

Package ‘TrendCatcher’

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Description Identify dynamic gene from longitudinal/time course RNA-seq dataset.

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URL <https://wangxinge.github.io/TrendCatcherWorkshopBioc2022/>

BugReports

<https://github.com/wangxinge/TrendCatcherWorkshopBioc2022/issues/new/choose>

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cal_p

Calculate the significance of the dynamic signal for a single gene's single time expression. Compared to baseline NB confidence interval.

Description

Calculate the significance of the dynamic signal for a single gene's single time expression. Compared to baseline NB confidence interval.

Usage

```
cal_p(obs.val, size, mu)
```

Arguments

obs.val,	estimated value from non-baseline model.
size,	the size of negative binomial model.
mu,	the estimated mean of constant negative binomial model

Value

a numeric p-value for time t.

cal_time_p_single_gene

Calculate the significance of the dynamic signal for a single gene's over all the time points.

Description

Calculate the significance of the dynamic signal for a single gene's over all the time points.

Usage

```
cal_time_p_single_gene(const.output, spline.output)
```

Arguments

const.output,	list returned from fit_single_gene_const function.
spline.output,	dataframe returned from fit_single_gene_spline function.

Value

p-value for a single point.

Check_CountTable_Format

Check the Input Count Table Format

Description

This function takes the CSV file path of the count table provided by user. The input count table must be a CSV file, includes integer count table, first column are the GENE SYMBOL or GENE ENSEMBL, and first row are SAMPLE NAME. The count table is a rounded up after normalization and batch correction. Please check TrendCatcher QC functions for normalization and batch correction details. The row names must be in the format of "ProjectName_Time_Rep1" format. ProjectName is a string, can be single letter. Time is a integer. Rep is the replicate ID. This function checked the count table format, and order the sample columns based on its time order. It will return a right formatted count table to run TrendCatcher.

Usage

```
Check_CountTable_Format(count.table.path, min.low.count = 1)
```

Arguments

count.table.path,
 string contain the absolute path of the CSV file count table, with first column as GENE SYMBOL or GENE ENSEMBL and first row as SAMPLE NAME (with format composed by project name,time and replicateID, such as "Lung_0_Rep1")
 min.low.count,
 one numeric variable, the minimal count threshold for filtering low count within each time group. By default it is 1.

Value

A list object, contains "raw.df", original count table ordered by time and replicate ID; "count.table", filtered out low genes for further fitting; "removed.genes", low count genes.

Examples

```
example.file.path<-system.file("extdata", "Lung_DemoCountTable.csv", package = "TrendCatcher")
## Not run:
count<-Check_CountTable_Format(example.file.path, min.low.count = 1)

## End(Not run)
```

combine_p_single_gene *Combine multiple p-value using Fisher's p-value combination method.*

Description

Combine multiple p-value using Fisher's p-value combination method.

Usage

```
combine_p_single_gene(p.arr)
```

Arguments

p.arr, a vector of p-values.

Value

a numeric combined p-value.

curveFitting.new	<i>CurveFitting</i>
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Description

CurveFitting

Usage

```
curveFitting.new(perm.dat.1, points)
```

draw_CurveComp	<i>Draw DDEGs trajectories from two master.list object and also show LOESS curve fitting</i>
----------------	--

Description

This funcitons takes the master.list output from run_TrendCatcher, and merge.df output from draw_TimeHeatmap_GO and draw_TimeHeatmap_enrichR. And showing all the genes used for enrichment analysis and their logFC compared to previous break point.

Usage

```
draw_CurveComp(  
  master.list.1,  
  master.list.2,  
  ht.1,  
  pathway,  
  group.1.name,  
  group.2.name  
)
```

Arguments

master.list.1,	a list object. The output from run_TrendCatcher function, contains master.table element.
master.list.2,	a list object. The output from run_TrendCatcher function, contains master.table element.
ht.1,	TimeHeatmap object. The output from draw_TimeHeatmap_GO function, contains GO.df object.
pathway,	characters. Must be a biological pathway from GO.df, Description column.
group.1.name,	characters. For example, severe group.
group.2.name,	characters. For example, moderate group.

Value

A ggplot object and plot.

draw_CurveComp_Perm	<i>Draw DDEGs trajectories from two master.list object and also show LOESS curve fitting with permuation</i>
---------------------	--

Description

Draw DDEGs trajectories from two master.list object and also show LOESS curve fitting with permuation

Usage

```
draw_CurveComp_Perm(
  master.list.1,
  master.list.2,
  ht.1,
  pathway,
  group.1.name,
  group.2.name,
  n.perm = 500,
  parall = F,
  pvalue.threshold = 0.05
)
```

draw_GeneTraj	<i>Draw gene(s) trajectory with observed data and fitted data</i>
---------------	---

Description

This function takes the master.list object output from run_TrendCatcher function, and an array of gene(s). It will draw gene(s) trajectory with observed data and fitted data.

Usage

```
draw_GeneTraj(
  master.list,
  gene.symbol.arr,
  savepdf.path = NA,
  ncol = 5,
  nrow = 3,
  fig.width = 15,
  fig.height = 10
)
```

Arguments

- master.list, the list object returned from run_TrendCatcher.
 gene.symbol.arr, a character array. It must be a subset of row names from the master.list\$master.table\$Symbol. The Symbol column need get_GeneEnsembl2Symbol function to convert original ensembl ID into gene symbol.
 savepdf.path, an absolute file path to save the figure as PDF file. By default is NA, it will be printed.
 ncol, an integer variable. If more than one gene need to be plotted, it will layout as grid structure. This represents the number of column of the grid layout.
 nrow, an integer variable. If more than one gene need to be plotted, it will layout as grid structure. This represents the number of row of the grid layout.
 fig.width, a numeric variable. If save figure as PDF file, the width of the PDF file. By default is 15.
 fig.height, a numeric variable. If save figure as PDF file, the height of the PDF file. By default is 10.
 master.list.new, a list object. Output from the run_TrendCatcher with ID conversion to add Symbol column to master table.
 count.table, the count table of mRNA.
 gene.name, name of the gene you would like to plot.

Value

"arrangeable" "list" object.

draw_GOHeatmap

Draw GOHeatmap contianing Terms from TimeHeatmap and included Genes

Description

This function takes the master.list output from run_TrendCatcher, and merge.df output from draw_TimeHeatmap_GO and draw_TimeHeatmap_enrichR. And showing all the genes used for enrichment analysis and their logFC compared to previous break point.

Usage

```
draw_GOHeatmap(
  master.list,
  time.window = "",
  go.terms = "",
  merge.df = NA,
  logFC.thres = 2,
```

```

    figure.title = "",
    save.tiff.path = NA,
    tiff.res = 100,
    tiff.width = 1500,
    tiff.height = 1500
)

```

Arguments

<code>master.list</code> ,	a list object. The output from run_TrendCatcher function, contains <code>master.table</code> element.
<code>go.terms</code> ,	a character array. Must be an array of go terms from the <code>merge.df\$Description</code> .
<code>merge.df</code> ,	a dataframe. The output dataframe from output list of <code>draw_TimeHeatmap_GO</code> or <code>draw_TimeHeatmap_enrichR</code> . Use <code>\$merge.df</code> to obtain it.
<code>logFC.thres</code> ,	a numeric variable. The logFC threshold compared to each genes previous break point expression level. By default is 2, meaning for each gene, the current time window's expression level is 2-fold compared to previous break point's expression level.
<code>time.window</code> ,	a character. Must be one of the <code>merge.df\$t.name</code> .
<code>id.ensembl</code> ,	a logic variable. If using <code>ensembl</code> as the keytype. This must match the row name of <code>master.table</code> . By default is TRUE.
<code>save.as.PDF</code> ,	a character variable. If need to save the figure into PDF file. This must be an absolute file path. If not needed save as PDF file, set it to NA. By default is NA.
<code>pdf.width</code> ,	a numeric variable. The width of PDF file. By default is 13.
<code>pdf.height</code> ,	a numeric variable. The height of PDF file. By default is 15.

Value

A list object, including elements names `merge.df` and `time.heatmap`. `time.heatmap` is the `ggplot` object. `merge.df` includes all the `enrichR` enrichment result and activation/deactivation time.

Examples

```

## Not run:
example.file.path<-system.file("extdata", "BrainMasterList.rda", package = "TrendCatcher")
load(example.file.path)
th.obj<-draw_TimeHeatmap_GO(master.list = master.list)
merge.df<-th.obj$merg.df

time.window<-"0h-6h"
go.terms<-c("regulation of defense response", "leukocyte migration", "myeloid leukocyte migration", "leukocyte ch
"granulocyte chemotaxis", "cellular response to chemokine", "chemokine-mediated signaling pathway", "angiogenesis",
go.df<-draw_GOHeatmap(master.list = master.list, time.window = "0h-6h", go.terms = go.terms, id.ensembl = TRUE, me

## End(Not run)

```

draw_TimeHeatmap_enrichR

Draw Time-Heatmap Using enrichR

Description

This function takes the master.list output from run_TrendCatcher. And apply a time window sliding strategy to capture all the genes increased/decreased compared to its previous break point, and apply enrichR enrichment analysis.

Usage

```
draw_TimeHeatmap_enrichR(  
  master.list,  
  logFC.thres = 1,  
  top.n = 10,  
  dyn.gene.p.thres = 0.05,  
  dbs = "BioPlanet_2019",  
  term.width = 80,  
  OrgDb = "org.Mm.eg.db",  
  GO.enrich.p = 0.05,  
  figure.title = "",  
  save.tiff.path = NA,  
  tiff.res = 100,  
  tiff.width = 1500,  
  tiff.height = 1500  
)
```

Arguments

- | | |
|-------------------|--|
| master.list, | a list object. The output from run_TrendCatcher function, contains master.table element. |
| logFC.thres, | a numeric variable. The logFC threshold compared to each genes previous break point expression level. By default is 1, meaning for each gene, the current time window's expression level is 2-fold compared to previous break point's expression level. |
| top.n, | an integer variable. The top N GO enrichment term need to be shown in the Time-Heatmap for up and down regulated pathway. By default is 10. Top 20 GO terms, 10 from up-regulated pathway and 10 from down-regulated pathway will shown in Time-Heatmap. |
| dyn.gene.p.thres, | a numeric variable. The DDEGs dynamic p-value threshold. By default is 0.05. |
| dbs, | must one of the enrichR supported database name. To check the list, run dbs <- listEnrichrDbs() command. By default is "BioPlanet_2019". |

term.width,	an integer variable. The character length for each GO term. If one GO term is super long, we can wrap it into term.width of strings into multiple rows. By default if 80.
GO.enrich.p,	an numeric variable. The GO enrichment p-value threshold. By default if 0.05.
figure.title,	a character variable. The main title of Time-Heatmap.
id.ensembl,	a logic variable. If using ensembl as the keytype. This must match the row name of master.table. By default is TRUE.
ont,	one of "BP", "MF", and "CC" subontologies, or "ALL" for all three. By default is "BP".
save.pdf.path,	a character variable. If need to save the figure into PDF file. This must be an absolute file path. If not needed save as PDF file, set it to NA. By defualt iS NA.
pdf.width,	a numeric variable. The width of PDF file. By default is 13.
pdf.height,	a numeric variable. The height of PDF file. By default is 15.

Value

A list object, including elements names merge.df and time.heatmap. time.heatmap is the ggplot object. merge.df includes all the enrichR enrichment result and activation/deactivation time.

Examples

```
## Not run:
example.file.path<-system.file("extdata", "BrainMasterList.rda", package = "TrendCatcher")
load(example.file.path)
th.obj<-draw_TimeHeatmap_enrichR(master.list = master.list)
print(th.obj$time.heatmap)
head(th.obj$merge.df)

## End(Not run)
```

draw_TimeHeatmap_GO *Draw Time-Heatmap Using Gene Ontology (GO) Enrichment*

Description

This funcitons takes the master.list output from run_TrendCatcher. And apply a time window sliding strategy to capture all the genes increased/decreased compared to its previous break point, and apply GO enrichment analysis.

Usage

```
draw_TimeHeatmap_GO(
  master.list,
  logFC.thres = 1,
  top.n = 10,
```

```

dyn.gene.p.thres = 0.05,
keyType = "SYMBOL",
OrgDb = "org.Mm.eg.db",
ont = "BP",
term.width = 80,
GO.enrich.p = 0.05,
figure.title = "",
save.tiff.path = NA,
tiff.res = 100,
tiff.width = 1500,
tiff.height = 1500
)

```

Arguments

master.list,	a list object. The output from run_TrendCatcher function, contains master.table element.
logFC.thres,	a numeric variable. The logFC threshold compared to each genes previous break point expression level. By default is 1, meaning for each gene, the current time window's expression level is 2-fold compared to previous break point's expression level.
top.n,	an integer variable. The top N GO enrichment term need to be shown in the Time-Heatmap for up and down regulated pathway. By default is 10. Top 20 GO terms, 10 from up-regulated pathway and 10 from down-regulated pathway will shown in Time-Heatmap.
dyn.gene.p.thres,	a numeric variable. The DDEGs dynamic p-value threshold. By default is 0.05.
keyType,	must be either ENSEMBL or SYMBOL. The row names of your master.list\$master.table.
OrgDb,	must be either "org.Mm.eg.db" or "org.Hg.eg.db". Currently only support mouse and human GO annotation database.
ont,	one of "BP", "MF", and "CC" subontologies, or "ALL" for all three. By default is "BP".
term.width,	an integer variable. The character length for each GO term. If one GO term is super long, we can wrap it into term.width of strings into multiple rows. By default if 80.
GO.enrich.p,	an numeric variable. The GO enrichment p-value threshold. By default if 0.05.
figure.title,	a character variable. The main title of Time-Heatmap.
save.pdf.path,	a character variable. If need to save the figure into PDF file. This must be an absolute file path. If not needed save as PDF file, set it to NA. By defualt iS NA.
pdf.width,	a numeric variable. The width of PDF file. By default is 15.
pdf.height,	a numeric variable. The height of PDF file. By default is 15.

Value

A list object, including elements names merge.df and time.heatmap. time.heatmap is the ggplot object. merge.df includes all the GO enrichment result and activation/deactivation time.

Examples

```
## Not run:
example.file.path<-system.file("extdata", "BrainMasterList.rda", package = "TrendCatcher")
load(example.file.path)
gene.symbol.df<-get_GeneEnsembl2Symbol(ensemble.arr = master.list$master.table$Gene)
th.obj<-draw_TimeHeatmap_G0(master.list = master.list)
print(th.obj$time.heatmap)
head(th.obj$merge.df)

## End(Not run)
```

draw_TimeHeatmap_selGO

Subset TimeHeatmap

Description

Subset TimeHeatmap

Usage

```
draw_TimeHeatmap_selGO(
  time_heatmap,
  sel.go,
  master.list,
  GO.perc.thres = 0,
  nDDEG.thres = 0,
  term.width = 80,
  figure.title = "",
  save.tiff.path = NA,
  tiff.res = 100,
  tiff.width = 1500,
  tiff.height = 1500
)
```

draw_TrajClusterGrid *Draw Grouped DDEGs Trajectories in Grid Plot*

Description

Group all DDEGs based on their sub-type trajectory patterns and plot their trajectories together, then layout all sub-type trajectory patterns which contains more than N genes in a grid plot. Each individual sub-grid plot is titled with sub-type trajectory pattern and number of genes included. X-axis is the time, Y-axis is log2 transformed fitted count trajectory.

Usage

```
draw_TrajClusterGrid(
  master.list,
  min.traj.n = 10,
  save.as.PDF = NA,
  pdf.width = 10,
  pdf.height = 10
)
```

Arguments

master.list,	a list object. The output from run_TrendCatcher function, contains master.table element.
min.traj.n,	an integer variable. The minimum number of genes from the same sub-type trajectory. By default is 10.
save.as.PDF,	a string. The absolute file path to save the figure as PDF file if needed. If set to NA, will print the figure instead of saving it as PDF file.
pdf.width,	a numeric variable. The PDF file width size. By default is 10.
pdf.height,	a numeric variable. The PDF file height size. By default is 10.

draw_TrajClusterPie *Draw Grouped DDEGs Main-type and Sub-type Composition in Hierarchical Pie Chart*

Description

Group all DDEGs based on their main-type and sub-type trajectory patterns and plot their composition in a hierarchical pie chart. Inner pie chart represents the main-type trajectory pattern composition. The outer pie chart represents sub-type trajectory pattern composition.

Usage

```
draw_TrajClusterPie(
  master.list,
  fig.title = "",
  inner.radius = 0.7,
  cex.out = 1,
  cex.in = 1
)
```

Arguments

master.list,	a list object. The output from run_TrendCatcher function, contains master.table element.
fig.title,	a string. The main title of the figure. By default is "".

inner.radius, a numeric variable. The inner pie chart radius size. By default is 0.7.
 cex.out, a numeric variable. The text size of label of outer pie chart. By default is 1.
 cex.in, a numeric variable. The text size of the label of inner pie chart. By default is 1.

fit_single_gene_const *Fit the baseline count data into a constant negative binomial model.*

Description

Fit the baseline count data into a constant negative binomial model.

Usage

```
fit_single_gene_const(
  count.arr,
  disp.var,
  MAXIT = 1000,
  RELTOL = 10^{(-8)},
  trace = 10
)
```

Arguments

count.arr, a data frame returned from transform_single_gene_df, only with the baseline time.
 disp.var, the dispersion value estimated from DESeq2.

Value

a list contain all the estimated value from NB model.

fit_single_gene_spline

Fit the non-baseline count data into a smoothed ANOVA model.

Description

Fit the non-baseline count data into a smoothed ANOVA model.

Usage

```
fit_single_gene_spline(data.trans)
```

Arguments

data.trans, a data frame returned from transform_single_gene_df, without the baseline time.

Value

a dataframe with all the fitted count from spline model.

get_GeneEnsembl2Symbol

ID convention (from ENSEMBL to SYMBOL)

Description

This function takes an array of ENSEMBL ID and convert it into GENE SYMBOL.

Usage

```
get_GeneEnsembl2Symbol(ensemble.arr, dataset = "mmusculus_gene_ensembl")
```

Arguments

ensemble.arr, a character array. The ENSEMBL ID array.

dataset, must be either "mmusculus_gene_ensembl" or "hsapiens_gene_ensembl".

Value

A data frame contains 3 columns, "Gene", "Symbol" and "description".

get_rep_array

Get the replicate array from count table

Description

It take the count table, grep the replicate element from column name. The column name of the count table must satisfy "Prj_Time_Rep1" format.

Usage

```
get_rep_array(raw.count.df)
```

Arguments

raw.count.df, a count table.

Value

a numeric array of each sample's replicate.

`get_Symbol2GeneEnsembl`

ID convention (from SYMBOL to ENSEMBL)

Description

This function takes an array of SYMBOL and convert it into GENE ENSEMBL.

Usage

```
get_Symbol2GeneEnsembl(symbol.arr, dataset = "mmusculus_gene_ensembl")
```

Arguments

`symbol.arr`, a character array. The SYMBOL array.

`dataset`, must be either "mmusculus_gene_ensembl" or "hsapiens_gene_ensembl".

Value

A data frame contains 3 columns, "Gene", original ID, "Symbol" and "description".

`get_time_array`

Get the time array from count table

Description

It take the count table, grep the time element from column name. The column name of the count table must satisfy Prj_Time_Rep1 format.

Usage

```
get_time_array(raw.count.df)
```

Arguments

`raw.count.df`, a count table.

Value

a numeric array of each sample's time.

permutation.new	<i>Permuation</i>
-----------------	-------------------

Description

Permuation

Usage

```
permutation.new(perm.dat, n.perm = 100, points, parallel = FALSE)
```

preprocess_TrendCatcher	<i>Preprocessing for TrendCatcher</i>
-------------------------	---------------------------------------

Description

This is the preprocessing function to prepare the input count table for run_TrendCatcher function. It takes the CSV file count table and logic variables to check if normalization and batch correction needed for preprocessing. It creates pdf figure report to show before and after of normalization and batch correction to assess quality control (QC).

Usage

```
preprocess_TrendCatcher(
  count.table.path = "",
  need.batch.correction = TRUE,
  need.normalization = TRUE,
  batch.arr = "",
  pdf.file.path = NA,
  pdf.width = 8,
  pdf.height = 10,
  n.low.count = 10
)
```

Arguments

count.table.path,	string contain the absolute path of the CSV file count table, with first column as GENE SYMBOL or GENE ENSEMBL and first row as SAMPLE NAME (with format composed by project name,time and replicateID, such as "Lung_0_Rep1")
need.batch.correction,	logic variable. If batch correction is needed. By default is TRUE.
need.normalization,	logic variable. If normalization is needed. By default is TRUE.

batch.arr, a numeric vector of batch number. Need to be the same length as the number of samples.
 pdf.file.path, an absolute file path for save the QC report file. If not need, set it to NA. The report will be printed. By default is NA.
 pdf.width, a numeric variable. The PDF file width size. By default is 8.
 pdf.height, a numeric variable. The PDF file height size. By default is 10.
 n.low.count, a numeric variable. The minimal number to filter low count genes. By default is 10.

Value

A matrix array object.

Examples

```

example.file.path<-system.file("extdata", "Brain_DemoCountRawTable.csv", package = "TrendCatcher")
## Not run:
count.table<-preprocess_TrendCatcher(count.table.path = example.file.path,
need.batch.correction = TRUE,
need.normalization = TRUE,
batch.arr ="",
pdf.file.path = NA,
pdf.width=8, pdf.height=10,
n.low.count = 10)

## End(Not run)
  
```

run_TrendCatcher

Run TrendCatcher Main Algorithm

Description

This is the main function to run TrendCatcher to identify Dynamic Differentially Expressed Genes (DDEGs). This function loads a rounded count matrix CSV file after the normalization and batch correction, run the core algorithm and output a list object contains all the genes dynamic information.

Usage

```

run_TrendCatcher(
  count.table.path = "~/Documents/TrendCatcher/inst/extdata/Lung_DemoCountTable.csv",
  baseline.t = 0,
  time.unit = "h",
  min.low.count = 1,
  para.core.n = NA,
  dyn.p.thres = 0.05,
  show.verbose = F
)
  
```

Arguments

count.table.path,	string contain the absolute path of the CSV file count table, with first column as GENE SYMBOL or GENE ENSEMBL and first row as SAMPLE NAME (with format composed by project name,time and replicateID, such as "Lung_0_Rep1")
baseline.t,	one numeric variable, the baseline time of the longitudinal study. By default it is 0.
time.unit,	one character variable, the time unit of longitudinal study. If choose hour, please transform all sample collecting time into hour.
min.low.count,	one numeric variable, the minimal count threshold for filtering low count within each time group. By default it is 1.
para.core.n,	one numeric variable, number of cores will be used for running TrendCatcher. By default it is NA, which will use N-1 cores from computer.
dyn.p.thres,	one numeric variable, the threshold of p-value of the dynamic gene. By default 0.05.
show.verbose,	logic variable. If gssanova fitting failed, users can set this to TRUE, it will print out which gene failed the fitting. This process takes only one CPU, so it may be slower than the multi-core version. Normally the fitting failure is caused by low count genes. Users can manually remove it from your count table. By default set to FALSE.

Value

A list object, including "time.unit", "baseline.t", "t.arr", "Project.name", "raw.df", "fitted.count" and "master.table".

Examples

```
example.file.path<-system.file("extdata", "Brain_DemoCountTable.csv", package = "TrendCatcher")
## Not run:
master.list<-run_TrendCatcher(count.table.path = example.file.path,
                                baseline.t = 0,
                                time.unit = "h",
                                min.low.count = 1,
                                para.core.n = NA,
                                dyn.p.thres = 0.05,
                                show.verbose = FALSE)

## End(Not run)
```

transform_single_gene_df

Convert a single gene's count row number into data frame with two columns.

Description

Convert a single gene's count row number into data frame with two columns.

Usage

```
transform_single_gene_df(gene.row.info, gene.name, time.arr, rep.arr)
```

Arguments

gene.row.info,	a single row from count table.
gene.name,	the gene name.
time.arr,	the return value from get_time_array function.
rep.arr,	the return value from get_rep_array function.

Value

a dataframe object ordered by time and replicate id.

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