R documentation

of all in 'man/'

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Contents

alla		Calculo script L	ea Ui	nder	Curv	e (A	UC)	and	Diff	eren	ces	of A	UC	for	Tr	an	-
Index			 														28
	universegroup		 													•	26
	tepr																
	retrieveanno																
	preprocessing		 														20
	plotmetagenes		 														19
	plothistoknee		 														17
	plotecdf																
	plotauc																
	meandifference																
	makewindows																
	kneeid																
	joinfiles																11
	genesECDF																9
	countna																8
	checkexptab																6 7
	blacklisthighmap																5
	averageandfilterexprs																4
	attenuation																2
	allauc																1

Description

This function computes the Area Under Curve (AUC) and the differences of AUC between two conditions for a list of transcript data. It supports parallel computation for efficiency.

2 allauc

Usage

```
allauc(bytranslistmean, expdf, nbwindows, nbcpu = 1, dontcompare = NULL,
controlcondname = "ctrl", stresscondname = "HS", showtime = FALSE,
verbose = TRUE)
```

Arguments

bytranslistmean

A list of data frames, each containing transcript level data with mean values for

one or more conditions.

expdf A data frame containing experiment data that should have columns named 'con-

dition', 'replicate', 'strand', and 'path'.

nbwindows An integer specifying the number of windows to consider for AUC calculations.

nbcpu An integer specifying the number of CPU cores to use for parallel processing on

bytranslistmean. Defaults to 1.

dontcompare An optional parameter to specify any conditions to exclude from the compari-

son. Defaults to NULL.

controlcondname

A string specifying the name of the control condition Defaults to "ctrl".

stresscondname A string specifying the name of the stress condition. Defaults to "HS".

showtime A logical value indicating if the duration of the function processing should be

indicated before ending. Defaults to FALSE.

verbose A logical value indicating whether to print progress messages Defaults to TRUE.

Details

The function first checks if exactly two conditions are present in 'expdf'. If so, it computes the differences in AUC between the two conditions using a Kolmogorov-Smirnov test and calculates the AUC for all conditions against a reference line (y=x). Results are merged by transcript and include adjusted p-values.

Value

A data frame containing the AUC and dAUC results for each transcript, along with associated statistical information.

See Also

```
[genesECDF]
```

```
# Example usage of allauc function
# results <- allauc(bytranslistmean, expdf, nbwindows = 100, nbcpu = 4)</pre>
```

attenuation 3

attenuation	Calculate Attenuation from AUC and Other Transcript Features

Description

This function computes the attenuation values for each window of each transcript based on the data frames obtained with the functions 'allauc', 'kneeid', and 'countna'.

Usage

```
attenuation(allaucdf, kneedf, matnatrans, bytranslistmean, expdf, dfmeandiff,
nbcpu = 1, significant = FALSE, replaceval = NA, pval = 0.1,
showtime = FALSE, verbose = TRUE)
```

Arguments

Suments	
allaucdf	A data frame containing AUC results for transcripts (see allauc).
kneedf	A data frame containing the inflection points (see kneeid).
matnatrans	A data frame containing the number of missing values per transcript (see countna).
bytranslistmea	n
	A list of data frames with mean values by transcripts.
expdf	A data frame containing experiment data that should have columns named 'condition', 'replicate', 'strand', and 'path'.
dfmeandiff	A data frame containing means and differences in mean values, if more than one condition. (see meandifference).
nbcpu	An integer specifying the number of CPU cores to use for parallel processing. The parallelization is done on bytranslistmean whose number of elements is equal to the number of lines provided as input of 'averageandfilterexprs'. Defaults to 1.
significant	A logical indicating whether to filter out non-significant attenuation values. Defaults to FALSE.
replaceval	A value to replace non-significant attenuation values Defaults to NA.
pval	A numeric value specifying the p-value threshold for significance of the KS test. Defaults to 0.1.
showtime	A logical value indicating if the duration of the function processing should be indicated before ending. Defaults to FALSE.
verbose	A logical value indicating whether to print progress messages Defaults to TRUE.

Details

The function merges several data frames to create a comprehensive dataset for each transcript. It computes mean values for the "up" and "down" segments of the transcript. The direction is determined by comparing the coordinates to the knee values. up = coord < knee and down = coord > knee. The up and down indexes are then retrieved and the attenuation scores are computed as: att <- 100 - downmean / upmean * 100

4 averageandfilterexprs

Value

A data frame containing the computed attenuation values along with associated transcript information.

See Also

```
[allauc()], [kneeid()], [countna()], [meandifference()]
```

Examples

```
# Example usage of attenuation function
#result <- attenuation(allaucdf, kneedf, matnatrans, bytranslistmean,
# expdf, dfmeandiff, nbcpu = 4)</pre>
```

averageandfilterexprs Calculate Average Expression and Filter Transcript Data

Description

This function calculates the average expression levels for transcripts from a provided expression data frame and filters out transcripts based on a specified expression threshold. The function also renames the columns in the output data frame to include mean expression values.

Usage

```
averageandfilterexprs(expdf, alldf, expthres, showtime = FALSE,
verbose = TRUE)
```

Arguments

expdf	A data frame containing experiment data that should have columns named 'condition', 'replicate', 'strand', and 'path'.
alldf	A data frame containing all transcript-related information, including biotype, chromosome, coordinates, transcript, gene, strand, window, ID and scores retrieved from the bedgraph files.
expthres	A numeric value specifying the expression threshold. Transcripts with average expression values below this threshold will be filtered out from the returned transcript vector.
showtime	A logical value indicating if the duration of the function processing should be indicated before ending. Defaults to FALSE.
verbose	A logical value indicating whether to print progress messages Defaults to TRUE.

Value

A list containing:

maintable The original data frame containing all transcript data.

exptranstab A character vector of transcripts that meet the filtering criteria.

blacklisthighmap 5

Examples

```
# Example usage of averageandfilterexprs
# result <- averageandfilterexprs(expdf, alldf, expthres = 10)</pre>
```

blacklisthighmap

Blacklist High Mappability Regions in Genomic Data

Description

This function processes genomic data to remove scores that fall within blacklisted regions or have low mappability, and computes weighted means for overlapping windows. The process ensures the integrity of genomic scores by focusing on high mappability regions and excluding blacklisted intervals.

Usage

```
blacklisthighmap(maptrackpath, blacklistshpath, exptabpath,
  nbcputrans, allwindowsbed, windsize, genomename, saveobjectpath = NA,
  tmpfold = "./tmp", reload = FALSE, showtime = FALSE, showmemory = FALSE,
  chromtab = NA, verbose = TRUE)
```

Arguments

maptrackpath	Character string. Path to the mappability track file.
exptabpath	Path to the experiment table file containing a table with columns named 'condition', 'replicate', 'strand', and 'path'.
nbcputrans	Number of CPU cores to use for transcript-level operations.
allwindowsbed	Data frame. BED-formatted data frame obtained with the function 'makewindows'.
windsize	An integer specifying the size of the genomic windows.
genomename	Character string. A valid UCSC genome name. It is used to retrieve chromosome metadata, such as names and lengths.
saveobjectpath	Path to save intermediate R objects. Default is 'NA' and R objects are not saved.
tmpfold	A character string specifying the temporary folder for saving output files. The temporary files contain the scores for each bedgraph on each chromosome.
reload	Logical. If 'TRUE', reloads existing saved objects to avoid recomputation. Default is 'FALSE'. If the function failed during object saving, make sure to delete the corresponding object.
showtime	A logical value indicating whether to display processing time.
showmemory	A logical value indicating whether to display memory usage during processing.
chromtab	A Seqinfo object retrieved with the rtracklayer method SeqinfoForUCSCGenome. If NA, the method is called automatically. Default is NA.
verbose	A logical value indicating whether to display detailed processing messages.
blacklistpath	Character string. Path to the blacklist regions file.

6 checkexptab

Details

The 'blacklisthighmap' function iterates through chromosomes, processes genomic scores by removing those overlapping with blacklisted regions, and ensures that scores within windows are computed using a weighted mean when overlaps occur. The function uses parallel processing for efficiency and supports saving (saveobjectpath) and reloading (reload) intermediate results to optimize workflow.

The main steps include: - Reading and processing bedGraph values. - Removing scores overlapping with blacklisted or low mappability regions. - Computing weighted means for overlapping scores in genomic windows. - Saving the processed results to specified path (tmpfold).

If chromtab is left to NA, the chromosome information is automatically retrieved from the UCSC server using 'genomename'. Otherwise, the Seqinfo object can be retrieved with: chromtab <- rtracklayer::SeqinfoForUCSCGenome(genomename)

Value

This function does not return a value directly. It saves intermediate results to 'tmpfold'. These intermediates files are then combined by the function 'createtablescores'.

See Also

[createtablescores][makewindows]

```
# Define paths to required files
maptrackpath <- "path/to/maptrack.bed"</pre>
blacklistshpath <- "path/to/blacklist.bed"</pre>
exptabpath <- "path/to/experiments.csv"</pre>
allwindowsbed <- data.frame(...)</pre>
# Run the function
results <- blacklisthighmap(</pre>
    maptrackpath = maptrackpath,
    blacklistshpath = blacklistshpath,
    exptabpath = exptabpath,
    nbcputrans = 4,
    allwindowsbed = allwindowsbed,
    windsize = 200,
    genomename = "hg38",
    saveobjectpath = "output/",
    tmpfold = "./tmp",
    reload = FALSE,
    showtime = TRUE,
    showmemory = FALSE,
    verbose = TRUE)
```

checkexptab 7

Description

The 'checkexptab' function verifies the structure and content of an experiment table to ensure it meets specific formatting requirements. It checks for the presence of required columns, and validates that the 'direction' and 'strand' columns contain only allowable values.

Usage

```
checkexptab(exptab)
```

Arguments

exptab

A data frame representing the experiment table. The table must contain the following columns: "condition", "replicate", "direction", and "strand".

Details

The function performs the following checks: - The column names of 'exptab' must match exactly: ""condition"', ""replicate"', ""direction", and ""strand"'. - The 'direction' column must contain only ""forward"' and ""reverse"'. - The 'strand' column must contain only ""plus" and ""minus"'.

Value

If the experiment table is valid, the function returns 'NULL'. If the table is invalid, the function throws an error specifying the issue.

```
## Not run:
  # Create a valid experiment table
  exptab <- data.frame(</pre>
    condition = c("cond1", "cond2"),
    replicate = c(1, 1),
    direction = c("forward", "reverse"),
    strand = c("plus", "minus")
  checkexptab(exptab) # Should pass without errors
  # Invalid experiment table (wrong column names)
  invalid_exptab <- data.frame(</pre>
    cond = c("cond1", "cond2"),
    rep = c(1, 1),
    dir = c("forward", "reverse"),
    str = c("+", "-")
  checkexptab(invalid_exptab) # Will throw an error
## End(Not run)
```

8 countna

countna	Count NA values per transcript and condition

Description

This function takes a list of expression data frames, a condition information data frame, and counts the number of NA values for each transcript based on strand and condition. NA represent missing scores that were filtered out from the black list and mappability track. The function operates in parallel on transcripts to speed up the process using multiple CPU cores.

Usage

```
countna(allexprsdfs, expdf, nbcpu = 1, showtime = FALSE, verbose = TRUE)
```

Arguments

allexprsdfs	A list of data frames containing expression data. The first element is assumed to be the main table. The second element is a vector of transcript names that passed the filtering of 'averageandfilterexprs'.
expdf	A data frame containing experiment data that should have columns named 'condition', 'replicate', 'strand', and 'path'.
nbcpu	An integer specifying the number of CPU cores to use for parallel computation on transcripts. The number of transcripts is equal to the number of lines provided as input of 'averageandfilterexprs'. Defaults to 1.
showtime	A logical value indicating if the duration of the function processing should be indicated before ending. Defaults to FALSE.
verbose	A logical flag indicating whether to print progress messages. Defaults to TRUE.

Value

A data frame where each row corresponds to a transcript, along with its associated gene, strand, and the count of NA values.

See Also

[averageandfilterexprs]

```
# Assuming allexprsdfs is a list of data frames and expdf contains the
# conditions:
# result <- countna(allexprsdfs, expdf, nbcpu = 4)</pre>
```

createtablescores 9

cre	eatetablescores	Create a Unified Table of Scores

Description

This function processes and combines table scores of each bedgraph and each chromosome stored in the temporary folder into a unified table.

Usage

```
createtablescores(tmpfold, exptabpath, showmemory = FALSE, showtime = TRUE,
    savefinaltable = TRUE, finaltabpath = "./", finaltabname = "anno.tsv",
    verbose)
```

Arguments

tmpfold	A string specifying the temporary folder containing the score files created with the function 'blacklisthighmap'.
exptabpath	Path to the experiment table file containing a table with columns named 'condition', 'replicate', 'strand', and 'path'.
showmemory	Logical; if 'TRUE', memory usage is printed during processing. Default is 'FALSE'.
showtime	Logical; if 'TRUE', the execution time of the function is printed. Default is 'TRUE'.
savefinaltable	Logical; if 'TRUE', the resulting table is saved to disk. Default is 'TRUE'.
finaltabpath	A string specifying the directory where the final table should be saved. Default is '"./"'.
finaltabname	A string specifying the name of the final table file. Default is "anno.tsv".
verbose	Logical; if 'TRUE', detailed messages are printed during execution.

Details

This function first merges files belonging to the same experiment and direction. These files are combined into a single table providing two columns per experiment. The first gives the name of the experiment and the second the scores. The resulting table also includes annotations for each transcript.

Value

A data frame containing the unified table of scores.

See Also

[blacklisthighmap]

10 genesECDF

Examples

```
# Example usage:
tmpfold <- "path/to/temp/folder"
exptabpath <- "path/to/experiment_table.csv"
finaltab <- createtablescores(tmpfold = tmpfold, exptabpath = exptabpath,
    showmemory = TRUE, showtime = TRUE, savefinaltable = TRUE,
    finaltabpath = "./results", finaltabname = "final_scores.tsv",
    verbose = TRUE)</pre>
```

genesECDF

Compute ECDF for Genes Based on Expression Data

Description

This function calculates the empirical cumulative distribution function (ECDF) for expressed genes across multiple transcripts. It processes the expression data to filter out non-expressed transcripts, compute ECDF values for each transcript, and combine the results into a unified data frame. The function operates in parallel for speed optimization.

Usage

```
genesECDF(allexprsdfs, expdf, nbcpu = 1, rounding = 10,
showtime = FALSE, verbose = TRUE)
```

Arguments

allexprsdfs	A list of data frames where the first element is the main expression data frame and the second element contains the names of the expressed transcripts (see 'averageandfilterexprs').
expdf	A data frame containing experiment data that should have columns named 'condition', 'replicate', 'strand', and 'path'.
nbcpu	An integer specifying the number of CPU cores to use for parallel computation. Default is 1.
rounding	An integer specifying the rounding factor for computing ECDF. Default is 10.
showtime	A logical value indicating if the duration of the function processing should be indicated before ending. Defaults to FALSE.
verbose	A logical flag indicating whether to print progress messages. Default is TRUE.

Details

The function performs several steps:

- 1. Filters the main expression table to retain only the expressed transcripts.
- 2. Splits the data by each transcript.
- 3. For each transcript, computes ECDF values for the score columns while respecting the strand orientation ("plus" or "minus").
- 4. Combines the ECDF results into a final data frame.

The function uses parallel processing to compute ECDF for each transcript simultaneously, making it faster on systems with multiple CPU cores.

joinfiles 11

Value

A list containing two elements:

concatdf A data frame with ECDF results for each transcript.

nbrows An integer indicating the number of rows in each transcript table.

See Also

[averageandfilterexprs]

Examples

```
# Assuming allexprsdfs is a list of data frames and expdf contains the
# conditions:
# result <- genesECDF(allexprsdfs, expdf, rounding = 10, nbcpu = 4,
# verbose = TRUE)</pre>
```

joinfiles

Join Bedgraph Files for Protein-Coding and IncRNA Data

Description

The 'joinfiles' function processes bedgraph files located in the specified working directory, joins data related to protein-coding and lncRNA annotations, and outputs a combined result as a TSV file. The function retrieves bedgraph files matching a specific pattern, processes the files to create windows-based summaries, and merges annotations for protein-coding and lncRNA biotypes. The resulting data is written to an output file.

Usage

```
joinfiles(workingdir = ".", window = 200, bgpattern = "*.bg",
   protscoredir = "protein_coding_score", lncscoredir = "lncRNA_score",
   outtsv = "dTAG_Cugusi_stranded_20230810.tsv", verbose = TRUE)
```

Arguments

workingdir

tory (".").
The window size used for joining the score files. Defaults to 200.
A file pattern to identify bedgraph files. Defaults to "*.bg".
Directory containing the protein-coding score files. Defaults to "protein_coding_score".

Incscoredir Directory containing the lncRNA score files. Defaults to "lncRNA_score".

The directory containing bedgraph files. Defaults to the current working direc-

outtsv The output TSV filename where the merged data will be saved. Defaults to

"dTAG_Cugusi_stranded_20230810.tsv".

nbcpu An integer specifying the number of CPU cores to use for parallel computation.

Default is 1.

verbose Logical flag to enable verbose output during the function execution. Defaults to

'TRUE'.

12 kneeid

Value

The data.frame with the complete set of annotations and scores.

Examples

```
## Not run:
    joinfiles(workingdir = "data", window = 100, bgpattern = "*.bedgraph",
        protscoredir = "prot_scores", lncscoredir = "lnc_scores",
        outtsv = "results.tsv")
## End(Not run)
```

kneeid

Identify the Knee and Max ECDF Differences for Each Transcript

Description

This function identifies the knee point (i.e., point of maximum change) and the maximum difference in the empirical cumulative distribution function (ECDF) for each transcript, across different experimental conditions.

Usage

```
kneeid(transdflist, expdf, nbcpu = 1, showtime = FALSE, verbose = TRUE)
```

Arguments

transdflist	A list of data frames where each data frame contains transcript data with ECDF
	values for each condition.
expdf	A data frame containing experiment data that should have columns named 'condition', 'replicate', 'strand', and 'path'.
nbcpu	An integer specifying the number of CPU cores to use for parallel computation. The parallelization is performed on the elements of transdflist. Defaults to 1.
showtime	A logical value indicating if the duration of the function processing should be indicated before ending. Defaults to FALSE.
verbose	A logical flag indicating whether to print progress messages. Defaults to TRUE.

Value

A data frame where each row corresponds to a transcript and contains the coordinates of the knee point and the maximum ECDF difference for each condition.

```
# Assuming transdflist is a list of transcript data frames and expdf contains
# conditions for each experiment:
# result <- kneeid(transdflist, expdf, nbcpu = 4, verbose = TRUE)</pre>
```

makewindows 13

Description

This functions uses the annotations filtered from gencode (see retrieveanno). It removes any ensembl names containing "PAR_Y". It filters out intervals smaller than windsize and splits each transcript into "windsize" windows.

Usage

```
makewindows(allannobed, windsize, nbcputrans = 1, verbose = TRUE,
    saveobjectpath = NA, showtime = FALSE)
```

Arguments

allannobed	A data frame which is the result of 'retrieveanno'.
windsize	An integer specifying the number of windows into which each gene annotation should be divided.
nbcputrans	Number of CPU cores to use for transcript-level operations. Defaults to 1.
verbose	A logical value indicating whether to display progress messages. Defaults to 'TRUE'.
saveobjectpath	A character string specifying the directory path where the output object should be saved as an '.rds' file. If 'NA', the object is not saved. Defaults to 'NA'.
showtime	A logical value indicating whether to display the runtime of the function. Defaults to 'FALSE'.

Details

The function filters out annotations with intervals smaller than the specified number of windows ('windsize'). It uses parallel processing to enhance performance when splitting transcripts into fixed-size windows. The result includes metadata for each window, such as its chromosome, start and end coordinates, associated gene, and the window number.

Intermediate functions, such as '.computewindflist' and '.divideannoinwindows', handle computation and validation of windows. Gene intervals with the "PAR_Y" tag are excluded from the analysis.

Value

A data frame containing the split windows for each gene annotation. The output includes fields such as 'biotype', 'chr', 'coor1', 'coor2', 'transcript', 'gene', 'strand', and 'window'.

See Also

[retrieveanno]

14 meandifference

Examples

```
# Example data
annotations <- data.frame(
    start = c(1, 1001, 2001),
    end = c(1000, 2000, 3000),
    strand = c("+", "-", "+"),
    chrom = c("chr1", "chr1", "chr2"),
    ensembl = c("ENSG000001", "ENSG000002", "ENSG000003"),
    symbol = c("Gene1", "Gene2", "Gene3"),
    biotype = c("protein_coding", "lncRNA", "protein_coding")
)
result <- makewindows(allannobed = annotations, windsize = 5, nbcputrans = 2)</pre>
```

meandifference

Compute Mean and Differences of Scores for Each Condition

Description

This function calculates the mean values, mean Fx (ECDF) and ECDF differences (Fx) for expression data, across different experimental conditions.

Usage

```
meandifference(resultsecdf, expdf, nbwindows, showtime = FALSE,
verbose = TRUE)
```

Arguments

resultsecdf	A data frame containing ECDF results for each transcript and condition (see genesECDF).
expdf	A data frame containing experiment data that should have columns named 'condition', 'replicate', 'strand', and 'path'.
nbwindows	An integer representing the number of windows (or segments) in each transcript.
showtime	A logical value indicating if the duration of the function processing should be indicated before ending. Defaults to FALSE.
verbose	A logical flag indicating whether to print progress messages. Defaults to TRUE.

Value

A data frame that contains, for each condition:

- Mean values for the "value" and "Fx" columns (e.g., mean_value_ctrl, mean_Fx_ctrl).
- Differences between the Fx column and coordinate ratios (e.g., diff_Fx_ctrl).

```
# Assuming resultsecdf is a data frame with ECDF results and expdf contains
# conditions:
# result <- meandifference(resultsecdf, expdf, nbwindows = 200,
# verbose = TRUE)</pre>
```

plotauc 15

plotauc	Plot AUC Comparison Between Conditions

Description

This function generates scatterplots comparing the area under the curve (AUC) for control and stress conditions, with an option to highlight specific genes or groups. The plot can be saved as a file or displayed interactively.

Usage

```
plotauc(tab, genevec = NA, auc_ctrlname = "AUC_ctrl",
auc_stressname = "AUC_HS",
pvalkstestcolname = "adjFDR_p_dAUC_Diff_meanFx_HS_ctrl",
labelx = "AUC in Control", labely = "AUC in Stress", axismin_x = -10,
axismax_x = 100, axismin_y = -10, axismax_y = 100, maintitle = "",
subtitle = "", legendpos = "bottom", formatname = "pdf", outfold = ".",
outfile = "AUCcompare_pval", plottype = "pval", plot = FALSE,
universename = "Universe", groupname = "Group", verbose = TRUE)
```

Arguments

outfile

ε	Guilleites	
	tab	A data frame containing the AUC values for control and stress conditions, and other columns required for plotting (e.g., p-values or group memberships, see allauc).
	genevec	A vector of gene names to highlight on the plot, applicable when plottype is set to "pval". Default is NA.
	auc_ctrlname	The column name in tab for the AUC under control conditions. Default is "AUC_ctrl".
	<pre>auc_stressname pvalkstestcolna</pre>	The column name in tab for the AUC under stress conditions. Default is "AUC_HS".
		The column name in tab for the adjusted FDR p-values from the KS test. Default is "adjFDR_p_dAUC_Diff_meanFx_HS_ctrl".
	labelx	Label for the x-axis. Default is "AUC in Control".
	labely	Label for the y-axis. Default is "AUC in Stress".
	axismin_x	Minimum value for the x-axis. Default is −10.
	axismax_x	Maximum value for the x-axis. Default is 100.
	axismin_y	Minimum value for the y-axis. Default is -10.
	axismax_y	Maximum value for the y-axis. Default is 100.
	maintitle	Main title of the plot. Default is an empty string.
	subtitle	Subtitle of the plot. Default is an empty string.
	legendpos	Position of the legend. Default is "bottom".
	formatname	Format of the saved plot (e.g., "pdf", "png"). Default is "pdf".
	outfold	Output folder where the plot will be saved. Default is ".".

Name of the output file. Default is "AUCcompare_pval".

16 plotecdf

plottype	Type of plot to generate. Can be "pval" for p-value based plots or "groups" for group-based plots. Default is "pval".
plot	A logical flag indicating whether to display the plot interactively (TRUE) or save it to a file (FALSE). Default is FALSE.
universename	Column name in tab representing the universe group in group-based plots. Default is "Universe".
groupname	Column name in tab representing specific groups in group-based plots. Default is "Group".
verbose	A logical flag indicating whether to display detailed messages about the function's progress. Default is TRUE.

Details

The function supports two plot types:

- "pval": The plot highlights genes based on adjusted FDR p-values and can highlight specific genes provided in genevec.
- "groups": The plot highlights predefined groups, such as "Attenuated" and "Outgroup", within the data.

If plot = TRUE, the plot is displayed interactively. If plot = FALSE, the plot is saved to a file in the specified format and output folder.

Value

A plot comparing AUC values between control and stress conditions, either displayed or saved to a file.

See Also

[allauc]

Examples

```
# Assuming `tab` contains AUC values and p-values:
# plotauc(tab, genevec = c("Gene1", "Gene2"), plottype = "pval")
```

plotecdf

Plot Empirical Cumulative Distribution Function (ECDF)

Description

This function generates an ECDF plot to analyze transcription density relative to the distance from the transcription start site (TSS) across different conditions. The plot displays AUC values, Kolmogorov-Smirnov (KS) statistics, and knee points, with options to display or save the plot.

Usage

```
plotecdf(dfmeandiff, unigroupdf, expdf, genename, colvec, outfold = NA,
digits = 2, middlewind = 100, pval = 0.01, plot = FALSE, verbose = TRUE)
```

plotecdf 17

Arguments

dfmeandiff	A data frame containing the mean differences of transcription levels and cumulative distribution values (Fx) for different windows around the TSS (see meandifference).
unigroupdf	A data frame containing gene-specific statistics, including their belonging to Universe or Group (see universegroup).
expdf	A data frame containing experiment data that should have columns named 'condition', 'replicate', 'strand', and 'path'.
genename	A string specifying the name of the gene of interest to plot.
colvec	A vector of colors used to distinguish different conditions in the plot.
outfold	A string specifying the output folder where the plot will be saved if plot = FALSE. Default is NA.
digits	The number of decimal places to round the AUC and KS values. Default is 2.
middlewind	The index of the middle window representing the region centered around the TSS. Default is 100.
pval	A numeric value for the p-value threshold to determine the significance of the KS test. Default is 0.01.
plot	A logical flag indicating whether to display the plot interactively (TRUE) or save it to a file (FALSE). Default is FALSE.
formatname	String of the format of the saved plot. Possible values are "eps", "ps", "tex" (pictex), "pdf", "jpeg", "tiff", "png", "bmp", and "svg". Default is "pdf".
verbose	A logical flag indicating whether to display detailed messages about the function's progress. Default is TRUE.

Details

The function processes data related to transcription levels and cumulative transcription density for a given gene across multiple experimental conditions. The ECDF plot is constructed with optional annotation of key statistics such as AUC values and significant KS test results. Knee points, representing significant changes in transcription density, are also displayed if the KS test passes the specified p-value threshold.

Value

An ECDF plot showing the transcription density across windows around the TSS, with highlights for significant KS test results and knee points. The plot can either be displayed or saved as a file.

See Also

[meandifference], [universegroup]

```
# Assuming `dfmeandiff`, `unigroupdf`, and `expdf` contain the necessary
# data:
# plotecdf(dfmeandiff, unigroupdf, expdf, genename = "GeneX",
# colvec = c("blue", "red"))
```

18 plothistoknee

plothistoknee	Plot Histogram of Distance from TSS to Knee Point

Description

This function generates a histogram showing the distribution of the distance from the transcription start site (TSS) to the knee point for attenuated genes. The distance can be plotted either as a percentage of the gene length or in kilobases (kb).

Usage

```
plothistoknee(unigroupdf, plottype = "percent", xlimvec = NA,
binwidthval = NA, kneename = "knee_AUC_HS", plot = FALSE, outfold = ".",
formatname = "pdf", universename = "Universe", groupname = "Group",
verbose = TRUE)
```

Arguments

unigroupdf	A data frame containing gene-level statistics, including knee point data and group classification (see universegroup).
plottype	A string specifying the type of distance to plot. Options are "percent" for the percentage of the gene or "kb" for distance in kilobases. Default is "percent".
xlimvec	A numeric vector of length 2 specifying the limits of the x-axis. Default is NA, which automatically sets the limits based on plottype.
binwidthval	A numeric value for the width of the bins in the histogram. Default is NA, which automatically selects a bin width based on plottype.
kneename	A string specifying the name of the column in unigroupdf that contains the knee point data. Default is "knee_AUC_HS".
plot	A logical flag indicating whether to display the plot interactively (TRUE) or save it to a file (FALSE). Default is FALSE.
outfold	A string specifying the output folder where the plot will be saved if plot = FALSE. Default is the current directory.
formatname	A string specifying the format of the saved plot file. Default is "pdf".
universename	A string specifying the name of the column in unigroupdf that defines the universe of genes. Default is "Universe".
groupname	A string specifying the name of the column in unigroupdf that defines the group classification of genes. Default is "Group".
verbose	A logical flag indicating whether to display detailed messages about the function's progress. Default is TRUE.

Value

A histogram showing the distribution of the distance from the TSS to the knee point for attenuated genes. The plot can either be displayed interactively or saved to a file.

See Also

[universegroup]

plotmetagenes 19

Examples

```
# Assuming `unigroupdf` contains the necessary data:
# plothistoknee(unigroupdf, plottype = "kb", xlimvec = c(0, 300),
# binwidthval = 10)
```

plotmetagenes

Plot Metagenes for Gene Groups

Description

This function plots metagene profiles based on transcript data, comparing transcription density across conditions (e.g., control vs. stress). The function allows the user to plot metagenes for different gene groups such as attenuated genes, outgroup genes, the entire universe of genes, or all genes.

Usage

```
plotmetagenes(unigroupdf, dfmeandiff, plottype = "attenuation",
daucname = "dAUC_Diff_meanFx_HS_ctrl", auc_ctrlname = "AUC_ctrl",
auc_stressname = "AUC_HS", plot = FALSE, formatname = "pdf", outfold = ".",
verbose = TRUE)
```

Arguments

unigroupdf	A data frame containing gene-level information, including group classifications and dAUC data for different conditions (see universegroup).
dfmeandiff	A data frame containing mean transcription values and coordinates for each transcript (see meandifference).
plottype	A string specifying the group of genes to plot. Options are "attenuation", "outgroup", "universe", or "all". Default is "attenuation".
daucname	A string specifying the column name for the delta AUC value (difference between conditions). Default is "dAUC_Diff_meanFx_HS_ctrl".
auc_ctrlname	A string specifying the column name for the control condition AUC values. Default is "AUC_ctrl".
auc_stressname	A string specifying the column name for the stress condition AUC values. Default is "AUC_HS".
plot	A logical flag indicating whether to display the plot interactively (TRUE) or save it to a file (FALSE). Default is FALSE.
formatname	A string specifying the format of the saved plot file. Default is "pdf".
outfold	A string specifying the output folder where the plot will be saved if plot = FALSE. Default is the current directory.
verbose	A logical flag indicating whether to display detailed messages about the func-

tion's progress. Default is TRUE.

20 preprocessing

Details

This function summarizes mean transcription levels across genomic coordinates for different gene groups and plots the transcription density from the transcription start site (TSS) to the transcription termination site (TTS). The function can generate metagene plots for different gene groups such as attenuated, outgroup, or all genes, and compares transcription profiles between conditions (e.g., control vs. stress). The resulting plot helps visualize differences in transcriptional response between groups of genes under different conditions.

Value

A metagene plot comparing transcription density across conditions (e.g., control vs. stress) for the selected group of genes. The plot can either be displayed interactively or saved to a file.

See Also

```
[universegroup], [meandifference]
```

Examples

```
# Assuming `unigroupdf` and `dfmeandiff` contain the necessary data:
# plotmetagenes(unigroupdf, dfmeandiff, plottype = "universe", plot = TRUE)
```

preprocessing

Preprocess Experimental Data for Genomic Analysis

Description

This function orchestrates a pipeline for preprocessing genomic data, including filtering annotations, splitting transcripts into windows, retrieving bedgraph values, and generating a final annotated table.

Usage

```
preprocessing(exptabpath, gencodepath, windsize, maptrackpath,
blacklistshpath, genomename, nbcputrans = 1, finaltabpath = "./",
finaltabname = "anno.tsv", tmpfold = "./tmp", saveobjectpath = NA,
savefinaltable = TRUE, reload = FALSE, showtime = FALSE, showmemory = FALSE,
deletetmp = TRUE, chromtab = NA, verbose = TRUE)
```

Arguments

exptabpath Character. Path to the experiment table file.

gencodepath Character. Path to the Gencode annotation file.

windsize Integer. Window size for splitting transcripts.

maptrackpath Character. Path to the mappability track file.

blacklistshpath

Character. Path to the blacklist file.

genomename Character. Name of the genome assembly (e.g., "hg38").

nbcputrans Integer. Number of CPUs to use for transcript processing. Default is 1.

preprocessing 21

finaltabpath Character. Path where the final annotated table will be saved. Default is "./". finaltabname Character. Name of the final annotated table file. Default is "anno.tsv". tmpfold Character. Path to a temporary folder for intermediate files. Default is "./tmp". saveobjectpath Character. Path to save intermediate objects. Default is NA. savefinaltable Logical. Whether to save the final table to disk. Default is TRUE. reload Logical. Whether to reload intermediate objects if available. Default is FALSE. showtime Logical. Whether to display timing information. Default is FALSE. showmemory Logical. Whether to display memory usage information. Default is FALSE. deletetmp Logical. Whether to delete temporary files after processing. Default is TRUE. A Seginfo object retrieved with the rtracklayer method SeginfoForUCSCGenome. chromtab If NA, the method is called automatically. Default is NA. Logical. Whether to display detailed progress messages. Default is TRUE. verbose

Details

The 'preprocessing' function performs several key tasks: 1. Filters Gencode annotations to retrieve "transcript" annotations. 2. Differentiates between protein-coding (MANE_Select) and long non-coding (lncRNA, Ensembl_canonical) transcripts. 3. Splits transcripts into windows of size 'windsize'. 4. Processes bedgraph files to retrieve values, exclude blacklisted regions, and retain high-mappability intervals. 5. Generates a final annotated table with scores derived from the above steps.

Temporary files created during processing are optionally deleted at the end.

Value

A data frame representing the final table containing transcript information and scores on 'windsize' windows for all experiments defined in the experiment table (exptabpath).

See Also

[retrieveanno][makewindows][blacklisthighmap][createtablescores]

```
# Example usage of preprocessing:
preprocessing(
  exptabpath = "./example_exptab.tsv",
  gencodepath = "./gencode.v38.annotation.gtf",
  windsize = 200,
  maptrackpath = "./mappability_track.bed",
  blacklistshpath = "./blacklist.bed",
  genomename = "hg38",
  nbcputrans = 2,
  finaltabpath = "./results/",
  finaltabname = "final_annotated_table.tsv",
  tmpfold = "./tmp",
  saveobjectpath = "./saved_objects",
  savefinaltable = TRUE,
  reload = FALSE,
  showtime = TRUE,
  showmemory = TRUE,
```

22 retrieveanno

```
deletetmp = TRUE,
  verbose = TRUE
)
```

retrieveanno

Retrieve and Combine Annotation Information

Description

This function filters gencode annotations to retrieve "transcript". It then distinguishes transcripts coming from protein coding genes (MANE_Select) and those coming from long non-coding genes (lncRNA, Ensembl_canonical).

Usage

```
retrieveanno(exptabpath, gencodepath, saveobjectpath = NA, showtime = FALSE,
verbose = TRUE)
```

Arguments

exptabpath Path to the experiment table file containing a table with columns named 'condi-

tion', 'replicate', 'strand', and 'path'.

gencodepath Path to the GENCODE annotation file.

saveobjectpath Path to save intermediate R objects. Default is 'NA' and R objects are not saved.

showtime Logical. If 'TRUE', displays timing information. Default is 'FALSE'.

verbose Logical. If 'TRUE', provides detailed messages during execution. Default is

'TRUE'.

Details

The function performs the following steps: 1. Reads experimental data from the provided CSV file and validates it. 2. Reads genomic annotations from the gencode file and filters for transcripts. 3. Separately processes protein-coding and long non-coding RNA transcripts: - For protein-coding genes, selects the most representative (MANE_Select or Ensembl_canonical) transcripts. - For long non-coding RNAs, filters out transcripts with undesirable evidence levels. 4. Combines these annotations into a single data frame, labeling each transcript with its biotype. 5. Optionally saves the resulting data frame as an RDS file in the specified directory. 6. Optionally reports the total time taken for analysis.

Value

A data frame containing the combined annotation information for protein-coding and long non-coding RNA transcripts. If 'saveobjectpath' is not 'NA', the object is also saved as an RDS file in the specified directory.

tepr 23

Examples

```
# Example usage:
exptab_file <- "path/to/experiment_table.csv"
gencode_file <- "path/to/gencode_annotations.gtf"
output_dir <- "path/to/output_directory"

# Run the function with verbose output and timing enabled
annotations <- retrieveanno(
    exptabpath = exptab_file,
    gencodepath = gencode_file,
    saveobjectpath = output_dir,
    showtime = TRUE,
    verbose = TRUE
)</pre>
```

tepr

Perform the tepr differential nascent rna-seq analysis

Description

This function wraps the different steps of tepr to identify transcripts with a significantly different nascent rna-seq signal.

Usage

```
tepr(expdf, alldf, expthres, nbcpu = 1, rounding = 10, dontcompare = NULL, controlcondname = "ctrl", stresscondname = "HS", replaceval = NA, pval = 0.1, significant = FALSE, windsizethres = 50, countnathres = 20, meanctrlthres = 0.5, meanstressthres = 0.5, pvaltheorythres = 0.1, aucctrlthreshigher = -10, aucctrlthreslower = 15, aucstressthres = 15, attenuatedpvalksthres = 2, outgrouppvalksthres = 0.2, showtime = FALSE, verbose = TRUE)
```

Arguments

expdf	A data frame containing experiment data that should have columns named 'condition', 'replicate', 'strand', and 'path'.
alldf	A data frame containing all transcript-related information, including biotype, chromosome, coordinates, transcript, gene, strand, window, ID and scores retrieved from the bedgraph files.
expthres	A numeric value specifying the expression threshold. Transcripts with average expression values below this threshold will be filtered out from the returned transcript vector.
nbcpu	An integer specifying the number of CPU cores to use for parallel computation on transcripts. The number of transcripts is equal to the number of lines provided as input of 'averageandfilterexprs'. Defaults to 1.
rounding	An integer specifying the rounding factor for computing ECDF. Default is 10.
dontcompare	An optional parameter to specify any conditions to exclude from the comparison. Defaults to NULL.

24 tepr

controlcondname

pval

A string specifying the name of the control condition Defaults to "ctrl".

stresscondname $\ A$ string specifying the name of the stress condition. Defaults to "HS".

replaceval A value to replace non-significant attenuation values Defaults to NA.

A numeric value specifying the p-value threshold for significance of the KS test. Defaults to 0.1.

significant A logical indicating whether to filter out non-significant attenuation values. De-

faults to FALSE.

windsizethres A numeric threshold for the minimum window size. Default is 50.

countnathres A numeric threshold for the maximum number of missing data points for an

experiment (NA values). Default is 20.

meanctrlthres A numeric threshold for the minimum mean transcription value in the control

condition. Default is 0.5.

meanstressthres

A numeric threshold for the minimum mean transcription value in the stress condition. Default is 0.5.

pvaltheorythres

A numeric threshold for the minimum p-value used to define the universe of genes. Default is 0.1.

aucctrlthreshigher

A numeric threshold for the lower bound of the control AUC value in the outgroup classification. Default is -10.

aucctrlthreslower

A numeric threshold for the upper bound of the control AUC value in the outgroup classification. Default is 15.

aucstressthres A numeric threshold for the minimum stress AUC value used to classify attenu-

ated genes. Default is 15.

attenuatedpvalksthres

A numeric threshold for the negative log10 of the p-value (from KS test) for defining attenuated genes. Default is 2.

outgrouppvalksthres

A numeric threshold for the maximum KS p-value used to define the outgroup.

Default is 0.2.

showtime A logical value indicating if the duration of the function processing should be

indicated before ending. Defaults to FALSE.

verbose A logical flag indicating whether to print progress messages. Defaults to TRUE.

Details

The tepr function calls successively:

- averageandfilterexprsThis function calculates the average expression levels for transcripts from alldf that was obtained with the 'preprocessing' function. It filters out transcripts based on the 'expthres' expression threshold. The function also renames the columns in the output data frame to include mean expression values. It returns a list containing the original alldf with the mean columns added and a character vector of transcripts that meet the filtering criteria.
- countnaIt takes the result of 'averageandfilterexprs' as input and counts the number of NA
 values for each transcript based on strand and condition. NA represent missing scores that
 were filtered out from the black list and mappability track. It returns a data frame where each
 row corresponds to a transcript, along with its associated gene, strand, and the count of NA
 values.

tepr 25

• genesECDFIt takes the result of 'averageandfilterexprs' as input and a) filters the main expression table to retain only the expressed transcripts; b) Splits the data by each transcript; c) For each transcript, computes ECDF values for the score columns while respecting the strand orientation ("plus" or "minus"); d) Combines the ECDF results into a final data frame. It returns a list containing two elements: A data frame with ECDF results for each transcript (concatdf) and an integer indicating the number of rows in each transcript table.

- meandifferenceIt takes the result of 'genesECDF' as input and calculates the mean values, mean Fx (ECDF) and ECDF differences (Fx) for expression data, across different experimental conditions. It returns a data frame that contains, for each condition: mean values for the "value" and "Fx" columns (e.g., mean_value_ctrl, mean_Fx_ctrl) and the differences between the Fx column and coordinate ratios (e.g., diff_Fx_ctrl).
- SplittingSplit the results of 'meandifference' by transcripts and stores the list into bytranslistmean.
- allaucIt uses the previously computed variable 'bytranslistmean' and it computes the Area Under Curve (AUC) and the differences of AUC between two conditions for a list of transcript data. It returns a data frame containing the AUC and dAUC results for each transcript, along with associated statistical information.
- kneeidIt uses the previously computed variable 'bytranslistmean' and identifies the knee point (i.e., point of maximum change) and the maximum difference in the empirical cumulative distribution function (ECDF) for each transcript, across different experimental conditions. It returns a data frame where each row corresponds to a transcript and contains the coordinates of the knee point and the maximum ECDF difference for each condition.
- attenuationIt uses the results of the previous functions and it computes the attenuation values for each window of each transcript. It returns a data frame containing the computed attenuation values along with associated transcript information.
- universegroupUsing the table produced by 'attenuation', it categorizes genes into a "Universe" and assigns them into groups such as "Attenuated" or "Outgroup" based on transcription data and thresholds. The universe is defined by thresholds for window size, missing data count, mean transcription levels, and p-values. Genes are further classified into groups based on conditions related to AUC and p-value thresholds. A transcript belongs to "Universe" if (window_size > windsizethres & Count_NA < countnathres & meanctrl > meanctrlthres & meanstress > meanstressthres & pvaltheory > pvaltheorythres). A transcript belongs to the groups: Attenuated: if Universe == TRUE & aucstress > aucstressthres & -log10(pvalks) > attenuatedpvalksthres. Outgroup: if Universe == TRUE & pvalks > outgrouppvalksthres & aucctrl > aucctrlthreshigher & aucctrl < aucctrlthreslower.

Value

A list of data.frame made of two elements. The first data.frame is the result of the function 'meandifference': For each condition, it provides the mean values for the "value" and "Fx" columns (e.g. mean_value_ctrl, mean_Fx_ctrl columns) as the differences between the Fx column and coordinate ratios (e.g., diff_Fx_ctrl column). The second data.frame is the result of the function 'universegroup': It contains columns concerning AUC, knee position, attenuation information, and columns defining the universe and groups (see details).

See Also

[averageandfilterexprs()], [countna()], [genesECDF()], [meandifference()], [allauc()], [kneeid()], [attenuation()], [universegroup()], [preprocessing()]

26 universegroup

Examples

```
# Example usage:
# exptabpath <- "exp.csv"
# alldfpath <- "result-preprocessing.tsv"
# expdf <- read.csv(exptabpath)
# alldf <- read.delim(alldfpath, header = FALSE)
# expthres <- 0.1
# reslist <- tepr(expdf, alldf, expthres)
# resmeandiff <- restlist[[1]]
# res <- reslist[[2]]</pre>
```

universegroup

Define Universe and Group of Genes Based on Expression Data

Description

This function categorizes genes into a "Universe" and assigns them into groups such as "Attenuated" or "Outgroup" based on transcription data and thresholds. The universe is defined by thresholds for window size, missing data count, mean transcription levels, and p-values. Genes are further classified into groups based on conditions related to AUC and p-value thresholds.

Usage

```
universegroup(completedf, controlname = "ctrl", stressname = "HS", windsizethres = 50, countnathres = 20, meanctrlthres = 0.5, meanstressthres = 0.5, pvaltheorythres = 0.1, aucctrlthreshigher = -10, aucctrlthreslower = 15, aucstressthres = 15, attenuatedpvalksthres = 2, outgrouppvalksthres = 0.2, showtime = FALSE, verbose = TRUE)
```

Arguments

completedf A data frame obtained with the function attenuation.

controlname A string representing the control condition name. Default is "ctrl".

 ${\tt stressname} \qquad \quad A \ string \ representing \ the \ stress \ condition \ name. \ Default \ is \ "HS".$

windsizethres A numeric threshold for the minimum window size. Default is 50.

countnathres A numeric threshold for the maximum number of missing data points (NA val-

ues). Default is 20.

meanctrlthres A numeric threshold for the minimum mean transcription value in the control

condition. Default is 0.5.

meanstressthres

A numeric threshold for the minimum mean transcription value in the stress

condition. Default is 0.5.

pvaltheorythres

A numeric threshold for the minimum p-value used to define the universe of genes. Default is 0.1.

aucctrlthreshigher

A numeric threshold for the lower bound of the control AUC value in the outgroup classification. Default is -10.

universegroup 27

aucctrlthreslower

A numeric threshold for the upper bound of the control AUC value in the outgroup classification. Default is 15.

aucstressthres A numeric threshold for the minimum stress AUC value used to classify attenuated genes. Default is 15.

attenuatedpvalksthres

A numeric threshold for the negative log10 of the p-value (from KS test) for defining attenuated genes. Default is 2.

outgrouppvalksthres

A numeric threshold for the maximum KS p-value used to define the outgroup.

Default is 0.2.

showtime A logical value indicating if the duration of the function processing should be

indicated before ending. Defaults to FALSE.

verbose A logical flag indicating whether to print progress messages. Defaults to TRUE.

Details

A transcript belongs to "Universe" if: window_size > windsizethres & Count_NA < countnathres & meanctrl > meanctrlthres & meanstress > meanstress thres & pvaltheory > pvaltheory thres

A transcript belongs to the groups: - **Attenuated**: if Universe == TRUE & aucstress > aucstressthres & $-\log 10$ (pvalks) > attenuatedpvalksthres - **Outgroup**: if Universe == TRUE & pvalks > outgroup-pvalksthres & aucctrl > aucctrlthreshigher & aucctrlthreshower

This function is useful for classifying genes in transcriptomics data based on their transcriptional response to different experimental conditions.

Value

A modified data frame with two additional columns: Universe, indicating whether each gene is part of the universe, and Group, classifying the genes into groups such as "Attenuated", "Outgroup", or NA.

See Also

[attenuation]

Examples

```
# Example usage:
```

```
# classified_df <- universegroup(completedf, controlname = "ctrl",</pre>
```

stressname = "HS")

Index

```
allauc, 1
attenuation, 2
average and filter exprs, 4
{\tt blacklisthighmap}, {\tt 5}
checkexptab, 6
countna, 7
createtablescores, 8
genesECDF, 9
joinfiles, 11
kneeid, 12
makewindows, 12
\  \, \text{meandifference}, \, 14
plotauc, 14
plotecdf, 16
plothistoknee, 17
plotmetagenes, 19
preprocessing, 20
retrieveanno, 22
tepr, 23
univer segroup, \color{red} \underline{26}
```