Package 'EpiCompare'

April 3, 2023

Type Package

Title Comparison, Benchmarking & QC of Epigenomic Datasets

Version 1.2.0

Description EpiCompare is used to compare and analyse epigenetic datasets for quality control and benchmarking purposes.

The package outputs an HTML report consisting of three sections:

- (1. General metrics) Metrics on peaks (percentage of blacklisted and non-standard peaks, and peak widths) and fragments (duplication rate) of samples,
- (2. Peak overlap) Percentage and statistical significance of overlapping and non-overlapping peaks. Also includes upset plot and
- (3. Functional annotation) functional annotation

(ChromHMM, ChIPseeker and enrichment analysis) of peaks.

Also includes peak enrichment around TSS.

License GPL-3

URL https://github.com/neurogenomics/EpiCompare

BugReports https://github.com/neurogenomics/EpiCompare/issues

Depends R (>= 4.1.0)

Imports AnnotationHub, BRGenomics, ChIPseeker, data.table, genomation, GenomicRanges, IRanges, GenomeInfoDb, ggplot2, htmltools, methods, plotly, reshape2, rmarkdown, rtracklayer, stats, stringr, utils, BiocGenerics

Suggests badger, BiocFileCache, BiocParallel, parallel, BiocStyle,

clusterProfiler, GenomicAlignments, grDevices, htmlwidgets,

knitr, org.Hs.eg.db, testthat (>= 3.0.0), tidyr,

TxDb.Hsapiens.UCSC.hg19.knownGene,

TxDb.Hsapiens.UCSC.hg38.knownGene,

TxDb.Mmusculus.UCSC.mm9.knownGene,

TxDb.Mmusculus.UCSC.mm10.knownGene,

BSgenome. Hsapiens. UCSC. hg19, BSgenome. Hsapiens. UCSC. hg38,

BSgenome.Mmusculus.UCSC.mm9, BSgenome.Mmusculus.UCSC.mm10,

UpSetR, plyranges, scales, Matrix, consensusSeekeR

VignetteBuilder knitr

```
biocViews Epigenetics, Genetics, QualityControl, ChIPSeq,
       MultipleComparison, FunctionalGenomics, ATACSeq, DNaseSeq
Config/testthat/edition 3
Encoding UTF-8
LazyData FALSE
RoxygenNote 7.2.1.9000
git_url https://git.bioconductor.org/packages/EpiCompare
git_branch RELEASE_3_16
git_last_commit c1e27ec
git_last_commit_date 2022-11-01
Date/Publication 2023-04-03
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bpplapply

Wrapper for bplapply

Description

Wrapper function for bplapply that automatically handles issues with **BiocParallel** related to different OS platforms.

Usage

```
bpplapply(
   X,
   FUN,
   apply_fun = parallel::mclapply,
   workers = 1,
   progressbar = workers > 1,
   verbose = workers == 1,
   use_snowparam = TRUE,
   register_now = FALSE,
   ...
)
```

Arguments

Χ	Any object for which methods length, [, and [[are implemented.
FUN	The function to be applied to each element of X.
apply_fun	Iterator function to use.
workers	Number of threads to parallelize across.
progressbar	logical(1) Enable progress bar (based on plyr:::progress_text).
verbose	Print messages.
use_snowparam	Whether to use SnowParam (default: TRUE) or MulticoreParam (FALSE) when parallelising across multiple workers.
register_now	Register the cores now with register (TRUE), or simply return the BPPARAM object (default: FALSE).
	Arguments passed on to BiocParallel::bplapply

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BPPARAM An optional BiocParallelParam instance determining the parallel back-end to be used during evaluation, or a list of BiocParallelParam instances, to be applied in sequence for nested calls to **BiocParallel** functions.

BPREDO A list of output from bplapply with one or more failed elements. When a list is given in BPREDO, bpok is used to identify errors, tasks are rerun and inserted into the original results.

BPOPTIONS Additional options to control the behavior of the parallel evaluation, see bpoptions.

Value

(Named) list.

Examples

```
X <- stats::setNames(seq_len(length(letters)), letters)
out <- bpplapply(X, print)</pre>
```

CnR_H3K27ac

Example CUT&Run peak file

Description

Human H3K27ac peak file generated with CUT&Run using K562 cell-line from Meers et al., (2019). Human genome build hg19 was used. Raw peak file (.BED) was obtained from GEO (https://trace.ncbi.nlm.nih.gov/Traces/sra/?run=SRR8581604). Peak calling was performed by Leyla Abbasova using MACS2. The peak file was then processed into GRanges object. Peaks located on chromosome 1 were subsetted to reduce the dataset size.

Usage

```
data("CnR_H3K27ac")
```

Format

An object of class GRanges of length 2707.

Source

The code to prepare the .Rda file from the raw peak file is:

```
# sequences were directly downloaded from https://trace.ncbi.nlm.nih.gov/Traces/sra/?run=SRR8581604
# and peaks (BED file) were generated by Leyla Abbasova (Neurogenomics Lab, Imperial College
London)
CnR_H3K27ac <- ChIPseeker::readPeakFile("path", as = "GRanges")
CnR_H3K27ac <- CnR_H3K27ac[seqnames(CnR_H3K27ac)== "chr1"]
my_label <- c("name", "score", "strand", "signalValue", "pValue", "qValue", "peak")</pre>
```

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```
colnames(GenomicRanges::mcols(CnR_H3K27ac)) <- my_label
usethis::use_data(CnR_H3K27ac, overwrite = TRUE)</pre>
```

CnR_H3K27ac_picard

Example Picard duplication metrics file 2

Description

Duplication metrics output on CUT&Run H3K27ac file (sample accession: SRR8581604). Raw sequences were aligned to hg19 genome and after, Picard was performed by Leyla Abbasova. The duplication summary output generated by Picard was processed to reduce the size of data.

Usage

```
data("CnR_H3K27ac_picard")
```

Format

An object of class data. frame with 1 rows and 10 columns.

Source

```
The code to prepare the .Rda file is:
```

```
picard <- read.table("path/to/picard/duplication/output", header = TRUE, fill = TRUE)
CnR_H3K27ac_picard <- picard[1,]
usethis::use_data(CnR_H3K27ac_picard, overwrite = TRUE)</pre>
```

CnT_H3K27ac

Example CUT&Tag peak file

Description

Human H3K27ac peak file generated with CUT&Tag using K562 cell-line from Kaya-Okur et al., (2019). Human genome build hg19 was used. Raw peak file (.BED) was obtained from GEO (https://trace.ncbi.nlm.nih.gov/Traces/sra/?run=SRR8383507). Peak calling was performed by Leyla Abbasova using MACS2. The peak file was then imported as an GRanges object. Peaks located on chromosome 1 were subsetted to reduce the dataset size.

Usage

```
data("CnT_H3K27ac")
```

Format

An object of class GRanges of length 1670.

Source

The code to prepare the .Rda file from the raw peak file is:

```
# sequences were directly downloaded from https://trace.ncbi.nlm.nih.gov/Traces/sra/?run=SRR8383507
# and peaks (BED file) were generated by Leyla Abbasova (Neurogenomics Lab, Imperial College
London)
CnT_H3K27ac <- ChIPseeker::readPeakFile("path", as = "GRanges")
CnT_H3K27ac <- CnT_H3K27ac[seqnames(CnT_H3K27ac)== "chr1"]
my_label <- c("name", "score", "strand", "signalValue", "pValue", "qValue", "peak")
colnames(GenomicRanges::mcols(CnT_H3K27ac)) <- my_label
usethis::use_data(CnT_H3K27ac)</pre>
```

CnT_H3K27ac_picard

Example Picard duplication metrics file 1

Description

Duplication metrics output of CUT&Tag H3K27ac file (sample accession: SRR8581604). Raw sequences were aligned to hg19 genome and Picard was performed by Leyla Abbasova. The duplication summary output generated by Picard was processed to reduce the size of data.

Usage

```
data("CnT_H3K27ac_picard")
```

Format

An object of class data. frame with 1 rows and 10 columns.

Source

The code to prepare the .Rda file is:

```
picard <- read.table("path/to/picard/duplication/output", header = TRUE, fill = TRUE)]
CnT_H3K27ac_picard <- picard[1,]
usethis::use_data(CnT_H3K27ac_picard, overwrite = TRUE)</pre>
```

compute_consensus_peaks

Compute consensus peaks

Description

Compute consensus peaks from a list of GRanges.

Usage

```
compute_consensus_peaks(
  grlist,
  groups = NULL,
  genome_build,
  lower = 2,
  upper = Inf,
  min.gapwidth = 1L,
  method = c("granges", "consensusseeker"),
  ...
)
```

Arguments

grlist Named list of GRanges objects.

groups A character vector of the same length as grlist defining how to group GRanges

objects when computing consensus peaks.

genome_build Genome build name.

lower, upper The lower and upper bounds for the slice.

min.gapwidth Ranges separated by a gap of at least min.gapwidth positions are not merged.

method Method to call peaks with:

• "granges" : Simple overlap procedure using GRanges functions. Faster but less accurate.

• "consensusseeker": Uses findConsensusPeakRegions to compute consensus peaks. Slower but more accurate.

Arguments passed on to consensusSeekeR::findConsensusPeakRegions

narrowPeaks a GRanges containing called peak regions of signal enrichment based on pooled, normalized data for all analyzed experiments. All GRanges entries must have a metadata field called "name" which identifies the region to the called peak. All GRanges entries must also have a row name which identifies the experiment of origin. Each peaks entry must have an associated narrowPeaks entry. A GRanges entry is associated to a narrowPeaks entry by having a identical metadata "name" field and a identical row name.

- peaks a GRanges containing called peaks of signal enrichment based on pooled, normalized data for all analyzed experiments. All GRanges entries must have a metadata field called "name" which identifies the called peak. All GRanges entries must have a row name which identifies the experiment of origin. Each peaks entry must have an associated narrowPeaks entry. A GRanges entry is associated to a narrowPeaks entry by having a identical metadata "name" field and a identical row name.
- chrInfo a Seqinfo containing the name and the length of the chromosomes to analyze. Only the chomosomes contained in this Seqinfo will be analyzed.
- extendingSize a numeric value indicating the size of padding on both sides of the position of the peaks median to create the consensus region. The minimum size of the consensus region is equal to twice the value of the extendingSize parameter. The size of the extendingSize must be a positive integer. Default = 250.
- expandToFitPeakRegion a logical indicating if the region size, which is set by the extendingSize parameter is extended to include the entire narrow peak regions of all peaks included in the unextended consensus region. The narrow peak regions of the peaks added because of the extension are not considered for the extension. Default: FALSE.
- shrinkToFitPeakRegion a logical indicating if the region size, which is set by the extendingSize parameter is shrinked to fit the narrow peak regions of the peaks when all those regions are smaller than the consensus region. Default: FALSE.
- minNbrExp a positive numeric or a positive integer indicating the minimum number of experiments in which at least one peak must be present for a potential consensus region. The numeric must be a positive integer inferior or equal to the number of experiments present in the narrowPeaks and peaks parameters. Default = 1.
- nbrThreads a numeric or a integer indicating the number of threads to use in parallel. The nbrThreads must be a positive integer. Default = 1.

Details

NOTE: If you get the error "Error in serialize(data, node\$con): error writing to connection", try running closeAllConnections and rerun compute_consensus_peaks. This error can sometimes occur when compute_consensus_peaks has been disrupted partway through.

Value

Named list of consensus peak GRanges.

Source

GenomicRanges tutorial consensusSeekeR

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Examples

compute_corr

Compute correlation matrix

Description

Compute correlation matrix on all peak files.

Usage

```
compute_corr(
  peakfiles,
  reference = NULL,
  genome_build,
  keep_chr = NULL,
  drop_empty_chr = FALSE,
  bin_size = 5000,
  method = "spearman",
  intensity_cols = c("total_signal", "qValue", "Peak Score", "score"),
  return_bins = FALSE,
  workers = 1
)
```

Arguments

peakfiles

A list of peak files as GRanges object and/or as paths to BED files. If paths are provided, EpiCompare imports the file as GRanges object. EpiCompare also accepts a list containing a mix of GRanges objects and paths. Files must be listed and named using list(). E.g. list("name1"=file1, "name2"=file2). If no names are specified, default file names will be assigned.

reference

A named list containing reference peak file(s) as GRanges object. Please ensure that the reference file is listed and named i.e. list("reference_name" = reference_peak). If more than one reference is specified, individual reports for each reference will be generated. However, please note that specifying more than one reference can take awhile. If a reference is specified, it enables two analyses: (1) plot showing statistical significance of overlapping/non-overlapping peaks; and (2) ChromHMM of overlapping/non-overlapping peaks.

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genome_build The build of **all** peak and reference files to calculate the correlation matrix

on. If all peak and reference files are not of the same build use liftover_grlist to convert them all before running. Genome build should be one of hg19, hg38,

mm9, mm10.

keep_chr Which chromosomes to keep.

drop_empty_chr Drop chromosomes that are not present in any of the peakfiles (default: FALSE).

bin_size Default of 100. Base-pair size of the bins created to measure correlation. Use

smaller value for higher resolution but longer run time and larger memory usage.

Default spearman (i.e. non-parametric). A character string indicating which correlation coefficient (or covariance) is to be computed. One of "pearson",

"kendall", or "spearman": can be abbreviated.

intensity_cols Depending on which columns are present, this value will be used to get quantiles and ultimately calculate the correlations:

- "total_signal": Used by the peak calling software SEACR. NOTE: Another SEACR column (e.g. "max_signal") can be used together or instead of "total_signal".
- "qValue"Used by the peak calling software MACS2/3. Should contain the negative log of the p-values after multiple testing correction.
- "Peak Score": Used by the peak calling software HOMER.

return_bins If TRUE, returns a named list with both the rebinned (standardised) peaks ("bin")

and the correlation matrix ("cor"). If FALSE (default), returns only the correlation

matrix (unlisted).

workers Number of cores to parallelise across (in applicable functions).

Value

method

correlation matrix

encode_H3K27ac

encode_H3K27ac

Example ChIP-seq peak file

Description

Human H3K27ac peak file generated with ChIP-seq using K562 cell-line. Human genome build hg19 was used. The peak file (.BED) was obtained from ENCODE project (https://www.encodeproject.org/files/ENCFF044JNJ/). The BED file was then imported as an GRanges object. Peaks located on chromosome 1 were subsetted to reduce the dataset size.

Usage

```
data("encode_H3K27ac")
```

Format

An object of class GRanges of length 5142.

Source

The code to prepare the .Rda file from the raw peak file is:

```
# dataset was directly downloaded from
# https://www.encodeproject.org/files/ENCFF044JNJ/encode_H3K27ac <- ChIPseeker::readPeakFile("path",
as = "GRanges")
encode_H3K27ac <- encode_H3K27ac[seqnames(encode_H3K27ac) == "chr1"]
my_label <- c("name", "score", "strand", "signalValue", "pValue", "qValue", "peak")
colnames(GenomicRanges::mcols(encode_H3K27ac)) <- my_label
usethis::use_data(encode_H3K27ac, overwrite = TRUE)</pre>
```

EpiCompare

Compare epigenomic datasets

Description

This function compares and analyses multiple epigenomic datasets and outputs an HTML report containing all results of the analysis. The report is mainly divided into three sections: (1) General Metrics on Peakfiles, (2) Peak Overlaps and (3) Functional Annotation of Peaks.

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Usage

```
EpiCompare(
  peakfiles,
  genome_build,
  genome_build_output = "hg19",
 blacklist,
  picard_files = NULL,
  reference = NULL,
  upset_plot = FALSE,
  stat_plot = FALSE,
  chromHMM_plot = FALSE,
  chromHMM_annotation = "K562",
  chipseeker_plot = FALSE,
  enrichment_plot = FALSE,
  tss_plot = FALSE,
  precision_recall_plot = FALSE,
  n_{threshold} = 15,
  corr_plot = FALSE,
  bin_size = 5000,
  interact = TRUE,
  save_output = FALSE,
  output_filename = "EpiCompare",
  output_timestamp = FALSE,
  output_dir,
  display = NULL,
 workers = 1
)
```

Arguments

peakfiles

A list of peak files as GRanges object and/or as paths to BED files. If paths are provided, EpiCompare imports the file as GRanges object. EpiCompare also accepts a list containing a mix of GRanges objects and paths. Files must be listed and named using list(). E.g. list("name1"=file1, "name2"=file2). If no names are specified, default file names will be assigned.

genome_build

A named list indicating the human genome build used to generate each of the following inputs:

- "peakfiles": Genome build for the peakfiles input. Assumes genome build is the same for each element in the peakfiles list.
- "reference": Genome build for the reference input.
- "blacklist": Genome build for the blacklist input.

Example input list:

```
genome_build = list(peakfiles="hg38", reference="hg19", blacklist="hg19")
```

Alternatively, you can supply a single character string instead of a list. This should *only* be done in situations where all three inputs (peakfiles, reference,

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blacklist) are of the same genome build. For example: genome_build = "hg19"

genome_build_output

Genome build to standardise all inputs to. Liftovers will be performed automatically as needed. Default: "hg19".

blacklist

A GRanges object containing blacklisted regions.

picard_files

A list of summary metrics output from Picard. Files must be in data.frame format and listed using list() and named using names(). To import Picard duplication metrics (.txt file) into R as data frame, use:

picard <- read.table("/path/to/picard/output", header = TRUE, fill =
TRUE).</pre>

reference

A named list containing reference peak file(s) as GRanges object. Please ensure that the reference file is listed and named i.e. list("reference_name" = reference_peak). If more than one reference is specified, individual reports for each reference will be generated. However, please note that specifying more than one reference can take awhile. If a reference is specified, it enables two analyses: (1) plot showing statistical significance of overlapping/non-overlapping peaks; and (2) ChromHMM of overlapping/non-overlapping peaks.

upset_plot

Default FALSE. If TRUE, the report includes upset plot of overlapping peaks.

stat_plot

Default FALSE. If TRUE, the function creates a plot showing the statistical significance of overlapping/non-overlapping peaks. Reference peak file must be provided.

chromHMM_plot

Default FALSE. If TRUE, the function outputs ChromHMM heatmap of individual peak files. If a reference peak file is provided, ChromHMM annotation of overlapping and non-overlapping peaks is also provided.

chromHMM_annotation

ChromHMM annotation for ChromHMM plots. Default K562 cell-line. Cell-line options are:

- "K562" = K-562 cells
- "Gm12878" = Cellosaurus cell-line GM12878
- "H1hesc" = H1 Human Embryonic Stem Cell
- "Hepg2" = Hep G2 cell
- "Hmec" = Human Mammary Epithelial Cell
- "Hsmm" = Human Skeletal Muscle Myoblasts
- "Huvec" = Human Umbilical Vein Endothelial Cells
- "Nhek" = Normal Human Epidermal Keratinocytes
- "Nhlf" = Normal Human Lung Fibroblasts

chipseeker_plot

Default FALSE. If TRUE, the report includes a barplot of ChIPseeker annotation of peak files.

enrichment_plot

Default FALSE. If TRUE, the report includes dotplots of KEGG and GO enrichment analysis of peak files.

tss_plot

Default FALSE. If TRUE, the report includes peak count frequency around transcriptional start site. Note that this can take awhile.

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precision_recall_plot Default is FALSE. If TRUE, creates a precision-recall curve plot and an F1 plot using plot_precision_recall. n_threshold Number of thresholds to test. corr_plot Default is FALSE. If TRUE, creates a correlation plot across all peak files using plot_corr. Default of 100. Base-pair size of the bins created to measure correlation. Use bin_size smaller value for higher resolution but longer run time and larger memory usage. interact Default TRUE. By default, all heatmaps are interactive. If set FALSE, all heatmaps in the report will be static. Default FALSE. If TRUE, all outputs (tables and plots) of the analysis will be save_output saved in a folder (EpiCompare file). output_filename Default EpiCompare.html. If otherwise, the html report will be saved in the specified name. output_timestamp Default FALSE. If TRUE, date will be included in the file name. output_dir Path to where output HTML file should be saved. After completion, automatically display the HTML report file in one of the foldisplay lowing ways: • "browser" : Display the report in your default web browser.

• "rsstudio": Display the report in Rstudio.

• NULL (default): Do not display the report.

workers Number of cores to parallelise across (in applicable functions).

Value

Path to one or more HTML report files.

```
### Load Data ###
data("encode_H3K27ac") # example dataset as GRanges object
data("CnT_H3K27ac") # example dataset as GRanges object
data("CnR_H3K27ac") # example dataset as GRanges object
data("hg19_blacklist") # hg38 blacklist dataset
data("CnT_H3K27ac_picard") # example Picard summary output
data("CnR_H3K27ac_picard") # example Picard summary output
#### Prepare Input ####
# create named list of peakfiles
peaks <- list(CnR=CnR_H3K27ac, CnT=CnT_H3K27ac)</pre>
# create named list of picard outputs
picard <- list(CnR=CnR_H3K27ac_picard, CnT=CnT_H3K27ac_picard)</pre>
# reference peak file
reference_peak <- list("ENCODE" = encode_H3K27ac)</pre>
```

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fragment_info

Summary on fragments

Description

This function outputs a summary on fragments using metrics generated by Picard. Provides the number of mapped fragments, duplication rate and number of unique fragments.

Usage

```
fragment_info(picard_list)
```

Arguments

picard_list

Named list of duplication metrics generated by Picard as data frame. Data frames must be named and listed using list(). e.g. list("name1"=file1, "name2"=file2). To import Picard duplication metrics (.txt file) into R as data frame, use picard <- read.table("/path/to/picard/output", header = TRUE, fill = TRUE).

Value

A table summarizing metrics on fragments.

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```
### Run ###
df <- fragment_info(picard_list = picard)</pre>
```

gather_files

Gather files

Description

Recursively find peak/picard files stored within subdirectories and import them as a list of GRanges objects.

Usage

```
gather_files(
  dir,
  type = "peaks.stringent",
  nfcore_cutandrun = FALSE,
  return_paths = FALSE,
  rbind_list = FALSE,
  workers = 1,
  verbose = TRUE
)
```

Arguments

dir

Directory to search within.

type

File type to search for. Options include:

- "<pattern>"Finds files matching an arbitrary regex pattern specified by user.
- "peaks.stringent"Finds files ending in "*.stringent.bed\$"
- "peaks.consensus"Finds files ending in "*.consensus.peaks.bed\$"
- "peaks.consensus.filtered" Finds files ending in"*.consensus.peaks.filtered.awk.bed\$"
- "picard"Finds files ending in "*.target.markdup.MarkDuplicates.metrics.txt\$"

nfcore_cutandrun

Whether the files were generated by the nf-core/cutandrun Nextflow pipeline. If TRUE, can use the standardised folder structure to automatically generate more

descriptive file names with sample IDs.

Return only the file paths without actually reading them in as GRanges. return_paths

rbind_list Bind all objects into one.

workers integer(1) Number of workers. Defaults to the maximum of 1 or the num-

> ber of cores determined by detectCores minus 2 unless environment variables R_PARALLELLY_AVAILABLECORES_FALLBACK or BIOCPARALLEL_WORKER_NUMBER

are set otherwise.

verbose Print messages. group_files 17

Details

For "peaks.stringent" files called with SEACR, column names will be automatically added:

- total_signal: Total signal contained within denoted coordinates.
- max_signalMaximum bedgraph signal attained at any base pair within denoted coordinates.
- max_signal_region Region representing the farthest upstream and farthest downstream bases within the denoted coordinates that are represented by the maximum bedgraph signal.

Value

A named list of GRanges objects.

Examples

```
#### Make example files ####
save_paths <- EpiCompare::write_example_peaks()
dir <- unique(dirname(save_paths))
#### Gather/import files ####
peaks <- EpiCompare::gather_files(dir=dir, type="peaks.narrow")</pre>
```

group_files

Group files

Description

Assign group names to each file in a named list based on a series of string searches based on combinations of relevant metadata factors.

Usage

```
group_files(peakfiles, searches)
```

Arguments

peakfiles

A list of peak files as GRanges object and/or as paths to BED files. If paths are provided, EpiCompare imports the file as GRanges object. EpiCompare also accepts a list containing a mix of GRanges objects and paths. Files must be listed and named using list(). E.g. list("name1"=file1, "name2"=file2). If no names are specified, default file names will be assigned.

searches

A named list of substrings to group peakfiles by.

hg19_blacklist

Examples

hg19_blacklist

Human genome hg19 blacklisted regions

Description

Obtained from https://www.encodeproject.org/files/ENCFF001TDO/. The ENCODE black-list includes regions of the hg19 genome that have anomalous and/or unstructured signals independent of the cell-line or experiment. Removal of ENCODE blacklist is recommended for quality measure.

Usage

```
data("hg19_blacklist")
```

Format

An object of class GRanges of length 411.

Source

The code to prepare the .Rda file is:

```
# blacklisted regions were directly downloaded
# from https://www.encodeproject.org/files/ENCFF001TDO/
hg19_blacklist <- ChIPseeker::readPeakFile(file.path(path), as = "GRanges")
usethis::use_data(hg19_blacklist, overwrite = TRUE)</pre>
```

hg38_blacklist

hg38_blacklist

Human genome hg38 blacklisted regions

Description

Obtained from https://www.encodeproject.org/files/ENCFF356LFX/. The ENCODE black-list includes regions of the hg38 genome that have anomalous and/ or unstructured signals independent of the cell-line or experiment. Removal of ENCODE blacklist is recommended for quality measure.

Usage

```
data("hg38_blacklist")
```

Format

An object of class GRanges of length 910.

Source

```
The code to prepare the .Rda file is:

## blacklisted regions were directly downloaded

## from https://www.encodeproject.org/files/ENCFF356LFX/
hg38_blacklist <- ChIPseeker::readPeakFile(file.path(path), as = "GRanges")
usethis::use_data(hg38_blacklist, overwrite = TRUE)
```

liftover_grlist

Liftover peak list

Description

Perform genome build liftover to one or more GRanges objects at once.

Usage

```
liftover_grlist(
  grlist,
  input_build,
  output_build = "hg19",
  style = "UCSC",
  keep_chr = paste0("chr", c(seq_len(22), "X", "Y")),
  as_grangeslist = FALSE,
  merge_all = FALSE,
  verbose = TRUE
)
```

20 overlap_heatmap

Arguments

grlist A named list of GRanges objects, or simply a single unlisted GRanges object.

Can perform liftover within species or across species.

input_build The genome build of grlist.

output_build Desired genome build for grlist to be lifted over to.

style Chromosome style, set by seqlevelsStyle.

"UCSC": Uses the chromosome style "chr1"."NCBI": Uses the chromosome style "1"

keep_chr Which chromosomes to keep.
as_grangeslist Return as a GRangesList.

merge_all Merge all GRanges into a single GRanges object.

verbose Print messages.

Value

Named list of lifted GRanges objects.

Examples

overlap_heatmap General

Generate heatmap of percentage overlap

Description

This function generates a heatmap showing percentage of overlapping peaks between peak files.

Usage

```
overlap_heatmap(peaklist, interact = TRUE)
```

Arguments

peaklist A list of peak files as GRanges object. Files must be listed and named using

list(). e.g. list("name1"=file1, "name2"=file2). If not named, default

file names will be assigned.

interact Default TRUE. By default heatmap is interactive. If FALSE, heatmap is static.

overlap_percent 21

Value

An interactive heatmap

Examples

```
### Load Data ###
data("encode_H3K27ac") # example peakfile GRanges object
data("CnT_H3K27ac") # example peakfile GRanges object

### Create Named List ###
peaks <- list("encode"=encode_H3K27ac, "CnT"=CnT_H3K27ac)

### Run ###
my_heatmap <- overlap_heatmap(peaklist = peaks)</pre>
```

overlap_percent

Calculate percentage of overlapping peaks

Description

This function calculates the percentage of overlapping peaks and outputs a table or matrix of results.

Usage

```
overlap_percent(
  peaklist1,
  peaklist2,
  invert = FALSE,
  precision_recall = TRUE,
  suppress_messages = TRUE)
```

Arguments

A list of peak files as GRanges object. Files must be listed and named using list(). e.g. list("name1"=file1, "name2"=file2). If not named, default file names will be assigned.

peaklist2 peaklist1 A list of peak files as GRanges object. Files must be listed and named using list(). e.g. list("name1"=file1, "name2"=file2).

invert If TRUE, keep only the ranges in x that do not overlap ranges.

precision_recall

Return percision-recall results for all combinations of peaklist1 (the "query") and peaklist2 (the "subject"). See subsetByOverlaps for more details on this terminology.

suppress_messages

Suppress messages.

22 overlap_stat_plot

Value

data frame

Examples

overlap_stat_plot

Statistical significance of overlapping peaks

Description

This function calculates the statistical significance of overlapping/ non-overlapping peaks against a reference peak file. If the reference peak file has the BED6+4 format (peak called by MACS2), the function generates a series of boxplots showing the distribution of q-values for sample peaks that are overlapping and non-overlapping with the reference. If the reference peak file does not have the BED6+4 format, the function uses 'enrichPeakOverlap()' from 'ChIPseeker' package to calculate the statistical significance of overlapping peaks only. In this case, please provide an annotation file as TxDb object.

Usage

```
overlap_stat_plot(reference, peaklist, annotation = NULL)
```

Arguments

reference A reference peak file as GRanges object.

peaklist A list of peak files as GRanges object. Files must be listed and named using

list(). E.g. list("name1"=file1, "name2"=file2). If not named, default

file names will be assigned.

annotation A TxDb annotation object from Bioconductor. This is required only if the refer-

ence file does not have BED6+4 format.

Value

A boxplot or barplot showing the statistical significance of overlapping/non-overlapping peaks.

overlap_upset_plot 23

Examples

overlap_upset_plot

Generate Upset plot for overlapping peaks

Description

This function generates upset plot (UpSetR package) of overlapping peaks.

Usage

```
overlap_upset_plot(peaklist)
```

Arguments

peaklist

A named list of peak files as GRanges object. Objects must be listed and named using list(). e.g. list("name1"=file1, "name2"=file2). If not named, default file names are assigned.

Value

Upset plot of overlapping peaks

```
### Load Data ###
data("encode_H3K27ac") # load example data
data("CnT_H3K27ac") # load example data

### Create Named List ###
peakfile <- list("encode"=encode_H3K27ac, "CnT"=CnT_H3K27ac)

### Run ###
my_plot <- overlap_upset_plot(peaklist = peakfile)</pre>
```

peak_info

Summary of Peak Information

Description

This function outputs a table summarizing information on the peak files. Provides the total number of peaks and the percentage of peaks in blacklisted regions.

Usage

```
peak_info(peaklist, blacklist)
```

Arguments

peaklist A named list of peak files as GRanges object. Objects listed using list("name1"

= peak, "name2" = peak2).

blacklist A GRanges object containing blacklisted regions.

Value

A summary table of peak information

Examples

```
plot_ChIPseeker_annotation
```

Create ChIPseeker annotation plot

Description

This function annotates peaks using ChIPseeker::annotatePeak. It outputs functional annotation of each peak file in a barplot.

plot_chromHMM 25

Usage

```
plot_ChIPseeker_annotation(peaklist, annotation)
```

Arguments

peaklist A list of peak files as GRanges object. Files must be listed and named using

list(). e.g. list("name1"=file1, "name2"=file2). If not named, default

file names will be assigned.

annotation A TxDb annotation object from Bioconductor.

Value

barplot

Examples

```
### Load Data ###
data("CnT_H3K27ac") # example peakfile GRanges object
data("CnR_H3K27ac") # example peakfile GRanges object

### Create Named Peaklist ###
peaks <- list("CnT"=CnT_H3K27ac, "CnR"=CnR_H3K27ac)

## not run
# txdb<-TxDb.Hsapiens.UCSC.hg19.knownGene::TxDb.Hsapiens.UCSC.hg19.knownGene
# my_plot <- plot_ChIPseeker_annotation(peaklist = peaks
# annotation = txdb)</pre>
```

plot_chromHMM

Plot ChromHMM heatmap

Description

Creates a heatmap using outputs from ChromHMM using ggplot2. The function takes a list of peakfiles, performs ChromHMM and outputs a heatmap. ChromHMM annotation file must be loaded prior to using this function. ChromHMM annotations are aligned to hg19, and will be automatically lifted over to the genome_build to match the build of the peaklist.

Usage

```
plot_chromHMM(
   peaklist,
   chromHMM_annotation,
   cell_line = NULL,
   genome_build,
   interact = TRUE,
   return_data = FALSE
)
```

26 plot_corr

Arguments

A named list of peak files as GRanges object. If list is not named, default names will be assigned.

chromHMM_annotation
ChromHMM annotation list.

cell_line
If not cell_line, will replace chromHMM_annotation by importing chromHMM data for a given cell line using get_chromHMM_annotation.

genome_build
The human genome reference build used to generate peakfiles. "hg19" or "hg38".

interact
Default TRUE. By default, the heatmaps are interactive. IfFALSE, the function generates a static ChromHMM heatmap.

return_data Return the plot data as in addition to the plot itself.

Value

ChromHMM heatmap, or a named list.

Examples

plot_corr

Plot correlation of peak files

Description

Plot correlation by binning genome and measuring correlation of peak quantile ranking. This ranking is based on p-value or other peak intensity measure dependent on the peak calling approach.

Usage

```
plot_corr(
  peakfiles,
  reference,
  genome_build,
  bin_size = 5000,
```

plot_corr 27

```
keep\_chr = NULL,
  drop_empty_chr = FALSE,
 method = "spearman",
  intensity_cols = c("total_signal", "qValue", "Peak Score", "score"),
  interact = FALSE,
 workers = 1,
  show_plot = TRUE
)
```

Arguments

peakfiles

A list of peak files as GRanges object and/or as paths to BED files. If paths are provided, EpiCompare imports the file as GRanges object. EpiCompare also accepts a list containing a mix of GRanges objects and paths. Files must be listed and named using list(). E.g. list("name1"=file1, "name2"=file2). If no names are specified, default file names will be assigned.

reference

A named list containing reference peak file(s) as GRanges object. Please ensure that the reference file is listed and named i.e. list("reference_name" = reference_peak). If more than one reference is specified, individual reports for each reference will be generated. However, please note that specifying more than one reference can take awhile. If a reference is specified, it enables two analyses: (1) plot showing statistical significance of overlapping/nonoverlapping peaks; and (2) ChromHMM of overlapping/non-overlapping peaks.

genome_build

The build of **all** peak and reference files to calculate the correlation matrix on. If all peak and reference files are not of the same build use liftover_grlist to convert them all before running. Genome build should be one of hg19, hg38, mm9, mm10.

bin_size

Default of 100. Base-pair size of the bins created to measure correlation. Use smaller value for higher resolution but longer run time and larger memory usage.

keep_chr

Which chromosomes to keep.

method

drop_empty_chr Drop chromosomes that are not present in any of the peakfiles (default: FALSE). Default spearman (i.e. non-parametric). A character string indicating which correlation coefficient (or covariance) is to be computed. One of "pearson", "kendall", or "spearman": can be abbreviated.

intensity_cols Depending on which columns are present, this value will be used to get quantiles and ultimately calculate the correlations:

- "total signal": Used by the peak calling software SEACR. NOTE: Another SEACR column (e.g. "max_signal") can be used together or instead of "total signal".
- "qValue"Used by the peak calling software MACS2/3. Should contain the negative log of the p-values after multiple testing correction.
- "Peak Score": Used by the peak calling software HOMER.

interact

Default TRUE. By default, all heatmaps are interactive. If set FALSE, all heatmaps in the report will be static.

workers

Number of cores to parallelise across (in applicable functions).

show_plot

Show the plot.

28 plot_enrichment

Value

list with correlation plot (corr_plot) and correlation matrix (data)

Examples

plot_enrichment

Generate enrichment analysis plots

Description

This function runs KEGG and GO enrichment analysis of peak files and generates dot plots.

Usage

```
plot_enrichment(peaklist, annotation)
```

Arguments

peaklist A list of peak files as GRanges object. Files must be listed and named using

list(). e.g. list("name1"=file1, "name2"=file2). If not named, default

file names will be assigned.

annotation A TxDb annotation object from Bioconductor.

Value

KEGG and GO dot plots

```
### Load Data ###
data("CnT_H3K27ac") # example peakfile GRanges object
data("CnR_H3K27ac") # example peakfile GRanges object
### Create Named Peaklist ###
peaks <- list("CnT"=CnT_H3K27ac, "CnR"=CnR_H3K27ac)</pre>
```

plot_precision_recall 29

```
## not run
# txdb<-TxDb.Hsapiens.UCSC.hg19.knownGene::TxDb.Hsapiens.UCSC.hg19.knownGene
# my_plot <- plot_enrichment(peaklist = peaks,
# annotation = txdb)</pre>
```

plot_precision_recall Plot precision-recall curves

Description

Plot precision-recall curves (and optionally F1 plots) by iteratively testing for peak overlap across a series of thresholds used to filter peakfiles. Each GRanges object in peakfiles will be used as the "query" against each GRanges object in reference as the subject. Will automatically use any columns that are specified with thresholding_cols and present within each GRanges object to create percentiles for thresholding. *NOTE*: Assumes that all GRanges in peakfiles and reference are already aligned to the same genome build.

Usage

```
plot_precision_recall(
  peakfiles,
  reference,
  thresholding_cols = c("total_signal", "qValue", "Peak Score"),
  initial_threshold = 0,
  n_{threshold} = 10,
 max_threshold = 1,
 workers = 1,
  plot_f1 = TRUE,
  subtitle = NULL,
  color = "peaklist1",
  shape = color,
  facets = "peaklist2 ~ .",
  interact = FALSE,
  show_plot = TRUE
)
```

Arguments

peakfiles

A list of peak files as GRanges object and/or as paths to BED files. If paths are provided, EpiCompare imports the file as GRanges object. EpiCompare also accepts a list containing a mix of GRanges objects and paths. Files must be listed and named using list(). E.g. list("name1"=file1, "name2"=file2). If no names are specified, default file names will be assigned.

reference

A named list containing reference peak file(s) as GRanges object. Please ensure that the reference file is listed and named i.e. list("reference_name"

30 plot_precision_recall

= reference_peak). If more than one reference is specified, individual reports for each reference will be generated. However, please note that specifying more than one reference can take awhile. If a reference is specified, it enables two analyses: (1) plot showing statistical significance of overlapping/non-overlapping peaks; and (2) ChromHMM of overlapping/non-overlapping peaks.

thresholding_cols

Depending on which columns are present, GRanges will be filtered at each threshold according to one or more of the following:

- "total_signal": Used by the peak calling software SEACR. NOTE: Another SEACR column (e.g. "max_signal") can be used together or instead of "total signal".
- "qValue"Used by the peak calling software MACS2/3. Should contain the negative log of the p-values after multiple testing correction.
- "Peak Score": Used by the peak calling software HOMER.

initial_threshold

Numeric threshold that was provided to SEACR (via the parameter --ctrl) when calling peaks without an IgG control.

n_threshold Number of thresholds to test.

max_threshold Maximum threshold to test.

workers Number of cores to parallelise across (in applicable functions).

plot_f1 Generate a plot with the F1 score vs. threshold as well.

subtitle Plot subtitle.

color Variable to color data points by.
shape Variable to set data point shapes by.

facets This argument is soft-deprecated, please use rows and cols instead.

interact Default TRUE. By default, all heatmaps are interactive. If set FALSE, all heatmaps

in the report will be static.

show_plot Show the plot.

Value

list with data and precision recall and F1 plots

precision_recall 31

precision_recall

Compute precision-recall

Description

Compute precision and recall using each GRanges object in peakfiles as the "query" against each GRanges object in reference as the subject.

Usage

```
precision_recall(
  peakfiles,
  reference,
  thresholding_cols = c("total_signal", "qValue", "Peak Score"),
  initial_threshold = 0,
  n_threshold = 15,
  max_threshold = 1,
  workers = 1,
  ...
)
```

Arguments

peakfiles

A list of peak files as GRanges object and/or as paths to BED files. If paths are provided, EpiCompare imports the file as GRanges object. EpiCompare also accepts a list containing a mix of GRanges objects and paths. Files must be listed and named using list(). E.g. list("name1"=file1, "name2"=file2). If no names are specified, default file names will be assigned.

reference

A named list containing reference peak file(s) as GRanges object. Please ensure that the reference file is listed and named i.e. list("reference_name" = reference_peak). If more than one reference is specified, individual reports for each reference will be generated. However, please note that specifying more than one reference can take awhile. If a reference is specified, it enables two analyses: (1) plot showing statistical significance of overlapping/non-overlapping peaks; and (2) ChromHMM of overlapping/non-overlapping peaks.

thresholding_cols

Depending on which columns are present, GRanges will be filtered at each threshold according to one or more of the following:

- "total_signal": Used by the peak calling software SEACR. NOTE: Another SEACR column (e.g. "max_signal") can be used together or instead of "total_signal".
- "qValue"Used by the peak calling software MACS2/3. Should contain the negative log of the p-values after multiple testing correction.
- "Peak Score": Used by the peak calling software HOMER.

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initial_threshold

Numeric threshold that was provided to SEACR (via the parameter --ctrl)

when calling peaks without an IgG control.

n_threshold Number of thresholds to test.
max_threshold Maximum threshold to test.

workers Number of cores to parallelise across (in applicable functions).

... Arguments passed on to bpplapply apply_fun Iterator function to use.

verbose Print messages.

register_now Register the cores now with register (TRUE), or simply return the BPPARAM object (default: FALSE).

use_snowparam Whether to use SnowParam (default: TRUE) or MulticoreParam

(FALSE) when parallelising across multiple workers.

progressbar logical(1) Enable progress bar (based on plyr:::progress_text).

X Any object for which methods length, [, and [[are implemented.

FUN The function to be applied to each element of X.

Value

Overlap

Examples

rebin_peaks

Rebin peaks

Description

Standardise a list of peak files by rebinning them into fixd-width tiles across the genome.

Usage

```
rebin_peaks(
  peakfiles,
  genome_build,
  intensity_cols = c("total_signal", "qValue", "Peak Score", "score"),
  bin_size = 5000,
```

33 rebin_peaks

```
keep\_chr = NULL,
  drop_empty_chr = FALSE,
  as\_sparse = TRUE,
 workers = 1,
  verbose = TRUE,
)
```

Arguments

peakfiles

A list of peak files as GRanges object and/or as paths to BED files. If paths are provided, EpiCompare imports the file as GRanges object. EpiCompare also accepts a list containing a mix of GRanges objects and paths. Files must be listed and named using list(). E.g. list("name1"=file1, "name2"=file2). If no names are specified, default file names will be assigned.

genome_build

The build of **all** peak and reference files to calculate the correlation matrix on. If all peak and reference files are not of the same build use liftover_grlist to convert them all before running. Genome build should be one of hg19, hg38, mm9, mm10.

intensity_cols Depending on which columns are present, this value will be used to get quantiles and ultimately calculate the correlations:

- "total signal": Used by the peak calling software SEACR. NOTE: Another SEACR column (e.g. "max_signal") can be used together or instead of "total signal".
- "qValue"Used by the peak calling software MACS2/3. Should contain the negative log of the p-values after multiple testing correction.
- "Peak Score": Used by the peak calling software HOMER.

bin_size

Default of 100. Base-pair size of the bins created to measure correlation. Use smaller value for higher resolution but longer run time and larger memory usage.

keep_chr

Which chromosomes to keep.

drop_empty_chr

Drop chromosomes that are not present in any of the peakfiles (default: FALSE).

as_sparse

Return the rebinned peaks as a sparse matrix (default: TRUE), which is more efficiently stored than a dense matrix (FALSE).

workers

Number of cores to parallelise across (in applicable functions).

verbose

Print messages.

. . .

Arguments passed on to bpplapply

apply_fun Iterator function to use.

register_now Register the cores now with register (TRUE), or simply return the BPPARAM object (default: FALSE).

use_snowparam Whether to use SnowParam (default: TRUE) or MulticoreParam (FALSE) when parallelising across multiple workers.

progressbar logical(1) Enable progress bar (based on plyr:::progress_text).

X Any object for which methods length, [, and [[are implemented.

FUN The function to be applied to each element of X.

34 tidy_peakfile

Value

Binned peaks matrix

Examples

tidy_peakfile

Tidy peakfiles in GRanges

Description

This function filters peak files by removing peaks in blacklisted regions and in non-standard chromosomes. It also checks that the input list of peakfiles is named. If no names are provided, default file names will be used.

Usage

```
tidy_peakfile(peaklist, blacklist)
```

Arguments

peaklist A named list of peak files as GRanges object. Objects must be named and listed

using list(). e.g. list("name1"=file1, "name2"=file2) If not named, de-

fault names are assigned.

blacklist Peakfile specifying blacklisted regions as GRanges object.

Value

list of GRanges object

```
### Load Data ###
data("encode_H3K27ac") # example peakfile GRanges object
data("CnT_H3K27ac") # example peakfile GRanges object
data("hg19_blacklist") # blacklist region for hg19 genome

### Create Named Peaklist ###
peaklist <- list("encode"=encode_H3K27ac, "CnT"=CnT_H3K27ac)</pre>
```

translate_genome 35

translate_genome

Translate genome

Description

Translate the name of a genome build from one format to another.

Usage

```
translate_genome(
  genome,
  style = c("UCSC", "Ensembl", "NCBI"),
  default_genome = NULL,
  omit_subversion = TRUE
)
```

Arguments

genome A character vector of genomes equivalent to UCSC version or Ensembl Assemblies

style A single value equivalent to "UCSC" or "Ensembl" specifying the output genome default_genome Default genome build when genome is NULL.

omit_subversion

Omit any subversion suffixes after the ".".

Value

Standardized genome build name as a character string.

```
genome <- translate_genome(genome="hg38", style="Ensembl")
genome2 <- translate_genome(genome="mm10", style="UCSC")</pre>
```

36 tss_plot

tss_plot

Read count frequency around TSS

Description

This function generates a plot of read count frequency around TSS.

Usage

```
tss_plot(
  peaklist,
  annotation,
  upstream = 3000,
  downstream = upstream,
  conf = 0.95,
  resample = 500,
  workers = 1
)
```

Arguments

A list of peak files as GRanges object. Files must be listed and named using peaklist list(). e.g. list("name1"=file1, "name2"=file2) If not named, default file names will be assigned. A TxDb annotation object from Bioconductor. annotation upstream upstream from TSS site downstream downstream from TSS site conf Confidence interval threshold estimated by bootstrapping (0.95 means 95 Argument passed to plotAvgProf. resample Number of bootstrapped iterations to run. Argument passed to plotAvgProf. Number of cores to parallelise bootstrapping across. Argument passed to plotAvgworkers

Value

profile plot in a list.

Prof.

width_boxplot 37

width_boxplot

Peak width boxplot

Description

This function creates boxplots showing the distribution of widths in each peak file.

Usage

```
width_boxplot(peaklist)
```

Arguments

peaklist

A list of peak files as GRanges object. Files must be named and listed using list(). e.g. list("name1"=file1, "name2"=file2)

Value

A boxplot of peak widths.

Examples

```
### Load Data ###
data("encode_H3K27ac") # example peaklist GRanges object
data("CnT_H3K27ac") # example peaklist GRanges object
### Create Named Peaklist ###
peaks <- list("encode"=encode_H3K27ac, "CnT"=CnT_H3K27ac)
### Run ###
my_plot <- width_boxplot(peaklist = peaks)</pre>
```

write_example_peaks

Write example peaks

Description

Write example peaks datasets to disk.

Usage

```
write_example_peaks(
  dir = file.path(tempdir(), "processed_results"),
  datasets = c("encode_H3K27ac", "CnT_H3K27ac", "CnR_H3K27ac")
)
```

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Arguments

dir Directory to save peak files to.

datasets Example datasets from **EpiCompare** to write.

Value

Named vector of paths to saved peak files.

```
save_paths <- EpiCompare::write_example_peaks()</pre>
```

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