Package ‘spkTools’

January 31, 2017

Version 1.30.0
Date 2016-02-10
Title Methods for Spike-in Arrays
Author Matthew N McCall <mccallm@gmail.com>, Rafael A Irizarry
<rafa@jhu.edu>
Maintainer Matthew N McCall <mccallm@gmail.com>
Depends R (>= 2.7.0), Biobase (>= 2.5.5)
Suggests xtable
Imports Biobase (>= 2.5.5), graphics, grDevices, gtools, methods,
RColorBrewer, stats, utils
Description The package contains functions that can be used to compare
expression measures on different array platforms.
License GPL (>= 2)
URL http://bioconductor.org
biocViews Software, Technology, Microarray
NeedsCompilation no

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**Description**

This is a SpikeInExpressionSet object containing the data from the Affymetrix HGU133A Spike-in Experiment.

**Usage**

data(affy)

**Format**

It contains a matrix of expression values and a matrix of nominal concentrations.

**Source**


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**plotSpkBox**

**Boxplots of Fold Changes Calculated by spkBox**

**Description**

Plots boxplots of the data resulting from a call to spkBox.

**Usage**

plotSpkBox(boxs, fc=2, box.names=NULL, ...)

**Arguments**

- **boxs** the output of a call to spkBox
- **fc** expected fold change
- **box.names** names to be printed below each boxplot
- **...** parameters passed to boxplot

**Value**

Boxplots for spike-in and non-spike-in comparisons stratified by ALE strata are produced.
SpikeInExpressionSet-class

Author(s)
Matthew N. McCall

Examples
data(affy)
affySlope <- spkSlope(affy)
affyBox <- spkBox(affy, affySlope)
plotSpkBox(affyBox)

SpikeInExpressionSet-class
Class to Contain and Describe High-Throughput Expression Level Assays with Spike-in Data

Description
This is a class representation for spike-in expression data. SpikeInExpressionSet class is derived
from ExpressionSet, and requires a matrix names exprs and a matrix named spikeIn.

Extends
Extends class ExpressionSet.

Creating Objects
createSpikeInExpressionSet(exprs, spikeIn, ...)
new("SpikeInExpressionSet", phenoData = new("AnnotatedDataFrame"), featureData = new("AnnotatedDataFrame"),
    experimentData = new("MIAME"), annotation = character(0), assayData = new("matrix"),spikeIn=new("matrix"))

This creates a SpikeInExpressionSet with assayData implicitly created to contain exprs and
spikeIn. Additional named matrix arguments with the same dimensions as exprs are added to
assayData; the row and column names of these additional matrices should match those of exprs
and spikeIn.

new("SpikeInExpressionSet", assayData = assayDataNew(exprs=new("matrix"),spikeIn=new("matrix"))
This creates a SpikeInExpressionSet with assayData provided explicitly. In this form, the only
required named argument is assayData.

Slots
Inherited from ExpressionSet:

assayData: Contains matrices with equal dimensions, and with column number equal to nrow(phenoData).
    assayData must contain a matrix exprs and a matrix spikeIn with rows representing features
    and columns representing samples.

phenoData: See eSet

annotation See eSet

featureData See eSet

experimentData: See eSet
Methods

Class-specific methods:

spikeIn(SpikeInExpressionSet), spikeIn(SpikeInExpressionSet)← Access and set elements named spikeIn in the AssayData-class slot.

spkSplit(SpikeInExpressionSet) creates two SpikeInExpressionSet objects – one with the spike-in probes and one with the non-spike-in probes.

For derived methods (see ExpressionSet).

See Also

eSet-class, ExpressionSet-class.

Examples

# create an instance of SpikeInExpressionSet
new("SpikeInExpressionSet")

new("SpikeInExpressionSet", exprs=matrix(runif(1000), nrow=100), spikeIn=matrix(rep(1:10,100), nrow=100))

# class specific methods
data(affy)
affySpikes ← spikeIn(affy)
affySplit ← spkSplit(affy)

---

spkAccSD Accuracy Standard Deviation

Description

Estimates the standard deviation for spike-ins at the lowest possible fold change in each bin.

Usage

spkAccSD(object, spkSlopeOut, tol=3)

Arguments

object a SpikeInExpressionSet object
spkSlopeOut the output from the spkSlope function
tol number of digits after decimal point

Value

returns the median absolute deviation (MAD) for each bin.

Author(s)

Matthew N. McCall
Examples
data(affy)
affySlope <- spkSlope(affy)
spkAccSD <- spkAccSD(affy, affySlope)

spkAll  

Spike-in Functions Wrapper

Description
A wrapper for the functions contained in the spkTools package, which calls each function.

Usage
spkAll(object, label, model=expr~spike+probe+array, fc=NULL, tol=3,
xrngs=NULL, yrngs=NULL, cuts=c(.6,.99), potQuantile=.995,
gnn=c(25,100,10000), pch=".", output="eps")

Arguments
object a SpikeInExpressionSet object
label a character string to insert into the graphs and tables produced
model model to be passed to spkAnova
fc the fold change for which fold change plots will be produced
tol the number of digits after the decimal point in fc
xrngs ranges for the x-axis of each plot. d=density, s=slope, v=box, m=M vs A
yrngs ranges for the y-axis of each plot. d=density, s=slope, v=box, m=M vs A
cuts quantiles used to make the low, medium, and high bins
potQuantile the desired quantile to compute the probability of being above
gnn a vector of 3 numbers passed to spkGNN: the desired number of true positives,
     the number of truly expressed genes, and the number of truly unexpressed genes
pch plotting point to be used in spkSlope
output the format in which to save the plots produced. Options are "pdf" and "eps"

Value
The full complement of plots and tables described in the vignette are created and saved in the current working directory.

Author(s)
Matthew N. McCall

Examples
data(affy)
spkAll(affy, label="affy", fc=2)
spkAnova  Anova Model for Microarray Spike-in Data

Description
Computes the mean squared errors of a microarray spike-in design due to concentration, probe, array, and error.

Usage
spkAnova(object, model=expr~spike+probe+array)

Arguments
- object: a SpikeInExpressionSet object
- model: the anova model

Value
A vector of the mean squared errors from the anova model.

Author(s)
Matthew N. McCall

Examples
data(affy)
spkAnova(affy)

spkBal  Quantify Microarray Spike-in Design Imbalance

Description
Computes the imbalance of a microarray spike-in design due to probes and arrays.

Usage
spkBal(object)

Arguments
- object: a SpikeInExpressionSet object

Value
The probe and array imbalances.
Author(s)
Matthew N. McCall

References

Examples
data(affy)
spkBal(affy)

spkBox

Fold Change Calculations

Description
A function to calculate the log-ratios stratified by which ALE groups yield the comparison. They are stratified by which bins are being compared to produce the given fold change.

Usage
spkBox(object, spkSlopeOut, fc = 2, tol = 3, reduce=TRUE)

Arguments
object a SpikeInExpressionSet object
spkSlopeOut the output of the spkSlope function
fc the fold change of interest
tol the precision (number of digits after decimal point) in fc
reduce if TRUE the number of points plotted in the null bins is reduced

Details
This function requires the output of spkSlope.

Value
A list with the log-ratios separated by ALE strata comparison.

Author(s)
Matthew N. McCall

Examples
data(affy)
affySlope <- spkSlope(affy)
spkBox(affy, affySlope)
spkDensity  

Spike-in Density Plot

Description

A density plot of the non-spike-in expression with a rug of the average expression at each spike-in level.

Usage

spkDensity(object, spkSlopeOut, cuts=TRUE, label = NULL, ...)

Arguments

object  
a SpikeInExpressionSet object

spkSlopeOut  
the output from the spkSlope function

cuts  
if TRUE vertical lines are drawn at the expression values separating low vs medium and medium vs high ALE strata

label  
a character string to insert into the plot title

...  
arguments passed to the plot function

Details

This function requires the output of spkSlope.

Value

Density plot is produced.

Author(s)

Matthew N. McCall

Examples

data(affy)
affySlope <- spkSlope(affy)
spkDensity(affy, affySlope)
spkGNN

Genes Needed to Detect N True Positives

Description

Computes the number of genes one would need to consider to obtain a given number of truly positive genes if one considered genes in order of decreasing observed fold change.

Usage

spkGNN(n, n.expr, n.unexpr, AccuracySlope, AccuracySD, nullfc)

Arguments

n                    the desired number of true positives
n.expr                the actual number of truly expressed genes
n.unexpr              the actual number of truly unexpressed genes
AccuracySlope         the signal detect slope from the spkSlope function
AccuracySD            the standard deviation of the signal detect slope from the spkAccSD function
nullfc                a vector of null fold changes from the spkBox function

Value

This function returns the expected number of genes one would have to consider to obtain N true positives under the given conditions.

Author(s)

Matthew N. McCall

Examples

data(affy)
spkSlopeOut <- spkSlope(affy)
spkBoxOut <- spkBox(affy, spkSlopeOut, fc=2)
AccuracySlope <- round(spkSlopeOut$slope[-1], digits=2)
AccuracySD <- round(spkAccSD(affy, spkSlopeOut), digits=2)
spkGNN(n=25, n.expr=100, n.unexpr=10000, AccuracySlope[2], AccuracySD[2], spkBoxOut[[2]])
spkMA

MA Plots

Description

Plots log-ratios (M) vs. average log expression (A) for a SpikeInExpressionSet object.

Usage

spkMA(object, spkSlopeOut, fc=2, tol=3, label=NULL, ylim=NULL, outlier=1, reduce=TRUE, plot.legend=TRUE)

Arguments

object                  a SpikeInExpressionSet object
spkSlopeOut            the output from the spkSlope function
fc                      the fold change of interest
tol                     the precision (number of digits after decimal point) in fc
label                   a character string to insert into the plot title
ylim                    limits of y-axis
outlier                 log fold change cut-off for outliers
reduce                  if TRUE some points are removed from the background to speed plotting
plot.legend             if TRUE a legend is plotted

Value

The MA plot is produced.

Author(s)

Matthew N. McCall

Examples

data(affy)
affySlope <- spkSlope(affy)
spkMA(affy, affySlope)
**spkPair**

**Pairwise Comparisons for Spike-in Genes**

**Description**
Compute log-ratios among spike-in genes.

**Usage**

```r
spkPair(object)
```

**Arguments**

- `object`: a SpikeInExpressionSet object

**Value**
An array containing either log-ratios (M), average log expression (A), and nominal concentrations (N1 & N2). Dimension one is genes, dimension two is array pairings, dimension three is M, A, N1, and N2.

**Author(s)**
Matthew N. McCall

**Examples**

```r
data(affy)
affyPair <- spkPair(affy)
```

---

**spkPairNS**

**Pairwise Comparisons for Non-Spike-in Genes**

**Description**
Compute log-ratios among non-spike-in genes.

**Usage**

```r
spkPairNS(object, output="M")
```

**Arguments**

- `object`: a SpikeInExpressionSet object
- `output`: what to return; either "M" for log-ratios or "A" for average log expression.

**Value**
A matrix containing either log-ratios (M) or average log expression (A). Rows are genes and columns are array pairings.
Author(s)

Matthew N. McCall

Examples

data(affy)
affyPairNS <- spkPairNS(affy)

---

spkPot  Probability of being in the Top

Description

Compute the probability that a spike-in with a nominal fold change of 2 appears in the top 0.5% (default) of log-ratios.

Usage

spkPot(object, spkSlopeOut, sig, SD, precisionQuantile)

Arguments

object    a SpikeInExpressionSet object
spkSlopeOut the output from the spkSlope function
sig     the signal detect slopes from a call to spkSlope
SD    the standard deviation from spkAccSD
precisionQuantile
      the desired quantile to compute the probability of being above

Value

A vector of probabilities for each ALE strata.

Author(s)

Matthew N. McCall

Examples

data(affy)
affySlope <- spkSlope(affy)
affyAccSD <- spkAccSD(affy, affySlope)
spkPot(affy, affySlope, affySlope$slopes, affyAccSD, .995)
spkQuantile

Empirical Quantiles

Description

An internal function called by spkSlope.

Usage

spkQuantile(amt, avgE, ens, p)

Arguments

amt a vector of nominal concentrations
avgE the observed average expression corresponding to each nominal concentration
ens the average expression across arrays of unexpressed genes
p the quantiles to make the bins

Author(s)

Matthew N. McCall

Examples

data(affy)
affySlope <- spkSlope(affy)

spkSlope

Signal Detect Slope Plot

Description

Plots observed expression vs. nominal concentration. The overall regression slope, as well as, regression slopes for low, medium, and high bins are computed and the regression lines plotted.

Usage

spkSlope(object, label = NULL, cuts=c(.6,.99), ...)

Arguments

object a SpikeInExpressionSet object
label a character string to insert into the plot title
cuts quantiles used to make the low, medium, and high bins
... arguments passed to the plot function
Details

The bins are created by computing the proportion of non-spike-in genes with expression values less than or equal to the average expression value at each nominal concentration. Using the default value of cuts, the high bin contains nominal concentrations with 99 percent or more of the non-spike-in expression values lower than it. The medium bin contains nominal concentrations with between 60 and 99 percent of the non-spike-in expression values lower than it. The low bin contains nominal concentrations with less than 60 percent of the non-spike-in expression values lower than it.

Value

- avgExp: average expression at each nominal concentration
- slopes: the regression slopes - overall and for each bin
- breaks: which spike-in levels fall in each bin
- brkpts: the expression value of the cut points between bins
- prop: the proportion of non-spike-in probes with expression less than the average expression at each nominal concentration

Author(s)

Matthew N. McCall

Examples

data(affy)
spkSlope(affy)

Description

A collection of functions to examine microarray datasets that include spike-ins. In particular, it allows one to explore the distribution of spike-ins within the range of possible expression values, the relationship between nominal concentration and expression, and the relationship between expected and observed fold change for different levels of comparison.

Details

Package: spkTools
Type: Package
Version: 0.0.1
Date: 2007-10-9
License: GPL version 2 or newer
spkVar

Author(s)

Matthew N. McCall

Maintainer: Matthew N. McCall <mmccall@jhsph.edu>

Examples

```r
## The Three Plots
data(affy)
par(mfrow=c(2,2))
affySlope <- spkSlope(affy)
spkDensity(affy, affySlope)
spkBox(affy, affySlope)

## The Full Wrapper
data(affy)
spkAll(affy, label="Affymetrix", fc=2)
```

---

<table>
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<tr>
<th>spkVar</th>
<th>Spike-in Variance</th>
</tr>
</thead>
</table>

Description

Compute an estimate of the standard deviation in expression at each nominal concentration.

Usage

`spkVar(object)`

Arguments

- **object**: a SpikeInExpressionSet object

Value

A matrix containing spike-in levels and corresponding MADs.

Author(s)

Matthew N. McCall

Examples

```r
data(affy)
spkVar(affy)
```
**summarySpkBox**  
*Summary of Fold Changes Calculated by spkBox*

**Description**
Prints a summary table of the data resulting from a call to `spkBox`.

**Usage**
```
summarySpkBox(boxs)
```

**Arguments**
- `boxs` the output of a call to `spkBox`

**Value**
A dataframe with 2 columns: the mean fold change and the median average distance of the fold changes.

**Author(s)**
Matthew N. McCall

**Examples**
```r
data(affy)
affySlope <- spkSlope(affy)
affyBox <- spkBox(affy, affySlope)
plotSpkBox(affyBox)
```
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