

# Package ‘cummeRbund’

June 22, 2018

**Title** Analysis, exploration, manipulation, and visualization of  
Cufflinks high-throughput sequencing data.

**Version** 2.22.0

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**Author** L. Goff, C. Trapnell, D. Kelley

**Description** Allows for persistent storage, access, exploration, and manipulation of Cufflinks high-throughput sequencing data. In addition, provides numerous plotting functions for commonly used visualizations.

**Imports** methods, plyr, BiocGenerics, S4Vectors (>= 0.9.25), Biobase

**Depends** R (>= 2.7.0), BiocGenerics (>= 0.3.2), RSQLite, ggplot2,  
reshape2, fastcluster, rtracklayer, Gviz

**Suggests** cluster, plyr, NMFN, stringr, GenomicFeatures, GenomicRanges,  
rjson

**Maintainer** Loyal A. Goff <lgoff@csail.mit.edu>

**License** Artistic-2.0

**Collate** AllGenerics.R AllClasses.R database-setup.R methods-CuffSet.R  
methods-CuffData.R methods-CuffDist.R methods-CuffGeneSet.R  
methods-CuffFeatureSet.R methods-CuffGene.R  
methods-CuffFeature.R tools.R

**LazyLoad** yes

**biocViews** HighThroughputSequencing, HighThroughputSequencingData,  
RNAseq, RNAseqData, GeneExpression, DifferentialExpression,  
Infrastructure, DataImport, DataRepresentation, Visualization,  
Bioinformatics, Clustering, MultipleComparisons, QualityControl

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cummeRbund-package     *cummeRbund: The finishing touch on your Tuxedo workflow. Analysis, manipulation, and visualization of Cufflinks HTS data. ~ package title ~*

---

### Description

Allows for persistent storage, access, and manipulation of Cufflinks high-throughput sequencing data. In addition, provides numerous plotting functions for commonly used visualizations. ~ A concise (1-5 lines) description of the package ~

### Details

Package: cummeRbund  
 Version: 0.1.3  
 Suggests:  
 Depends: R (>= 2.7.0), RSQLite, reshape2, ggplot2, methods  
 License: MIT License  
 Collate: AllGenerics.R AllClasses.R database-setup.R methods-CuffSet.R methods-CuffData.R methods-CuffDist.R  
 LazyLoad: yes  
 biocViews: HighThroughputSequencing, HighThroughputSequencingData, RNAseq, RNAseqData, GeneExpression, D  
 Packaged: 2011-08-05 18:03:50 UTC; lgoff  
 Built: R 2.12.1; ; 2011-08-05 18:03:57 UTC; unix

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CuffDist-class	Class "CuffDist"
CuffFeature-class	Class "CuffFeature"
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CuffGene-class	Class "CuffGene"
CuffGeneSet-class	Class "CuffGeneSet"
CuffSet-class	Class "CuffSet"
JSdist	Jensen-Shannon distance on columns
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createDB	createDB
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csDensity	Density plot of CuffData

```

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                    'cummeRbund' ~~
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                    'cummeRbund' ~~
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                    workflow. Analysis, manipulation, and
                    visualization of Cufflinks HTS data.
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                    ~~
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                    'base' ~~
makeprobs            Transform a matrix into probabilities by
                    columns

```

readCufflinks	readCufflinks
samples	Get sample list from CuffData object
samples-methods	~~ Methods for Function samples in Package 'cummeRbund' ~~
shannon.entropy	Shannon entropy

Further information is available in the following vignettes:

cummeRbund-manual An R package for visualization and analysis of Cufflinks high-throughput sequencing data (source,

~~ An overview of how to use the package, including the most important ~~ functions ~~

### Author(s)

L. Goff, C. Trapnell

Maintainer: Loyal A. Goff <lgoff@csail.mit.edu>

### References

~~ Literature or other references for background information ~~

---

addFeatures	<i>addFeatures</i>
-------------	--------------------

---

### Description

Adds a data.frame of features to a the SQLite backend database.

### Usage

```
## S4 method for signature 'CuffSet'
addFeatures(object, features, level="genes", ...)
```

### Arguments

object	An object of class ('CuffSet' or 'CuffData')
features	A data.frame of features to add. 1st column MUST contain ids (ie. gene_id for 'gene' features, isoform_id for 'isoform' features, etc)
level	One of c('genes', 'isoforms', 'TSS', 'CDS') to indicate which type of features you are being added, and to what data-level.
...	Additional arguments.

### Details

None

### Value

None

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
#None yet.
```

---

count-methods

*Retrieve count values (raw and normalized)*

---

**Description**

Returns a data.frame from @count slot

**Details**

Returns a data.frame of count values.

**Value**

A data.frame of count-level values for a set of features.

**Methods**

```
signature(object = "CuffData")  
signature(object = "CuffFeature")  
signature(object = "CuffFeatureSet")
```

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
data(sampleData)  
count(PINK1)
```

---

countMatrix	<i>countMatrix</i>
-------------	--------------------

---

## Description

Retrieve count values as gene by condition matrix

## Usage

```
## S4 method for signature 'CuffData'  
countMatrix(object, fullnames=FALSE, sampleIdList)  
## S4 method for signature 'CuffData'  
repCountMatrix(object, fullnames=FALSE, repIdList)
```

## Arguments

object	An object of class ('CuffData', 'CuffFeatureSet', 'CuffGeneSet', 'CuffGene', or 'Cuff-Feature')
fullnames	A logical value whether or not to concatenate gene_short_name and tracking_id values (easier to read labels)
sampleIdList	A vector of sample names to subset the resulting matrix.
repIdList	A vector of replicate names to subset the resulting replicate matrix.

## Details

None.

## Value

A feature x condition matrix of count values.

## Note

None

## Author(s)

Loyal A. Goff

## References

None.

## Examples

```
data(sampleData)  
countMatrix(sampleGeneSet)  
repCountMatrix(sampleGeneSet)
```

---

 csBoxplot

*csBoxplot*


---

### Description

Creates a ggplot2 plot object with a geom\_box layer displaying summary statistics for FPKM values across samples (x).

### Usage

```
## S4 method for signature 'CuffData'
csBoxplot(object, logMode=TRUE, pseudocount=0.0001, replicates=FALSE,...)
```

### Arguments

object	An object of class CuffData.
logMode	A logical argument to log10 -transform FPKM values.
pseudocount	Value added to FPKM to avoid log-transform issues.
replicates	A logical value whether or not to plot individual replicates or aggregate condition values.
...	Additional arguments to csBoxplot

### Details

None

### Value

A ggplot2 plot object with a geom\_box layer.

### Note

None

### Author(s)

Loyal A. Goff

### References

None

### Examples

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Read cufflinks data and create CuffSet object
genes<-a@genes #CuffData object for all genes
csBoxplot(genes)
```



---

`csCluster`*csCluster*

---

**Description**

Returns a ggplot2 plot object with geom\_line layer plotting FPKM values over conditions faceted by k-means clustering clusters. (Euclidean). This is very crude at this point. This does not return any of the clustering information directly, but if you want it, you can retrieve it from the ggplot object returned.

**Usage**

```
## S4 method for signature 'CuffFeatureSet'  
csCluster(object,k,logMode=T,method = "none",pseudocount=1,...)
```

**Arguments**

object	An object of class CuffFeatureSet.
k	Number of pre-defined clusters to attempt to find.
logMode	A logical value whether or not to log-transform the FPKM values prior to clustering.
method	Distance function to use when computing cluster solution. Default "none" will use the Jensen-Shannon distance (JSdist). Provide a function that returns a dist object on rows.
pseudocount	Value added to FPKM to avoid log-transform issues.
...	Additional arguments to pam.

**Details**

Uses 'kmeans' function.

**Author(s)**

Loyal A. Goff

**Source**

None

**References**

None.

**Examples**

```
data(sampleData)  
csCluster(sampleGeneSet,4)
```

---

csClusterPlot	<i>csClusterPlot</i>
---------------	----------------------

---

### Description

Replaces the default plotting behavior of the old csCluster. Takes as an argument the output of csCluster and plots expression profiles of features facet by cluster.

### Usage

```
csClusterPlot(clustering, pseudocount=1.0, logMode=FALSE, drawSummary=TRUE, sumFun=mean_cl_boot)
```

### Arguments

clustering	The output of csCluster. (Must be the output of csCluster. Only this data format contains the necessary information for csClusterPlot.)
pseudocount	Value added to FPKM to avoid log transformation issues.
logMode	Logical argument whether to plot FPKM with log axis (Y-axis).
drawSummary	Logical value whether or not to draw a summary line for each cluster (by default this is the cluster mean)
sumFun	Summary function used to by drawSummary (default: mean_cl_boot)

### Details

This replaces the default plotting behavior of the old csCluster() method. This was necessary so as to preserve the cluster information obtained by csCluster in a stable format. The output of csClusterPlot is a ggplot2 object of expressionProfiles faceted by cluster ID.

### Value

A ggplot2 object of expressionProfiles faceted by cluster ID.

### Note

None.

### Author(s)

Loyal A. Goff

### References

None.

### Examples

```
data(sampleData)
myClustering<-csCluster(sampleGeneSet,k=4)
csClusterPlot(myClustering)
```

---

csDendro

*csDendro*


---

**Description**

Creates a grid graphics plot of a dendrogram of Jensen-Shannon distances between conditions of a CuffFeatureSet or CuffGeneSet object.

**Usage**

```
## S4 method for signature 'CuffFeatureSet'
csDendro(object,logMode=T,pseudocount=1,replicates=FALSE)
## S4 method for signature 'CuffData'
csDendro(object,logMode=T,pseudocount=1,replicates=FALSE,...)
```

**Arguments**

object	An object of class 'CuffFeatureSet' or 'CuffGeneSet'
logMode	A logical argument to log10-transform FPKM values prior to plotting.
pseudocount	Value to be added to FPKM for appropriate log transformation and clustering. (Avoids zero-based errors)
replicates	A logical value whether or not to plot individual replicates or aggregate condition values.
...	Additional arguments to csHeatmap

**Details**

None

**Value**

Returns a dendrogram object and plots that object by default.

**Note**

None

**Author(s)**

Loyal A. Goff and Cole Trapnell

**References**

None.

**Examples**

```
data(sampleData)
csDendro(sampleGeneSet)
```

---

csDensity

*Density plot of CuffData*


---

**Description**

Creates a smoothed density plot, by sample, for log10 FPKM values from a cuffdiff run.

**Usage**

```
## S4 method for signature 'CuffData'
csDensity(object, logMode=TRUE, pseudocount=0, labels, features=FALSE, replicates=FALSE,...)
## S4 method for signature 'CuffFeatureSet'
csDensity(object, logMode=TRUE, pseudocount=0, labels, features=FALSE, replicates=FALSE,...)
```

**Arguments**

object	An object of class CuffData.
logMode	A logical value of whether or not to log10-transform FPKM values. By default this is TRUE.
pseudocount	Pseudocount value added to FPKM to avoid errors in log-transformation of true zero values.
labels	A list of tracking_id values or gene_short_name values used for 'callout' points on the density plot for reference. (Not implemented yet).
features	Will include all fields from 'features' slot in returned ggplot object. Useful for further manipulations of plot object using feature-level attributes (e.g. gene_type, class_code, etc)
replicates	A logical value whether or not to plot individual replicates or aggregate condition values.
...	Additional arguments

**Details**

Creates a density plot, by sample, for log10-transformed FPKM values from a cuffdiff run.

**Value**

A ggplot2 plot object

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Create CuffSet object from sample data
genes<-a@genes #Create CuffData object for all 'genes'
d<-csDensity(genes) #Create csDensity plot
d #Render plot
```

---

csDistHeat

*csDistHeat*


---

**Description**

Creates a ggplot plot object with a geom\_tile layer of JS Distance values between samples or genes.

**Usage**

```
## S4 method for signature 'CuffFeatureSet'
csDistHeat(object, replicates=F, samples.not.genes=T,
  logMode=T, pseudocount=1.0,
  heatscale=c(low='lightyellow',mid='orange',high='darkred'),
  heatMidpoint=NULL, ...)
```

**Arguments**

object	An object of class 'CuffFeatureSet' or 'CuffGeneSet'
replicates	A logical argument whether or not to use individual replicate FPKM values as opposed to condition FPKM estimates. (default: FALSE)
samples.not.genes	Compute distances between samples rather than genes. If False, compute distances between genes.
logMode	A logical argument to log10-transform FPKM values prior to plotting.
pseudocount	Value to be added to FPKM for appropriate log transformation and clustering. (Avoids zero-based errors)
heatscale	A list with min length=2, max length=3 that describe the the color scale.
heatMidpoint	Value for midpoint of color scale.
...	Additional arguments to csHeatmap

**Details**

None

**Value**

A ggplot2 plot object with a geom\_tile layer to display distance between samples or genes.

**Note**

None

**Author(s)**

Loyal A. Goff, Cole Trapnell, and David Kelley

## References

None

## Examples

```
data(sampleData)
csDistHeat(sampleGeneSet)
```

---

csHeatmap

*csHeatmap*

---

## Description

Creates a ggplot plot object with a geom\_tile layer of FPKM values per feature and sample.

## Usage

```
## S4 method for signature 'CuffFeatureSet'
csHeatmap(object, rescaling='none', clustering='none', labCol=T, labRow=T, logMode=T, pseudocount
border=FALSE, heatscale= c(low='lightyellow',mid='orange',high='darkred'), heatMidpoint=NULL, full
## S4 method for signature 'CuffFeatureSet'
csFoldChangeHeatmap(object, control_condition, replicate_num=NULL, clustering='none', labCol=T, l
border=FALSE, heatscale=c(low='steelblue',mid='white',high='tomato'), heatMidpoint=0,fullnames=T
```

## Arguments

object	An object of class 'CuffFeatureSet' or 'CuffGeneSet'
control_condition	A character argument indicating which condition should be used as the denominator for fold change. (e.g. "Day0", "Control", etc)
replicate_num	If replicates == TRUE, you must specify both a control condition and a replicate number to use as the denominator.
rescaling	Rescaling can either be 'row' or 'column' OR you can pass rescale a function that operates on a matrix to do your own rescaling. Default is 'none'.
clustering	Clustering can either be 'row','column','none', or 'both', in which case the appropriate indices are re-ordered based on the pairwise Jensen-Shannon distance of FPKM values.
labCol	A logical argument to display column labels.
labRow	A logical argument to display row labels.
logMode	A logical argument to log10-transform FPKM values prior to plotting.
pseudocount	Value to be added to FPKM for appropriate log transformation and clustering. (Avoids zero-based errors)
border	A logical argument to draw border around plot.
heatscale	A list with min length=2, max length=3 that detail the low,mid,and high colors to build the color scale.
heatMidpoint	Value for midpoint of color scale.
fullnames	A logical value whether to use 'fullnames' (concatenated gene_short_name and gene_id) for rows in heatmap. Default [ TRUE ].

replicates	A logical value whether or not to plot individual replicates or aggregate condition values.
method	Function to be used for clustering. Default is JS-distance. You can pass your own function to this argument as long as the output is an instance of the 'dist' class and is applied to the rows of the input matrix.
heatRange	Numerical argument for upper bound on log fold change to be visualized.
...	Additional arguments to csHeatmap

**Details**

None

**Value**

A ggplot2 plot object with a geom\_tile layer to display FPKM values by sample (x) and feature (y)

**Note**

None

**Author(s)**

Loyal A. Goff and Cole Trapnell

**References**

None.

**Examples**

```
data(sampleData)
csHeatmap(sampleGeneSet)
```

---

csScatter

*Scatter Plot*


---

**Description**

A scatter plot comparing the FPKM values from two samples in a cuffdiff run.

**Usage**

```
## S4 method for signature 'CuffData'
csScatter(object, x, y, logMode=TRUE, pseudocount=1.0, labels, smooth=FALSE, colorByStatus = FALSE)
## S4 method for signature 'CuffData'
csScatterMatrix(object, replicates=FALSE, logMode=TRUE, pseudocount=1.0, hexbin=FALSE, useCounts=FALSE)
```

**Arguments**

object	An object of class ('CuffData','CuffFeatureSet')
x	Sample name for x axis
y	Sample name for y axis
logMode	Logical argument to render axes on log10 scale (default: T)
replicates	Logical argument whether or not to draw individual replicate values instead of condition values. (default: T)
pseudocount	Value to add to zero FPKM values for log transformation (default: 0.0001)
smooth	Logical argument to add a smooth-fit regression line
labels	A list of tracking_ids or gene_short_names that will be 'callout' points in the plot for reference. Useful for finding genes of interest in the field. Not implemented yet.
colorByStatus	A logical argument whether or not to color the points by 'significant' Y or N. [Default = FALSE]
drawRug	A logical argument whether or not to draw the rug for x and y axes [Default = TRUE]
hexbin	Logical value whether or not to visualize overplotting with hexbin.
useCounts	Uses normalized counts instead of FPKM.
...	Additional arguments to csScatter

**Details**

None

**Value**

ggplot object with geom\_point and geom\_rug layers

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Create CuffSet object from sample data
genes<-a@genes #Create CuffData object for all genes
s<-csScatter(genes, 'hESC', 'Fibroblasts', smooth=TRUE) #Create plot object
s #render plot object
```



---

csSpecificity	<i>csSpecificity</i>
---------------	----------------------

---

**Description**

Returns a matrix of 'Specificity scores' ( $S$ ) defined as  $1 - \text{JSD}(p_g, q_i)$  where  $p_g$  is the  $\text{Log}_{10}+1$  expression profile of a gene  $g$  across all conditions  $j$ , collapsed into a probability distribution, and  $q_i$  is the unit vector of 'perfect expression' in a given condition  $i$ .

**Usage**

```
## S4 method for signature 'CuffFeatureSet'
csSpecificity(object, logMode=T, pseudocount=1, relative=FALSE, ...)
## S4 method for signature 'CuffData'
csSpecificity(object, logMode=T, pseudocount=1, relative=FALSE, ...)
```

**Arguments**

object	An object of class CuffFeatureSet, CuffGeneSet, or CuffData.
logMode	A logical argument to log10-transform FPKM values prior to plotting.
pseudocount	Value to be added to FPKM for appropriate log transformation and clustering. (Avoids zero-based errors)
relative	A logical argument that when TRUE, will scale the $S$ values from 0-1 by dividing by $\max(S)$
...	Additional arguments to fpkmMatrix.

**Details**

None

**Author(s)**

Loyal A. Goff

**Source**

None

**References**

None.

**Examples**

```
data(sampleData)
csSpecificity(sampleGeneSet)
```

csVolcano

*Volcano Plot***Description**

Creates a volcano plot of log fold change in expression vs  $-\log(\text{pval})$  for a pair of samples (x,y)

**Usage**

```
## S4 method for signature 'CuffData'
csVolcano(object, x, y, alpha=0.05, showSignificant=TRUE, features=FALSE, xlimits = c(-20, 20), ...)
## S4 method for signature 'CuffData'
csVolcanoMatrix(object, alpha=0.05, xlimits=c(-20,20), mapping=aes(), ...)
```

**Arguments**

object	An object of class CuffData, CuffFeatureSet, or CuffGeneSet
x	Sample name from 'samples' table for comparison
y	Sample name from 'samples' table for comparison
alpha	Provide an alpha cutoff for visualizing significant genes
showSignificant	A logical value whether or not to distinguish between significant features or not (by color).
features	Will include all fields from 'features' slot in returned ggplot object. Useful for further manipulations of plot object using feature-level attributes (e.g. gene_type, class_code, etc)
xlimits	Set boundaries for x limits to avoid infinity plotting errors. [Default c(-20,20)]
mapping	Passthrough argument for ggplot aesthetics. Can be ignored completely.
...	Additional arguments

**Details**

This creates a 'volcano' plot of fold change vs. significance for a pairwise comparison of genes or features across two different samples.

**Value**

A ggplot2 plot object

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Create CuffSet object
genes<-a@genes #Create cuffData object for all genes
v<-csVolcano(genes,"hESC","Fibroblasts") # Volcano plot of all genes for conditions x='hESC' and y='Fibroblasts'
v #print plot
```

---

CuffData-class	<i>Class "CuffData"</i>
----------------	-------------------------

---

**Description**

A 'pointer' class for all information (FPKM, annotation, differential expression) for a given feature type (genes, isoforms, TSS, CDS). The methods for this function communicate directly with the SQL backend to present data to the user.

**Objects from the Class**

Objects can be created by calls of the form `new("CuffData", DB, tables, filters, type, idField, ...)`.

**Slots**

DB: Object of class "SQLiteConnection" ~~  
 tables: Object of class "list" ~~  
 filters: Object of class "list" ~~  
 type: Object of class "character" ~~  
 idField: Object of class "character" ~~

**Methods**

**dim** signature(x = "CuffData"): ...  
**getFeatures** signature(object = "CuffData"): ...  
**DB** signature(object = "CuffData"): Accessor for @DB slot  
**diffTable** signature(object = "CuffData"): Create a Full table (wide format) of differential expression information for all pairwise comparisons  
**makeRnk** signature(object = "CuffData"): Internal method to create .rnk file. Should not be called directly  
**annotation** signature(object="CuffData"): Access annotation data

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**See Also**

None

**Examples**

```
showClass("CuffData")
```

---

CuffDist-class

*Class "CuffDist"*


---

**Description**

A 'pointer' class to information relative to the distribution-level tests (promoters, splicing, and relative CDS usage)

**Objects from the Class**

Objects can be created by calls of the form `new("CuffDist", DB, table, type, idField, ...)`.

**Slots**

DB: Object of class "SQLiteConnection" ~~

table: Object of class "character" ~~

type: Object of class "character" ~~

idField: Object of class "character" ~~

**Methods**

**dim** signature(x = "CuffDist"): ...

**samples** signature(x = "CuffDist"): ...

**DB** signature(object = "CuffDist"): Accessor for @DB slot

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**See Also**

None

**Examples**

```
showClass("CuffDist")
```

---

CuffFeature-class      *Class "CuffFeature"*

---

### Description

A 'data' container class for all FPKM, annotation, and differential expression data for a single feature (gene, isoform, TSS, or CDS).

### Objects from the Class

Objects can be created by calls of the form `new("CuffFeature", annotation, fpkm, diff, ...)`.

### Slots

**annotation:** Object of class "data.frame" ~~  
**fpkm:** Object of class "data.frame" ~~  
**diff:** Object of class "data.frame" ~~  
**repFpkm:** Object of class "data.frame" ~~  
**count:** Object of class "data.frame" ~~  
**genome:** Object of class "character" ~~

### Methods

**fpkmMatrix** signature(object="CuffFeature"): ...  
**repFpkmMatrix** signature(object = "CuffFeature"): ...  
**length** signature(x = "CuffFeature"): ...

### Accessors

**annotation** signature(object="CuffFeature"): Access @annotation slot  
**diffData** signature(object="CuffFeature"): Access @diff slot  
**samples** signature(object="CuffFeature"): Get vector of samples

### Note

'CuffGene' is a superclass of 'CuffFeature' that links gene information for a given gene with all isoform-, TSS-, and CDS-level data for the given gene.

### Author(s)

Loyal A. Goff

### References

None

### See Also

[CuffGene](#)

**Examples**

```
showClass("CuffFeature")
```

---

CuffFeatureSet-class    *Class "CuffFeatureSet"*

---

**Description**

A 'data' container class for all FPKM, annotation, and differential expression data for a set of features (genes, isoforms, TSS, CDS).

**Objects from the Class**

Objects can be created by calls of the form `new("CuffFeatureSet", annotation, fpkm, diff, ...)`.

**Slots**

**annotation:** Object of class "data.frame" ~~  
**fpkm:** Object of class "data.frame" ~~  
**diff:** Object of class "data.frame" ~~  
**repFpkm:** Object of class "data.frame" ~~  
**count:** Object of class "data.frame" ~~  
**genome:** Object of class "character" ~~

**Methods**

**diffData** signature(object = "CuffFeatureSet"): ...  
**featureNames** signature(object = "CuffFeatureSet"): ...  
**features** signature(object = "CuffFeatureSet"): ...  
**fpkmMatrix** signature(object = "CuffFeatureSet"): ...  
**repFpkmMatrix** signature(object = "CuffFeatureSet"): ...  
**countMatrix** signature(object = "CuffFeatureSet"): ...  
**samples** signature(object = "CuffFeatureSet"): ...  
**length** signature(object = "CuffFeatureSet"): ...

**Accessors**

**annotation** signature(object="CuffFeatureSet"): Access @annotation slot

**Note**

None.

**Author(s)**

Loyal A. Goff

**References**

None.

**See Also**

[CuffGeneSet](#)

**Examples**

```
showClass("CuffFeatureSet")
```

---

CuffGene-class	<i>Class "CuffGene"</i>
----------------	-------------------------

---

**Description**

A 'data' container class for all FPKM, annotation, and differential expression Data (as well as for all linked features) for a given gene.

**Objects from the Class**

Objects can be created by calls of the form `new("CuffGene", id, isoforms, TSS, CDS, promoters, splicing, relCDS, annotation, genome, fpkm, diff, features)`.

**Slots**

id: Object of class "character" ~~  
 isoforms: Object of class "CuffFeature" ~~  
 TSS: Object of class "CuffFeature" ~~  
 CDS: Object of class "CuffFeature" ~~  
 promoters: Object of class "CuffFeature" ~~  
 relCDS: Object of class "CuffFeature" ~~  
 splicing: Object of class "CuffFeature" ~~  
 annotation: Object of class "data.frame" ~~  
 genome: Object of class "character" ~~  
 fpkm: Object of class "data.frame" ~~  
 diff: Object of class "data.frame" ~~  
 features: Object of class "data.frame" ~~

**Extends**

Class "[CuffFeature](#)", directly.

**Methods**

**length** signature(object="CuffFeature"): Part of length validation (internal use only)  
**makeGeneRegionTrack** signature(object="CuffFeature"): Creates a GeneRegionTrack object (see package Gviz) from a CuffGene object.  
**genePlot** signature(object="CuffFeature"): Internal use only.  
**csPie** signature(object="CuffGene"): Allows for visualization of relative isoform proportion as a pie chart by condition (or optionally as stacked bar charts by adding `+ coord_cartesian()`)

**Accessors**

**genes** signature(object = "CuffGene"): Access @genes slot  
**isoforms** signature(object = "CuffGene"): Access @isoforms slot  
**TSS** signature(object = "CuffGene"): Access @TSS slot  
**CDS** signature(object = "CuffGene"): Access @CDS slot  
**promoters** signature(object = "CuffGene"): Access @CDS slot  
**relCDS** signature(object = "CuffGene"): Access @CDS slot  
**splicing** signature(object = "CuffGene"): Access @CDS slot  
**features** signature(object = "CuffGene"): Access @features slot

**Note**

None.

**Author(s)**

Loyal A. Goff

**References**

None.

**See Also**

[CuffFeature](#)

**Examples**

```
showClass("CuffGene")
```

---

CuffGeneSet-class	<i>Class "CuffGeneSet"</i>
-------------------	----------------------------

---

**Description**

A 'data' container class for all FPKM, annotation, and differential expression data (an associated features) for a given set of genes.

**Objects from the Class**

Objects can be created by calls of the form `new("CuffGeneSet", annotation, fpkm, diff, ...)`.



**Slots**

ids: Object of class "character" ~~  
isoforms: Object of class "CuffFeatureSet" ~~  
TSS: Object of class "CuffFeatureSet" ~~  
CDS: Object of class "CuffFeatureSet" ~~  
promoters: Object of class "CuffFeatureSet" ~~  
relCDS: Object of class "CuffFeatureSet" ~~  
splicing: Object of class "CuffFeatureSet" ~~  
annotation: Object of class "data.frame" ~~  
fpkm: Object of class "data.frame" ~~  
diff: Object of class "data.frame" ~~

**Extends**

Class "[CuffFeatureSet](#)", directly.

**Methods**

No methods defined with class "CuffGeneSet" in the signature.

**Accessors**

**genes** signature(object = "CuffGeneSet"): Access @genes slot  
**isoforms** signature(object = "CuffGeneSet"): Access @isoforms slot  
**TSS** signature(object = "CuffGeneSet"): Access @TSS slot  
**CDS** signature(object = "CuffGeneSet"): Access @CDS slot  
**promoters** signature(object = "CuffGeneSet"): Access @promoters slot  
**relCDS** signature(object = "CuffGeneSet"): Access @relCDS slot  
**splicing** signature(object = "CuffGeneSet"): Access @splicing slot

**Note**

None.

**Author(s)**

Loyal A. Goff

**References**

None.

**See Also**

[CuffFeatureSet](#)

**Examples**

```
showClass("CuffGeneSet")
```

---

CuffSet-class	<i>Class "CuffSet"</i>
---------------	------------------------

---

### Description

A 'pointer' class to connect to, and retrieve data from the SQLite backend database.

### Objects from the Class

Objects can be created by calls of the form `new("CuffSet", DB, conditions, genes, isoforms, TSS, CDS, promoters)`. Available methods are primary accessors to retrieve CuffGeneSet or CuffGene objects for manipulation.

### Slots

DB: Object of class "SQLiteConnection" ~~  
 conditions: Object of class "data.frame" ~~  
 genes: Object of class "CuffData" ~~  
 isoforms: Object of class "CuffData" ~~  
 phenoData: Object of class "data.frame" ~~  
 TSS: Object of class "CuffData" ~~  
 CDS: Object of class "CuffData" ~~  
 promoters: Object of class "CuffDist" ~~  
 splicing: Object of class "CuffDist" ~~  
 relCDS: Object of class "CuffDist" ~~  
 conditions: Object of class "data.frame" ~~

### Methods

[ signature(x = "CuffSet"): ...

### Accessors

**DB** signature(object="CuffSet"): Access @DB slot  
**genes** signature(object = "CuffSet"): Access @genes slot  
**isoforms** signature(object = "CuffSet"): Access @isoforms slot  
**TSS** signature(object = "CuffSet"): Access @TSS slot  
**CDS** signature(object = "CuffSet"): Access @CDS slot  
**promoters** signature(object = "CuffSet"): Access @promoters slot  
**splicing** signature(object = "CuffSet"): Access @splicing slot  
**relCDS** signature(object = "CuffSet"): Access @relCDS slot  
**varModel** signature(object = "CuffSet"): Access varModel info

### Note

None.

**Author(s)**

Loyal A. Goff

**References**

None.

**See Also**

None.

**Examples**

```
showClass("CuffSet")
```

---

diffData

*Differential comparison data*


---

**Description**

An accessor method to retrieve differential expression data from a 'CuffData', 'CuffFeatureSet', or 'CuffFeature' object

**Usage**

```
## S4 method for signature 'CuffData'
diffData(object, x, y, features=FALSE)
## S4 method for signature 'CuffData'
diffTable(object, logCutoffValue=99999)
```

**Arguments**

object	An object of class ('CuffData' or 'CuffFeatureSet')
x	Optional, if x and y are both missing, data for all pairwise differential comparisons are returned, otherwise if x and y are sample names from the 'samples' table, than only differential data pertaining to those two samples are returned.
y	See 'x'
features	A logical value that returns all feature-level data as part of data.frame when true. object must be of class 'CuffData'.
logCutoffValue	Cutoff value for FC estimates to convert to [-]Inf values. Should never really be needed...
...	Additional arguments.

**Details**

None

**Value**

A data.frame object

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
data(sampleData)
diff<-diffData(sampleGeneSet) #returns a dataframe of differential expression data from sample CuffGeneSet ob
```

---

 Dimensionality Reduction

*Dimensionality reduction utilities*


---

**Description**

Dimensionality reduction plots for feature selection and extraction for cummeRbund

**Usage**

```
## S4 method for signature 'CuffData'
MDSplot(object,replicates=FALSE,logMode=TRUE,pseudocount=1.0)
## S4 method for signature 'CuffData'
PCAplot(object,x="PC1",y="PC2",replicates=FALSE,pseudocount=1.0,scale=TRUE,showPoints = TRUE,..
```

**Arguments**

object	The output of class CuffData from which to draw expression estimates. (e.g. genes(cuff))
x	For PCAplot, indicates which principal component is to be presented on the x-axis (e.g. "PC1","PC2","PC3", etc)
y	See x.
pseudocount	Value added to FPKM to avoid log transformation issues.
logMode	Logical value whether or not to use log-transformed expression estimates (default: TRUE)
replicates	A logical value to indicate whether or not individual replicate expression estimates will be used.
scale	For PCAplot, a logical value passed directly to prcomp.
showPoints	For PCAplot, a logical value whether or not to display individual gene values on final PCA plot.
...	Additional passthrough arguments (may not be fully implemented yet).

**Details**

These methods attempt to project a matrix of expression estimates across conditions and/or replicates onto a smaller number of dimensions for feature selection, feature extraction, and can also be useful for outlier detection.

**Value**

A ggplot2 object.

**Note**

None.

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
cuff<-readCufflinks(system.file("extdata", package="cummeRbund")) #Create CuffSet object from sample data
p<-PCAPlot(genes(cuff),x="PC2",y="PC3",replicates=TRUE)
m<-MDSplot(genes(cuff),replicates=TRUE)
p #Render PCA plot
m #Render MDS plot
```

---

dispersionPlot	<i>Mean count vs dispersion plot</i>
----------------	--------------------------------------

---

**Description**

A scatter plot comparing the mean counts against the estimated dispersion for a given level of features from a cuffdiff run.

**Usage**

```
## S4 method for signature 'CuffData'
dispersionPlot(object)
## S4 method for signature 'CuffSet'
dispersionPlot(object)
```

**Arguments**

object            An object of class ('CuffData')

**Details**

None

**Value**

ggplot object with geom\_point layer

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Create CuffSet object from sample data
genes<-genes(a) #Create CuffData object for all genes
d<-dispersionPlot(genes) #Create plot object
d #render plot object
```

---

distValues

*distValues*

---

**Description**

Returns a data.frame of distribution-level test values from a CuffDist object (@promoters, @splicing, @relCDS)

**Usage**

```
## S4 method for signature 'CuffDist'
distValues(object)
```

**Arguments**

object	An object of class 'CuffDist'
...	Additional arguments to distValues

**Details**

None

**Value**

Returns a data.frame of distribution-level test values.

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**See Also**

None

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) # Read cufflinks data and create CuffSet object
distValues(a@promoters) # returns data.frame of values from CuffDist object in slot 'promoters'
```

---

Exploratory Analysis    *Methods for Exploratory Analysis in cummeRbund*

---

**Description**

Exploratory analysis methods for cummeRbund RNA-Seq data.

**Usage**

```
## S4 method for signature 'CuffData'
csNMF(object,k,logMode=T,pseudocount=1,maxiter=1000,replicates=FALSE,fullnames=FALSE)
## S4 method for signature 'CuffFeatureSet'
csNMF(object,k,logMode=T,pseudocount=1,maxiter=1000,replicates=FALSE,fullnames=FALSE)
```

**Arguments**

object	The output of class CuffData or CuffFeatureSet from which to draw expression estimates. (e.g. genes(cuff) or custom feature set via getGenes() or getFeatures())
k	rank value for factorization
logMode	Logical value whether or not to use log-transformed FPKM values. [Default: TRUE]
pseudocount	Value added to FPKM to avoid log transformation issues.
maxiter	Maximum number of iterations for factorization [Default: 1000]
replicates	A logical value to indicate whether or not individual replicate expression estimates will be used.
fullnames	Logical passthrough value to fpkmMatrix whether or not to concatenate gene_short_name with tracking_id. [Default: FALSE]

**Details**

csNMF is a convenience method to invoke the nnmf() method from package:NMFN. This performs non-negative matrix factorization on the provided data and can be useful for many downstream applications.

**Value**

csNMF returns W, H - decomposed matrices of input FPKM values. (See package:NMFN for details)

**Note**

None.

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
data(sampleData)
csNMF(sampleGeneSet, 4)
```

---

expressionBarplot      *Barplot*

---

**Description**

A barplot of FPKM values with confidence intervals for a given gene, set of genes, or features of a gene (e.g. isoforms, TSS, CDS, etc).

**Usage**

```
## S4 method for signature 'CuffFeatureSet'
expressionBarplot(object, logMode=TRUE, pseudocount=1.0, showErrorbars=TRUE, showStatus=TRUE, rep
```

**Arguments**

object	An object of class ('CuffFeatureSet','CuffGeneSet','CuffFeature','CuffGene')
logMode	A logical value whether or not to draw y-axis on log10 scale. Default = FALSE.
pseudocount	Numerical value added to each FPKM during log-transformation to avoid errors.
showErrorbars	A logical value whether or not to draw error bars. Default = TRUE
showStatus	A logical value whether or not to draw visual queues for quantification status of a given gene:condition. Default = TRUE
replicates	A logical value whether or not to plot individual replicates or aggregate condition values.
...	Additional arguments.

**Details**

None



**Value**

A ggplot2 plot object

**Note**

Need to implement logMode and features for this plotting method.

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
data(sampleData)
PINK1 # sample CuffFeature object
expressionBarplot(PINK1) #Barplot of PINK1 FPKM values
expressionBarplot(PINK1@isoforms) #Barplot of PINK1 FPKM values faceted by isoforms
```

---

expressionPlot

*Expression Plot*

---

**Description**

A line plot (optionally with confidence intervals) detailing FPKM expression levels across conditions for a given gene(s) or feature(s)

**Usage**

```
## S4 method for signature 'CuffFeature'
expressionPlot(object, logMode=FALSE, pseudocount=1.0, drawSummary=FALSE, sumFun=mean_cl_boot, sh
```

**Arguments**

object	An object of class ('CuffFeature' or 'CuffGene')
logMode	A logical value to draw y-axis (FPKM) on log-10 scale. Default = FALSE.
pseudocount	A numeric value added to FPKM to avoid errors on log-10 transformation.
drawSummary	A logical value. Draws a 'summary' line with mean FPKM values for each condition.
sumFun	Function used to determine values for summary line. Default = mean_cl_boot
showErrorbars	A logical value whether or not to draw error bars.
showStatus	A logical value whether or not to draw visual queues for quantification status of a given gene:condition. Default = TRUE
replicates	A logical value whether or not to plot individual replicates or aggregate condition values.
facet	A logical value whether or not to facet the plot by feature id (default=TRUE).
...	Additional arguments

**Details**

None

**Value**

A ggplot2 plot object

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```

data(sampleData)
PINK1 # sample CuffFeature object
expressionPlot(PINK1) #Line plot of PINK1 FPKM values
expressionPlot(PINK1@isoforms) #Line plot of PINK1 FPKM values faceted by isoforms

```

---

featureNames	<i>Feature names</i>
--------------	----------------------

---

**Description**

Retrieve a vector of feature names from a 'CuffData' or 'CuffFeatureSet' object

**Usage**

```

## S4 method for signature 'CuffData'
featureNames(object)

```

**Arguments**

object            An object of class ('CuffData' or 'CuffFeatureSet')

**Details**

None

**Value**

A list of feature names

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
data(sampleData)
featureNames(sampleGeneSet)
```

---

features

*Features*

---

**Description**

Returns a data frame of features from a CuffGene object

**Usage**

```
## S4 method for signature 'CuffGene'
features(object)
```

**Arguments**

object            An object of class ('CuffGene')

**Details**

None

**Value**

A data.frame of feature-level information

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
data(sampleData)
features(PINK1)
```

---

`findGene`*findGene*

---

**Description**

A helper function to retrieve the `gene_ids` given a 'lookup' value (e.g. `gene_short_name`, `isoform_id`, etc). Utility to search for `gene_id` and `gene_short_name` given a single 'query' string (e.g. `query='pink1'` will return all genes with 'pink1' (case-insensitive) in the `gene_short_name` field.

**Usage**

```
## S4 method for signature 'CuffSet'  
findGene(object, query)
```

**Arguments**

<code>object</code>	An object of class 'CuffSet' (Primary 'pointer' object for Cufflinks data).
<code>query</code>	A character string for which you would like to retrieve corresponding <code>gene_id</code> values.

**Details**

None.

**Value**

Returns a data.frame of `gene_id` and `gene_short_name` values corresponding to genes from which 'query' matches

**Note**

Right now, this does not return an error if it cannot find a gene. (this is probably a bad thing...)

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
cuff<-readCufflinks(system.file("extdata", package="cummeRbund")) #Read cufflinks data and create master Cufflinks object  
myQuery<-'pink1'  
findGene(cuff,myQuery) # Retrieve gene_id values for any genes matching 'pink1'
```

---

findSimilar	<i>findSimilar</i>
-------------	--------------------

---

**Description**

Returns a CuffGeneSet containing n genes with the most similar expression profiles to gene/profile x.

**Usage**

```
## S4 method for signature 'CuffSet'
findSimilar(object, x, n, distThresh, returnGeneSet=TRUE, ...)
```

**Arguments**

object	A object of class 'CuffSet'
x	A 'gene_id' or 'gene_short_name' from which to look up an expression profile OR a vector of expression values to compare all genes (vector must have same length and order of 'samples')
n	Number of similar genes to return
distThresh	A thresholding value on which to filter results based on JS-distance (e.g. A distThresh of 1.0 will return all genes, 0.0 will return those genes with 'perfect identity' to the gene of interest.)
returnGeneSet	A logical value whether to return a CuffGeneSet object [default] or a distance-ranked data frame of similar genes. The latter is useful if you want to explore the returned list based on distances.
...	Additional arguments to fpkmMatrix call within findSimilar (e.g. fullnames=T)

**Details**

By default, returns a CuffGeneSet object with n similar genes. This may change in the future.

**Value**

A CuffGeneSet object of n most similar genes to x.

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund"))
mySimilarGenes<-findSimilar(a, "PINK1", 10)
```

fpkm-methods

*Retrieve FPKM values*

---

**Description**

Returns a data.frame from @FPKM slot

**Details**

Returns a data.frame of FPKM values.

**Value**

A data.frame of FPKM-level values for a set of features.

**Methods**

```
signature(object = "CuffData")  
signature(object = "CuffFeature")  
signature(object = "CuffFeatureSet")
```

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
data(sampleData)  
fpkm(PINK1)
```

---

fpkmMatrix*fpkmMatrix*

---

**Description**

Retrieve FPKM values as gene by condition (fpkmMatrix) or gene by replicate (repFpkmMatrix) matrix

**Usage**

```
## S4 method for signature 'CuffData'  
fpkmMatrix(object, fullnames=FALSE, sampleIdList)  
## S4 method for signature 'CuffData'  
repFpkmMatrix(object, fullnames=FALSE, repIdList)
```

**Arguments**

object	An object of class ('CuffData', 'CuffFeatureSet', 'CuffGeneSet', 'CuffGene', or 'Cuff-Feature')
fullnames	A logical value whether or not to concatenate gene_short_name and tracking_id values (easier to read labels)
sampleIdList	A vector of sample names to subset the resulting matrix.
repIdList	A vector of sample names to subset the resulting matrix.

**Details**

None.

**Value**

A feature x condition matrix of FPKM values.

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
data(sampleData)  
fpkmMatrix(sampleGeneSet)  
repFpkmMatrix(sampleGeneSet)
```

---

getFeatures

*getFeatures*

---

**Description**

Primary accessor from a CuffSet object to retrieve all related information for >1 (MANY) given FEATURES, indexed by tracking id.

**Usage**

```
## S4 method for signature 'CuffSet'
getFeatures(object, featureIdList, sampleIdList=NULL, level='isoforms')
```

**Arguments**

object	An object of class 'CuffSet' (Primary 'pointer' object for Cufflinks data).
featureIdList	A vector of 'isoform_id', 'TSS_group_id', or 'CDS_id' to identify which features for which you would like to retrieve all information.
sampleIdList	A vector of sample names used to subset or re-order samples in returned object
level	Feature level to be queried for significance (must be one of c('isoforms', 'TSS', 'CDS'))

**Details**

None.

**Value**

Returns a CuffFeatureSet object containing all related information for a given set of tracking\_id values

**Note**

Right now, this does not return an error if it cannot find a gene. (this is probably a bad thing...)

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
cuff<-readCufflinks(system.file("extdata", package="cummeRbund")) #Read cufflinks data and create master Cufflinks object
sample.isoform.ids<-sample(featureNames(isoforms(cuff)),10)
myGene<-getFeatures(cuff,sample.isoform.ids) # Retrieve all information for a set of 10 sampled features.
```

---

getGene

*getGene*

---

**Description**

Primary accessor from a CuffSet object to retrieve all related information for 1 (one) given gene, indexed by gene\_id or gene\_short\_name.

**Usage**

```
## S4 method for signature 'CuffSet'
getGene(object, geneId, sampleIdList=NULL)
```



**Arguments**

object	An object of class 'CuffSet' (Primary 'pointer' object for Cufflinks data).
geneId	A character string to identify which gene for which you would like to retrieve all information.
sampleIdList	A vector of sample names used to subset or re-order samples in returned object

**Details**

None.

**Value**

Returns a CuffGene object containing all related information for a given gene\_id or gene\_short\_name

**Note**

Right now, this does not return an error if it cannot find a gene. (this is probably a bad thing...)

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Read cufflinks data and create master CuffSet
myGene<-getGene(a,"PINK1") # Retrieve all information for gene "PINK1"
```

---

getGeneId

*getGene*

---

**Description**

A helper function to retrieve the gene\_ids for a given list of feature ids (e.g. isoform\_ids, tss\_group\_ids, or CDS\_ids). This should not be called directly by the user

**Usage**

```
## S4 method for signature 'CuffSet'
getGeneId(object, idList)
```

**Arguments**

object	An object of class 'CuffSet' (Primary 'pointer' object for Cufflinks data).
idList	A character string to identify the identifiers for which you would like to retrieve corresponding gene_id values.

**Details**

None.

**Value**

Returns a vector of `gene_id` values corresponding to genes from which `idList` are sub-features.

**Note**

Right now, this does not return an error if it cannot find a gene. (this is probably a bad thing...)

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
cuff<-readCufflinks(system.file("extdata", package="cummeRbund")) #Read cufflinks data and create master CuffSet
sampleFeatureIds<-sample(featureNames(isoforms(cuff)),10)
correspondingGeneIds<-getGeneId(cuff,sampleFeatureIds) # Retrieve gene_id values for parent genes of sampleFeatureIds
```

---

getGenes

*getGenes*

---

**Description**

Primary accessor from a `CuffSet` object to retrieve all related information for >1 (MANY) given genes, indexed by `gene_id` or `gene_short_name`.

**Usage**

```
## S4 method for signature 'CuffSet'
getGenes(object, geneIdList, sampleIdList=NULL)
```

**Arguments**

<code>object</code>	An object of class 'CuffSet' (Primary 'pointer' object for Cufflinks data).
<code>geneIdList</code>	A vector of <code>gene_ids</code> or <code>gene_short_name</code> s to identify which genes for which you would like to retrieve all information.
<code>sampleIdList</code>	A vector of sample names used to subset or re-order samples in returned object

**Details**

None.

**Value**

Returns a `CuffGeneSet` object containing all related information for a given set of `gene_id` or `gene_short_name` values

**Note**

Right now, this does not return an error if it cannot find a gene. (this is probably a bad thing...)

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Read cufflinks data and create master CuffSe
data(sampleData)
sampleIDs
myGene<-getGenes(a,sampleIDs) # Retrieve all information for a set of 20 'sample' genes.
```

---

getLevels

*getLevels*

---

**Description**

Returns a list of samples as levels. This should not be called directly by user.

**Usage**

```
## S4 method for signature 'CuffData'
getLevels(object)
```

**Arguments**

object            An object of class 'CuffData' or 'CuffFeatureSet' or 'CuffFeature'

**Details**

For internal usage only.

**Value**

A vector of sample names as factors.

**Note**

None.

**Author(s)**

Loyal A. Goff

**References**

None.

getRepLevels                      *getRepLevels*

---

**Description**

Returns a list of replicate samples as levels. This should not be called directly by user.

**Usage**

```
## S4 method for signature 'CuffData'  
getRepLevels(object)
```

**Arguments**

object                      An object of class 'CuffSet' or 'CuffData'

**Details**

For internal usage only.

**Value**

A vector of replicate names as factors.

**Note**

None.

**Author(s)**

Loyal A. Goff

**References**

None.

---

getSig                              *getSig*

---

**Description**

Returns the identifiers of significant genes in a vector format.

**Usage**

```
## S4 method for signature 'CuffSet'  
getSig(object,x,y,alpha=0.05,level='genes',method="BH", useCuffMTC=FALSE)
```

**Arguments**

object	A CuffSet object (e.g. cuff)
x	Optional argument to restrict significance results to one pairwise comparison. Must be used with a 'y' argument to specify the other half of the pair.
y	See x.
alpha	An alpha value by which to filter multiple-testing corrected q-values to determine significance
level	Feature level to be queried for significance (must be one of c('genes', 'isoforms', 'TSS', 'CDS'))
method	Multiple testing method to be used for correction. (default: "BH")
useCuffMTC	Logical vector whether or not to use the multiple-testing corrected q-values from the cuffdiff analysis directly, or calculate new q-values from a subset of tests.

**Details**

This is a convenience function to quickly retrieve vectors of identifiers for genes or features that were determined to be significantly regulated between conditions by cuffdiff. This function only returns tracking IDs that correspond to tests with an 'OK' status from cuffdiff, NOTEST values are ignored. By default getSig returns a vector of IDs for all pairwise comparisons together. If you specify an 'x' AND 'y' values as sample names, then only the features that are significant in that particular pairwise comparison are reported, after appropriate multiple testing correction of output p-values.

**Value**

A vector of feature IDs.

**Note**

None.

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Read cufflinks data in sample directory and
mySig<-getSig(a,x='hESC',y='Fibroblasts',alpha=0.05,level='genes')
head(mySig)
```

---

getSigTable	<i>getSigTable</i>
-------------	--------------------

---

**Description**

Returns the identifiers of significant genes in a test table - like format.

**Usage**

```
## S4 method for signature 'CuffSet'
getSigTable(object,alpha=0.05,level='genes')
```

**Arguments**

object	A CuffSet object (e.g. cuff)
alpha	An alpha value by which to filter multiple-testing corrected q-values to determine significance
level	Feature level to be queried for significance (must be one of c('genes','isoforms','TSS','CDS'))

**Details**

This is a convenience function to quickly retrieve lists of identifiers for genes or features that were determined to be significantly regulated between conditions by cuffdiff. This function only returns tracking IDs that correspond to tests with an 'OK' status from cuffdiff, NOTEST values are ignored or reported as NA. By default getSig returns a table of genes x conditions, where the column names represent the pairwise comparisons from the cuffdiff analysis. The values in the table are 1 for features that are significant for this comparison and 0 for genes that are not, any failed tests are reported as <NA>. Only includes the features that are significant in at least one comparison.

**Value**

A data.frame of pairwise test results.

**Note**

None.

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Read cufflinks data in sample directory and
mySigTable<-getSigTable(a,alpha=0.05,level='genes')
head(mySigTable)
```

---

**JSdist***Jensen-Shannon distance on columns*

---

**Description**

JSdist takes a matrix of expression probabilities (calculated directly or output from `makeprobs()`) and returns a dist object of the pairwise Jensen-Shannon distances between columns

**Usage**

```
JSdist(mat, ...)
```

**Arguments**

<code>mat</code>	A matrix of expression probabilities (e.g. from <code>makeprobs()</code> )
<code>...</code>	Passthrough argument to <code>as.dist()</code>

**Details**

Returns pairwise Jensen-Shannon distance (in the form of a dist object) for a matrix of probabilities (by column)

**Value**

A dist object of pairwise J-S distances between columns.

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
mat<-matrix(sample(1:50,50),10)
probs<-makeprobs(mat)
js.distance<-JSdist(probs)
```

---

JSdistFromP	<i>Jensen-Shannon distance on rows from a pre-defined vector of probabilities</i>
-------------	---

---

**Description**

JSdist takes a matrix of expression probabilities (calculated directly or output from makeprobs()) and returns a matrix of Jensen-Shannon distances between individual rows and a specific vector of probabilities (q)

**Usage**

```
JSdistFromP(mat,q)
```

**Arguments**

mat	A matrix of expression probabilities (e.g. from makeprobs())
q	A vector of expression probabilities.

**Details**

Returns Jensen-Shannon distance for each row of a matrix of probabilities against a provided probability distribution (q)

**Value**

A vector of JS distances

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
mat<-matrix(sample(1:50,50),10)
q<-c(100,4,72,8,19)
q<-q/sum(q)
js.distance<-JSdistFromP(mat,q)
```



---

`JSdistVec`*JSdistVec*

---

**Description**

Returns the Jensen-Shannon Distance (square root of JS divergence) between two probability vectors.

**Usage**

```
JSdistVec(p, q)
```

**Arguments**

<code>p</code>	A vector of probabilities
<code>q</code>	A vector of probabilities

**Details**

Should not be called directly by user.

**Value**

Returns the JS distance as a numeric

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
p<-sample(1:5000,20)
q<-sample(1:5000,20)
p<-makeprobsvec(p)
q<-makeprobsvec(q)
JSdistVec(p,q)
```

---

`makeprobs`*Transform a matrix into probabilities by columns*

---

**Description**

This function takes a matrix of expression values (must be greater than 0) and returns a matrix of probabilities by column. This is a required transformation for the Jensen-Shannon distance which is a metric that operates on probabilities.

**Usage**

```
makeprobs(a)
```

**Arguments**

`a` A matrix of expression values (values must be greater than 0).

**Details**

To make a matrix of probabilities by row, use `t()` to transpose prior to calling `makeprobs`.

**Value**

A matrix of expression probabilities by column.

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
myMat<-matrix(sample(1:50,50),10)
probs<-makeprobs(myMat)
```

---

makeprobsvec	<i>makeprobsvec</i>
--------------	---------------------

---

**Description**

Sums a vector of numerics and divides by the sum

**Usage**

```
makeprobsvec(p)
```

**Arguments**

p                    A vector of numerics

**Details**

None

**Value**

A vector of probabilities

**Note**

Should not be called directly by user.

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
p<-sample(1:5000,20)
makeprobsvec(p)
```

MAplot

*MAplot***Description**

Creates an M vs A plot (Avg intensity vs log ratio) for a given pair of conditions across all fpkms

**Usage**

```
## S4 method for signature 'CuffData'
MAplot(object, x, y, logMode=T, pseudocount=1, smooth=FALSE, useCount=FALSE)
```

**Arguments**

object	An object of class 'CuffData'.
x	Sample name from 'samples' table for comparison
y	Sample name from 'samples' table for comparison
logMode	A logical argument to log10-transform FPKM values prior to plotting.
pseudocount	Value to be added to FPKM for appropriate log transformation and clustering. (Avoids zero-based errors)
smooth	Logical argument whether or not to draw a smoothed line fit through data.
useCount	Logical argument whether or not to use mean counts instead of FPKM values.

**Details**

None

**Value**

Returns a ggplot MvsA plot object.

**Note**

None

**Author(s)**

Loyal A. Goff and Cole Trapnell

**References**

None.

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Create CuffSet object from sample data
genes<-a@genes #Create CuffData object for all 'genes'
d<-MAplot(genes, 'hESC', 'Fibroblasts') #Create csDensity plot
d #Render plot
```

---

PINK1

*PINK1*

---

### Description

A sample 'CuffGene' dataset

### Usage

```
data(sampleData)
```

### Format

PINK1 is a CuffGene object (extends CuffFeature) with all sample gene-, isoform-, TSS-, and CDS-level data for the gene 'PINK1'.

### Details

Sample CuffGene data for gene 'PINK1'

### Source

None

### References

None

### Examples

```
data(sampleData)  
PINK1
```

---

QCplots

*Quality Control visualizations*

---

### Description

A collection of ggplot2 visualizations for quality control assessment of cuffdiff output.

- fpkmSCVPlot: A measure of cross-replicate variability, the squared coefficient of variation is a normalized measure of variance between empirical replicate FPKM values per condition, across the range of FPKM estimates.

### Usage

```
## S4 method for signature 'CuffData'  
fpkmSCVPlot(object, FPKMLowerBound=1, showPool = FALSE)
```

**Arguments**

object	An object of class CuffData.
FPKMLowerBound	A lower limit cutoff for FPKM values from which a fit of squared Coefficient of variation (default: 1)
showPool	Logical argument whether to display variability across all replicates independent of condition (TRUE) or the cross-replicate variability for each condition (FALSE)

**Details**

None

**Value**

A ggplot2 plot object with a geom\_box layer.

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Read cufflinks data and create CuffSet object
genes<-a@genes #CuffData object for all genes
csBoxplot(genes)
```

---

readCufflinks	<i>readCufflinks</i>
---------------	----------------------

---

**Description**

This initializes the backend SQLite table and provides a DB connection for all downstream data analysis.

**Usage**

```
readCufflinks(dir = getwd(), dbFile = "cuffData.db", gtfFile = NULL,
runInfoFile = "run.info", repTableFile = "read_groups.info",
geneFPKM = "genes.fpkm_tracking", geneDiff = "gene_exp.diff", geneCount="genes.count_tracking", g
isoformFPKM = "isoforms.fpkm_tracking", isoformDiff = "isoform_exp.diff", isoformCount="isoforms.
TSSFPKM = "tss_groups.fpkm_tracking", TSSDiff = "tss_group_exp.diff", TSSCount="tss_groups.count_
CDSFPKM = "cds.fpkm_tracking", CDSExpDiff = "cds_exp.diff", CDSCount="cds.count_tracking", CDSRep
CSDiff = "cds.diff",
```

```

promoterFile = "promoters.diff",
splicingFile = "splicing.diff",
varModelFile = "var_model.info",
driver = "SQLite",
genome = NULL,
rebuild = FALSE, verbose=FALSE, ...)

```

### Arguments

dir	Directory in which all CuffDiff output files can be located. Defaults to current working directory.
dbFile	Name of backend database. Default is 'cuffData.db'
gtfFile	Path to .gtf file used in cuffdiff analysis. This file will be parsed to retrieve transcript model information.
runInfoFile	run.info file
repTableFile	read_groups.info file
geneFPKM	genes.fpkm_tracking file
geneDiff	gene_exp.diff file
geneCount	genes.count_tracking file
geneRep	genes.read_group_tracking file
isoformFPKM	isoforms.fpkm_tracking file
isoformDiff	isoform_exp.diff file
isoformCount	isoforms.count_tracking file
isoformRep	isoform.read_group_tracking file
TSSFPKM	tss_groups.fpkm_tracking file
TSSDiff	tss_group_exp.diff file
TSSCount	tss_groups.count_tracking file
TSSRep	tss_groups.read_group_tracking file
CDSFPKM	cds.fpkm_tracking file
CDSExpDiff	cds_exp.diff file
CDSCount	cds.count_tracking file
CDSRep	cds.read_group_tracking file
CSDiff	cds.diff file (distribution tests on CDS)
promoterFile	promoters.diff file (distribution tests on promoters)
splicingFile	splicing.diff (distribution tests on isoforms)
varModelFile	varModel.info (emitted in cuffdiff >= v2.1)
driver	Driver for backend database. (Currently only "SQLite" is supported).
genome	A character string indicating to which genome build the .gtf annotations belong (e.g. 'hg19' or 'mm9')
rebuild	A logical argument to rebuild database backend.
verbose	A logical argument for super verbose reporting (As if it wasn't enough already!)
...	Additional arguments to readCufflinks

**Details**

This is the initialization function for the cummeRbund package. It creates the SQLite backend database, populates the data, and provides a connection object for all future interactions with the dataset. Once the initial build is complete, this function will default to using the database for all future sessions. **IMPORTANT:** - Each R session should begin with a call to readCufflinks to re-open the connection to the database. - Should any connectivity issues to the database arise, another call to readCufflinks should create a new connection object and repair any issue. - The database can always be rebuilt (using rebuild=TRUE) from the original CuffDiff output files.

**Value**

A CuffSet object. A 'pointer' class that allows interaction with cufflinks/cuffdiff data via a SQLite database backend.

**Note**

None.

**Author(s)**

Loyal A. Goff

**References**

None.

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Read cufflinks data in sample directory and
```

---

repFpkm-methods	<i>Retrieve FPKM values</i>
-----------------	-----------------------------

---

**Description**

Returns a data.frame from @repFpkm slot

**Details**

Returns a data.frame of replicate FPKM values and associated statistics.

**Value**

A data.frame of replicate-level FPKM values and associated statistics for a set of features.

**Methods**

```
signature(object = "CuffData")
signature(object = "CuffFeature")
signature(object = "CuffFeatureSet")
```



**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
data(sampleData)  
repFpkm(PINK1)
```

---

replicates

*Get replicate sample list from CuffData object*

---

**Description**

Returns a list of replicate names from a CuffData or CuffFeatureSet object

**Usage**

```
## S4 method for signature 'CuffData'  
replicates(object)
```

**Arguments**

object            An object of class ('CuffSet','CuffData')

**Details**

None

**Value**

A list of replicate sample names

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**See Also**

None

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Create CuffSet object
replicates(a@genes)
```

---

**runInfo***Retrieve run parameters and information from a CuffSet object*

---

**Description**

Returns a data.frame of cuffdiff run parameters and information

**Usage**

```
## S4 method for signature 'CuffSet'
runInfo(object)
```

**Arguments**

object            An object of class ('CuffSet')

**Details**

None

**Value**

A data.frame of run parameters

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**See Also**

None

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Create CuffSet object
runInfo(a)
```

---

sampleGeneSet	<i>sampleGeneSet</i>
---------------	----------------------

---

**Description**

A sample CuffGeneSet data set for 20 genes.

**Usage**

```
data(sampleData)
```

**Format**

sampleGeneSet is a CuffGeneSet (extends CuffFeatureSet) object containing all sample gene-, isoform-, TSS-, and CDS-level data for 20 different genes. These data were derived from a toy set of hESC-vs-iPSC-vs-Fibroblast RNA-Seq expression data.

**Details**

None

**Source**

None

**References**

None

**Examples**

```
data(sampleData)
```

---

sampleIDs	<i>sampleIDs</i>
-----------	------------------

---

**Description**

A vector of gene\_ids used to create 'sampleGeneSet' example

**Usage**

```
data(sampleData)
```

**Format**

The format is: chr "sampleIDs"

**Details**

None

**Source**

None

**References**

None

**Examples**

```
data(sampleData)
```

---

samples

*Get sample list from CuffData object*

---

**Description**

Returns a list of sample names from a CuffData or CuffFeatureSet object

**Usage**

```
## S4 method for signature 'CuffData'  
samples(object)
```

**Arguments**

object            An object of class ('CuffData','CuffFeatureSet','CuffFeature')

**Details**

None

**Value**

A list of sample names

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**See Also**

None

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Create CuffSet object
samples(a@genes)
```

---

shannon.entropy	<i>Shannon entropy</i>
-----------------	------------------------

---

**Description**

Calculates the Shannon entropy for a probability distribution

**Usage**

```
shannon.entropy(p)
```

**Arguments**

p                    A vector of probabilities (must sum to ~1)

**Details**

None

**Value**

Returns a numeric value for the Shannon entropy of the supplied probability distribution

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
x<-sample(1:500,50)
p<-x/sum(x)
shannon.entropy(p)
```

---

`sigMatrix`*sigMatrix*

---

**Description**

Returns a ggplot2 plot object representing a matrix of significant features. This is a useful synopsis of all significant pairwise comparisons within the dataset.

**Usage**

```
## S4 method for signature 'CuffSet'  
sigMatrix(object,alpha=0.05,level='genes',orderByDist=FALSE)
```

**Arguments**

<code>object</code>	An object of class CuffSet.
<code>alpha</code>	An alpha value by which to filter multiple-testing corrected q-values to determine significance
<code>level</code>	Feature level to be queried for significance (must be one of c('genes','isoforms','TSS','CDS'))
<code>orderByDist</code>	Logical. If TRUE then samples are re-ordered based on JS-distance from one another (fairly useless unless you have a specific need for this).

**Details**

Creates a matrix plot to illustrate the number of significant features of type 'level' at a given alpha from a cuffdiff run.

**Value**

A ggplot2 plot object

**Note**

None

**Author(s)**

Loyal A. Goff

**References**

None

**Examples**

```
a<-readCufflinks(system.file("extdata", package="cummeRbund")) #Create CuffSet object from sample data  
d<-sigMatrix(a) #Create csDensity plot  
d #Render plot
```

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