# Package 'BRGenomics'

December 26, 2019

Title Tools for the Efficient Analysis of High-Resolution Genomics Data

```
Version 0.2.0
Description This package provides useful and efficient utilites for the
     analysis of high-resolution genomic data using standard Bioconductor
License Artistic-2.0
Encoding UTF-8
LazyData FALSE
RoxygenNote 7.0.2
Depends rtracklayer,
     GenomicFeatures,
     BiocParallel
Imports GenomicRanges,
     stats4,
     BiocGenerics,
     parallel,
     S4Vectors,
     IRanges,
     AnnotationDbi,
     GenomeInfoDb
Suggests BiocStyle,
     knitr,
     rmarkdown,
     TxDb.Hsapiens.UCSC.hg38.knownGene,
     TxDb.Hsapiens.UCSC.hg19.knownGene,
     TxDb.Mmusculus.UCSC.mm10.knownGene,
     TxDb.Mmusculus.UCSC.mm9.knownGene,
     TxDb.Dmelanogaster.UCSC.dm6.ensGene,
     TxDb.Dmelanogaster.UCSC.dm3.ensGene,
     testthat
Enhances DESeq2
biocViews Software,
     DataImport,
     PROseq,
     RNAseq,
```

Type Package

2 binNdimensions

ATACseq, NETseq, ChIPseq, CutAndRun, Transcription, GeneRegulation, Normalization

### VignetteBuilder knitr

# **R** topics documented:

hinN	dimensions N-dimensional binning of data by quantiles	
Index		23
	txs_dm6_chr4	22
	subsetRegionsBySignal	
	subsampleGRanges	
	PROseq_paired	
	PROseq	
	metaSubsampleMatrix	
	metaSubsample	
	mergeGRangesData	
	makeGRangesBPres	
	importTxsUCSC	
	importGenesUCSC	
	import.PROseq	
	import.CoPRO	
	import.bw_trim	
	getStrandedCoverage	
	getPausingIndices	
	getMaxPositionsBySignal	9
	getDESeqResultsInBatch	9
	getDESeqResults	7
	getDESeqDataSet	6
	getCountsByRegions	5
	getCountsByPositions	4
	genebodies	3
	biningimensions	

# Description

This function takes in data along 1 or more dimensions, and for each dimension the data is divided into evenly-sized quantiles from the minimum value to the maximum value, and bin numbers are returned. For instance, if each index of the input data were a gene, the input dimensions would be various quantitative measures of that gene, e.g. expression level, number of exons, length, etc. If plotted in cartesian coordinates, each gene would be a single datapoint, and each measurement would be a separate dimension. The bin numbers for each datapoint in each dimension are returned in a dataframe, with a column for each dimension and a row for each index.

genebodies 3

### Usage

```
binNdimensions(..., quantiles = 10)
```

### **Arguments**

. . .

A single dataframe, or any number of lists or vectors containing different measurements across the same datapoints. If a dataframe is given, columns should correspond to measurements (dimensions). If lists or vectors are given, they must all have the same lengths. Other input classes will be coerced into a single dataframe.

quantiles

Either a number giving the number of quantiles to use for all dimensions (default = 10), or a vector containing the number of quantiles to use for each dimension of input data given.

#### Value

A dataframe containing indices in 1: quantiles for each datapoint in each dimension.

### Author(s)

Mike DeBerardine

genebodies

Extract Genebodies

### **Description**

This function returns ranges that are defined relative to the strand-specific start and end sites of regions of interest (usually genes). Unlike GenomicRanges::promoters, distances can be upstream or downstream based on the sign, and both the start and end of the returned regions can be defined in terms of either the start of end site of the input ranges. For example, genebodies(txs,-50,150,fix\_end = "start") is equivalent to promoters(txs,50,150). The default arguments return ranges that begin 300 bases downstream of the original start positions, and end 300 bases upstream of the original end positions.

```
genebodies(
  genelist,
  start = 300,
  end = -300,
  fix_start = "start",
  fix_end = "end",
  min_window = 500
)
```

### **Arguments**

genelist	A GRanges object containing genes of interest.
start	Depending on fix_start, the distance from either the strand-specific start or end site to begin the returned ranges. If positive, the returned range will begin downstream of the reference position; negative numbers are used to return sites upstream of the reference. Set start = 0 to return the reference position.
end	Identical to the start argument, but defines the strand-specific end position of returned ranges. end must be downstream of start.
fix_start	The reference point to use for defining the strand-specific start positions of returned ranges, either "start" or "end".
fix_end	The reference point to use for defining the strand-specific end positions of returned ranges, either "start" or "end". Cannot be set to "start" if fix_start = "end".
min_window	When fix_start = "start" and fix_end = "end", min_window defines the minimum size (width) of a returned range. However, when fix_end = fix_start, all returned ranges have the same width, and min_window filters the input ranges based on the size of the excluded region downstream (when fix_end = "start") or upstream (when fix_start = "end") of the returned region.

### Value

A GRanges object that may be shorter than genelist due to loss of short ranges.

# Author(s)

Mike DeBerardine

getCountsByPositions Get signal count matrix within regions of interest

# Description

Get signal count matrix within regions of interest

```
getCountsByPositions(
  dataset.gr,
  regions.gr,
  binsize = 1,
  bin_FUN = sum,
  simplify_multi_widths = c("list", "pad zero", "pad NA"),
  field = "score",
  ncores = detectCores()
)
```

getCountsByRegions 5

### **Arguments**

dataset.gr	A GRanges object in which signal is contained in metadata (typically in the "score" field).			
regions.gr	A GRanges object containing all the regions of interest. All ranges must have the same width!			
binsize	Size of bins (in bp) to use for counting within each range of regions.gr. Note that counts will <i>not</i> be length-normalized.			
bin_FUN	If binsize > 1, the function used to aggregate the signal at each base within each bin. The default is to sum the signal in each bin, but any function operating on a numeric vector can be used.			
simplify_multi_widths				
	A string indicating the output format if the ranges in regions.gr have variable widths. Default = "list". See details below.			
field	The metadata field of dataset.gr to be counted. If length(field) > 1, the output is a list whose elements contain the output for generated each field.			
ncores	Multiple cores can only be used if length(field) > 1.			

### **Details**

If the widths of all ranges in regions.gr are equal, a matrix is returned containing a row for each range in regions.gr, and a column for each bin. For input regions.gr with varying widths, setting simplify\_multi\_widths = "list" will output a list of variable-length vectors, with each vector corresponding to an input region. If simplify\_multi\_widths = "pad zero" or simplify\_multi\_widths = "pad NA", the output is a matrix containing a row for each range in regions.gr, and a column for each position in each range. The number of columns is determined by the largest range in regions.gr, and columns corresponding to positions outside of each range are either set to 0 or NA, depending on the argument.

### Author(s)

Mike DeBerardine

getCountsByRegions Signal counts in regions of interest

# **Description**

Count signal (e.g. read coverage) of data in each region of interest.

```
getCountsByRegions(
  dataset.gr,
  regions.gr,
  field = "score",
  ncores = detectCores()
)
```

6 getDESeqDataSet

### **Arguments**

A GRanges object in which signal is contained in metadata (typically in the "score" field).

Regions.gr A GRanges object containing all the regions of interest.

The metadata field of dataset.gr to be counted. If length(field) > 1, a dataframe is returned containing the counts for each region in each field.

ncores Multiple cores can only be used if length(field) > 1.

### Value

Returns a vector the same length as regions.gr containing signal found in each range.

#### Author(s)

Mike DeBerardine

getDESeqDataSet Get DESeqDataSet objects for downstream analysis

### **Description**

This is a convenience function for generating DESeqDataSet objects, but this function also adds support for counting reads across non-contiguous regions.

### Usage

```
getDESeqDataSet(
  dataset.list,
  regions.gr,
  sample_names = names(dataset.list),
  gene_names = NULL,
  sizeFactors = NULL,
  field = "score",
  ncores = detectCores(),
  quiet = FALSE
)
```

### **Arguments**

dataset.list A list of GRanges datasets that can be individually passed to getCountsByRegions.

regions.gr A GRanges object containing regions of interest.

sample\_names Names for each dataset in dataset.list are required, and by default the names

of the list elements are used. The names must each contain the string "\_rep#", where "#" is a single character (usually a number) indicating the replicate. Sam-

ple names across different replicates must be otherwise identical.

gene\_names An optional character vector giving gene names, or any other identifier over

which reads should be counted. Gene names are required if counting is to be performed over non-contiguous ranges, i.e. if any genes have multiple ranges. If supplied, gene names are added to the resulting DESeqDataSet object.

getDESeqResults 7

sizeFactors DESeq2 sizeFactors can be optionally applied in to the DESeqDataSet object

in this function, or they can be applied later on, either by the user or in a call to getDESeqResults. Applying the sizeFactors later is useful if multiple sets of factors will be explored, although sizeFactors can be overwritten at any time.

field Argument passed to getCountsByRegions.

ncores Number of cores to use for read counting across all samples. Default is the total

number of cores available.

quiet If TRUE, all output messages from call to DESeqDataSet will be suppressed.

#### Value

A DESeqData object in which rowData are given as rowRanges, which are equivalent to regions.gr, unless there are non-contiguous gene regions (see note below). Samples (as seen in colData) are factored so that samples are grouped by replicate and condition, i.e. all non-replicate samples are treated as distinct, and the DESeq2 design = ~condition.

### Use of non-contiguous gene regions

In DESeq2, genes must be defined by single, contiguous chromosomal locations. This function allows individual genes to be encompassed by multiple distinct ranges in regions.gr. To use non-contiguous gene regions, provide gene\_names in which some names are duplicated. For each unique gene in gene\_names, this function will generate counts across all ranges for that gene, but be aware that it will only keep the largest range for each gene in the resulting DESeqDataSet object's rowRanges.

### A note on DESeq2 sizeFactors

DESeq2 sizeFactors are sample-specific normalization that are applied by division, i.e.  $counts_{norm,i} = counts_i/sizeFactor_i$ . This is in contrast to normalization factors as defined in this package (and commonly elsewhere), which are applied by multiplication. Also note that DESeq2's "normalizationFactors" are not sample specific, but rather gene specific factors used to correct for ascertainment bias across different genes (e.g. as might be relevant for GSEA or Go analysis).

#### Author(s)

Mike DeBerardine

### See Also

DESeqDataSet, getDESeqResults, getDESeqResultsInBatch

getDESeqResults Get DESeq2 results using reduced dispersion matrices

# Description

This function calls DESeq2::DESeq and DESeq2::results on a pre-existing DESeqDataSet object and returns a DESeqResults table for one or more pairwise comparisons. However, unlike a standard call to DESeq2::results using the contrast argument, this function subsets the dataset so that DESeq2 only estimates dispersion for the samples being compared, and not for all samples present.

8 getDESeqResults

#### **Usage**

```
getDESeqResults(
  dds,
  contrast.numer,
  contrast.denom,
  comparisons.list = NULL,
  sizeFactors = NULL,
  alpha = 0.1,
  args_DESeq = NULL,
  args_results = NULL,
 ncores = detectCores(),
  quiet = FALSE
)
```

### **Arguments**

dds

A DESeqDataSet object, produced using either getDESeqDataSet from this package or DESeqDataSet from DESeq2. If dds was not created using getDESeqDataSet, dds must be made with design = ~condition such that a unique condition level exists for each sample/treatment condition.

contrast.numer

A string naming the condition to use as the numerator in the DESeq2 comparison, typically the perturbative condition.

contrast.denom A string naming the condition to use as the denominator in the DESeq2 comparison, typically the control condition.

comparisons.list

As an optional alternative to supplying a single contrast. numer and contrast. denom, users can supply a list of character vectors containing numerator-denominator pairs, e.g. list(c("B", "A"), c("C", "A"), c("C", "B")).

sizeFactors

A vector containing DESeq2 sizeFactors to apply to each sample. Each sample's readcounts are *divided* by its respective DESeq2 sizeFactor. A warning will be generated if the DESeqDataSet already contains sizeFactors, and the previous sizeFactors will be over-written.

alpha

The significance threshold passed to DESeqResults. This won't affect the output results, but is used as a performance optimization by DESeq2.

args\_DESeq

Additional arguments passed to DESeq, given as a list of argument-value pairs, e.g. list(test = "LRT", fitType = "local"). All arguments given here will be passed to DESeq except for object and parallel. If no arguments are given, all defaults will be used.

args\_results

Additional arguments passed to DESeq2::results, given as a list of argumentvalue pairs, e.g. list(altHypothesis = "greater", lfcThreshold = 1.5). All arguments given here will be passed to results except for object, contrast, alpha, and parallel. If no arguments are given, all defaults will be used.

The number of cores to use for parallel processing. Multicore processing is only used if more than one comparison is being made (i.e. argument comparisons.list is used), and the number of cores utilized will not be greater than the number of comparisons being performed.

quiet

If TRUE, all output messages from calls to DESeq and results will be suppressed, although passing option quiet in args\_DESeq will supersede this option for the call to DESeq.

ncores

### Value

For a single comparison, the output is the DESeqResults result table. If a comparisons.list is used to make multiple comparisons, the output is a named list of DESeqResults objects, with elements named following the pattern "X\_vs\_Y", where X is the name of the numerator condition, and Y is the name of the denominator condition.

### Author(s)

Mike DeBerardine

### See Also

DESeq2::results, getDESeqResultsInBatch, getDESeqDataSet

```
getDESeqResultsInBatch
```

Automate batch calls to getDESeqResults

### **Description**

This function can automate the generation numerous pairwise DESeq2 comparisons using several logical schemes.

### Usage

```
getDESeqResultsInBatch(
  dds,
  sizeFactors = NULL,
  alpha = 0.05,
  anchor = NULL,
  permutations = FALSE,
  additional_comparisons = NULL,
  ncores = detectCores()
)
```

# Arguments

ncores

getMaxPositionsBySignal

Find sites with max signal in regions of interest

### **Description**

For each signal-containing region of interest, find the single site with the most signal. Sites can be found at base-pair resolution, or defined for larger bins.

10 getPausingIndices

#### **Usage**

```
getMaxPositionsBySignal(
  regions.gr,
  dataset.gr,
  binsize = 1,
  field = "score",
  keep.score = F
)
```

### Arguments

regions.gr A GRanges object containing regions of interest.

dataset.gr A GRanges object in which signal is contained in metadata (typically in the

"score" field).

binsize The size of bin in which to calculate signal scores. field The metadata field of dataset.gr to be counted.

keep.score Logical indicating if the signal value at the max site should be reported. If set to

TRUE, the values are kept as a new metadata column in regions.gr.

### Value

Output is a GRanges object with regions.gr metadata, but each range only contains the site within each regions.gr range that had the most signal. If binsize != 1, a single site is still returned, but its position is set to the center of the bin. If the binsize is even, the site is rounded to be closer to the beginning of the range. If keep.score = TRUE, then the output will also have metadata for score at the max site. The output is *not* necessarily same length as regions.gr, as regions without signal are not returned. If *no regions* have signal (e.g. as could happen if running this function on a single region), the function will return an empty GRanges object with intact metadata columns.

### Author(s)

Mike DeBerardine

getPausingIndices

Calculate pausing indices from user-supplied promoters & genebodies

### **Description**

Pausing index (PI) is calculated for each gene (within matched promoters.gr and genebodies.gr) as promoter signal counts divided by genebody signal counts. If length.normalize = TRUE (recommended), the signal counts within each range in promoters.gr and genebodies.gr are divided by their respective range widths (region lengths) before pausing indices are calculated.

```
getPausingIndices(
  dataset.gr,
  promoters.gr,
  genebodies.gr,
  field = "score",
```

getStrandedCoverage 11

```
length_normalize = TRUE,
remove_empty = FALSE
)
```

#### **Arguments**

dataset.gr A GRanges object in which signal is contained in metadata (typically in the

"score" field).

promoters.gr A GRanges object containing all the regions of interest. The sum of all signal

counts within is the pause index numerator.

genebodies.gr A GRanges object containing all the regions of interest. The sum of all signal

counts within is the pause index denominator.

field The metadata field of dataset.gr to be counted, i.e. that contains the read-

counts of interest.

length\_normalize

A logical indicating if signal counts within regions of interest should be length normalized. The default is TRUE, which is recommended, especially if input

regions don't all have the same width.

remove\_empty A logical indicating if genes without any signal should be removed. The default

is FALSE.

#### Value

A vector of length given by the length of the genelist (or possibly shorter if remove\_empty = TRUE).

#### Author(s)

Mike DeBerardine

getStrandedCoverage
Get strand-specific coverage

### **Description**

Computes strand-specific coverage signal, and returns a GRanges object with signal in the "score" metadata column. Function also works for non-strand-specific data. Note that output is not automatically converted into a "basepair-resolution" GRanges object.

### Usage

```
getStrandedCoverage(dataset.gr, field = "score")
```

### **Arguments**

dataset.gr A GRanges object either containing ranges for each read, or one in which read-

counts for individual ranges are contained in metadata (typically in the "score"

field).

field The name of the field that contains readcounts. If no metadata field contains

readcounts, and each range represents a single read, set to NULL.

# Author(s)

Mike DeBerardine

12 import.CoPRO

import.bw\_trim

Import bigWig files (general)

### **Description**

General function for importing a single bigWig file as a GRanges object. The added functionality over rtracklayer::import.bw is in trimming odd chromosomes.

# Usage

```
import.bw_trim(
  file,
  genome = NULL,
  keep_X = TRUE,
  keep_Y = TRUE,
  keep_M = FALSE,
  keep_nonstandard = FALSE
)
```

### **Arguments**

file Path of a bigWig file (non-stranded).

genome Optional string for UCSC reference genome, e.g. "hg38". If given, non-standard chromosomes are trimmed.

keep\_X Logical indicating whether the X chromosome should be kept.

keep\_Y Logical indicating whether the Y chromosome should be kept.

keep\_M Logical indicating whether mitochondrial chromosomes should be kept.

### Value

Imports a GRanges object

# Author(s)

Mike DeBerardine

 ${\tt import.CoPRO}$ 

Import CoPRO (or similar) bedGraph files

# Description

This function imports plus/minus pairs of bedGraph files. This function is useful for when both 5'- and 3'-end information is to be maintained for each sequenced molecule.

import.PROseq 13

### Usage

```
import.CoPRO(
  plus_file,
  minus_file,
  genome = NULL,
  keep_X = TRUE,
  keep_Y = TRUE,
  keep_M = FALSE,
  keep_nonstandard = FALSE
)
```

### Arguments

genome	Optional string for UCSC reference genome, e.g. "hg38". If given, non-standard chromosomes are trimmed.
keep_X	Logical indicating whether the X chromosome should be kept.
keep_Y	Logical indicating whether the Y chromosome should be kept.
keep_M	Logical indicating whether mitochondrial chromosomes should be kept.
plus.file	Path of plus strand bedGraph file.
minus.file	Path of minus strand bedGraph file.

### Value

Imports a GRanges object containing entire strand-specific reads. Each range is unique, and the score metadata column indicates the number of identical reads (which share the same 5' and 3' ends).

# Author(s)

Mike DeBerardine

# Examples

```
md.import.CoPRO("~/LacZ_RNAi_plus.bedGraph",
"~/LacZ_RNAi_minus.bedGraph", "dm6")
```

import.PROseq

Import PRO-seq (or similar) bigWig files

# Description

This function imports plus/minus pairs of bigWig files containing basepair-resolution data, e.g. PRO-seq or PRO-cap data.

14 importGenesUCSC

### Usage

```
import.PROseq(
  plus_file,
  minus_file,
  genome = NULL,
  keep_X = TRUE,
  keep_Y = TRUE,
  keep_M = FALSE,
  keep_nonstandard = FALSE
)
```

### Arguments

genome	Optional string for UCSC reference genome, e.g. "hg38". If given, non-standard chromosomes are trimmed, and options for sex and mitochondrial chromosomes are applied.
keep_X	Logical indicating whether the X chromosome should be kept.
keep_Y	Logical indicating whether the Y chromosome should be kept.
keep_M	Logical indicating whether mitochondrial chromosomes should be kept.
plus_bw	Path of plus strand bigWig file.
minus_bw	Path of minus strand bigWig file.

# Value

Imports a GRanges object containing base-pair resolution data, with the score metadata column indicating readcounts at each base. All ranges are of width = 1.

## Author(s)

Mike DeBerardine

# **Examples**

```
md.import.PROseq("~/LacZ_RNAi_plus.bw", "~/LacZ_RNAi_minus.bw", "dm6")
```

 ${\tt importGenesUCSC}$ 

Import genes from UCSC

### **Description**

Imports all annotated genes by calling GenomicFeatures::genes, which provides a single range for each annotated gene. Currently supports hg38, hg19, mm10, mm9, dm6, and dm3. This script filters non-standard and mitochondrial chromosomes.

importTxsUCSC 15

### Usage

```
importGenesUCSC(
  genome,
  keep_X = TRUE,
  keep_Y = TRUE,
  keep_M = FALSE,
  keep_nonstandard = FALSE
)
```

# Arguments

genome A string indicating the UCSC reference genome, e.g. "hg38".

keep\_X A logical indicating if X chromosome transcripts should be kept.

keep\_Y A logical indicating if Y chromosome transcripts should be kept.

### Value

A GRanges object, including metadata for unique identifiers.

### Author(s)

Mike DeBerardine

importTxsUCSC

Import transcripts from UCSC

# Usage

```
importTxsUCSC(
  genome,
  keep_X = TRUE,
  keep_Y = TRUE,
  keep_M = FALSE,
  keep_nonstandard = FALSE
)
```

# Arguments

genome A string indicating the UCSC reference genome, e.g. "hg38".

keep\_X A logical indicating if X chromosome transcripts should be kept.

keep\_Y A logical indicating if Y chromosome transcripts should be kept.

### Value

A GRanges object, including metadata for unique identifiers.

# Author(s)

Mike DeBerardine

16 mergeGRangesData

makeGRangesBPres

Make base-pair resolution GRanges object

### **Description**

Splits up all ranges in gr to be each 1 basepair wide. All information is preserved, including all metadata. To wit, length(output.gr) = sum(width(dataset.gr)).

### Usage

makeGRangesBPres(dataset.gr)

### **Arguments**

gr

A disjoint GRanges object.

Note that this function doesn't perform any transformation on the metadata in the input; for any ranges of width > 1, the metadata is simply copied to the daughters of that range (whose widths are all equal to 1).

This function is intended to work on datasets at single-base resolution. Data of this type is often formatted as a bigWig file, and any data imported from a big-Wig file by rtracklayer is suitable for processing. bigWig files will typically use run-length compression on the data signal (the 'score' column), such that when imported by rtracklayer, adjacent bases sharing the same signal will combined into a single range. The base-pair resolution GRanges objects produced by this function remove this compression, resulting in each index (each range) of the GRanges object addressing a single genomic position.

To properly use base-pair resolution information, the user should be selecting a single-base from each read, which can be accomplished using GenomicRanges::resize(). Then, single-base coverage can be calculated using get-StrandedCoverage.

### Author(s)

Mike DeBerardine

mergeGRangesData

Merge base-pair resolution GRanges objects

#### **Description**

Merges 2 or more GRanges objects. For each object, the range widths must all be 1, and the score metadata column contains coverage information at each site.

```
mergeGRangesData(..., field = "score", ncores = detectCores())
```

metaSubsample 17

#### **Arguments**

Any number of GRanges objects in which signal (e.g. readcounts) are contained within metadata.

#### Value

A single GRange object containing all sites of the input objects, and the sum of all scores at all sites.

### Author(s)

Mike DeBerardine

metaSubsample

Iterative Subsampling for Metaplotting

### Description

This function performs bootstrap subsampling of mean readcounts at different positions within regions of interest. Mean signal counts can be estimated at base-pair resolution, or smoothed over larger bins.

### Usage

```
metaSubsample(
  dataset.gr,
  regions.gr,
  binsize = 1,
  first_output_xval = 1,
  sample_name = deparse(substitute(dataset.gr)),
  n_iter = 1000,
  prop_subsample = 0.1,
  lower = 0.125,
  upper = 0.875,
  NF = 1,
  field = "score",
  remove_empty = FALSE,
  ncores = 1
)
```

# **Arguments**

dataset.gr A GRanges object in which signal is contained in metadata (typically in the

"score" field).

regions.gr A GRanges object containing intervals over which to metaplot. All ranges must

have the same width.

binsize The size of bin (number of columns, e.g. basepairs) to use for metaplotting.

Especially important for metaplots over large/sparse regions.

first\_output\_xval

The relative start position of the first bin, e.g. if regions.gr begins at 50 bases upstream of the TSS, set first\_output\_xval = -50. This number only affects the x-values that are returned, which are provided as a convenience.

sample\_name Defaults to the name of dataset.gr. This is included in the output as a conve-

nience for row-binding outputs from different samples.

n\_iter Number of random subsampling iterations to perform. Default is 1000.

prop\_subsample The proportion of the ranges in regions.gr (e.g. the proportion of genes) to

subsample in each iteration. The default is 0.1 (10 percent).

10wer The lower quantile of subsampled signal means to return. The default is 0.125

(12.5th percentile).

upper The upper quantile of subsampled signal means to return. The default is 0.875

(85.5th percentile).

NF Optional normalization factor by which to multiply the counts.

field The metadata field of dataset.gr to be counted.

remove\_empty A logical indicating whether regions without signal should be removed from the

analysis.

ncores Number of cores to use for parallel computation. No parallel processing is used

by default, as there's no performance benefit for typical usage with short com-

putation times.

#### Value

Dataframe containing x-values, means, lower quantiles, upper quantiles, and the sample name (as a convenience for row-binding multiple of these dataframes).

### Author(s)

Mike DeBerardine

metaSubsampleMatrix

Iterative Subsampling for Metaplotting (On Count Matrices)

### **Description**

In the most general sense, this function performs iterations of randomly subsampling rows of a matrix, and returns a summary of mean values calculated for each column. The typical application is for generating metaplots, with the typical input being a matrix in which each row is a gene or other region of interest, each column is a position within that gene (either a specific basepair or a bin), and element values are signal (e.g. read counts) within those positions.

```
metaSubsampleMatrix(
  counts.mat,
  binsize = 1,
  first_output_xval = 1,
  sample_name = deparse(substitute(counts.mat)),
  n_iter = 1000,
  prop_subsample = 0.1,
  lower = 0.125,
  upper = 0.875,
  NF = 1,
  ncores = 1
)
```

PROseq 19

### **Arguments**

counts.mat A matrix of signal counts in which rows are regions of interest and columns are

sites/bins in each region.

binsize The size of bin (number of columns, e.g. basepairs) to use for metaplotting.

Especially important for metaplots over large/sparse regions.

first\_output\_xval

The relative start position of the first bin, e.g. if regions.gr begins at 50 bases upstream of the TSS, set first\_output\_xval = -50. This number only affects

the x-values that are returned, which are provided as a convenience.

sample\_name Defaults to the name of dataset.gr.

n\_iter Number of random subsampling iterations to perform. Default is 1000.

prop\_subsample The proportion of rows to subsample in each iteration. The default is 0.1.

10wer The lower quantile of subsampled signal means to return. The default is 0.125

(12.5th percentile).

upper The upper quantile of subsampled signal means to return. The default is 0.875

(85.5th percentile).

NF Optional normalization factor by which to multiply the counts.

ncores Number of cores to use for parallel computation. As of writing, parallel pro-

cessing doesn't show any benefit for short computation times (e.g. <1 minute

for our typical experience on a laptop).

### Value

Dataframe containing x-values, means, lower quantiles, upper quantiles, and the sample name (as a convenience for row-binding multiple of these dataframes).

### Author(s)

Mike DeBerardine

PROseq PRO-seq data from Drosophila S2 cells

### **Description**

PRO-seq data of Drosophila S2 cells, chromosome 4.

### Usage

**PROseq** 

#### **Format**

A disjoint GRanges object with 47533 ranges with 1 metadata column:

score coverage of PRO-seq read 3'-ends ...

20 subsampleGRanges

#### **Details**

Hojoong Kwak, Nicholas J. Fuda, Leighton J. Core, John T. Lis (2013). Precise Maps of RNA Polymerase Reveal How Promoters Direct Initiation and Pausing. Science, 339(6122), 950–953. https://doi.org/10.1126/science.1229386

#### **Source**

GEO Accession GSM1032758, run SRR611828.

PROseq\_paired

Paired PRO-seq data from Drosophila S2 cells

### **Description**

PRO-seq data of Drosophila S2 cells, chromosome 4. Entire mapped reads kept.

### Usage

PROseq\_paired

#### **Format**

A GRanges object with 52464 ranges with 1 metadata column:

**score** number of reads sharing the same mapped 5' and 3' ends ...

### **Details**

Hojoong Kwak, Nicholas J. Fuda, Leighton J. Core, John T. Lis (2013). Precise Maps of RNA Polymerase Reveal How Promoters Direct Initiation and Pausing. Science, 339(6122), 950–953. https://doi.org/10.1126/science.1229386

#### Source

GEO Accession GSM1032758, run SRR611828.

subsampleGRanges

Randomly subsample reads from GRanges dataset

### **Description**

Random subsampling is not performed on ranges, but on reads. Readcounts should be given as a metadata field (usually "score"), and should normally be integers. If normalized readcounts are given, an attempt will be made to infer the normalization factor based on the least-common-multiple of the signal found in the specified field.

```
subsampleGRanges(dataset.gr, n = NULL, prop = NULL, field = "score")
```

### **Arguments**

dataset.gr A GRanges object in which signal (e.g. readcounts) are contained within meta-

data.

n Number of reads to subsample. Either n or prop can be given.

prop Proportion of total signal to subsample.

field The metadata field of dataset.gr that contains readcounts for reach position.

If each range represents a single read, set field = NULL

#### Author(s)

Mike DeBerardine

subsetRegionsBySignal Subset regions of interest by quantiles of overlapping signal

### **Description**

A convenience function to subset regions of interest by the amount of signal they contain, according to their quantile (i.e. their signal ranks).

### Usage

```
subsetRegionsBySignal(
  regions.gr,
  dataset.gr,
  quantiles = c(0.5, 1),
  field = "score",
  order_by_rank = FALSE,
  density = FALSE
)
```

### **Arguments**

regions.gr A GRanges object containing regions of interest.

dataset.gr A GRanges object in which signal is contained in metadata (typically in the

"score" field).

quantiles A value pair giving the lower quantile and upper quantile of regions to keep.

Regions with signal quantiles below than the lower quantile are removed, while regions with signal quantiles above the upper quantile are removed. Quantiles must be in range (0,1). An empty GRanges object is returned if lower

quantile = 1 or upper quantile = 0.

field The metadata field of dataset.gr to be counted.

(in decreasing order). If FALSE (the default), genes are sorted by their positions.

density A logical indicating whether signal counts should be normalized to the width of

ranges in regions.gr. By default, the function only considers the total signal

in each range.

 $txs_dm6_chr4$ 

#### **Details**

Typical uses may include removing the 5 signal (lower\_quantile = 0.05) and the 5 (upper\_quantile = 0.95), or returning the middle 50 signal (lower\_quantile = 0.25, upper\_quantile = 0.75). If lower\_quantile = 0 and upper\_quantile = 1, all regions are returned, but the returned regions will be sorted by position, or by score if order\_by\_rank = TRUE.

### Value

A GRanges object of length length(regions.gr) \* (upper\_quantile -lower\_quantile).

### Author(s)

Mike DeBerardine

txs\_dm6\_chr4

Ensembl transcripts for Drosophila melanogaster, dm6, chromosome 4.

### **Description**

Transcripts obtained from annotation package TxDb.Dmelanogaster.UCSC.dm6.ensGene, which was in turn made by the Bioconductor Core Team from UCSC resources on 2019-04-25. Metadata columns were obtained from "TXNAME" and "GENEID" columns. Data exported from the TxDb package using GenomicFeatures version 1.35.11 on 2019-12-19.

### Usage

txs\_dm6\_chr4

# **Format**

A GRanges object with 339 ranges and 2 metadata columns:

tx\_name Flybase unique identifiers for transcriptsgene\_id FLybase unique identifiers for the associated genes

#### Source

TxDb.Dmelanogaster.UCSC.dm6.ensGene version 3.4.6

# **Index**

```
* datasets
    PROseq, 19
    PROseq_paired, 20
    txs_dm6_chr4, 22
binNdimensions, 2
DESeq, 8
DESeq2::DESeq, 7
DESeq2::results, 7-9
DESeqDataSet, 7, 8
genebodies, 3
GenomicFeatures::genes, 14
GenomicRanges::promoters, 3
getCountsByPositions, 4
getCountsByRegions, 5, 6
getDESeqDataSet, 6, 8, 9
getDESeqResults, 7, 7
getDESeqResultsInBatch, 7, 9, 9
getMaxPositionsBySignal, 9
getPausingIndices, 10
getStrandedCoverage, 11
{\tt import.bw\_trim},\, 12
import.CoPRO, 12
import.PROseq, 13
importGenesUCSC, 14
{\tt importTxsUCSC},\, {\color{red}15}
makeGRangesBPres, 16
mergeGRangesData, 16
metaSubsample, 17
metaSubsampleMatrix, 18
PROseq, 19
PROseq_paired, 20
subsampleGRanges, 20
subsetRegionsBySignal, 21
txs_dm6_chr4, 22
```