

Package ‘ChromSCape’

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Title Analysis of single-cell epigenomics datasets with a Shiny App

Version 1.2.62

Description ChromSCape - Chromatin landscape profiling for Single Cells - is a ready-to-launch user-friendly Shiny Application for the analysis of single-cell epigenomics datasets (scChIP-seq, scATAC-seq, scCUT&Tag, ...) from aligned data to differential analysis & gene set enrichment analysis. It is highly interactive, enables users to save their analysis and covers a wide range of analytical steps: QC, preprocessing, filtering, batch correction, dimensionality reduction, visualization, clustering, differential analysis and gene set analysis.

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biocViews Software, SingleCell, ChIPSeq, ATACSeq, MethylSeq, Classification, Clustering, Epigenetics, PrincipalComponent, SingleCell, ATACSeq, ChIPSeq, Annotation, BatchEffect, MultipleComparison, Normalization, Pathways, Preprocessing, QualityControl, ReportWriting, Visualization, GeneSetEnrichment, DifferentialPeakCalling

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BugReports <https://github.com/vallotlab/ChromSCape/issues>

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annotation_from_merged_peaks
Find nearest peaks of each gene and return refined annotation

Description

Find nearest peaks of each gene and return refined annotation

Usage

```
annotation_from_merged_peaks(scExp, odir, merged_peaks, geneTSS_annotation)
```

Arguments

scExp A SingleCellExperiment object
odir An output directory where to write the mergedpeaks BED file
merged_peaks A list of GRanges object containing the merged peaks
geneTSS_annotation
 A GRanges object with reference genes

Value

A data.frame with refined annotation

annotToCol2 *annotToCol2*

Description

annotToCol2

Usage

```
annotToCol2(  
  annotS = NULL,  
  annotT = NULL,  
  missing = c("", NA),  
  anotype = NULL,  
  maxnumcateg = 2,  
  categCol = NULL,  
  quantitCol = NULL,  
  plotLegend = TRUE,  
  plotLegendFile = NULL  
)
```

Arguments

| | |
|----------------|------------------------------|
| annotS | A color matrix |
| annotT | A color matrix |
| missing | Convert missing to NA |
| anotype | Annotation type |
| maxnumcateg | Maximum number of categories |
| categCol | Categorical columns |
| quantitCol | Quantitative columns |
| plotLegend | Plot legend ? |
| plotLegendFile | Which file to plot legend ? |

Value

A matrix of continuous or discrete colors

Examples

```
data("scExp")
annotToCol2(SingleCellExperiment::colData(scExp), plotLegend = FALSE)
```

| | |
|---------------|---|
| anocol_binary | <i>Helper binary column for anocol function</i> |
|---------------|---|

Description

Helper binary column for anocol function

Usage

```
anocol_binary(anocol, anotype, plotLegend, annotS)
```

Arguments

| | |
|------------|--------------------------|
| anocol | The color feature matrix |
| anotype | The feature types |
| plotLegend | Plot legend ? |
| annotS | A color matrix |

Value

A color matrix similar to anocol with binary columns colored

anocol_categorical *Helper binary column for anocol function*

Description

Helper binary column for anocol function

Usage

```
anocol_categorical(anocol, categCol, anotype, plotLegend, annotS)
```

Arguments

| | |
|------------|---------------------------------|
| anocol | The color feature matrix |
| categCol | Colors for categorical features |
| anotype | The feature types |
| plotLegend | Plot legend ? |
| annotS | A color matrix |

Value

A color matrix similar to anocol with binary columns colored

bams_to_matrix_indexes

Count bam files on interval to create count indexes

Description

Count bam files on interval to create count indexes

Usage

```
bams_to_matrix_indexes(dir, which, BPPARAM = BiocParallel::bpparam())
```

Arguments

| | |
|---------|---|
| dir | A directory containing single cell BAM files and BAI files |
| which | Genomic Range on which to count |
| BPPARAM | BPPARAM object for multiprocessing. See bpparam for more informations. Will take the default BPPARAM set in your R session. |

Value

A list containing a "feature index" data.frame and a count vector for non 0 entries, both used to form the sparse matrix

beds_to_matrix_indexes

Count bed files on interval to create count indexes

Description

Count bed files on interval to create count indexes

Usage

```
beds_to_matrix_indexes(dir, which, BPPARAM = BiocParallel::bpparam())
```

Arguments

| | |
|---------|---|
| dir | A directory containing the single cell BED files |
| which | Genomic Range on which to count |
| BPPARAM | BPPARAM object for multiprocessing. See bpparam for more informations. Will take the default BPPARAM set in your R session. |

Value

A list containing a "feature index" data.frame and a names of cells as vector both used to form the sparse matrix

calculate_CNA

Estimate copy number alterations in cytobands

Description

Cytobands are considered large enough in order that a variation at the cytoband level is not considered as an epigenetic event but as a genetic event, e.g. Copy Number Alterations. The function successively :

- Calculates the fraction of reads in each cytoband (FrCyto). See [calculate_cyto_mat](#)
- Calculates the log2-ratio FrCyto of each cell by the average FrCyto in normal cells. See [calculate_logRatio_CNA](#)
- Estimates if there was a gain or a loss of copy in each cyto band. See [calculate_gain_or_loss](#)

The corresponding matrices are accessibles in the reducedDim slots "cytoBands", "logRatio_cytoBands" and "gainOrLoss_cytoBands" respectively.

Usage

```
calculate_CNA(
  scExp,
  control_samples = unique(scExp$sample_id)[1],
  ref_genome = c("hg38", "mm10")[1],
  quantiles_to_define_gol = c(0.05, 0.95)
)
```

Arguments

`scExp` A SingleCellExperiment with "logRatio_cytoBand" reducedDim slot filled. See [calculate_logRatio_CNA](#)

`control_samples` Sample IDs of the normal sample to take as reference.

`ref_genome` Reference genome ('hg38' or 'mm10')

`quantiles_to_define_gol` Quantiles of normal log2-ratio distribution below/above which cytoband is considered to be a loss/gain. (c(0.05,0.95)). See [calculate_gain_or_loss](#)

Value

The SCE with the fraction of reads, log2-ratio and gain or loss in each cytobands in each cells (of dimension cell x cytoband) in the reducedDim slots.

Examples

```
data("scExp")
scExp = calculate_CNA(scExp, control_samples = unique(scExp$sample_id)[1],
  ref_genome="hg38", quantiles_to_define_gol = c(0.05,0.95))
SingleCellExperiment::reducedDim(scExp, "cytoBand")
SingleCellExperiment::reducedDim(scExp, "logRatio_cytoBand")
SingleCellExperiment::reducedDim(scExp, "gainOrLoss_cytoBand")
```

calculate_cyto_mat *Calculate Fraction of reads in each cytobands*

Description

Re-Count binned reads onto cytobands and calculate the fraction of reads in each of the cytoband in each cell. For each cell, the fraction of reads in any given cytoband is calculated. Cytobands are considered large enough in order that a variation at the cytoband level is not considered as an epigenetic event but as a genetic event, e.g. Copy Number Alterations.

Usage

```
calculate_cyto_mat(scExp, ref_genome = c("hg38", "mm10")[1])
```

Arguments

scExp A SingleCellExperiment with genomic coordinate as features (peaks or bins)
 ref_genome Reference genome ('hg38' or 'mm10')

Value

The SCE with the fraction of reads in each cytobands in each cells (of dimension cell x cytoband) in the reducedDim slot "cytoBand".

Examples

```
data("scExp")
scExp = calculate_cyto_mat(scExp, ref_genome="hg38")
SingleCellExperiment::reducedDim(scExp, "cytoBand")
```

calculate_gain_or_loss

Estimate the copy gains/loss of tumor vs normal based on log2-ratio of fraction of reads

Description

Given a SingleCellExperiment object with the slot "logRatio_cytoBand" containing the log2-ratio of the fraction of reads in each cytoband, estimate if the cytoband was lost or acquired a gain in a non-quantitative way. To do so, the quantiles distribution of the normal cells are calculated, and any cytoband below or above will be considered as a loss/gain. The False Discovery Rate is directly proportional to the quantiles.

Usage

```
calculate_gain_or_loss(scExp, controls, quantiles = c(0.05, 0.95))
```

Arguments

scExp A SingleCellExperiment with "logRatio_cytoBand" reducedDim slot filled. See [calculate_logRatio_CNA](#)
 controls Sample IDs or Cell IDs of the normal sample to take as reference.
 quantiles Quantiles of normal log2-ratio distribution below/above which cytoband is considered to be a loss/gain. (c(0.05,0.95))

Value

The SCE with the gain or loss in each cytobands in each cells (of dimension cell x cytoband) in the reducedDim slot "gainOrLoss_cytoBand".

Examples

```
data("scExp")
scExp = calculate_cyto_mat(scExp, ref_genome="hg38")
scExp = calculate_logRatio_CNA(scExp, controls=unique(scExp$sample_id)[1])
scExp = calculate_gain_or_loss(scExp, controls=unique(scExp$sample_id)[1])
SingleCellExperiment::reducedDim(scExp, "gainOrLoss_cytoBand")
```

```
calculate_logRatio_CNA
```

Calculate the log2-ratio of tumor vs normal fraction of reads in cyto-bands

Description

Given a SingleCellExperiment object with the slot "cytoBand" containing the fraction of reads in each cytoband, calculates the log2-ratio of tumor vs normal fraction of reads in cytobands, cell by cell. If the average signal in normal sample in a cytoband is 0, set this value to 1 so that the ratio won't affect the fraction of read value.

Usage

```
calculate_logRatio_CNA(scExp, controls)
```

Arguments

| | |
|----------|--|
| scExp | A SingleCellExperiment with "cytoBand" reducedDim slot filled. • see calculate_cyto_mat |
| controls | Sample IDs or Cell IDs of the normal sample to take as reference. |

Value

The SCE with the log2-ratio of fraction of reads in each cytobands in each cells (of dimension cell x cytoband) in the reducedDim slot "logRatio_cytoBand".

Examples

```
data("scExp")
scExp = calculate_cyto_mat(scExp, ref_genome="hg38")
scExp = calculate_logRatio_CNA(scExp, controls=unique(scExp$sample_id)[1])
SingleCellExperiment::reducedDim(scExp, "logRatio_cytoBand")
```

 call_mac2_merge_peaks

Calling MACS2 peak caller and merging resulting peaks

Description

Calling MACS2 peak caller and merging resulting peaks

Usage

```
call_mac2_merge_peaks(
  affectation,
  odir,
  p.value,
  format = c("scBED", "BAM")[1],
  ref,
  peak_distance_to_merge
)
```

Arguments

| | |
|------------------------|---|
| affectation | Annotation data.frame with cell cluster and cell id information |
| odir | Output directory to write MACS2 output |
| p.value | P value to detect peaks, passed to MACS2 |
| format | File format, either "BAM" or "scBED" |
| ref | Reference genome to get chromosome information from. |
| peak_distance_to_merge | Distance to merge peaks |

Value

A list of merged GRanges peaks

 changeRange

changeRange

Description

changeRange

Usage

```
changeRange(v, newmin = 1, newmax = 10)
```

Arguments

| | |
|--------|------------------|
| v | A numeric vector |
| newmin | New min |
| newmax | New max |

Value

A matrix with values scaled between newmin and newmax

check_correct_datamatrix

Check if matrix rownames are well formatted and correct if needed

Description

Throws warnings / error if matrix is in the wrong format

Usage

```
check_correct_datamatrix(datamatrix_single, sample_name = "")
```

Arguments

| | |
|-------------------|---------------------------------|
| datamatrix_single | A sparse matrix |
| sample_name | Matrix sample name for warnings |

Value

A sparseMatrix in the right rownames format

choose_cluster_scExp *Choose a number of clusters*

Description

This functions takes as input a SingleCellExperiment object with consclust and a number of cluster to select. It outputs a SingleCellExperiment object with each cell assigned to a correlation cluster in colData. Also calculates a hierarchical clustering of the consensus associations calculated by ConsensusClusterPlus.

Usage

```
choose_cluster_scExp(
  scExp,
  nclust = 3,
  consensus = FALSE,
  hc_linkage = "ward.D"
)
```

Arguments

| | |
|------------|--|
| scExp | A SingleCellExperiment object containing consclust in metadata. |
| nclust | Number of cluster to pick (3) |
| consensus | Use consensus clustering results instead of simple hierarchical clustering ? (FALSE) |
| hc_linkage | A linkage method for hierarchical clustering. See cor . ('ward.D') |

Value

Returns a SingleCellExperiment object with each cell assigned to a correlation cluster in colData.

Examples

```
data("scExp")
scExp_cf = correlation_and_hierarchical_clust_scExp(scExp)
scExp_cf = choose_cluster_scExp(scExp_cf, nclust=3, consensus=FALSE)
table(scExp_cf$cell_cluster)

scExp_cf = consensus_clustering_scExp(scExp)
scExp_cf_consensus = choose_cluster_scExp(scExp_cf, nclust=3, consensus=TRUE)
table(scExp_cf_consensus$cell_cluster)
```

choose_perplexity *Choose perplexity depending on number of cells for Tsne*

Description

Choose perplexity depending on number of cells for Tsne

Usage

```
choose_perplexity(dataset)
```

Arguments

| | |
|---------|---|
| dataset | A matrix of features x cells (rows x columns) |
|---------|---|

Value

A number between 5 and 30 to use in Rtsne function

| | |
|---------|----------------|
| col2hex | <i>Col2Hex</i> |
|---------|----------------|

Description

Transform character color to hexadecimal color code.

Usage

```
col2hex(cname)
```

Arguments

| | |
|-------|------------|
| cname | Color name |
|-------|------------|

Value

The HEX color code of a particular color

| | |
|--------------|--|
| colors_scExp | <i>Adding colors to cells & features</i> |
|--------------|--|

Description

Adding colors to cells & features

Usage

```
colors_scExp(
  scExp,
  annotCol = "sample_id",
  color_by = "sample_id",
  color_df = NULL
)
```

Arguments

| | |
|----------|---|
| scExp | A SingleCellExperiment Object |
| annotCol | Column names to color |
| color_by | If specifying color_df, column names to color |
| color_df | Color data.frame to specify which color for which condition |

Value

A SingleCellExperiment with additionnal "color" columns in colData

Examples

```

data("scExp")
scExp = colors_scExp(scExp,annotCol = c("sample_id",
"total_counts"),
  color_by = c("sample_id","total_counts"))

#Specific colors using a manually created data.frame :
color_df = data.frame(sample_id=unique(scExp$sample_id),
  sample_id_color=c("red","blue","green","yellow"))
scExp = colors_scExp(scExp,annotCol="sample_id",
  color_by="sample_id",color_df=color_df)

```

| | |
|--------------------|--|
| combine_datamatrix | <i>Combine two matrices and emit warning if no regions are in common</i> |
|--------------------|--|

Description

Combine two matrices and emit warning if no regions are in common

Usage

```
combine_datamatrix(datamatrix, datamatrix_single, file_names, i)
```

Arguments

| | |
|-------------------|--|
| datamatrix | A sparse matrix or NULL if empty |
| datamatrix_single | Another sparse matrix |
| file_names | File name corresponding to the matrix for warnings |
| i | file number |

Value

A combined sparse matrix

`combine_enrichmentTests`*Run enrichment tests and combine into list*

Description

Run enrichment tests and combine into list

Usage

```
combine_enrichmentTests(  
  diff,  
  enrichment_qval,  
  qval.th,  
  cdiff.th,  
  annotFeat_long,  
  peak_distance,  
  refined_annotation,  
  GeneSets,  
  GeneSetsDf,  
  GenePool,  
  progress = NULL  
)
```

Arguments

| | |
|---------------------------------|---|
| <code>diff</code> | Differential list |
| <code>enrichment_qval</code> | Adjusted p-value threshold above which a pathway is considered significant list |
| <code>qval.th</code> | Differential analysis adjusted p.value threshold |
| <code>cdiff.th</code> | Differential analysis log-fold change threshold |
| <code>annotFeat_long</code> | Long annotation |
| <code>peak_distance</code> | Maximum gene to peak distance |
| <code>refined_annotation</code> | Refined annotation data.frame if peak calling is done |
| <code>GeneSets</code> | List of pathways |
| <code>GeneSetsDf</code> | Data.frame of pathways |
| <code>GenePool</code> | Pool of possible genes for testing |
| <code>progress</code> | A shiny Progress instance to display progress bar. |

Value

A list of list of pathway enrichment data.frames for Both / Over / Under and for each cluster

| | |
|-----------------|---|
| CompareedgeRGLM | <i>Creates a summary table with the number of genes under- or overexpressed in each group and outputs several graphical representations</i> |
|-----------------|---|

Description

Creates a summary table with the number of genes under- or overexpressed in each group and outputs several graphical representations

Usage

```
CompareedgeRGLM(
  dataMat = NULL,
  annot = NULL,
  ref_group = NULL,
  groups = NULL,
  featureTab = NULL,
  norm_method = "TMMwsp"
)
```

Arguments

| | |
|-------------|--|
| dataMat | reads matrix |
| annot | selected annotation of interest |
| ref_group | List containing one or more vectors of reference samples. Name of the vectors will be used in the results table. The length of this list should be 1 or the same length as the groups list |
| groups | List containing the IDs of groups to be compared with the reference samples. Names of the vectors will be used in the results table |
| featureTab | Feature annotations to be added to the results table |
| norm_method | Which method to use for normalizing ('upperquantile') |

Value

A dataframe containing the foldchange and p.value of each feature

Author(s)

Eric Letouze & Celine Vallot

Examples

```
data("scExp")
scExp_cf = correlation_and_hierarchical_clust_scExp(scExp)
scExp_cf = choose_cluster_scExp(scExp_cf, nclust=2, consensus=FALSE)
featureTab = as.data.frame(SummarizedExperiment::rowRanges(scExp_cf))
```

```

rownames(featureTab) = featureTab$ID
ref_group = list("C1"=scExp_cf$cell_id[which(scExp_cf$cell_cluster=="C1")])
groups = list("C2"=scExp_cf$cell_id[which(scExp_cf$cell_cluster=="C2")])
myres = CompareedgeRGLM(as.matrix(SingleCellExperiment::counts(scExp_cf)),
  annot=as.data.frame(SingleCellExperiment::colData(scExp_cf)),
  ref_group=ref_group,groups=groups, featureTab=featureTab)

```

CompareWilcox

CompareWilcox

Description

CompareWilcox

Usage

```

CompareWilcox(
  dataMat = NULL,
  annot = NULL,
  ref_group = NULL,
  groups = NULL,
  featureTab = NULL,
  block = NULL,
  BPPARAM = BiocParallel::bpparam()
)

```

Arguments

| | |
|------------|---|
| dataMat | A raw count matrix |
| annot | A cell annotation data.frame |
| ref_group | List with cells in reference group(s) |
| groups | List with cells in group(s) to test |
| featureTab | data.frame with feature annotation |
| block | Use a blocking factor to contract batch effect ? |
| BPPARAM | BPPARAM object for multiprocessing. See bpparam for more informations. Will take the default BPPARAM set in your R session. |

Value

A dataframe containing the foldchange and p.value of each feature

Author(s)

Eric Letouze & Celine Vallot & Pacome Prompsy

Examples

```

data("scExp")
scExp_cf = correlation_and_hierarchical_clust_scExp(scExp)
scExp_cf = choose_cluster_scExp(scExp_cf, nclust=2, consensus=FALSE)
featureTab = as.data.frame(SummarizedExperiment::rowRanges(scExp_cf))
rownames(featureTab) = featureTab$ID
ref_group = list("C1"=scExp_cf$cell_id[which(scExp_cf$cell_cluster=="C1")])
groups = list("C2"=scExp_cf$cell_id[which(scExp_cf$cell_cluster=="C2")])
myres = CompareWilcox(as.matrix(SingleCellExperiment::normcounts(scExp_cf)),
  annot=as.data.frame(SingleCellExperiment::colData(scExp_cf)),
  ref_group=ref_group, groups=groups, featureTab=featureTab)

```

concatenate_scBed_into_clusters

Concatenate single-cell BED into clusters

Description

Concatenate single-cell BED into clusters

Usage

```
concatenate_scBed_into_clusters(affectation, files_list, odir)
```

Arguments

| | |
|-------------|--|
| affectation | Annotation data.frame containing cluster information |
| files_list | Named list of scBED file paths to concatenate. List Names must match affectation\$sample_id and basenames must match affectation\$barcode. |
| odir | Output directory to write concatenate pseudo-bulk BEDs. |

Value

Merge single-cell BED files into cluster BED files. Ungzip file if BED is gzipped.

 consensus_clustering_scExp

Wrapper to apply ConsensusClusterPlus to scExp object

Description

Runs consensus hierarchical clustering on PCA feature space of scExp object. Plot consensus scores for each number of clusters. See [ConsensusClusterPlus](#) - Wilkerson, M.D., Hayes, D.N. (2010). ConsensusClusterPlus: a class discovery tool with confidence assessments and item tracking. *Bioinformatics*, 2010 Jun 15;26(12):1572-3.

Usage

```
consensus_clustering_scExp(
  scExp,
  prefix = NULL,
  maxK = 10,
  reps = 100,
  pItem = 0.8,
  pFeature = 1,
  distance = "pearson",
  clusterAlg = "hc",
  innerLinkage = "ward.D",
  finalLinkage = "ward.D",
  plot_consclust = "pdf",
  plot_icl = "png"
)
```

Arguments

| | |
|------------|--|
| scExp | A SingleCellExperiment object containing 'PCA' in reducedDims. |
| prefix | character value for output directory. Directory is created only if plot_consclust is not NULL. This title can be an absolute or relative path. |
| maxK | integer value. maximum cluster number to evaluate. (10) |
| reps | integer value. number of subsamples. (100) |
| pItem | numerical value. proportion of items to sample. (0.8) |
| pFeature | numerical value. proportion of features to sample. (1) |
| distance | character value. 'pearson': (1 - Pearson correlation), 'spearman' (1 - Spearman correlation), 'euclidean', 'binary', 'maximum', 'canberra', 'minkowski' or custom distance function. ('pearson') |
| clusterAlg | character value. cluster algorithm. 'hc' heirarchical (hclust), 'pam' for partitioning around medoids, 'km' for k-means upon data matrix, 'kmdist' ('hc') for k-means upon distance matrices (former km option), or a function that returns a clustering. ('hc') |

| | |
|----------------|---|
| innerLinkage | hierarchical linkage method for subsampling. ('ward.D') |
| finalLinkage | hierarchical linkage method for consensus matrix. ('ward.D') |
| plot_consclust | character value. NULL - print to screen, 'pdf', 'png', 'pngBMP' for bitmap png, helpful for large datasets. ('pdf') |
| plot_icl | same as above for item consensus plot. ('png') |

Details

This functions takes as input a SingleCellExperiment object that must have 'PCA' in reducedDims and outputs a SingleCellExperiment object containing consclust list calculated cluster consensus and item consensus scores in metadata.

Value

Returns a SingleCellExperiment object containing consclust list, calculated cluster consensus and item consensus scores in metadata.

References

ConsensusClusterPlus package by Wilkerson, M.D., Hayes, D.N. (2010). ConsensusClusterPlus: a class discovery tool with confidence assessments and item tracking. *Bioinformatics*, 2010 Jun 15;26(12):1572-3.

Examples

```
data("scExp")
scExp_cf = correlation_and_hierarchical_clust_scExp(scExp)
scExp_cf = consensus_clustering_scExp(scExp)
```

correlation_and_hierarchical_clust_scExp
Correlation and hierarchical clustering

Description

Calculates cell to cell correlation matrix based on the PCA feature space and runs hierarchical clustering taking 1 - correlation scores as distance.

Usage

```
correlation_and_hierarchical_clust_scExp(scExp, hc_linkage = "ward.D")
```

Arguments

| | |
|------------|--|
| scExp | A SingleCellExperiment object, containing 'PCA' in reducedDims. |
| hc_linkage | A linkage method for hierarchical clustering. See cor . ('ward.D') |

Details

This functions takes as input a SingleCellExperiment object that must have PCA calculated and outputs a SingleCellExperiment object with correlation matrix and hierarchical clustering.

Value

Return a SingleCellExperiment object with correlation matrix & hierarchical clustering.

Examples

```
data("scExp")
scExp_cf = correlation_and_hierarchical_clust_scExp(scExp)
```

| | |
|----------------|---|
| count_coverage | <i>Create a smoothed and normalized coverage track from a BAM file and given a bin GenomicRanges object (same as deepTools bamCoverage)</i> |
|----------------|---|

Description

Normalization is CPM, smoothing is done by averaging on n_smoothBin regions left and right of any given region.

Usage

```
count_coverage(
  filename,
  format = "BAM",
  bins,
  canonical_chr,
  n_smoothBin = 5,
  ref = "hg38",
  read_size = 101
)
```

Arguments

| | |
|---------------|--|
| filename | Path towards the BAM to create coverage from |
| format | File format, either "BAM" or "BED" |
| bins | A GenomicRanges object of binned genome |
| canonical_chr | GenomicRanges of the chromosomes to read the BAM file. |
| n_smoothBin | Number of bins left and right to smooth the signal. |
| ref | Genomic reference |
| read_size | Length of the reads |

Value

A binned GenomicRanges that can be readily exported into bigwig file.

create_project_folder *Create ChromSCape project folder*

Description

Creates a project folder that will be recognizable by ChromSCape Shiny application.

Usage

```
create_project_folder(  
  output_directory,  
  analysis_name = "Analysis_1",  
  ref_genome = c("hg38", "mm10")[1]  
)
```

Arguments

output_directory Path towards the directory to create the 'ChromSCape_Analyses' folder and the analysis subfolder. If this path already contains the 'ChromSCape_Analyses' folder, will only create the analysis subfolder.

analysis_name Name of the analysis. Must only contain alphanumerical characters or '_'.

ref_genome Reference genome, either 'hg38' or 'mm10'.

Value

Creates the project folder and returns the root of the project.

Examples

```
dir = tempdir()  
create_project_folder(output_directory = dir,  
  analysis_name = "Analysis_1")  
list.dirs(file.path(dir))
```

`create_sample_name_mat`*Create a sample name matrix*

Description

Create a sample name matrix

Usage

```
create_sample_name_mat(nb_samples, samples_names)
```

Arguments

`nb_samples` Number of samples
`samples_names` Character vector of sample names

Value

A matrix

`create_scDataset_raw` *Create a simulated single cell datamatrix & cell annotation*

Description

Create a simulated single cell datamatrix & cell annotation

Usage

```
create_scDataset_raw(  
  cells = 300,  
  features = 600,  
  featureType = c("window", "peak", "gene"),  
  sparse = TRUE,  
  nsamp = 4,  
  ref = "hg38",  
  batch_id = factor(rep(1, nsamp))  
)
```

Arguments

| | |
|-------------|----------------------------------|
| cells | Number of cells (300) |
| features | Number of features (600) |
| featureType | Type of feature (window) |
| sparse | Is matrix sparse ? (TRUE) |
| nsamp | Number of samples (4) |
| ref | Reference genome ('hg38') |
| batch_id | Batch origin (factor((1,1,1,1))) |

Value

A list composed of * mat : a sparse matrix following an approximation of the negative binomial law (adapted to scChIPseq) * annot : a data.frame of cell annotation * batches : an integer vector with the batch number for each cell

Examples

```
# Creating a basic sparse 600 genomic bins x 300 cells matrix and annotation
l = create_scDataset_raw()
head(l$mat)
head(l$annot)
head(l$batches)

# Specifying number of cells, features and samples
l2 = create_scDataset_raw(cells = 500, features = 500, nsamp=2)

# Specifying species
mouse_l = create_scDataset_raw(ref="mm10")

# Specifying batches
batch_l = create_scDataset_raw(nsamp=4, batch_id = factor(c(1,1,2,2)))

# Peaks of different size as features
peak_l = create_scDataset_raw(featureType="peak")
head(peak_l$mat)

# Genes as features
gene_l = create_scDataset_raw(featureType="gene")
head(gene_l$mat)
```

create_scExp

Wrapper to create the single cell experiment from count matrix and feature dataframe

Description

Create the single cell experiment from (sparse) datamatrix and feature dataframe containing feature names and location. Also optionally removes zero count Features, zero count Cells, non canonical chromosomes, and chromosome M. Calculates QC Metrics (scrn).

Usage

```
create_scExp(  
  datamatrix,  
  annot,  
  remove_zero_cells = TRUE,  
  remove_zero_features = TRUE,  
  remove_non_canonical = TRUE,  
  remove_chr_M = TRUE,  
  mainExpName = "main",  
  verbose = TRUE  
)
```

Arguments

| | |
|----------------------|--|
| datamatrix | A matrix or sparseMatrix of raw counts. Features x Cells (rows x columns). |
| annot | A data.frame containing informations on cells. Should have the same number of rows as the number of columns in datamatrix. |
| remove_zero_cells | remove cells with zero counts ? (TRUE) |
| remove_zero_features | remove cells with zero counts ? (TRUE) |
| remove_non_canonical | remove non canonical chromosomes ?(TRUE) |
| remove_chr_M | remove chromosomes M ? (TRUE) |
| mainExpName | Name of the mainExpName e.g. 'bins', 'peaks'... ("default") |
| verbose | (TRUE) |

Value

Returns a SingleCellExperiment object.

Examples

```
scExp = create_scExp(create_scDataset_raw())$mat, create_scDataset_raw())$annot)  
scExp
```

 DA_custom

Differential Analysis in 'One vs Rest' mode

Description

Differential Analysis in 'One vs Rest' mode

Usage

```
DA_custom(
  affectation,
  counts,
  method,
  feature,
  block,
  ref,
  group,
  progress = NULL,
  BPPARAM = BiocParallel::bpparam()
)
```

Arguments

| | |
|-------------|---|
| affectation | An annotation data.frame with cell_id and cell_cluster columns |
| counts | Count matrix |
| method | DA method : Wilcoxon or EdgeR |
| feature | Feature tables |
| block | Blocking feature |
| ref | If de_type is custom, the reference to compare (data.frame), must be a one-column data.frame with cell_clusters or sample_id as character in rows |
| group | If de_type is custom, the group to compare (data.frame), must be a one-column data.frame with cell_clusters or sample_id as character in rows |
| progress | A shiny Progress instance to display progress bar. |
| BPPARAM | BPPARAM object for multiprocessing. See bpparam for more informations. Will take the default BPPARAM set in your R session. |

Value

A list of results, groups compared and references

DA_one_vs_rest_fun *Differential Analysis in 'One vs Rest' mode*

Description

Differential Analysis in 'One vs Rest' mode

Usage

```
DA_one_vs_rest_fun(  
  affectation,  
  nclust,  
  counts,  
  method,  
  feature,  
  block,  
  progress = NULL,  
  BPPARAM = BiocParallel::bpparam()  
)
```

Arguments

| | |
|-------------|--|
| affectation | An annotation data.frame with cell_id and cell_cluster columns |
| nclust | Number of clusters |
| counts | Count matrix |
| method | DA method : Wilcoxon or EdgeR |
| feature | Feature tables |
| block | Blocking feature |
| progress | A shiny Progress instance to display progress bar. |
| BPPARAM | BPPARAM object for multiprocessing. See bpparam for more informations. Will take the default BPPARAM set in your R session. |

Value

A list of results, groups compared and references

| | |
|-------------|---|
| DA_pairwise | <i>Run differential analysis in Pairwise mode</i> |
|-------------|---|

Description

Run differential analysis in Pairwise mode

Usage

```
DA_pairwise(  
  affectation,  
  nclust,  
  counts,  
  method,  
  feature,  
  block,  
  progress = NULL,  
  BPPARAM = BiocParallel::bpparam()  
)
```

Arguments

| | |
|-------------|--|
| affectation | An annotation data.frame with cell_cluster and cell_id columns |
| nclust | Number of clusters |
| counts | Count matrix |
| method | DA method, Wilcoxon or edgeR |
| feature | Feature data.frame |
| block | Blocking feature |
| progress | A shiny Progress instance to display progress bar. |
| BPPARAM | BPPARAM object for multiprocessing. See bpparam for more informations. Will take the default BPPARAM set in your R session. |

Value

A list of results, groups compared and references

| | |
|----------------|---|
| define_feature | <i>Define the features on which reads will be counted</i> |
|----------------|---|

Description

Define the features on which reads will be counted

Usage

```
define_feature(ref = c("hg38", "mm10")[1],
  peak_file = NULL,
  bin_width = NULL,
  genebody = FALSE,
  extendPromoter = 2500)
```

Arguments

| | |
|----------------|---|
| ref | Reference genome |
| peak_file | A bed file if counting on peaks |
| bin_width | A number of bins if dividing genome into fixed width bins |
| genebody | A logical indicating if feature should be counted in genebodies and promoter. |
| extendPromoter | Extension length before TSS (2500). |

Value

A GRanges object

Examples

```
gr_bins = define_feature("hg38", bin_width = 50000)
gr_genes = define_feature("hg38", genebody = TRUE, extendPromoter = 5000)
```

| | |
|----------------|--|
| detect_samples | <i>Heuristic discovery of samples based on cell labels</i> |
|----------------|--|

Description

Identify a fixed number of common string (samples) in a set of varying strings (cells). E.g. in the set "Sample1_cell1", "Sample1_cell2", "Sample2_cell1", "Sample2_cell2" and with nb_samples=2, the function returns "Sample1", "Sample1", "Sample2", "Sample2".

Usage

```
detect_samples(barcodes, nb_samples = 1)
```

Arguments

barcodes Vector of cell barcode names (e.g. Sample1_cell1, Sample1_cell2...)
nb_samples Number of samples to find

Value

character vector of sample names the same length as cell labels

Examples

```
barcodes = c(paste0("HBCx22_BC_", seq_len(100)),  
             paste0("mouse_sample_XX", 208:397))  
samples = detect_samples(barcodes, nb_samples=2)
```

differential_analysis_scExp

Runs differential analysis between cell clusters

Description

Based on clusters of cell defined previously, runs non-parametric Wilcoxon Rank Sum test to find significantly depleted or enriched features, in 'one_vs_rest' mode or 'pairwise' mode. In pairwise mode, each cluster is compared to all other cluster individually, and then pairwise comparisons between clusters are combined to find overall differential features using combineMarkers function from scan.

Usage

```
differential_analysis_scExp(  
  scExp,  
  de_type = "one_vs_rest",  
  method = "wilcox",  
  qual.th = 0.01,  
  cdiff.th = 1,  
  block = NULL,  
  group = NULL,  
  ref = NULL,  
  prioritize_genes = nrow(scExp) > 20000,  
  max_distanceToTSS = 1000,  
  progress = NULL,  
  BPPARAM = BiocParallel::bpparam()  
)
```

Arguments

| | |
|-------------------|--|
| scExp | A SingleCellExperiment object containing consclust with selected number of cluster. |
| de_type | Type of comparisons. Either 'one_vs_rest', to compare each cluster against all others, or 'pairwise' to make 1 to 1 comparisons. ('one_vs_rest') |
| method | Differential testing method, either 'wilcox' for Wilcoxon non- parametric testing or 'neg.binomial' for edgeRGLM based testing. ('wilcox') |
| qval.th | Adjusted p-value threshold. (0.01) |
| cdiff.th | Fold change threshold. (1) |
| block | Use batches as blocking factors ? If TRUE, block will be taken as the column "batch_id" from the SCE. Cells will be compared only within samples belonging to the same batch. |
| group | If de_type = "custom", the sample / cluster of interest as a one- column data.frame. The name of the column is the group name and the values are character either cluster ("C1", "C2", ...) or sample_id. |
| ref | If de_type = "custom", the sample / cluster of reference as a one- column data.frame. The name of the column is the group name and the values are character either cluster ("C1", "C2", ...) or sample_id. |
| prioritize_genes | First filter by loci being close to genes ? E.g. for differential analysis, it is more relevant to keep features close to genes |
| max_distanceToTSS | If prioritize_genes is TRUE, the maximum distance to consider a feature close to a gene. |
| progress | A shiny Progress instance to display progress bar. |
| BPPARAM | BPPARAM object for multiprocessing. See bpparam for more informations. Will take the default BPPARAM set in your R session. |

Details

This functions takes as input a SingleCellExperiment object with consclust, the type of comparison, either 'one_vs_rest' or 'pairwise', the adjusted p-value threshold (qval.th) and the fold-change threshold (cdiff.th). It outputs a SingleCellExperiment object containing a differential list.

Value

Returns a SingleCellExperiment object containing a differential list.

Examples

```
data("scExp")
scExp_cf = differential_analysis_scExp(scExp)
```

| | |
|-------------|--------------------|
| distPearson | <i>distPearson</i> |
|-------------|--------------------|

Description

distPearson

Usage

```
distPearson(m)
```

Arguments

| | |
|---|----------|
| m | A matrix |
|---|----------|

Value

A dist object

| | |
|----------------|-----------------------|
| enrichmentTest | <i>enrichmentTest</i> |
|----------------|-----------------------|

Description

enrichmentTest

Usage

```
enrichmentTest(gene.sets, mylist, possibleIds, sep = ";", silent = FALSE)
```

Arguments

| | |
|-------------|----------------------------------|
| gene.sets | A list of reference gene sets |
| mylist | A list of genes to test |
| possibleIds | All existing genes |
| sep | Separator used to collapse genes |
| silent | Silent mode ? |

Value

A dataframe with the gene sets and their enrichment p.value

 exclude_features_scExp

Remove specific features (CNA, repeats)

Description

Remove specific features (CNA, repeats)

Usage

```
exclude_features_scExp(
  scExp,
  features_to_exclude,
  by = "region",
  verbose = TRUE
)
```

Arguments

| | |
|---------------------|---|
| scExp | A SingleCellExperiment object. |
| features_to_exclude | A GenomicRanges object or data.frame containing genomic regions or features to exclude or path towards a BED file containing the features to exclude. |
| by | Type of features. Either 'region' or 'feature_name'. If 'region', will look for genomic coordinates in columns 1-3 (chr,start,stop). If 'feature_name', will look for a genes in first column. ('region') |
| verbose | (TRUE) |

Value

A SingleCellExperiment object without features to exclude.

Examples

```
scExp = create_scExp(create_scDataset_raw()$mat, create_scDataset_raw()$annot)
features_to_exclude = data.frame(chr=c("chr4", "chr7", "chr17"),
  start=c(50000, 8000000, 2000000),
  end=c(100000, 16000000, 2500000))
features_to_exclude = as(features_to_exclude, "GRanges")
scExp = exclude_features_scExp(scExp, features_to_exclude)
scExp
```

```
feature_annotation_scExp
```

Add gene annotations to features

Description

Add gene annotations to features

Usage

```
feature_annotation_scExp(scExp, ref = "hg38", reference_annotation = NULL)
```

Arguments

| | |
|----------------------|---|
| scExp | A SingleCellExperiment object. |
| ref | Reference genome. Either 'hg38' or 'mm10'. ('hg38') |
| reference_annotation | A data.frame containing gene (or else) annotation with genomic coordinates. |

Value

A SingleCellExperiment object with annotated rowData.

Examples

```
scExp = create_scExp(create_scDataset_raw()$mat, create_scDataset_raw()$annot)
scExp = feature_annotation_scExp(scExp)
head(SummarizedExperiment::rowRanges(scExp))

# Mouse
scExp = create_scExp(create_scDataset_raw(ref="mm10")$mat,
  create_scDataset_raw(ref="mm10")$annot)
scExp = feature_annotation_scExp(scExp, ref="mm10")
head(SummarizedExperiment::rowRanges(scExp))
```

```
filter_correlated_cell_scExp
```

Filter lowly correlated cells

Description

Remove cells that have a correlation score lower than what would be expected by chance with other cells.

Usage

```
filter_correlated_cell_scExp(scExp, random_iter = 5,
  corr_threshold = 99, percent_correlation = 1,
  downsample = 2500, verbose = TRUE, n_process = 250,
  BPPARAM = BiocParallel::bpparam())
```

Arguments

| | |
|---------------------|---|
| scExp | A SingleCellExperiment object containing 'Cor', a correlation matrix, in reducedDims. |
| random_iter | Number of random matrices to create to calculate random correlation scores. (50) |
| corr_threshold | Quantile of random correlation score above which a cell is considered to be 'correlated' with another cell. (99) |
| percent_correlation | Percentage of the cells that any cell must be 'correlated' to in order to not be filtered. (1) |
| downsample | Number of cells to calculate correlation filtering threshold ? (2500) |
| verbose | Print messages ? (TRUE) |
| n_process | Number of cell to proceed at a time. Increase this number to increase speed at memory cost |
| BPPARAM | BPPARAM object for multiprocessing. See bpparam for more informations. Will take the default BPPARAM set in your R session. |

Details

This functions takes as input a SingleCellExperiment object that must have correlation matrix calculated and outputs a SingleCellExperiment object without lowly correlated cells. TSNE is recalculated.

Value

Returns a SingleCellExperiment object without lowly correlated cells. The calculated correlation score limit threshold is saved in metadata.

Examples

```
data("scExp")
dim(scExp)
scExp_cf = filter_correlated_cell_scExp(scExp,
  corr_threshold = 99, percent_correlation = 1)
dim(scExp_cf)
```

filter_genes_with_refined_peak_annotation

Filter genes based on peak calling refined annotation

Description

Filter genes based on peak calling refined annotation

Usage

```
filter_genes_with_refined_peak_annotation(
  refined_annotation,
  peak_distance,
  signific,
  over,
  under
)
```

Arguments

| | |
|--------------------|---|
| refined_annotation | A data.frame containing each gene distance to real peak |
| peak_distance | Minimum distance to an existing peak to accept a given gene |
| signific | Indexes of all significantly differential genes |
| over | Indexes of all significantly overexpressed genes |
| under | Indexes of all significantly underexpressed genes |

Value

List of significantly differential, overexpressed and underexpressed genes close enough to existing peaks

filter_scExp

Filter cells and features

Description

Function to filter out cells & features from SingleCellExperiment based on total count per cell, number of cells 'ON' in features and top covered cells that might be doublets.

Usage

```
filter_scExp(  
  scExp,  
  min_cov_cell = 1600,  
  quant_removal = 95,  
  min_count_per_feature = 10,  
  verbose = TRUE  
)
```

Arguments

| | |
|-----------------------|--|
| scExp | A SingleCellExperiment object. |
| min_cov_cell | Minimum counts for each cell. (1600) |
| quant_removal | Centile of cell counts above which cells are removed. (95) |
| min_count_per_feature | Minimum number of reads per feature (10). |
| verbose | (TRUE) |

Value

Returns a filtered SingleCellExperiment object.

Examples

```
scExp = create_scExp(create_scDataset_raw()$mat, create_scDataset_raw()$annot)  
scExp. = filter_scExp(scExp)  
  
# No feature filtering (all features are valuable)  
scExp. = filter_scExp(scExp, min_count_per_feature=30)  
  
# No cell filtering (all features are valuable)  
scExp. = filter_scExp(scExp, min_cov_cell=0, quant_removal=100)
```

| | |
|-------------------|-----------------------------------|
| find_top_features | <i>Find most covered features</i> |
|-------------------|-----------------------------------|

Description

Find the top most covered features that will be used for dimensionality reduction. Optionally remove non-top features.

Usage

```

find_top_features(
  scExp,
  n = 20000,
  keep_others = FALSE,
  prioritize_genes = FALSE,
  max_distanceToTSS = 10000,
  verbose = TRUE
)

```

Arguments

| | |
|-------------------|---|
| scExp | A SingleCellExperiment. |
| n | Either an integer indicating the number of top covered regions to find or a character vector of the top percentile of features to keep (e.g. 'q20' to keep top 20% features). |
| keep_others | Logical indicating if non-top regions are to be removed from the SCE or not (FALSE). |
| prioritize_genes | First filter by loci being close to genes ? E.g. for differential analysis, it is more relevant to keep features close to genes |
| max_distanceToTSS | If prioritize_genes is TRUE, the maximum distance to consider a feature close to a gene. |
| verbose | Print ? |

Value

A SCE with top features

Examples

```

data(scExp)
scExp_top = find_top_features(scExp, n = 4000, keep_others = FALSE)

```

| | |
|-------------------|--|
| generate_analysis | <i>Generate a complete ChromSCape analysis</i> |
|-------------------|--|

Description

Generate a complete ChromSCape analysis

Usage

```

generate_analysis(input_data_folder,
analysis_name = "Analysis_1",
output_directory = "./",
input_data_type = c("scBED", "DenseMatrix", "SparseMatrix",
                    "scBAM")[1],
feature_count_on = c("bins", "genebody", "peaks")[1],
feature_count_parameter = 50000,
ref_genome = c("hg38", "mm10")[1],
run = c("filter", "CNA", "cluster", "consensus", "peak_call", "coverage",
        "DA", "GSA", "report")[c(1,3,6,7,8,9)],
min_reads_per_cell = 1000,
max_quantile_read_per_cell = 99,
n_top_features = 40000,
norm_type = "CPM",
subsample_n = NULL,
exclude_regions = NULL,
n_clust = NULL,
corr_threshold = 99,
percent_correlation = 1,
maxK = 10,
qval.th = 0.1,
logFC.th = 1,
enrichment_qval = 0.1,
doBatchCorr = FALSE,
batch_sels = NULL,
control_samples_CNA = NULL,
genes_to_plot = c("Krt8", "Krt5", "Tgfb1", "Foxq1", "Cdkn2b",
                  "Cdkn2a", "chr7:15000000-20000000")
)

```

Arguments

input_data_folder Directory containing the input data.

analysis_name Name given to the analysis.

output_directory Directory where to create the analysis and the HTML report.

input_data_type The type of input data.

feature_count_on For raw data type, on which features to count the cells.

feature_count_parameter Additional parameter corresponding to the 'feature_count_on' parameter. E.g. for 'bins' must be a numeric, e.g. 50000, for 'peaks' must be a character containing path towards a BED peak file.

ref_genome The genome of reference.

| | |
|---|--|
| <code>run</code> | What steps to run. By default runs everything. Some steps are required in order to run downstream steps. |
| <code>min_reads_per_cell</code> | Minimum number of reads per cell. |
| <code>max_quantile_read_per_cell</code> | Upper quantile above which to consider cells doublets. |
| <code>n_top_features</code> | Number of features to keep in the analysis. |
| <code>norm_type</code> | Normalization type. |
| <code>subsample_n</code> | Number of cells per condition to downsample to, for performance principally. |
| <code>exclude_regions</code> | Path towards a BED file containing CNA to exclude from the analysis (optional). |
| <code>n_clust</code> | Number of clusters to force choice of clusters. |
| <code>corr_threshold</code> | Quantile of correlation above which two cells are considered as correlated. |
| <code>percent_correlation</code> | Percentage of the total cells that a cell must be correlated with in order to be kept in the analysis. |
| <code>maxK</code> | Upper cluster number to rest for ConsensusClusterPlus. |
| <code>qval.th</code> | Adjusted p-value below which to consider features differential. |
| <code>logFC.th</code> | Log2-fold-change above/below which to consider a feature depleted/enriched. |
| <code>enrichment_qval</code> | Adjusted p-value below which to consider a gene set as significantly enriched in differential features. |
| <code>doBatchCorr</code> | Logical indicating if batch correction using fastMNN should be run. |
| <code>batch_sels</code> | If <code>doBatchCorr</code> is TRUE, a named list containing the samples in each batch. |
| <code>control_samples_CNA</code> | If running CopyNumber Analysis, a character vector of the sample names that are 'normal'. |
| <code>genes_to_plot</code> | A character vector containing genes of interest of which to plot the coverage. |

Value

Creates a ChromSCape-readable directory and saved objects, as well as a multi-tabbed HTML report resuming the analysis.

Examples

```
## Not run:
generate_analysis("/path/to/data/", "Analysis_1")

## End(Not run)
```

generate_count_matrix *Generate count matrix*

Description

Generate count matrix

Usage

```
generate_count_matrix(cells, features, sparse, cell_names, feature_names)
```

Arguments

| | |
|---------------|--------------------|
| cells | Number of cells |
| features | Number of features |
| sparse | Is matrix sparse ? |
| cell_names | Cell names |
| feature_names | Feature names |

Value

A matrix or a sparse matrix

generate_coverage_tracks

Generate cell cluster pseudo-bulk coverage tracks

Description

Generate cell cluster pseudo-bulk coverage tracks. First, scBED files are concatenated into cell clusters contained in the 'cell_cluster' column of your SingleCellExperiment object. To do so, for each sample in the given list, the barcodes of each cluster are grepped and BED files are merged into pseudo-bulk of clusters (C1,C2...). Two cells from different can have the same barcode ID as cell affectation is done sample by sample. Then coverage of pseudo-bulk BED files is calculated by averaging & smoothing reads on small genomic window (150bp per default). The pseudo bulk BED and BigWigs coverage tracks are writtend to the output directory. This functionality is not available on Windows as it uses the 'cat' and 'gzip' utilities from Unix OS.

Usage

```
generate_coverage_tracks(
  scExp_cf,
  input_files_coverage,
  odir,
  ref_genome = c("hg38", "mm10")[1],
  bin_width = 150,
  n_smoothBin = 5,
  read_size = 101,
  progress = NULL
)
```

Arguments

| | |
|----------------------|---|
| scExp_cf | A SingleCellExperiment with cluster selected. (see choose_cluster_scExp). It is recommended having a minimum of ~100 cells per cluster in order to obtain smooth tracks. |
| input_files_coverage | A named list of character vector of path towards single-cell BED files. The names MUST correspond to the 'sample_id' column in your SingleCellExperiment object. The single-cell BED files names MUST match the barcode names in your SingleCellExperiment (column 'barcode'). The scBED files can be gzipped or not. |
| odir | The output directory to write the cumulative BED and BigWig files. |
| ref_genome | The genome of reference, used to constrain to canonical chromosomes. Either 'hg38' or 'mm10'. 'hg38' per default. |
| bin_width | The width of the bin to create the coverage track. The smaller the greater the resolution & runtime. Default to 150. |
| n_smoothBin | Number of bins left & right to average ('smooth') the signal on. Default to 5. |
| read_size | The estimated size of reads. Default to 101. |
| progress | A Progress object for Shiny. Default to NULL. |

Value

Generate coverage tracks (.bigwig) for each cluster in the SingleCellExperiment ("cell_cluster" column).

Examples

```
## Not run:
data(scExp)
input_files_coverage = list(
  "scChIP_Jurkat_K4me3" = paste0("/path/to/", scExp$barcode[1:51], ".bed"),
  "scChIP_Ramos_K4me3" = paste0("/path/to/", scExp$barcode[52:106], ".bed")
)
generate_coverage_tracks(scExp, input_files_coverage, "/path/to/output",
  ref_genome = "hg38")
```

```
## End(Not run)
```

```
generate_feature_names
    Generate feature names
```

Description

Generate feature names

Usage

```
generate_feature_names(featureType, ref, features)
```

Arguments

| | |
|-------------|--------------------------------|
| featureType | Type of feature |
| ref | Reference genome |
| features | Number of features to generate |

Value

A character vector of feature names

```
gene_set_enrichment_analysis_scExp
    Runs Gene Set Enrichment Analysis on genes associated with differential features
```

Description

This function takes previously calculated differential features and runs hypergeometric test to look for enriched gene sets in the genes associated with differential features, for each cell cluster. This functions takes as input a SingleCellExperiment object with consclust, the type of comparison, either 'one_vs_rest' or 'pairwise', the adjusted p-value threshold (qval.th) and the fold-change threshold (cdiff.th). It outputs a SingleCellExperiment object containing a differential list.

Usage

```

gene_set_enrichment_analysis_scExp(
  scExp,
  enrichment_qval = 0.1,
  ref = "hg38",
  GeneSets = NULL,
  GeneSetsDf = NULL,
  GenePool = NULL,
  qval.th = 0.01,
  cdiff.th = 1,
  peak_distance = 1000,
  use_peaks = FALSE,
  GeneSetClasses = c("c1_positional", "c2_curated", "c3_motif", "c4_computational",
    "c5_G0", "c6_oncogenic", "c7_immunologic", "hallmark"),
  progress = NULL
)

```

Arguments

| | |
|-----------------|---|
| scExp | A SingleCellExperiment object containing list of differential features. |
| enrichment_qval | Adjusted p-value threshold for gene set enrichment. (0.1) |
| ref | A reference annotation, either 'hg38' or 'mm10'. ('hg38') |
| GeneSets | A named list of gene sets. If NULL will automatically load MSigDB list of gene sets for specified reference genome. (NULL) |
| GeneSetsDf | A dataframe containing gene sets & class of gene sets. If NULL will automatically load MSigDB dataframe of gene sets for specified reference genome. (NULL) |
| GenePool | The pool of genes to run enrichment in. If NULL will automatically load Gen-code list of genes fro specified reference genome. (NULL) |
| qval.th | Adjusted p-value threshold to define differential features. (0.01) |
| cdiff.th | Fold change threshold to define differential features. (1) |
| peak_distance | Maximum distanceToTSS of feature to gene TSS to consider associated, in bp. (1000) |
| use_peaks | Use peak calling method (must be calculated beforehand). (FALSE) |
| GeneSetClasses | Which classes of MSIGdb to look for. |
| progress | A shiny Progress instance to display progress bar. |

Value

Returns a SingleCellExperiment object containing list of enriched Gene Sets for each cluster, either in depleted features, enriched features or simply differential features (both).

Examples

```
data("scExp")

#Usually recommending qual.th = 0.01 & cdiff.th = 1 or 2
## Not run: scExp_cf = gene_set_enrichment_analysis_scExp(scExp,
  qual.th = 0.4, cdiff.th = 0.3)
## End(Not run)
```

getExperimentNames *Get experiment names from a SingleCellExperiment*

Description

Get experiment names from a SingleCellExperiment

Usage

```
getExperimentNames(scExp)
```

Arguments

scExp A SingleCellExperiment with named mainExp and altExps.

Value

Character vector of unique experiment names

Examples

```
data(scExp)
getExperimentNames(scExp)
```

getMainExperiment *Get Main experiment of a SingleCellExperiment*

Description

Get Main experiment of a SingleCellExperiment

Usage

```
getMainExperiment(scExp)
```

Arguments

scExp A SingleCellExperiment with named mainExp and altExps.

Value

The swapped SingleCellExperiment towards "main" experiment

Examples

```
data(scExp)
getMainExperiment(scExp)
```

```
get_color_dataframe_from_input
```

Get color dataframe from shiny::colorInput

Description

Get color dataframe from shiny::colorInput

Usage

```
get_color_dataframe_from_input(  
  input,  
  levels_selected,  
  color_by = c("sample_id", "total_counts"),  
  input_id_prefix = "color_"  
)
```

Arguments

input Shiny input object
levels_selected Names of the features
color_by Which feature color to retrieve
input_id_prefix Prefix in front of the feature names

Value

A data.frame with the feature levels and the colors of each level of this feature.

get_cyto_features *Map features onto cytobands*

Description

Map the features of a SingleCellExperiment onto the cytobands of a given genome. Some features might not be mapped to any cytobands (e.g. if they are not in the canonical chromosomes), and are removed from the returned object.

Usage

```
get_cyto_features(scExp, ref_genome = c("hg38", "mm10")[1])
```

Arguments

scExp A SingleCellExperiment with genomic coordinate as features (peaks or bins)
ref_genome Reference genome ('hg38' or 'mm10')

Details

The cytobands are an arbitrary cutting of the genome that dates back to staining metaphase chromosomes with Giemsa.

Value

A data.frame of the SCE features with their corresponding cytoband name

Examples

```
data("scExp")  
matching_cyto = get_cyto_features(scExp, ref_genome="hg38")
```

get_genomic_coordinates *Get SingleCellExperiment's genomic coordinates*

Description

Get SingleCellExperiment's genomic coordinates

Usage

```
get_genomic_coordinates(scExp)
```

Arguments

scExp A SingleCellExperiment object.

Value

A GRanges object of genomic coordinates.

Examples

```
scExp = create_scExp(create_scDataset_raw()$mat, create_scDataset_raw()$annot)
feature_GRanges = get_genomic_coordinates(scExp)
```

get_most_variable_cyto

Retrieve the cytobands with the most variable fraction of reads

Description

Given a SingleCellExperiment object with the slot "cytoBand" containing the fraction of reads in each cytoband, calculates the variance of each cytoband and returns a data.frame with the top variables cytobands. Most cytobands are expected to be unchanged between normal and tumor samples, therefore focusing on the top variable cytobands enable to focus on the most interesting regions.

Usage

```
get_most_variable_cyto(scExp, top = 50)
```

Arguments

scExp A SingleCellExperiment with "cytoBand" reducedDim slot filled.
top Number of cytobands to return (50).

Value

A data.frame of the top variable cytoBands and their variance

Examples

```
data("scExp")
scExp = calculate_cyto_mat(scExp, ref_genome="hg38")
get_most_variable_cyto(scExp, top=50)
```

gg_fill_hue

gg_fill_hue

Description

gg_fill_hue

Usage

gg_fill_hue(n)

Arguments

n num hues

Value

A color in HEX format

groupMat

groupMat

Description

groupMat

Usage

groupMat(mat = NA, margin = 1, groups = NA, method = "mean")

Arguments

mat A matrix
margin By row or columns ?
groups Groups
method Method to group

Value

A grouped matrix

| | |
|--------------|---------------------|
| H1proportion | <i>H1proportion</i> |
|--------------|---------------------|

Description

H1proportion

Usage

```
H1proportion(pv = NA, lambda = 0.5)
```

Arguments

| | |
|--------|----------------|
| pv | P.value vector |
| lambda | Lambda value |

Value

H1 proportion value

has_genomic_coordinates

Does SingleCellExperiment has genomic coordinates in features ?

Description

Does SingleCellExperiment has genomic coordinates in features ?

Usage

```
has_genomic_coordinates(scExp)
```

Arguments

| | |
|-------|-------------------------------|
| scExp | A SingleCellExperiment object |
|-------|-------------------------------|

Value

TRUE or FALSE

Examples

```
scExp = create_scExp(create_scDataset_raw()$mat, create_scDataset_raw()$annot)
has_genomic_coordinates(scExp)
scExp_gene = create_scExp(create_scDataset_raw(featureType="gene")$mat,
  create_scDataset_raw(featureType="gene")$annot)
has_genomic_coordinates(scExp_gene)
```

`hclustAnnotHeatmapPlot`*hclustAnnotHeatmapPlot*

Description`hclustAnnotHeatmapPlot`**Usage**

```
hclustAnnotHeatmapPlot(  
  x = NULL,  
  hc = NULL,  
  hmColors = NULL,  
  anocol = NULL,  
  xpos = c(0.1, 0.9, 0.114, 0.885),  
  ypos = c(0.1, 0.5, 0.5, 0.6, 0.62, 0.95),  
  dendro.cex = 1,  
  xlab.cex = 0.8,  
  hmRowNames = FALSE,  
  hmRowNames.cex = 0.5  
)
```

Arguments

| | |
|-----------------------------|----------------------|
| <code>x</code> | A correlation matrix |
| <code>hc</code> | An hclust object |
| <code>hmColors</code> | A color palette |
| <code>anocol</code> | A matrix of colors |
| <code>xpos</code> | Xpos |
| <code>ypos</code> | Ypos |
| <code>dendro.cex</code> | Size of denro names |
| <code>xlab.cex</code> | Size of x label |
| <code>hmRowNames</code> | Write rownames ? |
| <code>hmRowNames.cex</code> | Size of rownames ? |

Value

A heatmap

hg38.chromosomes *Data.frame of chromosome length - hg38*

Description

This data frame provides the length of each "canonical" chromosomes of Homo Sapiens genome build hg38.

Usage

```
data("hg38.chromosomes")
```

Format

hg38.chromosomes - a data frame with 24 rows and 3 variables:

chr Chromosome - character

start Start of the chromosome (bp) - integer

end End of the chromosome (bp) - integer

hg38.cytoBand *Data.frame of cytoBandlocation - hg38*

Description

This data frame provides the location of each cytoBands of Homo Sapiens genome build hg38.

Usage

```
data("hg38.cytoBand")
```

Format

hg38.cytoBand - a data frame with 862 rows and 4 variables:

chr Chromosome - character

start Start of the chromosome (bp) - integer

end End of the chromosome (bp) - integer

cytoBand Name of the cytoBand - character

| | |
|--------------|--------------------------------------|
| hg38.GeneTSS | <i>Data.frame of gene TSS - hg38</i> |
|--------------|--------------------------------------|

Description

This dataframe was extracted from Gencode v25 and report the Transcription Start Site of each gene in the Homo Sapiens genome build hg38.

Usage

```
data("hg38.GeneTSS")
```

Format

hg38.GeneTSS - a data frame with 24 rows and 3 variables:

chr Chromosome - character
start Start of the gene (TSS) - integer
end End of the gene - integer
gene Gene symbol - character

| | |
|----------|-----------------|
| imageCol | <i>imageCol</i> |
|----------|-----------------|

Description

imageCol

Usage

```
imageCol(  
  matcol = NULL,  
  strat = NULL,  
  xlab.cex = 0.5,  
  ylab.cex = 0.5,  
  drawLines = c("none", "h", "v", "b")[1],  
  ...  
)
```

Arguments

| | |
|-----------|-----------------------|
| matcol | A matrix of colors |
| strat | Strat |
| xlab.cex | X label size |
| ylab.cex | Y label size |
| drawLines | Draw lines ? |
| ... | Additional parameters |

Value

A rectangular image

import_count_input_files

Import and count input files depending on their format

Description

Import and count input files depending on their format

Usage

```
import_count_input_files(  
  files_dir_list,  
  file_type,  
  which,  
  ref,  
  verbose,  
  progress,  
  BPPARAM = BiocParallel::bpparam()  
)
```

Arguments

| | |
|----------------|---|
| files_dir_list | A named list of directories containing the input files. |
| file_type | Input file type. |
| which | A GRanges object of features. |
| ref | Reference genome. |
| verbose | Print ? |
| progress | A progress object for Shiny. |
| BPPARAM | BPPARAM object for multiprocessing. See bpparam for more informations. Will take the default BPPARAM set in your R session. |

Value

A list with the feature indexes data.frame containing non-zeroes entries in the count matrix and the cell names

| | |
|--------------|--|
| import_scExp | <i>Read single-cell matrix(ces) into scExp</i> |
|--------------|--|

Description

Combine one or multiple matrices together to create a sparse matrix and cell annotation data.frame.

Usage

```
import_scExp(file_paths, remove_pattern = "", temp_path = NULL)
```

Arguments

| | |
|----------------|---|
| file_paths | A character vector of file names towards single cell epigenomic matrices (features x cells) (must be .txt / .tsv) |
| remove_pattern | A string pattern to remove from the sample names. Can be a regexp. |
| temp_path | In case matrices are stored in temporary folder, a character vector of path towards temporary files. (NULL) |

Value

A list containing:

- datamatrix: a sparseMatrix of features x cells
- annot_raw: an annotation of cells as data.frame

Examples

```
mat1 = mat2 = create_scDataset_raw()$mat
tmp1 = tempfile(fileext = ".tsv")
tmp2 = tempfile(fileext = ".tsv")
write.table(as.matrix(mat1),file=tmp1,sep = "\t",
row.names = TRUE,col.names = TRUE,quote = FALSE)
write.table(as.matrix(mat2),file=tmp2, sep = "\t",
row.names = TRUE,col.names = TRUE,quote = FALSE)
file_paths = c(tmp1,tmp2)
out = import_scExp(file_paths)
```

```
index_peaks_barcode_to_matrix_indexes
```

Read index-peaks-barcode trio files on interval to create count indexes

Description

Read index-peaks-barcode trio files on interval to create count indexes

Usage

```
index_peaks_barcode_to_matrix_indexes(
  feature_file,
  matrix_file,
  barcode_file,
  binarize = FALSE
)
```

Arguments

| | |
|--------------|--|
| feature_file | A file containing the features genomic locations |
| matrix_file | A file containing the indexes of non-zeroes values and their value (respectively i,j,x,see sparseMatrix) |
| barcode_file | A file containing the barcode ids |
| binarize | Binarize matrix ? |

Value

A list containing a "feature index" data.frame, name_cells, and a region GenomicRange object used to form the sparse matrix

```
inter_correlation_scExp
```

Calculate inter correlation between cluster or samples

Description

Calculate inter correlation between cluster or samples

Usage

```
inter_correlation_scExp(
  scExp_cf,
  by = c("sample_id", "cell_cluster")[1],
  reference_group = unique(scExp_cf[[by]][1]),
  other_groups = unique(scExp_cf[[by]])
)
```

Arguments

| | |
|-----------------|--|
| scExp_cf | A SingleCellExperiment |
| by | On which feature to calculate correlation ("sample_id" or "cell_cluster") |
| reference_group | Reference group to calculate correlation with. Must be in accordance with "by". |
| other_groups | Groups on which to calculate correlation (can contain multiple groups, and also reference_group). Must be in accordance with "by". |

Value

A data.frame of average inter-correlation of cells in other_groups with cells in reference_group

Examples

```
data(scExp)
inter_correlation_scExp(scExp)
```

```
intra_correlation_scExp
```

Calculate intra correlation between cluster or samples

Description

Calculate intra correlation between cluster or samples

Usage

```
intra_correlation_scExp(scExp_cf, by = c("sample_id", "cell_cluster")[1])
```

Arguments

| | |
|----------|---|
| scExp_cf | A SingleCellExperiment |
| by | On which feature to calculate correlation ("sample_id" or "cell_cluster") |

Value

A data.frame of cell average intra-correlation

Examples

```
data(scExp)
intra_correlation_scExp(scExp, by = "sample_id")
intra_correlation_scExp(scExp, by = "cell_cluster")
```

| | |
|-----------|--------------------------|
| launchApp | <i>Launch ChromSCape</i> |
|-----------|--------------------------|

Description

Main function to launch ChromSCape in your favorite browser. You can pass additional parameters that you would pass to shiny::runApp ([runApp](#))

Usage

```
launchApp(launch.browser = TRUE, ...)
```

Arguments

| | |
|----------------|--|
| launch.browser | Wether to launch browser or not |
| ... | Additional parameters passed to runApp |

Value

Launches the shiny application

Examples

```
## Not run:
launchApp()

## End(Not run)
```

| | |
|-------------|---|
| load_MSIGdb | <i>Load and format MSIGdb pathways using msigdb package</i> |
|-------------|---|

Description

Load and format MSIGdb pathways using msigdb package

Usage

```
load_MSIGdb(ref, GeneSetClasses)
```

Arguments

| | |
|----------------|---------------------------------------|
| ref | Reference genome, either mm10 or hg38 |
| GeneSetClasses | Which classes of MSIGdb to load |

Value

A list containing the GeneSet (list), GeneSetDf (data.frame) and GenePool character vector of all possible genes

| | |
|-------------------|--|
| merge_MACS2_peaks | <i>Merge peak files from MACS2 peak caller</i> |
|-------------------|--|

Description

Merge peak files from MACS2 peak caller

Usage

```
merge_MACS2_peaks(odir, class, peak_distance_to_merge, ref)
```

Arguments

| | |
|------------------------|-------------------------------------|
| odir | Output directory |
| class | Cell cluster |
| peak_distance_to_merge | Maximum distance to merge two peaks |
| ref | Reference genome |

Value

Peaks as GRanges

| | |
|------------------|---|
| mm10.chromosomes | <i>Data.frame of chromosome length - mm10</i> |
|------------------|---|

Description

This data frame provides the length of each "canonical" chromosomes of Mus Musculus (Mouse) genome build mm10.

Usage

```
data("mm10.chromosomes")
```

Format

mm10.chromosomes - a data frame with 24 rows and 3 variables:

chr Chromosome - character
start Start of the chromosome (bp) - integer
end End of the chromosome (bp) - integer

| | |
|---------------|--|
| mm10.cytoBand | <i>Data.frame of cytoBandlocation - mm10</i> |
|---------------|--|

Description

This data frame provides the location of each cytoBands of Homo Sapiens genome build mm10.

Usage

```
data("mm10.cytoBand")
```

Format

mm10.cytoBand - a data frame with 862 rows and 4 variables:

chr Chromosome - character
start Start of the chromosome (bp) - integer
end End of the chromosome (bp) - integer
cytoBand Name of the cytoBand - character

| | |
|--------------|--------------------------------------|
| mm10.GeneTSS | <i>Data.frame of gene TSS - mm10</i> |
|--------------|--------------------------------------|

Description

This dataframe was extracted from Gencode v25 and report the Transcription Start Site of each gene in the Mus Musculus genome build mm10 (Mouse).

Usage

```
data("mm10.GeneTSS")
```

Format

mm10.GeneTSS - a data frame with 24 rows and 3 variables:

chr Chromosome name - character
start Start of the gene (TSS) - integer
end End of the gene - integer
gene Gene symbol - character

| | |
|-----------------|-------------------------|
| normalize_scExp | <i>Normalize counts</i> |
|-----------------|-------------------------|

Description

Normalize counts

Usage

```
normalize_scExp(
  scExp,
  type = c("CPM", "TFIDF", "RPKM", "TPM", "feature_size_only")
)
```

Arguments

| | |
|-------|--|
| scExp | A SingleCellExperiment object. |
| type | Which normalization to apply. Either 'CPM', 'TFIDF', 'RPKM', 'TPM' or 'feature_size_only'. Note that for all normalization by size (RPKM, TPM, feature_size_only), the features must have defined genomic coordinates. |

Value

A SingleCellExperiment object containing normalized counts. (See ?normcounts())

Examples

```
scExp = create_scExp(create_scDataset_raw())$mat, create_scDataset_raw())$annot)
scExp = normalize_scExp(scExp)
head(SingleCellExperiment::normcounts(scExp))
```

| | |
|-------------------------------|---|
| num_cell_after_cor_filt_scExp | <i>Number of cells before & after correlation filtering</i> |
|-------------------------------|---|

Description

Number of cells before & after correlation filtering

Usage

```
num_cell_after_cor_filt_scExp(scExp, scExp_cf)
```

Arguments

scExp SingleCellExperiment object before correlation filtering.
 scExp_cf SingleCellExperiment object after correlation filtering.

Value

A colored kable with the number of cells per sample before and after filtering for display

Examples

```
data("scExp")
scExp_cf = correlation_and_hierarchical_clust_scExp(scExp)
scExp_cf = filter_correlated_cell_scExp(scExp_cf,
  corr_threshold = 99, percent_correlation = 1)
## Not run: num_cell_after_cor_filt_scExp(scExp, scExp_cf)
```

```
num_cell_after_QC_filt_scExp
```

Table of cells before / after QC

Description

Table of cells before / after QC

Usage

```
num_cell_after_QC_filt_scExp(scExp, annot, datamatrix)
```

Arguments

scExp A SingleCellExperiment object.
 annot A raw annotation data.frame of cells before filtering.
 datamatrix A matrix of cells per regions before filtering.

Value

A formatted kable in HTML.

Examples

```
scExp = create_scExp(create_scDataset_raw())$mat, create_scDataset_raw())$annot)
scExp_filtered = filter_scExp(scExp)
## Not run: num_cell_after_QC_filt_scExp(
  scExp_filtered, SingleCellExperiment::colData(scExp))
## End(Not run)
```

`num_cell_before_cor_filt_scExp`*Table of number of cells before correlation filtering*

Description

Table of number of cells before correlation filtering

Usage

```
num_cell_before_cor_filt_scExp(scExp)
```

Arguments

scExp A SingleCellExperiment Object

Value

A colored kable with the number of cells per sample for display

Examples

```
data("scExp")  
## Not run: num_cell_before_cor_filt_scExp(scExp)
```

`num_cell_in_cluster_scExp`*Number of cells in each cluster*

Description

Number of cells in each cluster

Usage

```
num_cell_in_cluster_scExp(scExp)
```

Arguments

scExp A SingleCellExperiment object containing chromatin groups.

Value

A formatted kable of cell assignation to each cluster.

Examples

```
data("scExp")
scExp_cf = correlation_and_hierarchical_clust_scExp(scExp)
scExp_cf = choose_cluster_scExp(scExp_cf, nclust=3, consensus=FALSE)
## Not run: num_cell_in_cluster_scExp(scExp_cf)
```

| | |
|----------------|-----------------------|
| num_cell_scExp | <i>Table of cells</i> |
|----------------|-----------------------|

Description

Table of cells

Usage

```
num_cell_scExp(annot, datamatrix)
```

Arguments

| | |
|------------|---|
| annot | An annotation of cells. Can be obtain through 'colData(scExp)'. |
| datamatrix | A matrix of cells per regions before filtering. |

Value

A formatted kable in HTML.

Examples

```
scExp = create_scExp(create_scDataset_raw()$mat, create_scDataset_raw()$annot)
## Not run: num_cell_scExp(SingleCellExperiment::colData(scExp))
```

| | |
|----------------------------|---------------------------------------|
| pca_irlba_for_sparseMatrix | <i>Run sparse PCA using irlba SVD</i> |
|----------------------------|---------------------------------------|

Description

This function allows to run a PCA using IRLBA Singular Value Decomposition in a fast & memory efficient way. The incremental Lanczos bidiagonalisation algorithm allows to keep the matrix sparse as the "loci" centering is implicit. The function then multiplies by the approximate singular values (svd\$d) in order to get more importance to the first PCs proportionally to their singular values. This step is crucial for downstream approaches, e.g. UMAP or T-SNE.

Usage

```
pca_irlba_for_sparseMatrix(x, n_comp, work = 3 * n_comp)
```

Arguments

| | |
|--------|---|
| x | A sparse normalized matrix (features x cells) |
| n_comp | The number of principal components to keep |
| work | Working subspace dimension, larger values can speed convergence at the cost of more memory use. |

Value

The rotated data, e.g. the cells x PC column in case of sc data.

| | |
|---------------|--|
| peaks_to_bins | <i>Transforms a peaks x cells count matrix into a bins x cells count matrix.</i> |
|---------------|--|

Description

This function is best used to re-count large number of small peaks (e.g. ≤ 5000 bp) into equal or larger bins. The genome is either cut in fixed bins (e.g. 50,000bp) or into an user defined number of bins. Bins are calculated based on the canonical chromosomes. Note that if peaks are larger than bins, or if peaks are overlapping multiple bins, the signal is added to each bin. Users can increase the minimum overlap to consider peaks overlapping bins (by default 150bp, size of a nucleosome) to diminish the number of peaks overlapping multiple region. Any peak smaller than the minimum overlap threshold will be dismissed. Therefore, library size might be slightly different from peaks to bins if signal was duplicated into multiple bins or omitted due to peaks smaller than minimum overlap.

Usage

```
peaks_to_bins(
  mat,
  bin_width = 50000,
  n_bins = NULL,
  minoverlap = 150,
  verbose = TRUE,
  ref = "hg38"
)
```

Arguments

| | |
|-----------|--|
| mat | A matrix of peaks x cells |
| bin_width | width of bins to produce in base pairs (minimum 500) (50000) |
| n_bins | number of bins (exclusive with bin_width) |

| | |
|-------------------------|---|
| <code>minoverlap</code> | Minimum overlap between a peak and a bin to consider the peak as overlapping the bin (150). |
| <code>verbose</code> | Verbose |
| <code>ref</code> | reference genome to use (hg38) |

Value

A sparse matrix of bins instead of peaks

Examples

```
mat = create_scDataset_raw()$mat
binned_mat = peaks_to_bins(mat, bin_width = 10e6)
dim(binned_mat)
```

`plot_cluster_consensus_scExp`
Plot cluster consensus

Description

Plot cluster consensus score for each k as a bargraph.

Usage

```
plot_cluster_consensus_scExp(scExp)
```

Arguments

`scExp` A SingleCellExperiment

Value

The consensus score for each cluster for each k as a barplot

Examples

```
data("scExp")
plot_cluster_consensus_scExp(scExp)
```

plot_coverage_BigWig *Coverage plot using Sushi*

Description

Coverage plot using Sushi

Usage

```
plot_coverage_BigWig(  
  coverages,  
  label_color_list,  
  peaks = NULL,  
  chrom,  
  start,  
  end,  
  ref = "hg38"  
)
```

Arguments

| | |
|------------------|---|
| coverages | A list containing sample coverage as GenomicRanges |
| label_color_list | List of colors, list names are labels |
| peaks | A GRanges object containing peaks location to plot (optional) |
| chrom | Chromosome |
| start | Start |
| end | End |
| ref | Genomic Reference |

Value

A coverage plot annotated with genes

Examples

```
data(scExp)  
plot_intra_correlation_scExp(scExp)
```

plot_differential_H1_scExp

Differential H1 distribution plot

Description

Differential H1 distribution plot

Usage

```
plot_differential_H1_scExp(scExp_cf, cell_cluster = "C1")
```

Arguments

scExp_cf A SingleCellExperiment object
cell_cluster Which cluster to plot

Value

A barplot of H1 distribution

Examples

```
data("scExp")  
plot_differential_H1_scExp(scExp)
```

plot_differential_summary_scExp

Differential summary barplot

Description

Differential summary barplot

Usage

```
plot_differential_summary_scExp(scExp_cf)
```

Arguments

scExp_cf A SingleCellExperiment object

Value

A barplot summary of differential analysis

Examples

```
data("scExp")
plot_differential_summary_scExp(scExp)
```

```
plot_differential_volcano_scExp
      Volcano plot of differential features
```

Description

Volcano plot of differential features

Usage

```
plot_differential_volcano_scExp(
  scExp_cf,
  cell_cluster = "C1",
  cdiff.th = 1,
  qval.th = 0.01
)
```

Arguments

| | |
|---------------------------|-------------------------------|
| <code>scExp_cf</code> | A SingleCellExperiment object |
| <code>cell_cluster</code> | Which cluster to plot |
| <code>cdiff.th</code> | Fold change threshold |
| <code>qval.th</code> | Adjusted p.value threshold |

Value

A volcano plot of differential analysis of a specific cluster

Examples

```
data("scExp")
plot_differential_volcano_scExp(scExp, "C1")
```

 plot_distribution_scExp

Plotting distribution of signal

Description

Plotting distribution of signal

Usage

```
plot_distribution_scExp(
  scExp,
  raw = TRUE,
  log10 = FALSE,
  pseudo_counts = 1,
  bins = 150
)
```

Arguments

| | |
|---------------|------------------------------------|
| scExp | A SingleCellExperiment Object |
| raw | Use raw counts ? |
| log10 | Transform using log10 ? |
| pseudo_counts | Pseudo-count to add if using log10 |
| bins | Number of bins in the histogram |

Value

A ggplot histogram representing the distribution of count per cell

Examples

```
data("scExp")
plot_distribution_scExp(scExp)
```

 plot_gain_or_loss_barplots

Plot Gain or Loss of cytobands of the most variables cytobands

Description

Plot Gain or Loss of cytobands of the most variables cytobands

Plot Gain or Loss of cytobands of the most variables cytobands

Usage

```
plot_gain_or_loss_barplots(scExp, cells = NULL, top = 20)
```

```
plot_gain_or_loss_barplots(scExp, cells = NULL, top = 20)
```

Arguments

| | |
|-------|--|
| scExp | A SingleCellExperiment with "logRatio_cytoBand" reducedDim slot filled. See calculate_logRatio_CNA |
| cells | Cell IDs of the tumor samples to |
| top | Number of most variables cytobands to plot |

Value

Plot the gains/lost in the selected cells of interest as multiple barplots

Plot the gains/lost in the selected cells of interest as multiple barplots

Examples

```
data("scExp")
scExp = calculate_CNA(scExp, control_samples = unique(scExp$sample_id)[1],
  ref_genome="hg38", quantiles_to_define_gol = c(0.05,0.95))
plot_gain_or_loss_barplots(scExp, cells = scExp$cell_id[which(
  scExp$sample_id %in% unique(scExp$sample_id)[2])])
```

```
data("scExp")
scExp = calculate_CNA(scExp, control_samples = unique(scExp$sample_id)[1],
  ref_genome="hg38", quantiles_to_define_gol = c(0.05,0.95))
plot_gain_or_loss_barplots(scExp, cells = scExp$cell_id[which(
  scExp$sample_id %in% unique(scExp$sample_id)[2])])
```

| | |
|--------------------|---|
| plot_heatmap_scExp | <i>Plot cell correlation heatmap with annotations</i> |
|--------------------|---|

Description

Plot cell correlation heatmap with annotations

Usage

```
plot_heatmap_scExp(
  scExp,
  name_hc = "hc_cor",
  corColors = (grDevices::colorRampPalette(c("royalblue", "white", "indianred1")))(256),
  color_by = NULL,
```

```

    downsample = 1000,
    hc_linkage = "ward.D"
  )

```

Arguments

| | |
|------------|--|
| scExp | A SingleCellExperiment Object |
| name_hc | Name of the hclust contained in the SingleCellExperiment object |
| corColors | A palette of colors for the heatmap |
| color_by | Which features to add as additional bands on top of plot |
| downsample | Number of cells to downsample |
| hc_linkage | A linkage method for hierarchical clustering. See cor . ('ward.D') |

Value

A heatmap of cell to cell correlation, grouping cells by hierarchical clustering.

Examples

```

data("scExp")
plot_heatmap_scExp(scExp)

```

plot_inter_correlation_scExp

Violin plot of inter-correlation distribution between one or multiple groups and one reference group

Description

Violin plot of inter-correlation distribution between one or multiple groups and one reference group

Usage

```

plot_inter_correlation_scExp(
  scExp_cf,
  by = c("sample_id", "cell_cluster")[1],
  jitter_by = NULL,
  reference_group = unique(scExp_cf[[by]][1]),
  other_groups = unique(scExp_cf[[by]]),
  downsample = 5000
)

```

Arguments

| | |
|-----------------|---|
| scExp_cf | A SingleCellExperiment |
| by | Color by sample_id or cell_cluster |
| jitter_by | Add jitter points of another layer (cell_cluster or sample_id) |
| reference_group | Character containing the reference group name to calculate correlation from. |
| other_groups | Character vector of the other groups for which to calculate correlation with the reference group. |
| downsample | Downsample for plotting |

Value

A violin plot of inter-correlation

Examples

```
data(scExp)
plot_intra_correlation_scExp(scExp)
```

```
plot_intra_correlation_scExp
```

Violin plot of intra-correlation distribution

Description

Violin plot of intra-correlation distribution

Usage

```
plot_intra_correlation_scExp(
  scExp_cf,
  by = c("sample_id", "cell_cluster")[1],
  jitter_by = NULL,
  downsample = 5000
)
```

Arguments

| | |
|------------|--|
| scExp_cf | A SingleCellExperiment |
| by | Color by sample_id or cell_cluster |
| jitter_by | Add jitter points of another layer (cell_cluster or sample_id) |
| downsample | Downsample for plotting |

Value

A violin plot of intra-correlation

Examples

```
data(scExp)
plot_intra_correlation_scExp(scExp)
```

```
plot_most_contributing_features
    Plot Top/Bottom most contributing features to PCA
```

Description

Plot Top/Bottom most contributing features to PCA

Usage

```
plot_most_contributing_features(
  scExp,
  component = "Component_1",
  n_top_bot = 10
)
```

Arguments

| | |
|-----------|---|
| scExp | A SingleCellExperiment containing "PCA" in reducedDims and gene annotation in rowRanges |
| component | The name of the component of interest |
| n_top_bot | An integer number of top and bot regions to plot |

Details

If a gene TSS is within 10,000bp of the region, the name of the gene(s) will be displayed instead of the region

Value

A barplot of top and bottom features with the largest absolute value in the component of interest

Examples

```
data(scExp)
plot_most_contributing_features(scExp, component = "Component_1")
```

`plot_pie_most_contributing_chr`

Pie chart of top contribution of chromosomes in the 100 most contributing features to PCA #'

Description

Pie chart of top contribution of chromosomes in the 100 most contributing features to PCA #'

Usage

```
plot_pie_most_contributing_chr(  
  scExp,  
  component = "Component_1",  
  n_top_bot = 100  
)
```

Arguments

| | |
|-----------|---|
| scExp | A SingleCellExperiment containing "PCA" in reducedDims and gene annotation in rowRanges |
| component | The name of the component of interest |
| n_top_bot | An integer number of top and bot regions to plot (100) |

Value

A pie chart showing the distribution of chromosomes in the top features with the largest absolute value in the component of interest

Examples

```
data(scExp)  
plot_pie_most_contributing_chr(scExp, component = "Component_1")
```

`plot_reduced_dim_scExp`

Plot reduced dimensions (PCA, TSNE, UMAP)

Description

Plot reduced dimensions (PCA, TSNE, UMAP)

Usage

```
plot_reduced_dim_scExp(
  scExp,
  color_by = "sample_id",
  reduced_dim = c("PCA", "TSNE", "UMAP"),
  select_x = NULL,
  select_y = NULL,
  downsample = 5000,
  transparency = 0.6,
  size = 1,
  max_distanceToTSS = 1000,
  annotate_clusters = "cell_cluster" %in% colnames(colData(scExp)),
  min_quantile = 0.01,
  max_quantile = 0.99
)
```

Arguments

| | |
|-------------------|---|
| scExp | A SingleCellExperiment Object |
| color_by | Character of feature used for coloration. Can be cell metadata ('total_counts', 'sample_id', ...) or a gene name. |
| reduced_dim | Reduced Dimension used for plotting |
| select_x | Which variable to select for x axis |
| select_y | Which variable to select for y axis |
| downsample | Number of cells to downsample |
| transparency | Alpha parameter, between 0 and 1 |
| size | Size of the points. |
| max_distanceToTSS | The maximum distance to TSS to consider a gene linked to a region. Used only if "color_by" is a gene name. |
| annotate_clusters | A logical indicating if clusters should be labelled. The 'cell_cluster' column should be present in metadata. |
| min_quantile | The lower threshold to remove outlier cells, as quantile of cell embeddings (between 0 and 0.5). |
| max_quantile | The upper threshold to remove outlier cells, as quantile of cell embeddings (between 0.5 and 1). |

Value

A ggplot geom_point plot of reduced dimension 2D representation

Examples

```
data("scExp")
plot_reduced_dim_scExp(scExp, color_by = "sample_id")
plot_reduced_dim_scExp(scExp, color_by = "total_counts")
plot_reduced_dim_scExp(scExp, reduced_dim = "UMAP")
plot_reduced_dim_scExp(scExp, color_by = "CD52", reduced_dim = "UMAP")
```

plot_reduced_dim_scExp_CNA

Plot UMAP colored by Gain or Loss of cytobands

Description

Plot UMAP colored by Gain or Loss of cytobands

Usage

```
plot_reduced_dim_scExp_CNA(scExp, cytoBand)
```

Arguments

| | |
|----------|--|
| scExp | A SingleCellExperiment with "gainOrLoss_cytoBand" reducedDim slot filled. See calculate_gain_or_loss |
| cytoBand | Which cytoBand to color cells by |

Value

Plot the gains/lost of the cytoband overlayed on the epigenetic UMAP.

Examples

```
data("scExp")
scExp = calculate_CNA(scExp, control_samples = unique(scExp$sample_id)[1],
ref_genome="hg38", quantiles_to_define_gol = c(0.05,0.95))
plot_reduced_dim_scExp_CNA(scExp, get_most_variable_cyto(scExp)$cytoBand[1])
```

preprocess_CPM *Preprocess scExp - Counts Per Million (CPM)*

Description

Preprocess scExp - Counts Per Million (CPM)

Usage

```
preprocess_CPM(scExp)
```

Arguments

scExp A SingleCellExperiment Object

Value

A SingleCellExperiment object.

Examples

```
scExp = create_scExp(create_scDataset_raw())$mat, create_scDataset_raw())$annot)
scExp = preprocess_CPM(scExp)
head(SingleCellExperiment::normcounts(scExp))
```

preprocess_feature_size_only
 Preprocess scExp - size only

Description

Preprocess scExp - size only

Usage

```
preprocess_feature_size_only(scExp)
```

Arguments

scExp A SingleCellExperiment Object

Value

A SingleCellExperiment object.

Examples

```
scExp = create_scExp(create_scDataset_raw())$mat, create_scDataset_raw())$annot)
scExp = preprocess_feature_size_only(scExp)
head(SingleCellExperiment::normcounts(scExp))
```

```
preprocess_RPKM          Preprocess scExp - Read per Kilobase Per Million (RPKM)
```

Description

Preprocess scExp - Read per Kilobase Per Million (RPKM)

Usage

```
preprocess_RPKM(scExp)
```

Arguments

scExp A SingleCellExperiment Object

Value

A SingleCellExperiment object.

Examples

```
scExp = create_scExp(create_scDataset_raw())$mat, create_scDataset_raw())$annot)
scExp = preprocess_RPKM(scExp)
head(SingleCellExperiment::normcounts(scExp))
```

```
preprocess_TFIDF       Preprocess scExp - TF-IDF
```

Description

Preprocess scExp - TF-IDF

Usage

```
preprocess_TFIDF(scExp, scale = 10000, log = TRUE)
```

Arguments

scExp A SingleCellExperiment Object

scale A numeric to multiply the matrix in order to have human readable numbers.
Has no impact on the downstream analysis

log Wether to use neperian log on the TF-IDF normalized data or not.

Value

A SingleCellExperiment object.

Examples

```
scExp = create_scExp(create_scDataset_raw())$mat, create_scDataset_raw())$annot)
scExp = preprocess_TFIDF(scExp)
head(SingleCellExperiment::normcounts(scExp))
```

preprocess_TPM

Preprocess scExp - Transcripts per Million (TPM)

Description

Preprocess scExp - Transcripts per Million (TPM)

Usage

```
preprocess_TPM(scExp)
```

Arguments

scExp A SingleCellExperiment Object

Value

A SingleCellExperiment object.

Examples

```
scExp = create_scExp(create_scDataset_raw())$mat, create_scDataset_raw())$annot)
scExp = preprocess_TPM(scExp)
head(SingleCellExperiment::normcounts(scExp))
```

| | |
|------------------|---|
| rawfile_ToBigWig | <i>rawfile_ToBigWig : reads in BAM file and write out BigWig coverage file, normalized and smoothed</i> |
|------------------|---|

Description

rawfile_ToBigWig : reads in BAM file and write out BigWig coverage file, normalized and smoothed

Usage

```
rawfile_ToBigWig(
  filename,
  BigWig_filename,
  format = "BAM",
  bin_width = 150,
  n_smoothBin = 5,
  ref = "hg38",
  read_size = 101
)
```

Arguments

| | |
|-----------------|---|
| filename | Path to the BAM file (with index) or BED file |
| BigWig_filename | Path to write the output BigWig file |
| format | File format, either "BAM" or "BED" |
| bin_width | Bin size for coverage |
| n_smoothBin | Number of bins for smoothing values |
| ref | Reference genome. |
| read_size | Length of the reads. |

Value

Writes a BigWig file as output

| | |
|-----------------------------|--|
| raw_counts_to_sparse_matrix | <i>Create a sparse count matrix from various format of input data.</i> |
|-----------------------------|--|

Description

This function takes three different type of single-cell input: - Single cell BAM files (sorted) - Single cell BED files (gzipped) - A combination of an index file, a peak file and cell barcode file (The index file is composed of three column: index i, index j and value x for the non zeroes entries in the sparse matrix.)

Usage

```
raw_counts_to_sparse_matrix(
  files_dir_list,
  file_type = c("scBED", "scBAM", "FragmentFile"),
  use_Signac = TRUE,
  peak_file = NULL,
  n_bins = NULL,
  bin_width = NULL,
  genebody = NULL,
  extendPromoter = 2500,
  verbose = TRUE,
  ref = c("hg38", "mm10")[1],
  progress = NULL,
  BPPARAM = BiocParallel::bpparam()
)
```

Arguments

| | |
|-----------------------------|---|
| <code>files_dir_list</code> | A named character vector of directories containing the files. The names correspond to sample names. |
| <code>file_type</code> | Input file(s) type(s) ('scBED', 'scBAM', 'FragmentFile') |
| <code>use_Signac</code> | Use Signac wrapper function 'FeatureMatrix' if the Signac package is installed (TRUE). |
| <code>peak_file</code> | A file containing genomic location of peaks (NULL) |
| <code>n_bins</code> | The number of bins to tile the genome (NULL) |
| <code>bin_width</code> | The size of bins to tile the genome (NULL) |
| <code>genebody</code> | Count on genes (body + promoter) ? (NULL) |
| <code>extendPromoter</code> | If counting on genes, number of base pairs to extend up or downstream of TSS (2500). |
| <code>verbose</code> | Verbose (TRUE) |
| <code>ref</code> | reference genome to use (hg38) |
| <code>progress</code> | Progress object for Shiny |
| <code>BPPARAM</code> | BPPARAM object for multiprocessing. See bpparam for more informations. Will take the default BPPARAM set in your R session. |

Details

This functions re-counts signal on either fixed genomic bins, a set of user-defined peaks or around the TSS of genes.

Value

A sparse matrix of features x cells

References

Stuart et al., Multimodal single-cell chromatin analysis with Signac bioRxiv <https://doi.org/10.1101/2020.11.09.373613>

read_count_mat_with_separated_chr_start_end

Read a count matrix with three first columns (chr,start,end)

Description

Read a count matrix with three first columns (chr,start,end)

Usage

```
read_count_mat_with_separated_chr_start_end(
  path_to_matrix,
  format_test,
  separator
)
```

Arguments

| | |
|----------------|--------------------------|
| path_to_matrix | Path to the count matrix |
| format_test | Sample of the read.table |
| separator | Separator character |

Value

A sparseMatrix with rownames in the form "chr1:1222-55555"

read_sparse_matrix *Read in one or multiple sparse matrices (10X format)*

Description

Given one or multiple directories, look in each directory for a combination of the following files :

- A 'features' file containing unique feature genomic locations -in tab separated format (*_features.bed / .txt / .tsv / .gz), e.g. chr, start and end
- A 'barcodes' file containing unique barcode names (_barcode.txt / .tsv / .gz)
- A 'matrix' A file containing indexes of non zero entries (_matrix.mtx / .gz)

Usage

```
read_sparse_matrix(files_dir_list, ref = c("hg38", "mm10")[1], verbose = TRUE)
```

Arguments

`files_dir_list` A named character vector containing the full path towards folders. Each folder should contain only the Feature file, the Barcode file and the Matrix file (see description).

`ref` Reference genome (used to filter non-canonical chromosomes).

`verbose` Print ?

Value

Returns a list containing a datamatrix and cell annotation

Examples

```
## Not run:
sample_dirs = c("/path/to/folder1/", "/path/to/folder2/")
names(sample_dirs) = c("sample_1", "sample_2")
out <- read_sparse_matrix(sample_dirs, ref = "hg38")
head(out$datamatrix)
head(out$annot_raw)

## End(Not run)
```

`reduce_dims_scExp` *Reduce dimensions (PCA, TSNE, UMAP)*

Description

Reduce dimensions (PCA, TSNE, UMAP)

Usage

```
reduce_dims_scExp(
  scExp,
  dimension_reductions = c("PCA", "UMAP"),
  n = 50,
  batch_correction = FALSE,
  batch_list = NULL,
  remove_PC1 = FALSE,
  verbose = TRUE
)
```

Arguments

| | |
|----------------------|---|
| scExp | A SingleCellExperiment object. |
| dimension_reductions | A character vector of methods to apply. (c('PCA','TSNE','UMAP')) |
| n | Numbers of dimensions to keep for PCA. (50) |
| batch_correction | Do batch correction ? (FALSE) |
| batch_list | List of characters. Names are batch names, characters are sample names. |
| remove_PC1 | Remove PC1 before UMAP & T-SNE, as probably correlated to library size ? Recommended when using 'TFIDF' normalization method. (FALSE) |
| verbose | Print messages ?(TRUE) |

Value

A SingleCellExperiment object containing feature spaces. See ?reduceDims().

Examples

```
scExp = create_scExp(create_scDataset_raw()$mat,create_scDataset_raw()$annot)
scExp = reduce_dims_scExp(scExp,dimension_reductions=c("PCA", "UMAP"))
scExp = normalize_scExp(scExp)
scExp = reduce_dims_scExp(scExp,dimension_reductions=c("PCA", "UMAP"))
```

reduce_dim_batch_correction

Reduce dimension with batch corrections

Description

Reduce dimension with batch corrections

Usage

```
reduce_dim_batch_correction(scExp, mat, batch_list, n)
```

Arguments

| | |
|------------|-----------------------------|
| scExp | SingleCellExperiment |
| mat | The normalized count matrix |
| batch_list | List of batches |
| n | Number of PCs to keep |

Value

A list containing the SingleCellExperiment with batch info and the corrected pca

remove_chr_M_fun *Remove chromosome M from scExprownames*

Description

Remove chromosome M from scExprownames

Usage

```
remove_chr_M_fun(scExp, verbose)
```

Arguments

| | |
|---------|------------------------|
| scExp | A SingleCellExperiment |
| verbose | Print ? |

Value

A SingleCellExperiment without chromosome M (mitochondrial chr)

remove_non_canonical_fun
Remove non canonical chromosomes from scExp

Description

Remove non canonical chromosomes from scExp

Usage

```
remove_non_canonical_fun(scExp, verbose)
```

Arguments

| | |
|---------|------------------------|
| scExp | A SingleCellExperiment |
| verbose | Print ? |

Value

A SingleCellExperiment without non canonical chromosomes (random,unknown, contigs etc...)

`results_enrichmentTest`*Results of hypergeometric gene set enrichment test*

Description

Run hypergeometric enrichment test and combine significant pathways into a data.frame

Usage

```
results_enrichmentTest(  
  differentialGenes,  
  enrichment_qval,  
  GeneSets,  
  GeneSetsDf,  
  GenePool  
)
```

Arguments

| | |
|--------------------------------|--|
| <code>differentialGenes</code> | Genes significantly over / under expressed |
| <code>enrichment_qval</code> | Adjusted p-value threshold above which a pathway is considered significant |
| <code>GeneSets</code> | List of pathways |
| <code>GeneSetsDf</code> | Data.frame of pathways |
| <code>GenePool</code> | Pool of possible genes for testing |

Value

A data.frame with pathways passing q.value threshold

`retrieve_top_bot_features_pca`*Retrieve Top and Bot most contributing features of PCA*

Description

Retrieve Top and Bot most contributing features of PCA

Usage

```
retrieve_top_bot_features_pca(
  pca,
  counts,
  component,
  n_top_bot,
  absolute = FALSE
)
```

Arguments

| | |
|-----------|--|
| pca | A matrix/data.frame of rotated data |
| counts | the normalized counts used for PCA |
| component | the component of interest |
| n_top_bot | the number of top & bot features to take |
| absolute | If TRUE, return the top features in absolute values instead. |

Value

a data.frame of top bot contributing features in PCA

| | |
|--------------------|---------------------------|
| run_pairwise_tests | <i>Run pairwise tests</i> |
|--------------------|---------------------------|

Description

Run pairwise tests

Usage

```
run_pairwise_tests(
  affectation,
  nclust,
  counts,
  feature,
  method,
  progress = NULL,
  BPPARAM = BiocParallel::bpparam()
)
```

Arguments

| | |
|-------------|---|
| affectation | An annotation data.frame with cell_cluster and cell_id columns |
| nclust | Number of clusters |
| counts | Count matrix |
| feature | Feature data.frame |
| method | DA method, Wilcoxon or edgeR |
| progress | A shiny Progress instance to display progress bar. |
| BPPARAM | BPPARAM object for multiprocessing. See bpparam for more informations. Will take the default BPPARAM set in your R session. |

Value

A list containing objects for DA function

| | |
|----------------|---|
| run_tsne_scExp | <i>Run tsne on single cell experiment</i> |
|----------------|---|

Description

Run tsne on single cell experiment

Usage

```
run_tsne_scExp(scExp, verbose = FALSE)
```

Arguments

| | |
|---------|-------------------------------|
| scExp | A SingleCellExperiment Object |
| verbose | Print ? |

Value

A colored kable with the number of cells per sample for display

 scExp

A SingleCellExperiment outputed by ChromSCape

Description

Data from a single-cell ChIP-seq experiment against H3K4me3 active mark from two cell lines, Jurkat B cells and Ramos T cells from Grosselin et al., 2019. The count matrices, on 5kbp bins, were given to ChromSCape and the filtering parameter was set to 3% of cells active in regions and subsampled down to 150 cells per sample. After correlation filtering, the experiment is composed of respectively 51 and 55 cells from Jurkat & Ramos and 5499 5kbp-genomic bins where signal is located.

Usage

```
data("scExp")
```

Format

scExp - a SingleCellExperiment with 106 cells and 5499 features (genomic bins) in hg38:

chr A SingleCellExperiment

Details

The scExp is composed of :

- counts and normcounts assays, PCA, UMAP, and Correlation matrix in `reducedDims(scExp)`
- Assignment of genes to genomic bins in `rowRanges(scExp)`
- Cluster information in `colData(scExp)` correlation
- Hierarchical clustering dendrogram in `metadata$hc_cor`
- Consensus clustering raw data in `metadata$consclust`
- Consensus clustering cluster-consensus and item consensus dataframes in `metadata$icl`
- Differential analysis in `metadata$diff`
- Gene Set Analysis in `metadata$enr`

Examples

```
data("scExp")
plot_reduced_dim_scExp(scExp)
plot_reduced_dim_scExp(scExp, color_by = "cell_cluster")
plot_heatmap_scExp(scExp)
plot_differential_volcano_scExp(scExp, cell_cluster = "C1")
plot_differential_summary_scExp(scExp)
```

```
separate_BAM_into_clusters
    Separate BAM files into cell cluster BAM files
```

Description

Separate BAM files into cell cluster BAM files

Usage

```
separate_BAM_into_clusters(affectation, odir, merged_bam)
```

Arguments

| | |
|-------------|---|
| affectation | An annotation data.frame containing cell_id and cell_cluster columns |
| odir | A valid output directory path |
| merged_bam | A list of merged bam file paths @importFrom Rsamtools filterBam ScanBamParam |

Value

Create one BAM per cluster from one BAM per condition

```
separator_count_mat    Determine Count matrix separator ("tab" or ",")
```

Description

Determine Count matrix separator ("tab" or ",")

Usage

```
separator_count_mat(path_to_matrix)
```

Arguments

| | |
|----------------|--|
| path_to_matrix | A path towards the count matrix to check |
|----------------|--|

Value

A character separator

| | |
|-----------|---|
| smoothBin | <i>Smooth a vector of values with nb_bins left and right values</i> |
|-----------|---|

Description

Smooth a vector of values with nb_bins left and right values

Usage

```
smoothBin(bin_score, nb_bins = 10)
```

Arguments

| | |
|-----------|--|
| bin_score | A numeric vector of values to be smoothed |
| nb_bins | Number of values to take left and right @importFrom BiocParallel bvec |

Value

A smooth vector of the same size

| | |
|-----------------|------------------------|
| subsample_scExp | <i>Subsample scExp</i> |
|-----------------|------------------------|

Description

Randomly sample x cells from each sample in a SingleCellExperiment to return a subsampled SingleCellExperiment with all samples having maximum n cells. If n is higher than the number of cell in a sample, this sample will not be subsampled.

Usage

```
subsample_scExp(scExp, n_cells = 500)
```

Arguments

| | |
|---------|---|
| scExp | A SingleCellExperiment |
| n_cells | An integer number of cells to subsample for each sample (500) |

Value

A subsampled SingleCellExperiment

Examples

```
scExp = create_scExp(create_scDataset_raw())$mat,create_scDataset_raw())$annot)
scExp_sub = subsample_scExp(scExp,50)
## Not run: num_cell_scExp(scExp_sub)
```

subset_bam_call_peaks *Peak calling on cell clusters*

Description

This functions does peak calling on each cell population in order to refine gene annotation for large bins. For instance, a 50000bp bins might contain the TSS of several genes, while in reality only one or two of these genes are overlapping the signal (peak). To do so, first in-silico cell sorting is applied based on previously defined clusters contained in the SingleCellExperiment. Taking BAM files of each sample as input, samtools pools then splits reads from each cell barcode into 1 BAM file per cell cluster (pseudo-bulk). Then MACS2 calls peaks on each cluster. The peaks are aggregated and merged if closer to a certain distance defined by user (10000bp). Then,

This function takes as input a SingleCellExperiment, that must contain a 'cell_cluster' column in it's colData, an output directory where to store temporary files, the list of BAM files corresponding to each sample and containing the cell barcode information as a tag (for instance tag CB:Z:xxx, XB:Z:xxx or else...) or single-cell BED files containing the raw reads and corresponding to the 'barcode' column metadata, the p.value used by MACS2 to distinguish significant peaks, the reference genome (either hg38 or mm10), the maximal merging distance in bp and a data.frame containing gene TSS genomic coordinates of corresponding genome (if set to NULL, will automatically load geneTSS). The output is a SingleCellExperiment with GRanges object containing ranges of each merged peaks that falls within genomic bins of the SingleCellExperiment, saving the bin range as additional column (window_chr, window_start, window_end), as well as the closest genes and their distance relative to the peak. The peaks may be present in several rows if multiple genes are close / overlap to the peaks.

Note that the user must have MACS2 installed and available in the PATH. Users can open command terminal and type 'which macs2' to verify the availability of these programs. Will only work on unix operating system. Check operating system with 'print(.Platform)'.

Usage

```
subset_bam_call_peaks(
  scExp,
  odir,
  input,
  format = "BAM",
  p.value = 0.05,
  ref = "hg38",
  peak_distance_to_merge = 10000,
  geneTSS_annotation = NULL,
  run_coverage = FALSE,
```

```

    progress = NULL
  )

```

Arguments

| | |
|------------------------|---|
| scExp | A SingleCellExperiment object |
| odir | Output directory where to write temporary files and each cluster's BAM file |
| input | A character vector of file paths to each sample's BAM file, containing cell barcode information as tags. BAM files can be paired-end or single-end. |
| format | Format of the input data, either "BAM" or "scBED". |
| p.value | a p-value to use for MACS2 to determine significant peaks. (0.05) |
| ref | A reference genome, either hg38 or mm10. ('hg38') |
| peak_distance_to_merge | Maximal distance to merge peaks together after peak calling , in bp. (10000) |
| geneTSS_annotation | A data.frame annotation of genes TSS. If NULL will automatically load Gene-code list of genes fro specified reference genome. |
| run_coverage | Create coverage tracks (.bw) for each cluster ? |
| progress | A shiny Progress instance to display progress bar. |

Details

The BED files of the peaks called for each clusters, as well as the merged peaks are written in the output directory.

Value

A SingleCellExperiment with refinded annotation

Examples

```

## Not run:
data("scExp")
subset_bam_call_peaks(scExp, "path/to/out/", list("sample1" =
  "path/to/BAM/sample1.bam", "sample2" = "path/to/BAM/sample2.bam"),
  p.value = 0.05, ref = "hg38", peak_distance_to_merge = 10000,
  geneTSS_annotation = NULL)

## End(Not run)

```

`swapAltExp_sameColData`*Swap main & alternative Experiments, with fixed colData*

Description

Swap main & alternative Experiments, with fixed colData

Usage

```
swapAltExp_sameColData(scExp, alt)
```

Arguments

| | |
|--------------------|------------------------------------|
| <code>scExp</code> | A SingleCellExperiment |
| <code>alt</code> | Name of the alternative experiment |

Value

A swapped SingleCellExperiment with the exact same colData.

Examples

```
data(scExp)
swapAltExp_sameColData(scExp, "peaks")
```

`table_enriched_genes_scExp`*Creates table of enriched genes sets*

Description

Creates table of enriched genes sets

Usage

```
table_enriched_genes_scExp(
  scExp,
  set = "Both",
  group = "C1",
  enr_class_sel = c("c1_positional", "c2_curated", "c3_motif", "c4_computational",
    "c5_G0", "c6_oncogenic", "c7_immunologic", "hallmark")
)
```

Arguments

| | |
|---------------|--|
| scExp | A SingleCellExperiment object containing list of enriched gene sets. |
| set | A character vector, either 'Both', 'Overexpressed' or 'Underexpressed'. ('Both') |
| group | The "group" name from differential analysis. Can be the cluster name or the custom name in case of a custom differential analysis. |
| enr_class_sel | Which classes of gene sets to show. (c('c1_positional', 'c2_curated', ...)) |

Value

A DT::data.table of enriched gene sets.

Examples

```
data("scExp")
## Not run: table_enriched_genes_scExp(scExp)
```

| | |
|------------|--|
| warning_DA | <i>Warning for differential_analysis_scExp</i> |
|------------|--|

Description

Warning for differential_analysis_scExp

Usage

```
warning_DA(scExp, de_type, method, qval.th, cdiff.th, block, group, ref)
```

Arguments

| | |
|----------|---|
| scExp | A SingleCellExperiment object containing consclust with selected number of cluster. |
| de_type | Type of comparisons. Either 'one_vs_rest', to compare each cluster against all others, or 'pairwise' to make 1 to 1 comparisons. ('one_vs_rest') |
| method | Wilcoxon or edgeRGLM |
| qval.th | Adjusted p-value threshold. (0.01) |
| cdiff.th | Fold change threshold. (1) |
| block | Use batches as blocking factors ? |
| group | If de_type is custom, the group to compare (data.frame), must be a one-column data.frame with cell_clusters or sample_id as character in rows |
| ref | If de_type is custom, the reference to compare (data.frame), must be a one-column data.frame with cell_clusters or sample_id as character in rows |

Value

Warnings or Errors if the input are not correct

```
warning_filter_correlated_cell_scExp
      warning_filter_correlated_cell_scExp
```

Description

warning_filter_correlated_cell_scExp

Usage

```
warning_filter_correlated_cell_scExp(  
  scExp,  
  random_iter,  
  corr_threshold,  
  percent_correlation,  
  run_tsne,  
  downsample,  
  verbose  
)
```

Arguments

| | |
|---------------------|--|
| scExp | A SingleCellExperiment object containing 'Cor', a correlation matrix, in reducedDims. |
| random_iter | Number of random matrices to create to calculate random correlation scores. (50) |
| corr_threshold | Quantile of random correlation score above which a cell is considered to be 'correlated' with another cell. (99) |
| percent_correlation | Percentage of the cells that any cell must be 'correlated' to in order to not be filtered. (1) |
| run_tsne | Re-run tsne ? (FALSE) |
| downsample | Number of cells to calculate correlation filtering threshold ? (2500) |
| verbose | (TRUE) |

Value

Warnings or Errors if the input are not correct

```
warning_plot_reduced_dim_scExp
```

A warning helper for plot_reduced_dim_scExp

Description

A warning helper for plot_reduced_dim_scExp

Usage

```
warning_plot_reduced_dim_scExp(  
  scExp,  
  color_by,  
  reduced_dim,  
  downsample,  
  transparency,  
  size,  
  max_distanceToTSS,  
  annotate_clusters,  
  min_quantile,  
  max_quantile  
)
```

Arguments

| | |
|-------------------|---|
| scExp | A SingleCellExperiment Object |
| color_by | Feature used for coloration |
| reduced_dim | Reduced Dimension used for plotting |
| downsample | Number of cells to downsample |
| transparency | Alpha parameter, between 0 and 1 |
| size | Size of the points. |
| max_distanceToTSS | Numeric. Maximum distance to a gene's TSS to consider a region linked to a gene. |
| annotate_clusters | A logical indicating if clusters should be labelled. The 'cell_cluster' column should be present in metadata. |
| min_quantile | The lower threshold to remove outlier cells, as quantile of cell embeddings (between 0 and 0.5). |
| max_quantile | The upper threshold to remove outlier cells, as quantile of cell embeddings (between 0.5 and 1). |

Value

Warning or errors if the inputs are not correct

 warning_raw_counts_to_sparse_matrix

Warning for raw_counts_to_sparse_matrix

Description

Warning for raw_counts_to_sparse_matrix

Usage

```

warning_raw_counts_to_sparse_matrix(
  files_dir_list,
  file_type = c("scBAM", "scBED", "SparseMatrix"),
  peak_file = NULL,
  n_bins = NULL,
  bin_width = NULL,
  genebody = NULL,
  extendPromoter = 2500,
  verbose = TRUE,
  ref = "hg38"
)

```

Arguments

| | |
|----------------|--|
| files_dir_list | A named character vector of directory containing the raw files |
| file_type | Input file(s) type(s) ('scBED','scBAM','SparseMatrix') |
| peak_file | A file containing genomic location of peaks (NULL) |
| n_bins | The number of bins to tile the genome (NULL) |
| bin_width | The size of bins to tile the genome (NULL) |
| genebody | Count on genes (body + promoter) ? (NULL) |
| extendPromoter | If counting on genes, number of base pairs to extend up or downstream of TSS (2500). |
| verbose | Verbose (TRUE) |
| ref | reference genome to use (hg38) |

Value

Error or warnings if the input are not correct

 wrapper_Signac_FeatureMatrix

Wrapper around 'FeatureMatrix' function from Signac Package

Description

Wrapper around 'FeatureMatrix' function from Signac Package

Usage

```

wrapper_Signac_FeatureMatrix(
  files_dir_list,
  which,
  ref = "hg38",
  process_n = 2000,
  set_future_plan = TRUE,
  verbose = TRUE,
  progress = NULL
)

```

Arguments

| | |
|-----------------|---|
| files_dir_list | A named character vector of directories containing the files. The names correspond to sample names. |
| which | A GenomicRanges containing the features to count on. |
| ref | Reference genome to use (hg38). Chromosomes that are not present in the canonical chromosomes of the given reference genome will be excluded from the matrix. |
| process_n | Number of regions to load into memory at a time, per thread. Processing more regions at once can be faster but uses more memory. (2000) |
| set_future_plan | Set 'multisession' plan within the function (TRUE). If TRUE, the previous plan (e.g. future::plan()) will be set back on exit. |
| verbose | Verbose (TRUE). |
| progress | Progress object for Shiny. |

Details

Signac & future are not required packages for ChromSCape as they are required only for the fragment matrix calculations. To use this function, install Signac package first (future will be installed as a dependency). For the simplicity of the application & optimization, the function by defaults sets future::plan("multisession") with workers = future::availableCores() - 1 in order to allow parallel processing with Signac. On exit the plan is re-set to the previously set future plan. Note that future multisession may have trouble running when VPN is on. To run in parallel, first deactivate your VPN if you encounter long runtimes.

Value

A sparse matrix of features x cells

References

Stuart et al., Multimodal single-cell chromatin analysis with Signac bioRxiv <https://doi.org/10.1101/2020.11.09.373613>

Examples

```
## Not run:  
gr_bins = define_feature("hg38", bin_width = 50000)  
wrapper_Signac_FeatureMatrix("/path/to/dir_containing_fragment_files",  
  gr_bins, ref = "hg38")  
  
## End(Not run)
```

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