on the smoothed curve

### Author(s)

Simon Lin, Pan Du

monoSpline 63

### References

Lin, S.M., Du, P., Kibbe, W.A., (2008) 'Model-based Variance-stabilizing Transformation for Illumina Microarray Data', Nucleic Acids Res. 36, e11

# See Also

monoSpline

monoSpline

Fitting a curve with monotonic spline

## **Description**

Fitting a curve with monotonic spline

## Usage

```
monoSpline(x, y, newX=NULL, nKnots = 6, ifPlot = FALSE)
```

## **Arguments**

x a vector represents x valuesy a vector represents y values

newX the new values to be transformed. If not provided, "x" will be used.

nKnots parameter used by function smoothCon in package mgcv

ifPlot determine whether to plot intermediate results

#### **Details**

Function internally called by monoSmu

#### Value

return the transformed "newX" based on the smoothed curve

# Author(s)

Simon Lin, Pan Du

## See Also

monoSmu

normalizeMethylation.quantile

Quantile normalization of Illumina Infinium methylation data at probe level

## **Description**

Quantile normalization of Illumina Infinium methylation data at probe level. Input data is a Methy-LumiM object

## Usage

```
normalizeMethylation.quantile(methyLumiM, separateColor = FALSE, reference = NULL, ...)
```

## **Arguments**

methyLumiM a MethyLumiM object includes Illumina Infinium methylation data

separateColor determine whether separately normalize two color channels

reference the reference vector (same length as the number of matrix rows) for quanitle

normalization

... other parameters used by normalize.quantiles.robust method

# Value

Return an object (same class as input methyLumiM) with updated "methylated" and "unmethylated" data matrix after background level adjustment.

### Author(s)

Pan DU

#### See Also

See Also lumiMethyN, and normalizeMethylation.ssn

```
data(example.lumiMethy)
lumiMethy.norm = normalizeMethylation.quantile(example.lumiMethy)
```

```
normalizeMethylation.ssn
```

Shift and scaling normalization of Illumina Infinium methylation data at probe level

## **Description**

Shift and scaling normalization of Illumina Infinium methylation data at probe level

### Usage

```
normalizeMethylation.ssn(methyLumiM, separateColor = FALSE)
```

# Arguments

methyLumiM a MethyLumiM object includes Illumina Infinium methylation data

separateColor determine whether separately normalize two color channels

#### Value

Return an object (same class as input methyLumiM) with updated "methylated" and "unmethylated" data matrix after background level adjustment.

### Author(s)

Pan DU

## See Also

 $See \ Also \ lumiMethyN, and \ normalize Methylation. quantile$ 

```
data(example.lumiMethy)
lumiMethy.norm = normalizeMethylation.ssn(example.lumiMethy)
```

66 nuID2EntrezID

nuID2EntrezID Map nuID to Entrez ID
-------------------------------------

### Description

Map nuID to EntrezID through RefSeq ID based on IDMapping libraries.

## Usage

```
nuID2EntrezID(nuID = NULL, lib.mapping, filterTh = c(Strength1 = 95, Uniqueness = 95), returnAllInfo = F
```

### **Arguments**

nuID a vector of nuIDs. If it is NULL, all mappings will be returned.

lib.mapping the ID mapping library

filterTh the mapping quality filtering threshold used to filter the ID mapping.

returnAllInfo determine to return the detailed mapping information or just the matched RefSeq

IDs

#### **Details**

This function is based on the return of <code>getNuIDMappingInfo</code> function. The mapping from nuID to EntrezID was based on the mapping from nuID to RefSeqID and RefSeqID to EntrezID. It uses mapping quality information to filter out the bad mappings from nuID to RefSeqID. The parameter "filterTh" is obsolete for lumi ID mapping package > version 1.3, which only keeps the perfect mapping. For the old version of ID mapping package (< 1.3), the names of "filterTh" are basically the field names of "nuID\\_MappingInfo" table, which include 'Strength1', 'Strength2', 'Uniqueness' and 'Total hits'. For the definition of these metrics, please refer to the IDMapping library or see the reference website.

### Value

returns the matched Entrez IDs or a data.frame with each row corresponding to an input nuID (when "returnAllInfo" is TRUE).

### Author(s)

Warren Kibbe, Pan Du, Simon Lin

## References

https://prod.bioinformatics.northwestern.edu/nuID/

#### See Also

See Also getNuIDMappingInfo

nuID2IlluminaID 67

### **Examples**

```
## load example data
data(example.lumi)
if (require(lumiHumanIDMapping)) {
  nuIDs <- featureNames(example.lumi)
  mappingInfo <- nuID2EntrezID(nuIDs, lib.mapping='lumiHumanIDMapping')
  head(mappingInfo)
}</pre>
```

nuID2IlluminaID

Matching nuIDs to Illumina IDs based on Illumina ID mapping library

#### **Description**

Matching nuIDs to Illumina IDs based on Illumina ID mapping library

#### **Usage**

```
nuID2IlluminaID(nuID, lib.mapping=NULL, species = c("Human", "Mouse", "Rat", "Unknown"), idType=c('All
```

### **Arguments**

```
nuID a vector of nuIDs

lib.mapping the ID mapping library. If it is provided, the parameter "species" will be ignored.

species the species of the chip designed for. If users do not know it, it can be set as "Unknown".

idType the Illumina ID type

chipVersion chipVersion information returned by function getChipInfo

other parameters of getChipInfo
```

#### **Details**

The parameter "idType" represents different types of Illumina IDs. It returns the entire table when idType = "All". When idType = 'Probe', it returns "ProbeId" or "Probe\\_Id". When idType = 'Gene', it returns "Target" or "ILMN\\_Gene" IDs.

This function basically returned the "idMapping" item returned by function getChipInfo. If nuID is NULL and chipVersion is provided, it will return all mapping information of the chip.

### Value

The mapping information from nuID to Illumina ID. It will be a matrix with each column corresponding to one matched manifest file when parameter "returnAllMatches" is TRUE. In this case, the columns are sorted from the best match to worst.

## Author(s)

Pan Du

nuID2probeID

### See Also

```
getChipInfo, IlluminaID2nuID
```

### **Examples**

```
## load example data
data(example.lumi)
nuIDs <- featureNames(example.lumi)
if (require(lumiHumanIDMapping)) {
  illuminaID <- nuID2IlluminaID(nuIDs[1:5], lib='lumiHumanIDMapping')
  illuminaID
}</pre>
```

nuID2probeID

Mapping nuID into Illumina ProbeID

# Description

Mapping nuID into Illumina ProbeID.

## Usage

```
nuID2probeID(nuID, lib.mapping = "lumiHumanIDMapping", ...)
```

### **Arguments**

```
nuID a vector of nuID

lib.mapping an Illumina ID mapping library

other parameters of nuID2IlluminaID
```

#### **Details**

The function will call nuID2IlluminaID when ID mapping library were provided.

#### Value

```
see function nuID2IlluminaID
```

# Author(s)

Pan Du

### References

Du, P., Kibbe, W.A. and Lin, S.M., "nuID: A universal naming schema of oligonucleotides for Illumina, Affymetrix, and other microarrays", Biology Direct 2007, 2:16 (31May2007).

nuID2RefSeqID 69

### See Also

```
probeID2nuID, nuID2IlluminaID
```

### **Examples**

```
if (require(lumiHumanIDMapping)) {
    nuID2probeID("B2J6WGhV.RevOJYff4", lib.mapping = "lumiHumanIDMapping")
}
```

nuID2RefSeqID

Map nuID to RefSeq ID

### **Description**

Map nuID to RefSeq ID based on IDMapping libraries.

### Usage

```
nuID2RefSeqID(nuID = NULL, lib.mapping, filterTh = c(Strength1 = 95, Uniqueness = 95), returnAllInfo = F
```

### **Arguments**

nuID a vector of nuIDs. If it is NULL, all mappings will be returned.

lib.mapping the ID mapping library

filterTh the mapping quality filtering threshold used to filter the ID mapping. Obsolete

for lumi ID mapping package > version 1.3!

returnAllInfo determine to return the detailed mapping information or just the matched RefSeq

**IDs** 

#### Details

This function is based on the return of <code>getNuIDMappingInfo</code> function. It uses mapping quality information to filter out the bad mappings. The parameter "filterTh" is obsolete for lumi ID mapping package > version 1.3, which only keeps the perfect mapping. For the old version of ID mapping package (< 1.3), the names of "filterTh" are basically the field names of "nuID\\_MappingInfo" table, which include 'Strength1', 'Strength2', 'Uniqueness' and 'Total hits'. For the definition of these metrics, please refer to the IDMapping library or see the reference website.

# Value

returns the matched RefSeq IDs or a data.frame with each row corresponding to an input nuID (when "returnAlIInfo" is TRUE).

## Author(s)

Warren Kibbe, Pan Du, Simon Lin

70 nuID2targetID

### References

https://prod.bioinformatics.northwestern.edu/nuID/

#### See Also

See Also getNuIDMappingInfo

## **Examples**

```
## load example data
data(example.lumi)
if (require(lumiHumanIDMapping)) {
  nuIDs <- featureNames(example.lumi)
  mappingInfo <- nuID2RefSeqID(nuIDs, lib.mapping='lumiHumanIDMapping')
  head(mappingInfo)
}</pre>
```

nuID2targetID

Mapping nuID into Illumina TargetID

## Description

Mapping nuID into Illumina TargetID or GeneID.

# Usage

```
nuID2targetID(nuID, lib.mapping = "lumiHumanIDMapping", ...)
```

### **Arguments**

```
nuID a vector of nuID

lib.mapping an Illumina ID mapping library

... other parameters of nuID2IlluminaID
```

## **Details**

The function will call nuID2IlluminaID when ID mapping library were provided.

### Value

```
see function nuID2IlluminaID
```

### Author(s)

Pan Du

pairs-methods 71

### References

Du, P., Kibbe, W.A. and Lin, S.M., "nuID: A universal naming schema of oligonucleotides for Illumina, Affymetrix, and other microarrays", Biology Direct 2007, 2:16 (31May2007).

### See Also

```
targetID2nuID, nuID2IlluminaID
```

### **Examples**

```
if (require(lumiHumanIDMapping)) {
    nuID2targetID("B2J6WGhV.RevOJYff4", lib.mapping = "lumiHumanIDMapping")
}
```

pairs-methods

Pair plot of an ExpressionSet object

## **Description**

Creating pairs plot of sample intensities in an ExpressionSet object

### Usage

```
## S4 method for signature 'ExpressionSet'
pairs(x, ..., smoothScatter = FALSE, logMode = TRUE, subset = 5000, fold=2, dotColor=1,
highlight = NULL, highlightColor = 2, main = NULL, checkTransform = TRUE)
```

### **Arguments**

x a ExpressionSet object
... optional arguments to pairs.

smoothScatter whether use smoothScatter function to plot points

logMode whether plot the data in log2 scale

subset subset of rows used to plot. It can be an index vector, or the length of a random

subset

fold The fold-change threshold used to estimate the number of probes having high

fold-changes

dotColor color of points in the scatter plot
highlight the subset dots need to be highlighted
highlightColor the color for those highlighted dots

main title of the plot

checkTransform whether to check the data is log2-transformed or not

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### **Details**

To increase the plot efficiency, by default, we only plot RANDOMLY selected subset of points (based on parameter "subset"). If users want to plot all the points, they can set the parameter "subset = NULL". When smoothScatter is set as TRUE, the subsetting will be suppressed because smoothScatter function has good plot efficiency for large number of points.

### See Also

```
LumiBatch-class, pairs
```

### **Examples**

```
## load example data
data(example.lumi)
pairs(example.lumi)
pairs(example.lumi, smoothScatter=TRUE)
```

plot-methods

Plot of a ExpressionSet object

### **Description**

Creating quality control plots of a ExpressionSet object

## Usage

```
## S4 method for signature 'ExpressionSet,missing'
plot(x, what = c("density", "boxplot", "pair", "MAplot", "sampleRelation", "outlier", "cv"), main, ...)
```

### **Arguments**

```
x a ExpressionSet object returned by lumiQ
what one of the six kinds of QC plots
main the title of the QC plot
... additional parameters for the corresponding QC plots
```

### **Details**

The parameter "what" of plot function controls the type of QC plots, which includes:

- density: the density plot of the chips, see hist-methods
- boxplot: box plot of the chip intensities, see boxplot-methods
- pair: the correlation among chips, plot as a hierarchical tree, see pairs-methods
- MAplot: the MAplot between chips, see MAplot-methods

plotCDF 73

- sampleRelation: plot the sample relations. See plotSampleRelation
- outlier: detect the outliers based on the sample distance to the center. See detectOutlier
- cv: the density plot of the coefficients of variance of the chips. See estimateLumiCV

#### See Also

LumiBatch-class, hist-methods, boxplot-methods, MAplot-methods, pairs-methods, plotSampleRelation, estimateLumiCV, detectOutlier

# **Examples**

```
## load example data
data(example.lumi)
## Quality control estimation
lumi.Q <- lumiQ(example.lumi)</pre>
## summary
summary(lumi.Q)
## plot the density
plot(lumi.Q, what='density')
## plot the pairwise sample correlation
plot(lumi.Q, what='pair')
## plot the pairwise MAplot
plot(lumi.Q, what='MAplot')
## sample relations
plot(lumi.Q, what='sampleRelation', method='mds', color=c('100US', '95US:5P', '100US', '95US:5P'))
## detect outlier based on the distance to the mean profile
plot(lumi.Q, what='outlier')
## Density plot of coefficient of variance
plot(lumi.Q, what='cv')
```

plotCDF

plot the cumulative distribution function of a ExpressionSet object

# Description

plot the cumulative distribution function of a ExpressionSet object from high to low value or in reverse

74 plotColorBias1D

#### Usage

```
plotCDF(x, reverse=TRUE, logMode=TRUE, xlab = NULL, ylab = "Cumulative density", col=1:dim(x)[2], lwd=1, xlim = NULL, index.highlight = NULL, color.highlight = 2, addLegend = TRUE, main="", ...)
```

#### **Arguments**

x a numeric or ExpressionSet object
reverse determine whether plot the CDF from high

reverse determine whether plot the CDF from high to low value or not logMode determine whether the CDF plot is based on a log2 scale

xlabxlab of the plotCDF plotylabylab of the plotCDF plotcolline colors of the plotCDF plotlwdline width of plot functionxlimparameter of the plot function

index.highlight

the column index of the highlighted plotCDF curve

color.highlight

color of highlighted plotCDF curve

addLegend whether add legend to the plot or not

main title for the plot

... additional parameters for plot.ecdf function

# See Also

```
LumiBatch-class, ecdf
```

# Examples

```
## load example data
data(example.lumi)
plotCDF(example.lumi)
```

plotColorBias1D

Plot the color bias density plot of Illumina Infinium Methylation data

# Description

Plot the color bias density plot of Illumina Infinium Methylation data in one dimension (comparing with 2D scatter plot)

## Usage

```
plotColorBias1D(methyLumiM, channel = c("both", "unmethy", "methy", "sum"), colorMode=TRUE, removeGence
```

plotColorBias1D 75

# **Arguments**

methyLumiM MethyLumiM-class object or eSet-class object, which include methylated and

unmethylated probe intensities

channel estimate the intensity in different methods

colorMode whether separate two color channels or not

removeGenderProbes

determine whether exclude probes on X and Y chromosomes if the chromosome

information is provided in the methyLumiM object.

logMode Whether plot the intensities in log-scale

subset plot subset of randomly selected rows. All data will be plotted if it is NULL.

... other parameters used by density and plot

#### **Details**

Plot the color bias density plot of Illumina Infinium Methylation data. There are four options using "channel" parameter to plot the density plot. "both": estimate the density by pooling together methylated and unmethylated probe intensities. "unmethy" and "methy": plot either unmethylated or methylated probe density. "sum" plot the density of the sum of methylated and unmethylated probe intensitys.

#### Value

Invisibly return TRUE if plot successfully.

# Author(s)

Pan DU

## See Also

See Also as plotColorBias2D and boxplotColorBias

```
data(example.lumiMethy)
# before adjustment
plotColorBias1D(example.lumiMethy)
```

76 plotColorBias2D

plotColorBias2D Plot the color bias of Illumina Infinium Methylation data in two mensions	o di-
---	-------

# **Description**

Plot the color bias (red and green channel) of Illumina Infinium Methylation data of one selected sample in two dimensions (methylated and unmethylated probe intensities)

## Usage

```
plotColorBias2D (methyLumiM, selSample = 1, combineMode = F, layoutRatioWidth = c(0.75, 0.25), layoutRatio
```

#### **Arguments**

methyLumiM MethyLumiM-class object or eSet-class object, which include methylated and

unmethylated probe intensities

selSample The index of sample name of the selected sample to plot color bias combineMode Whether combine two color channels together and plot as one color

layoutRatioWidth

the plot figure ratio between scatter plot and density plot

layoutRatioHeight

the plot figure ratio between scatter plot and density plot

margins margin of the plot

cex A numerical value giving the amount by which plotting text and symbols should

be magnified relative to the default. See par

logMode Whether plot the intensities in log-scale

subset plot subset of randomly selected rows. All data will be plotted if it is NULL.

... other parameters used by plot

# **Details**

The function basically plots the probe intensities in 2-dimension (methylated vs unmethylated), and colors the dots in Red and Green based on their color channel information. The related density plot will also be plotted at the right and top of the scatter plot.

#### Value

Invisibly return TRUE if plot successfully.

## Author(s)

Pan DU

plotControlData 77

#### See Also

See Also as plotColorBias1D

#### **Examples**

```
data(example.lumiMethy)
# plot in 2D plot of one selected sample
plotColorBias2D(example.lumiMethy, selSample = 1)
```

plotControlData Plot the mean expression (with standard deviation bar) of different

type of control probes

# Description

Plot the mean intensity (with standard deviation bar) of different type of control probes. Multiple control types can be plotted in a single plot. The available control types can be get by running getControlType(controlData).

#### Usage

```
plotControlData(controlData, type = NULL, slideIndex = NULL, logMode = FALSE, new = TRUE, ...)
```

## **Arguments**

controlData a LumiBatch object including control data, a control data data.frame, a Methy-

LumiQC object or a MethyLumiM object including MethyLumiQC control data

type the control probe type (case insensitive), which can be get by running getCon-

trolType(controlData)

slideIndex the slide index or ID corresponding to each sample

logMode whether show the data in log2 scale

new whether refresh the new plot or add it on the old one

... other parameters used by default plot function

#### **Details**

When multiple control types are selected, they will be plotted in a two-column plot. For methylation data, the red and green channels will be plotted respectively in red and green colors.

#### Value

plot the picture and invisibly return TRUE if everything is OK

# Author(s)

Pan Du

78 plotDensity

#### See Also

addControlData2lumi and addControlData2methyLumiM

#### **Examples**

```
controlFile <- system.file('doc', 'Control_Probe_Profile.txt', package='lumi')
if (file.exists(controlFile)) {
   controlData <- getControlData(controlFile)
   getControlType(controlData)
   plotControlData(controlData, type='NEGATIVE')
}</pre>
```

plotDensity

plot the density distribution

# Description

plot the density distribution of a dataMatrix or ExpressionSet object

# Usage

```
plotDensity(dataMatrix, logMode=TRUE, addLegend=TRUE, legendPos="topright", subset = NULL, ...)
```

# **Arguments**

dataMatrix a data matrix or ExpressionSet object
logMode determine whether the CDF plot is based on a log2 scale
addLegend whether add legend to the plot or not
legendPos the position of the legend. If its length is two, then it specifies the x and y location of legend.

subset plot subset of randomly selected rows. All data will be plotted if it is NULL.
additional parameters for matplot function

## See Also

```
LumiBatch-class, density
```

```
## load example data
data(example.lumi)
plotDensity(example.lumi)
```

plotGammaFit 79

plotGammaFit	plot the fitting results of gammaFitEM

# **Description**

plot the fitting results of gammaFitEM. It basically plot the histogram of M-values plus the estimated gamma density plots and their mixture.

# Usage

```
plotGammaFit(x, gammaFit = NULL, k = NULL, theta = NULL, shift = NULL, proportion = NULL, plotType = c("h
```

# Arguments

X	a vector of M-values covering the whole genome
gammaFit	a "gammaFit" class object returned by gammaFitEM
k	parameter k of gamma distribution
theta	parameter theta of gamma distribution
shift	parameter shift of gamma distribution
proportion	the proportion of two components (gamma distributions)
plotType	determine the way to show the distribution of the input data, either histogram or density plot
	Other parameters used by hist or plot (for "density" plotType) function.

# **Details**

This function is to visualize the fitting results, which helps us understand how well the fitting is.

#### Value

Invisibly return TRUE if the plot is successful.

# Author(s)

Pan DU

# See Also

```
gammaFitEM
```

```
data(example.lumiMethy)
M <- exprs(example.lumiMethy)
fittedGamma <- gammaFitEM(M[,1], initialFit=NULL, maxIteration=50, tol=0.0001, plotMode=FALSE, verbose=FALSE)
plotGammaFit(M[,1], gammaFit=fittedGamma)</pre>
```

plotHousekeepingGene Plot the housekeeping gene expression profile

# Description

Plot the housekeeping gene expression profile

# Usage

```
plotHousekeepingGene(controlData, lib = NULL, slideIndex = NULL, addLegend = TRUE, logMode = TRUE, ...)
```

# **Arguments**

```
controlData a LumiBatch object including control data or a control data data.frame
```

the annotation library (for retrieving the gene name) slideIndex the slide index or ID corresponding to each sample

addLegend whether add legend or not

logMode whether show the data in log2 scale

... other parameters used by default matplot function

# Value

plot the picture and return TRUE if everything is OK

# Author(s)

Pan Du

#### See Also

```
addControlData2lumi, plotControlData
```

```
controlFile <- system.file('doc', 'Control_Probe_Profile.txt', package='lumi')
if (file.exists(controlFile)) {
  controlData <- getControlData(controlFile)
  plotHousekeepingGene(controlData)
}</pre>
```

plotSampleRelation 81

plotSampleRelation	visualize the sample relations
	- 1

# Description

plot the sample relations based on MDS or hierarchical clustering

# Usage

```
plotSampleRelation(x, subset = NULL, cv.Th = 0.1, standardize = TRUE, method = c("cluster", "mds"), dimensional content of the standardize in the standardize is the standardize in the standardize is the standardize in the standardize is th
```

# Arguments

8	
х	a LumiBatch object, ExpressionSet object or a matrix with each column corresponding to a sample
subset	the subset probes used to determine the sample relations. If it is one number, then randomly selected "number" of probes will be used. If not provide, all the probes will be used.
cv.Th	the threshold of the coefficient of variance of probes used to select probes to estimate sample relations
standardize	standardize the expression profiles or not
method	"MDS" or "hierarchical clustering"
dimension	the principle components to visualize the MDS plot
color	the color for each sample during plot. Only support the "mds" method
main	the title of the plot
pch	use symbols instead of text to label the samples
addLegend	Whether to add legend to MDS (two-dimensional PCA) plot
	Other parameters used by plot function.

# **Details**

Estimate the sample relations based on selected probes (based on large coefficient of variance (mean / standard variance)). Two methods can be used: MDS (Multi-Dimensional Scaling) or hierarchical clustering methods.

# Value

Invisibly return the hierarchical clustering results (if 'cluster' method used) or coordinates of the mds plot (if 'mds' method used) .

# Author(s)

Pan Du

82 plotStringencyGene

#### See Also

```
lumiQ, LumiBatch, , plot, ExpressionSet-method
```

#### **Examples**

```
## load example data
data(example.lumi)

## plot the sample relations with MDS

## the color of sample is automatically set based on the sample type
plotSampleRelation(example.lumi, col=c('100US', '95US:5P', '100US', '95US:5P'))

## plot the sample relations with hierarchical clustering
plotSampleRelation(example.lumi, method='cluster')
```

plotStringencyGene

plot the Stringency related control probe profiles

## Description

Plot the Stringency related control probe (Low-Stringency, Medium-Stringency and High-Stringency) profiles. Using getControlType function to view available stringency types.

# Usage

```
plotStringencyGene(controlData, lib = NULL, slideIndex = NULL, addLegend = TRUE, logMode = TRUE, ...)
```

# **Arguments**

controlData a LumiBatch object including control data or a control data data.frame

lib the annotation library (for retrieving the gene name) slideIndex the slide index or ID corresponding to each sample

addLegend whether add legend or not

logMode whether show the data in log2 scale

... other parameters used by default matplot function

#### Value

plot the picture and return TRUE if everything is OK

## Author(s)

Pan Du

plotVST 83

#### See Also

```
addControlData2lumi, plotControlData
```

# **Examples**

```
controlFile <- system.file('doc', 'Control_Probe_Profile.txt', package='lumi')
if (file.exists(controlFile)) {
  controlData <- getControlData(controlFile)
  plotStringencyGene(controlData)
}</pre>
```

plotVST

plot the VST (Variance Stabilizing Transform) function

# Description

plot the VST (Variance Stabilizing Transform) function of VST transformed LumiBatch object or parameters of VST function.

# Usage

```
plotVST(x, transFun = NULL, plotRange = NULL, addLegend = TRUE, ...)
```

# **Arguments**

x a LumiBatch object after lumiT transform, or a matrix or data.frame with VST parameter

transFun a character vector of transformation function (asinh or log2)

plotRange the plot range of untransformed data

addLegend add legend or not

other parameter used by plot function

## Value

invisibly return the untransformed and transformed values.

# Author(s)

Pan Du

#### See Also

vst

probeID2nuID

#### **Examples**

```
## load example data
data(example.lumi)

## Do default VST variance stabilizing transform
lumi.T <- lumiT(example.lumi, ifPlot=TRUE)

## plot the transform function
plotVST(lumi.T)</pre>
```

probeID2nuID

Mapping Illumina ProbeID as nuID

# Description

Mapping Illumina ProbeID as nuID.

# Usage

```
probeID2nuID(probeID, lib.mapping = "lumiHumanIDMapping", ...)
```

# **Arguments**

```
probeID a vector of Illumina ProbeID lib.mapping an Illumina ID mapping library
```

... other parameters of IlluminaID2nuID

# **Details**

The function will call IlluminaID2nuID when ID mapping library were provided.

## Value

```
see function IlluminaID2nuID
```

# Author(s)

Pan Du

#### References

Du, P., Kibbe, W.A. and Lin, S.M., "nuID: A universal naming schema of oligonucleotides for Illumina, Affymetrix, and other microarrays", Biology Direct 2007, 2:16 (31May2007).

## See Also

```
nuID2probeID, IlluminaID2nuID
```

#### **Examples**

```
if (require(lumiHumanIDMapping)) {
   probeID2nuID('0001240020', lib='lumiHumanIDMapping')
}
```

produceGEOPlatformFile

Produce GEO Platform Submission File in SOFT format

# Description

Produce GEO Sample Submission File in SOFT format based on the provided LumiBatch object and Illumina ID Mapping library

#### Usage

```
produceGEOPlatformFile(x.lumi, lib.mapping = NULL, nuIDMode = TRUE, includeAllChipProbe=FALSE, fileNam
```

#### **Arguments**

x. lumi The LumiBatch object keeping all probes

1 ib.mapping The Illumina ID Mapping library, e.g., "lumiHumanIDMapping" nuIDMode Determine whether producing the platform indexed by nuID

includeAllChipProbe

Determine whether including all probes in the Manifest file or just the probes

used in the x.lumi object

fileName Filename of the GEO Platform File name

#### **Details**

The function produces the GEO platform submission file based on the chip information kept in the Illumina ID Mapping library (specified by lib.mapping parameter). The determination of chip type will be automatically done by selecting the best matching of the probe IDs with individual chips.

# Value

Save the result as a text file in SOFT platform submission format.

#### Author(s)

Pan Du

## References

http://www.ncbi.nlm.nih.gov/projects/geo/info/soft2.html

#### See Also

produceGEOSubmissionFile

## **Examples**

```
# data(example.lumi)
# produceGEOPlatformFile(example.lumi, lib.mapping='lumiHumanIDMapping')
```

produceGEOSampleInfoTemplate

Produce the template of GEO sample information

# **Description**

Produce the template of GEO sample information, which is used for function produceGEOSubmissionFile.

#### Usage

```
produceGEOSampleInfoTemplate(lumiNormalized, lib.mapping = NULL, fileName = "GEOsampleInfo.txt")
```

# **Arguments**

lumiNormalized The normalized data (LumiBatch object)

1ib.mapping The Illumina ID Mapping library, e.g., "lumiHumanIDMapping"

fileName The file name of Tab separated sample information file

#### Details

This function just produces a template of sample information with some default fillings. Users need to fill in the detailed sample descriptions, especially the Sample\\_title, Sample\\_description and some protocols. No blank fields are allowed. Function produceGEOSubmissionFile will produce the file GEO submission file based on this sample information. The users should not use "\#" in the description as it is a reserved character.

#### Value

Save the result as a Tab separated text file or return a data.frame if the fileName is NULL.

# Author(s)

Pan Du

#### References

http://www.ncbi.nlm.nih.gov/projects/geo/info/soft2.html

#### See Also

 ${\tt produce} {\tt GEOSubmissionFile}$ 

produceGEOSubmissionFile

Produce GEO Sample Submission File in SOFT format

#### **Description**

Produce GEO Sample Submission File in the SOFT format based on the provided LumiBatch object and sample information

## Usage

produceGEOSubmissionFile(lumiNormalized, lumiRaw, lib.mapping = NULL, idType = 'Probe', sampleInfo = NULL)

# **Arguments**

lumiNormalized The normalized data (LumiBatch object)

lumiRaw The raw data (LumiBatch object), e.g., returned by lumiR

1ib.mapping The Illumina ID Mapping library, e.g., "lumiHumanIDMapping"

idType the idType parameter of function nuID2IlluminaID

sampleInfo The sample information filename or data.frame, which is returned by produceGEOSampleInfoTemplate

fileName The file name of GEO Submission file

supplementaryRdata

determine whether produce the Rdata supplement data, which include both lu-

miNormalized and lumiRaw R objects.

... other parameters used by function nuID2IlluminaID

## Details

The function produces the GEO sample submission file including both normalized and raw data information in the SOFT format. The sample information should be provided by the user as a data.frame or Tab separated text file following the format of the template, which can be produced by function produceGEOSampleInfoTemplate. Users need to fill in the detailed sample descriptions in the template, especially the Sample\\_title, Sample\\_description and some protocols. Users are also required to fill in the "Sample\\_platform\\_id" by checking information of the GEO Illumina platform.

When the parameter "supplementaryRdata" is TRUE, the R objects, lumiNormalized, lumiRaw and sampleInfo, will be saved in a file named 'supplementaryData.Rdata'.

#### Value

Save the result as a text file in SOFT sample submission format. The supplementary Rdata will be saved in a file 'supplementaryData.Rdata'.

# Author(s)

Pan Du

#### References

http://www.ncbi.nlm.nih.gov/projects/geo/info/soft2.html

#### See Also

produceGEOSampleInfoTemplate, produceGEOPlatformFile

# **Examples**

```
## Not run
## Produce the sample information template
# produceGEOSampleInfoTemplate(lumiNormalized, lib.mapping = NULL, fileName = "GEOsampleInfo.txt")
## After editing the 'GEOsampleInfo.txt' by filling in sample information
# produceGEOSubmissionFile(lumiNormalized, lumiRaw, lib='lumiHumanIDMapping', sampleInfo='GEOsampleInfo.txt')
```

 $\verb|produceMethylationGEOSubmissionFile| \\$ 

Produce GEO Sample Submission File of Illumina methylation microarray data in SOFT format

#### **Description**

Produce GEO Sample Submission File in the SOFT format based on the provided MethyLumiM object and sample information

The normalized data in MethyLumiM class

#### Usage

```
produceMethylationGEOSubmissionFile(methyLumiM, methyLumiM.raw = NULL, lib.mapping = NULL, idType = "P
```

# Arguments

methyLumiM

methyLumiM.raw The raw data in MethyLumiM class

lib.mapping Currently not used for Illumina methylation data

idType Currently no other options for Illumina methylation data

sampleInfo The sample information filename or data.frame, which is returned by produceGEOSampleInfoTemplate

fileName The file name of GEO Submission file

supplementaryRdata

determine whether produce the Rdata supplement data, which include both methyLumiM and methyLumiM.raw R objects.

other parameters used by function nuID2IlluminaID, but not implemented for

methylation data

rankinvariant 89

#### **Details**

The function produces the GEO sample submission file including both normalized and raw data information in the SOFT format. The sample information should be provided by the user as a data.frame or Tab separated text file following the format of the template, which can be produced by function produceGEOSampleInfoTemplate. Users need to fill in the detailed sample descriptions in the template, especially the Sample\\_title, Sample\\_description and some protocols. Users are also required to fill in the "Sample\\_platform\\_id" by checking information of the GEO Illumina platform.

When the parameter "supplementaryRdata" is TRUE, the R objects, methyLumiM, methyLumiM.raw and sampleInfo, will be saved in a file named 'supplementaryData.Rdata'.

#### Value

Save the result as a text file in SOFT sample submission format. The supplementary Rdata will be saved in a file 'supplementaryData.Rdata'.

## Author(s)

Pan Du

#### References

http://www.ncbi.nlm.nih.gov/projects/geo/info/soft2.html

# See Also

produceGEOSampleInfoTemplate, produceGEOPlatformFile

# **Examples**

```
## Not run
## Produce the sample information template
# produceGEOSampleInfoTemplate(methyLumiM, fileName = "GEOsampleInfo.txt")
## After editing the 'GEOsampleInfo.txt' by filling in sample information
# produceMethylationGEOSubmissionFile(methyLumiM, methyLumiM.raw, sampleInfo='GEOsampleInfo.txt')
```

rankinvariant

Rank Invariant Normalization

#### **Description**

This function basically adjusts the samples to the same background level and then optionally scales to the same foreground level.

## Usage

```
rankinvariant(x.lumi, targetArray = NULL, rrc = .05, lowRank = seq(.5, .25, -.05), highRank = .9, minSiz
```

90 rsn

#### **Arguments**

x.lumi an ExpressionSet inherited object or a data matrix with columns as samples and rows as genes

targetArray A target chip is the model for other chips to normalize. It can be a column index,

a vector or a LumiBatch object with one sample.

rrc The relative rank change allowed for a gene to be selected as rank invariant

lowRank A vector with, in decreasing order, the minimum ranks where candidate genes

can be selected as rank invariant

highRank The maximum rank where candidate genes can be selected as rank invariant

minSize Fraction of genes required to be selected as rank invariant

Maximum number of iterations for rlm to reach convergence

#### **Details**

Rank invariant normalization uses a set of genes that are rank invariant between a given sample and a target sample. The target sample can be predefined by setting the targetArray argument. If targetArray is NULL the average expression of all samples will be the target. Rank invariant genes are found for each sample seperately by calculation the relative rank change for each gene. Furthermore, only genes with ranks between the lowRank and highRank are considered. If the number of probes is less than minSize multiplies by the number of genes the next lowRank value tried. If no rank invariant set can be found an error is thrown.

The default settings of this function are the same as used Genomstudio (Illumina). The results produced by this method are similar, but not identical to Genomestudio.

## Value

Return an object with expression values normalized. The class of the return object is the same as the input object x.lumi.

#### Author(s)

Arno Velds (contact: a.velds (at) nki.nl)

#### See Also

lumiN

rsn Robust Spline Normalization between chips

# Description

Robust spline normalization (monotonic curves) between chips

rsn 91

#### Usage

```
rsn(x.lumi, targetArray = NULL, excludeFold = 2, span = 0.03, ifPlot = FALSE, ...)
```

#### **Arguments**

x.lumi an ExpressionSet inherited object or a data matrix with columns as samples and

rows as genes

targetArray A target chip is the model for other chips to normalize. It can be a column index,

a vector or a LumiBatch object with one sample.

excludeFold exclude the genes with fold change larger than "excludeFold" during fitting the

curve in normalization

span the span parameter used by monoSmu

ifPlot determine whether to plot intermediate results

... other parameters used by monoSmu

#### **Details**

The robust spline normalization (RSN) algorithm combines the features of quantile and loess normalization. It is designed to normalize the variance-stabilized data. The function will check whether the data is variance stabilized (vst or log2 transform), if not, it will automatically run lumiT before run rsn. For details of the algorithm, please see the reference.

The targetArray can be a column index, a vector or a LumiBatch object with one sample, which corresponds to an external sample to be normalized with. This is very useful for handling large data set or normalizing the data set with a common reference (targetArray).

#### Value

Return an object with expression values normalized. The class of the return object is the same as the input object x.lumi. If it is a LumiBatch object, it also includes the VST transform function and its parameters as attributes: "transformFun", "parameter". See inverseVST for details.

#### Author(s)

Pan Du, Simon Lin

#### See Also

lumiN, monoSmu

92 seq2id

seq2id

Transfer a nucleotide sequence as a nuID

# **Description**

The nuID (nucleotide universal identifier) is uniquely corresponding to probe sequence. The nuID is also self-identification and error checking

# Usage

```
seq2id(seq)
```

# **Arguments**

seq

a nucleotide sequence composed of A, C, G, T (U).

#### **Details**

The nuID is a exact mapping of nucleotide sequence based on Base64 encoding scheme. A character set A-Z, a-z, 0-9, "\\_" and "." is used to represent to the base-64 numbers of 0-63. The first character of nuID is a checking code, which provide information of both the number of padded "A"s at the nucleotide sequence and error checking. Please refer to reference for more details.

#### Value

A string represents nuID

#### Author(s)

Pan Du

#### References

Du, P., Kibbe, W.A. and Lin, S.M., "nuID: A universal naming schema of oligonucleotides for Illumina, Affymetrix, and other microarrays", Biology Direct 2007, 2:16 (31May2007).

# See Also

id2seq

```
seq <- 'ACGTAAATTTCAGTTTAAAACCCCCCG'
id <- seq2id(seq)
id
id2seq(id)</pre>
```

smooth Quantile Normalization

Smooth quantile normalization

# **Description**

Smooth quantile normalization with a reference sample

# Usage

smoothQuantileNormalization(dataMatrix, ref = NULL, adjData=NULL, logMode = TRUE, bandwidth = NULL, deg

# Arguments

dataMatrix a matrix of microarray intensity data

ref a vector of reference sample intensity, which matches the dataMatrix

adjData data to be adjusted based on the ref and dataMatrix distribution

logMode whether perform the analysis in log2 scale

bandwidth a parameter used by locpoly degree a parameter used by locpoly

verbose whether print the processing sample names

... other parameters used by locpoly

# Value

a data matrix with intensity normalized.

# Author(s)

Pan DU

#### See Also

See Also adjColorBias.quantile

94 ssn

ssn	Simple Scaling Normalization
3311	Simple Seating Normalization

## **Description**

This function basically adjusts the samples to the same background level and then optionally scales to the same foreground level.

## Usage

```
ssn(x.lumi, targetArray = NULL, scaling = TRUE, bgMethod=c('density', 'mean', 'median', 'none'), fgMeth
```

## Arguments

x.lumi	an ExpressionSet inherited object or a data matrix with columns as samples and rows as genes
targetArray	A target chip is the model for other chips to normalize. It can be a column index, a vector or a LumiBatch object with one sample.
scaling	determine whether do scaling or just background shift
bgMethod	optional methods of determining the background level
fgMethod	optional methods of determining the foreground level
	other parameters used by density function

#### **Details**

This function basically adjusts the samples to the same background level and then optionally scales to the same foreground level. The adjustment is based on the raw scale data (For the transformed data, it still estimates the parameters in the raw scale by inverse transformation.).

Comparing with other normalization methods, like quantile and curve-fitting methods, SSN is a more conservative method. The only assumption is that each sample has the same background levels and the same scale (if do scaling). There are three methods ('density', 'mean' and 'median') for background estimation. If bgMethod is 'none', then the background level will be set as 0, i.e., no background adjustment. For the 'density' bgMethod, it estimates the background based on the mode of probe intensities based on the assumption that the background level intensity is the most frequent value across all the probes in the chip. For the foreground level estimation, it also provides three methods ('mean', 'density', 'median'). For the 'density' fgMethod, it assumes the background probe levels are symmetrically distributed. Then we estimate the foreground levels by taking the intensity mean of all other probes except from the background probes. For the 'mean' and 'median' methods (for both bgMethod and fgMethod), it basically estimates the level based on the mean or median of all probes of the sample. If the fgMethod is the same as bgMethod (except 'density' method), no scaling will be performed.

# Value

Return an object with expression values normalized. The class of the return object is the same as the input object x.lumi.

targetID2nuID 95

#### Author(s)

Pan Du, Simon Lin

#### See Also

**lumiN** 

targetID2nuID

Mapping Illumina TargetID (GeneID) into nuID

# **Description**

Mapping Illumina TargetID (GeneID) into nuID.

#### Usage

```
targetID2nuID(targetID, lib.mapping = "lumiHumanIDMapping", ...)
```

# Arguments

```
targetID a vector of Illumina TargetID (GeneID)
```

lib.mapping an Illumina ID mapping library

... other parameters of IlluminaID2nuID

#### **Details**

The function will call IlluminaID2nuID when ID mapping library were provided.

# Value

```
see function IlluminaID2nuID
```

# Author(s)

Pan Du

#### References

Du, P., Kibbe, W.A. and Lin, S.M., "nuID: A universal naming schema of oligonucleotides for Illumina, Affymetrix, and other microarrays", Biology Direct 2007, 2:16 (31May2007).

#### See Also

```
nuID2targetID, IlluminaID2nuID
```

96 vst

#### **Examples**

```
if (require(lumiHumanIDMapping)) {
   targetID2nuID('GI_21389350-S', lib='lumiHumanIDMapping')
}
```

vst

Variance Stabilizing Transformation

#### **Description**

Stabilizing the expression variance based on the bead level expression variance and mean relations

#### **Usage**

```
vst(u, std, nSupport = min(length(u), 500), backgroundStd=NULL, fitMethod = c('linear', 'quadratic'), l
```

#### **Arguments**

u mean expression of the beads with same sequence

std expression standard deviation of the beads with same sequence

nSupport the number of down-sampling to speed processing backgroundStd pre-estimated background standard deviation level

fitMethod methods of fitting the relations between expression variance and mean relations lowCutoff cutoff ratio to determine the low expression range. Do not change this until you

now what you are doing.

ifPlot plot intermediate results or not

# **Details**

The variance-stabilizing transformation (VST) takes the advantage of larger number of technical replicates available on the Illumina microarray. It models the mean-variance relationship of the within-array technical replicates at the bead level of Illumina microarray. An arcsinh transform is then applied to stabilize the variance. See reference for more details.

For the methods of fitting the relations between expression variance and mean relations, the 'linear' method is more robust and provides detailed parameters for inverseVST.

#### Value

Return the transformed (variance stabilized) expression values.

# Author(s)

Pan Du, Simon Lin

vst 97

#### References

Lin, S.M., Du, P., Kibbe, W.A., "Model-based Variance-stabilizing Transformation for Illumina Mi-croarray Data", submitted

# See Also

```
lumiT, inverseVST
```

```
## load example data
data(example.lumi)

## get the gene expression mean for one chip
u <- exprs(example.lumi)[,1]

## get the gene standard deviation for one chip
std <- se.exprs(example.lumi)[,1]

## do variance stabilizing transform
transformedU <- vst(u, std)

## do variance stabilizing transform with plotting intermediate result
transformedU <- vst(u, std, ifPlot=TRUE)</pre>
```

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