

Package ‘mosdef’

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Title MOST frequently used and useful Differential Expression Functions

Version 1.3.0

Description This package provides functionality to run a number of tasks in the differential expression analysis workflow. This encompasses the most widely used steps, from running various enrichment analysis tools with a unified interface to creating plots and beautifying table components linking to external websites and databases. This streamlines the generation of comprehensive analysis reports.

Depends R (>= 4.4.0)

Imports DT, ggplot2, ggforce, ggrepel, graphics, grDevices, htmltools, methods, AnnotationDbi, topGO, GO.db, clusterProfiler, goseq, utils, RColorBrewer, rlang, DESeq2, scales, SummarizedExperiment, S4Vectors, stats

Suggests knitr, rmarkdown, macrophage, org.Hs.eg.db, GeneTonic, testthat (>= 3.0.0), TxDb.Hsapiens.UCSC.hg38.knownGene, BiocStyle

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.info_enrichrun *Printing some info before the enrichment runs*

Description

Printing some info before the enrichment runs

Usage

```
.info_enrichrun(n_de, n_de_selected, de_type, res_de = NULL)
```

Arguments

| | |
|---------------|----------------------------------------------------------------------|
| n_de | Numeric, number of DE genes (in total) |
| n_de_selected | Character vector, containing the selected DE genes |
| de_type | Character string, specifying up/down/both direction of DE regulation |
| res_de | The res_de container as expected in most mosdef functions. |

Value

Prints out an informative summary message.

Examples

```
# .info_enrichrun(10, length(c("geneA", "geneB")), "up")
```

buttonifier *Create sets of buttons for gene symbols*

Description

A function to turn Gene Symbols into buttons in an Rmarkdown linking to various portals for further info about these genes.

Usage

```
buttonifier(  
  df,  
  create_buttons_to = c("PUBMED", "GC", "UNIPROT"),  
  col_to_use = "SYMBOL",  
  output_format = "DT",  
  ens_col = NULL,  
  ens_species = NULL  
)
```

Arguments

| | |
|-------------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| df | A dataframe with at least on column with gene Symbols named: SYMBOL |
| create_buttons_to | At least one of: "GC", "NCBI", "GTEX", "UNIPROT", "dbPTM", "HPA" "PUBMED" |
| col_to_use | name of the columns were the gene symbols are stored. Default is SYMBOL |
| output_format | a parameter deciding which output format to return, either a "DT" (<code>DT::datatable()</code> , recommended), or a simple dataframe ("DF"). In the latter case it is important that if the data is visualized with the <code>DT::datatable</code> function the parameter <code>escape</code> must be set to FALSE |
| ens_col | Character string, name of the columns were the ENSEMBL IDs are stored. |
| ens_species | The species you are working with to link to the correct gene on ENSEMBL |

Details

Current supported portals are: GeneCards, NCBI, GTEEx, Uniprot, dbPTM, Human Protein Atlas

Value

A data.frame or a `DT::datatable` object with columns adding HTML objects that link to websites with further information on the genes in question.

Examples

```
data(res_de_macrophage, package = "mosdef")

res_de <- res_macrophage_IFNg_vs_naive
res_df <- dresult_to_df(res_de)

## Subsetting for quicker run
res_df <- res_df[1:100, ]
buttonifier(res_df)

buttonifier(res_df,
  create_buttons_to = c("NCBI", "HPA"),
  ens_col = "id",
  ens_species = "Homo_sapiens"
)
```

create_link_dbPTM *Link to dbPTM database*

Description

Link to dbPTM database

Usage

```
create_link_dbPTM(val)
```

Arguments

`val` Character, the gene symbol

Value

HTML for an action button

Examples

```
create_link_dbPTM("Oct4")

data(res_de_macrophage, package = "mosdef")
res_macrophage_IFNg_vs_naive$SYMBOL <-
  create_link_dbPTM(res_macrophage_IFNg_vs_naive$SYMBOL)
```

`create_link_ENSEMBL` *Link to ENSEMBL database*

Description

Link to ENSEMBL database

Usage

```
create_link_ENSEMBL(val, species = "Mus_musculus")
```

Arguments

`val` Character, the gene symbol
`species` The species to be analyzed e.g "Mus_musculus"

Value

HTML for an action button

Examples

```
create_link_ENSEMBL("ENSMUSG00000024406")

data(res_de_macrophage, package = "mosdef")
rownames(res_macrophage_IFNg_vs_naive) <- create_link_ENSEMBL(
  rownames(res_macrophage_IFNg_vs_naive))
```

create_link_GeneCards *Link to the GeneCards database*

Description

Link to the GeneCards database

Usage

```
create_link_GeneCards(val)
```

Arguments

val Character, the gene symbol of interest

Value

HTML for an action button

Examples

```
create_link_GeneCards("Oct4")

data(res_de_macrophage, package = "mosdef")
res_macrophage_IFNg_vs_naive$SYMBOL <-
  create_link_GeneCards(res_macrophage_IFNg_vs_naive$SYMBOL)
```

create_link_GO *Link to AMIGO database*

Description

Link to AMIGO database

Usage

```
create_link_GO(val)
```

Arguments

val Character, the GOID

Value

HTML for an action button

Examples

```
create_link_GO("GO:0008150")
```

create_link_GTEX *Link to the GTEX Portal*

Description

Link to the GTEX Portal

Usage

```
create_link_GTEX(val)
```

Arguments

val Character, the gene symbol of interest

Value

HTML for an action button

Examples

```
create_link_GTEX("Oct4")

data(res_de_macrophage, package = "mosdef")
res_macrophage_IFNg_vs_naive$SYMBOL <-
  create_link_GTEX(res_macrophage_IFNg_vs_naive$SYMBOL)
```

create_link_HPA *Link to the Human Protein Atlas*

Description

Link to the Human Protein Atlas

Usage

```
create_link_HPA(val)
```

Arguments

val Character, the gene symbol

Value

HTML for an action button

Examples

```
create_link_HPA("Oct4")

data(res_de_macrophage, package = "mosdef")
res_macrophage_IFNg_vs_naive$SYMBOL <-
  create_link_HPA(res_macrophage_IFNg_vs_naive$SYMBOL)
```

| | |
|------------------|------------------------------|
| create_link_NCBI | <i>Link to NCBI database</i> |
|------------------|------------------------------|

Description

Link to NCBI database

Usage

```
create_link_NCBI(val)
```

Arguments

val Character, the gene symbol

Value

HTML for an action button

Examples

```
create_link_NCBI("Oct4")

data(res_de_macrophage, package = "mosdef")
res_macrophage_IFNg_vs_naive$SYMBOL <-
  create_link_NCBI(res_macrophage_IFNg_vs_naive$SYMBOL)
```

`create_link_PubMed` *Link to Pubmed*

Description

Link to Pubmed

Usage

```
create_link_PubMed(val)
```

Arguments

val Character, the gene symbol

Value

HTML for an action button

Examples

```
create_link_PubMed("Oct4")

data(res_de_macrophage, package = "mosdef")
res_macrophage_IFNg_vs_naive$SYMBOL <-
  create_link_PubMed(res_macrophage_IFNg_vs_naive$SYMBOL)
```

`create_link_UniProt` *Link to UniProt database*

Description

Link to UniProt database

Usage

```
create_link_UniProt(val)
```

Arguments

val Character, the gene symbol

Value

HTML for an action button

Examples

```
create_link_UniProt("Oct4")

data(res_de_macrophage, package = "mosdef")
res_macrophage_IFNg_vs_naive$SYMBOL <-
  create_link_UniProt(res_macrophage_IFNg_vs_naive$SYMBOL)
```

| | |
|----------------|-------------------------------------------------|
| deresult_to_df | <i>Generate a table from the DESeq2 results</i> |
|----------------|-------------------------------------------------|

Description

Generate a tidy table with the results of DESeq2

Usage

```
deresult_to_df(res_de, FDR = NULL)
```

Arguments

| | |
|--------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| res_de | An object containing the results of the Differential Expression analysis workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqResults object created using the DESeq2 framework. |
| FDR | Numeric value, specifying the significance level for thresholding adjusted p-values. Defaults to NULL, which would return the full set of results without performing any subsetting based on FDR. |

Value

A tidy data.frame with the results from differential expression, sorted by adjusted p-value. If FDR is specified, the table contains only genes with adjusted p-value smaller than the value.

Examples

```
library("DESeq2")
library("macrophage")
data(res_de_macrophage, package = "mosdef")
head(res_macrophage_IFNg_vs_naive)
res_df <- deresult_to_df(res_macrophage_IFNg_vs_naive)
head(res_df)
```

| | |
|-----------------|-------------------------|
| de_tablePainter | <i>DE table painter</i> |
|-----------------|-------------------------|

Description

Beautifying the aspect and looks of a DE results table

Usage

```
de_tablePainter(  
  res_de,  
  rounding_digits = NULL,  
  signif_digits = NULL,  
  up_DE_color = "darkred",  
  down_DE_color = "navyblue",  
  logfc_column = "log2FoldChange",  
  basemean_column = "baseMean",  
  lfcse_column = "lfcSE",  
  stat_column = "stat",  
  pvalue_column = "pvalue",  
  padj_column = "padj"  
)
```

Arguments

| | |
|-----------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| res_de | An object containing the results of the Differential Expression analysis workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqResults object created using the DESeq2 framework. Or a data frame obtained from such an object through deresult_to_df() |
| rounding_digits | Numeric value, specifying the number of digits to round the numeric values of the DE table (except the p-values) |
| signif_digits | Numeric value, specifying the number of significant digits to display for the p-values in the DE table |
| up_DE_color | Character string, specifying the color to use for coloring the bar of upregulated genes. |
| down_DE_color | Character string, specifying the color to use for coloring the bar of downregulated genes. |
| logfc_column | Character string, defining the name of the column in which to find the log2 fold change. |
| basemean_column | Character string, defining the name of the column in which to find the average expression value. |
| lfcse_column | Character string, defining the name of the column in which to find the standard error of the log2 fold change. |

| | |
|---------------|------------------------------------------------------------------------------------------------------|
| stat_column | Character string, defining the name of the column in which to find the values of the test statistic. |
| pvalue_column | Character string, defining the name of the column in which to find the unadjusted p-values. |
| padj_column | Character string, defining the name of the column in which to find the adjusted p-values. |

Details

Feeding on the classical results of DE workflows, this function formats and tries to prettify the representation of the key values in it.

Value

A datatable object, ready to be rendered as a widget inside an analysis Rmarkdown report.

Examples

```
data(res_de_macrophage, package = "mosdef")
de_tablePainter(res_macrophage_IFNg_vs_naive,
                rounding_digits = 3,
                signif_digits = 5)

## It is also possible to pass the "buttonified" table,
res_df_small <- deresult_to_df(res_macrophage_IFNg_vs_naive)[1:100, ]

buttonified_df <- buttonifier(res_df_small,
                              create_buttons_to = c("NCBI", "HPA"),
                              ens_col = "id",
                              ens_species = "Homo_sapiens",
                              output_format = "DF"
)

de_tablePainter(buttonified_df,
                rounding_digits = 3,
                signif_digits = 5)
```

de_volcano

Generates a volcano plot using ggplot2

Description

This function generates a base volcano plot for differentially expressed genes that can then be expanded upon using further ggplot functions.

Usage

```
de_volcano(  
  res_de,  
  mapping = "org.Mm.eg.db",  
  logfc_cutoff = 1,  
  FDR = 0.05,  
  labeled_genes = 30  
)
```

Arguments

| | |
|---------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| res_de | An object containing the results of the Differential Expression analysis workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqResults object created using the DESeq2 framework. |
| mapping | Which org.XX.eg.db package to use for annotation - select according to the species |
| logfc_cutoff | A numeric value that sets the cutoff for the xintercept argument of ggplot |
| FDR | The pvalue threshold to use for counting genes as de and therefore also where to draw the line in the plot. Default is 0.05 |
| labeled_genes | A numeric value describing the amount of genes to be labeled. This uses the Top(x) highest differentially expressed genes |

Value

A ggplot2 volcano plot object that can be extended upon by the user

Examples

```
library("ggplot2")  
library("RColorBrewer")  
library("ggrepel")  
library("DESeq2")  
library("org.Hs.eg.db")  
  
data(res_de_macrophage, package = "mosdef")  
  
p <- de_volcano(res_macrophage_IFNg_vs_naive,  
  logfc_cutoff = 1,  
  labeled_genes = 20,  
  mapping = "org.Hs.eg.db"  
)  
  
p
```

| | |
|------------------|------------------------------|
| geneinfo_to_html | <i>Information on a gene</i> |
|------------------|------------------------------|

Description

Assembles information, in HTML format, regarding a gene symbol identifier

Usage

```
geneinfo_to_html(gene_id, res_de = NULL, col_to_use = "SYMBOL")
```

Arguments

| | |
|------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| gene_id | Character specifying the gene identifier for which to retrieve information |
| res_de | An object containing the results of the Differential Expression analysis workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqResults object created using the DESeq2 framework. If not provided, the experiment-related information is not shown, and only some generic info on the identifier is displayed. The information about the gene is retrieved by matching on the SYMBOL column, which should be provided in res_de. |
| col_to_use | The column of your res_de object containing the gene symbols. Default is "SYMBOL" |

Details

Creates links to the NCBI and the GeneCards databases

Value

HTML content related to a gene identifier, to be displayed in web applications (or inserted in Rmd documents)

Examples

```
geneinfo_to_html("ACTB")  
geneinfo_to_html("Pf4")
```

| | |
|-----------|------------------------------------------|
| gene_plot | <i>Plot expression values for a gene</i> |
|-----------|------------------------------------------|

Description

Plot expression values (e.g. normalized counts) for a gene of interest, grouped by experimental group(s) of interest

Usage

```
gene_plot(
  de_container,
  gene,
  intgroup = "condition",
  assay = "counts",
  annotation_obj = NULL,
  normalized = TRUE,
  transform = TRUE,
  labels_display = TRUE,
  labels_repel = TRUE,
  plot_type = "auto",
  return_data = FALSE
)
```

Arguments

| | |
|----------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| de_container | An object containing the data for a Differential Expression workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqDataSet object, normally obtained after running your data through the DESeq2 framework. |
| gene | Character, specifies the identifier of the feature (gene) to be plotted |
| intgroup | A character vector of names in colData(de_container) to use for grouping. Note: the vector components should be categorical variables. |
| assay | Character, specifies with assay of the de_container object to use for reading out the expression values. Defaults to "counts". |
| annotation_obj | A data.frame object with the feature annotation information, with at least two columns, gene_id and gene_name. |
| normalized | Logical value, whether the expression values should be normalized by their size factor. Defaults to TRUE, applies when assay is "counts" |
| transform | Logical value, corresponding whether to have log scale y-axis or not. Defaults to TRUE. |
| labels_display | Logical value. Whether to display the labels of samples, defaults to TRUE. |
| labels_repel | Logical value. Whether to use ggrepel's functions to place labels; defaults to TRUE |

| | |
|-------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| plot_type | Character, one of "auto", "jitteronly", "boxplot", "violin", or "sina". Defines the type of geom_ to be used for plotting. Defaults to auto, which in turn chooses one of the layers according to the number of samples in the smallest group defined via intgroup |
| return_data | Logical, whether the function should just return the data.frame of expression values and covariates for custom plotting. Defaults to FALSE. |

Details

The result of this function can be fed directly to `plotly::ggplotly()` for interactive visualization, instead of the static `ggplot` viz.

Value

A `ggplot` object

Examples

```
library("macrophage")
library("DESeq2")
library("org.Hs.eg.db")

# dds object
data(gse, package = "macrophage")
dds_macrophage <- DESeqDataSet(gse, design = ~ line + condition)
rownames(dds_macrophage) <- substr(rownames(dds_macrophage), 1, 15)
keep <- rowSums(counts(dds_macrophage) >= 10) >= 6
dds_macrophage <- dds_macrophage[keep, ]
# dds_macrophage <- DESeq(dds_macrophage)

# annotation object
anno_df <- data.frame(
  gene_id = rownames(dds_macrophage),
  gene_name = mapIds(org.Hs.eg.db,
    keys = rownames(dds_macrophage),
    column = "SYMBOL",
    keytype = "ENSEMBL"
  ),
  stringsAsFactors = FALSE,
  row.names = rownames(dds_macrophage)
)

gene_plot(
  de_container = dds_macrophage,
  gene = "ENSG00000125347",
  intgroup = "condition",
  annotation_obj = anno_df
)
```

get_annotation_orgdb *Get an annotation data frame from org db packages*

Description

Get an annotation data frame from org db packages

Usage

```
get_annotation_orgdb(  
  de_container,  
  orgdb_package,  
  id_type,  
  key_for_genenames = "SYMBOL"  
)
```

Arguments

| | |
|-------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| de_container | An object containing the data for a Differential Expression workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqDataSet object, normally obtained after running your data through the DESeq2 framework. |
| orgdb_package | Character string, named as the org.XX.eg.db package which should be available in Bioconductor |
| id_type | Character, the ID type of the genes as in the row names of the de_container, to be used in the call to mapIds() |
| key_for_genenames | Character, corresponding to the column name for the key in the orgDb package containing the official gene name (often called gene symbol). This parameter defaults to "SYMBOL", but can be adjusted in case the key is not found in the annotation package (e.g. for org.Sc.sgd.db). |

Value

A data frame to be used for annotation of genes, with the main information encoded in the gene_id and gene_name columns.

Examples

```
library("macrophage")  
library("DESeq2")  
library("org.Hs.eg.db")  
  
# dds object  
data(gse, package = "macrophage")  
dds_macrophage <- DESeqDataSet(gse, design = ~ line + condition)  
rownames(dds_macrophage) <- substr(rownames(dds_macrophage), 1, 15)
```

```
anno_df <- get_annotation_orfdb(dds_macrophage, "org.Hs.eg.db", "ENSEMBL")
head(anno_df)
```

get_expr_values *Get expression values*

Description

Extract expression values, with the possibility to select other assay slots

Usage

```
get_expr_values(
  de_container,
  gene,
  intgroup,
  assay = "counts",
  normalized = TRUE
)
```

Arguments

| | |
|--------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| de_container | An object containing the data for a Differential Expression workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqDataSet object, normally obtained after running your data through the DESeq2 framework. |
| gene | Character, specifies the identifier of the feature (gene) to be extracted |
| intgroup | A character vector of names in colData(de_container) to use for grouping. |
| assay | Character, specifies with assay of the de_container object to use for reading out the expression values. Defaults to "counts". |
| normalized | Logical value, whether the expression values should be normalized by their size factor. Defaults to TRUE, applies when assay is "counts" |

Value

A tidy data.frame with the expression values and covariates for further processing

Examples

```
library("macrophage")
library("DESeq2")
library("org.Hs.eg.db")
library("AnnotationDbi")

# dds object
data(gse, package = "macrophage")
dds_macrophage <- DESeqDataSet(gse, design = ~ line + condition)
```

```
rownames(dds_macrophage) <- substr(rownames(dds_macrophage), 1, 15)
keep <- rowSums(counts(dds_macrophage) >= 10) >= 6
dds_macrophage <- dds_macrophage[keep, ]
# dds_macrophage <- DESeq(dds_macrophage)

df_exp <- get_expr_values(
  de_container = dds_macrophage,
  gene = "ENSG00000125347",
  intgroup = "condition"
)
head(df_exp)
```

go_to_html

Information on a Gene Ontology identifier

Description

Assembles information, in HTML format, regarding a Gene Ontology identifier

Usage

```
go_to_html(go_id, res_enrich = NULL)
```

Arguments

| | |
|------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| go_id | Character, specifying the GeneOntology identifier for which to retrieve information |
| res_enrich | A data.frame object, storing the result of the functional enrichment analysis. If not provided, the experiment-related information is not shown, and only some generic info on the identifier is displayed. |

Details

Also creates a link to the AmiGO database

Value

HTML content related to a GeneOntology identifier, to be displayed in web applications (or inserted in Rmd documents)

Examples

```
go_to_html("GO:0002250")
go_to_html("GO:0043368")
```

| | |
|------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| go_volcano | <i>Generates a volcano plot using ggplot2 This function generates a base volcano plot highlighting genes associated with a certain GOterm that can then be expanded upon using further ggplot functions.</i> |
|------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|

Description

Generates a volcano plot using ggplot2 This function generates a base volcano plot highlighting genes associated with a certain GOterm that can then be expanded upon using further ggplot functions.

Usage

```
go_volcano(
  res_de,
  res_enrich,
  mapping = "org.Hs.eg.db",
  term_index,
  logfc_cutoff = 1,
  FDR = 0.05,
  col_to_use = NULL,
  enrich_col = "genes",
  gene_col_separator = ",",
  down_col = "black",
  up_col = "black",
  highlight_col = "tomato",
  n_overlaps = 20
)
```

Arguments

| | |
|--------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| res_de | An object containing the results of the Differential Expression analysis workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqResults object created using the DESeq2 framework. |
| res_enrich | A enrichment result object created by for example using run_topGO() |
| mapping | Which org.XX.eg.db package to use for annotation - select according to the species |
| term_index | The location (row) of your GO term of interest in your enrichment result |
| logfc_cutoff | A numeric value that sets the cutoff for the xintercept argument of ggplot |
| FDR | The pvalue threshold to us for counting genes as de and therefore also where to draw the line in the plot. Default is 0.05 |
| col_to_use | The column in your differential expression results containing your gene symbols. If you don't have one it is created automatically |
| enrich_col | column name from your res_enrich where the genes associated with your GOterm are stored (for example see the run_topGO() result in mosdef) |

| | |
|--------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| gene_col_separator | The separator used to split the genes. If you used topGO or goseq this is a "," which is the default. (For an example see the run_topGO() result in mosdef) If you used clusterProfiler this has to be set to "/". (For example see the run_cluPro() result in mosdef) |
| down_col | The colour for your downregulated genes, default is "gray" |
| up_col | The colour for your upregulated genes, default is "gray" |
| highlight_col | The colour for the genes associated with your GOterm default is "tomato" |
| n_overlaps | Number of overlaps ggrepel is supposed to allow when labeling (for more info check ggrepel documentation) |

Value

A ggplot2 volcano plot object that can be extended upon by the user

Examples

```
library("org.Hs.eg.db")

data(res_de_macrophage, package = "mosdef")
data(res_enrich_macrophage_topGO, package = "mosdef")

p <- go_volcano(
  res_macrophage_IFNg_vs_naive,
  res_enrich = res_enrich_macrophage_topGO,
  term_index = 1,
  logfc_cutoff = 1,
  mapping = "org.Hs.eg.db",
  n_overlaps = 20
)

p
```

map_to_color

Maps numeric values to color values

Description

Maps numeric continuous values to values in a color palette

Usage

```
map_to_color(x, pal, symmetric = TRUE, limits = NULL)
```

Arguments

| | |
|------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <code>x</code> | A character vector of numeric values (e.g. <code>log2FoldChange</code> values) to be converted to a vector of colors |
| <code>pal</code> | A vector of characters specifying the definition of colors for the palette, e.g. obtained via <code>RColorBrewer::brewer.pal()</code> |
| <code>symmetric</code> | Logical value, whether to return a palette which is symmetrical with respect to the minimum and maximum values - "respecting" the zero. Defaults to <code>TRUE</code> . |
| <code>limits</code> | A vector containing the limits of the values to be mapped. If not specified, defaults to the range of values in the <code>x</code> vector. |

Value

A vector of colors, each corresponding to an element in the original vector

Examples

```
a <- 1:9
pal <- RColorBrewer::brewer.pal(9, "Set1")
map_to_color(a, pal)
plot(a, col = map_to_color(a, pal), pch = 20, cex = 4)

b <- 1:50
pal2 <- grDevices::colorRampPalette(
  RColorBrewer::brewer.pal(name = "RdYlBu", 11)
)(50)
plot(b, col = map_to_color(b, pal2), pch = 20, cex = 3)
```

mosdef-pkg

mosdef: mostly useful differential expression functions

Description

mostly useful differential expression functions

Details

This package provides functionality to run a number of tasks in the differential expression analysis workflow. This encompasses the most widely used steps, from running various enrichment analysis tools with a unified interface to creating plots and beautifying table components linking to external websites and databases. This streamlines the generation of comprehensive analysis reports.

Author(s)

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Authors:

- Leon Dammer <lc.dammer@gmail.com> ([ORCID](#))

See Also

Useful links:

- <https://github.com/imbeimainz/mosdef>
- Report bugs at <https://github.com/imbeimainz/mosdef/issues>

mosdef_de_container_check

A function checking if your de_container contains everything you need

Description

A function checking if your de_container contains everything you need

Usage

```
mosdef_de_container_check(de_container, verbose = FALSE)
```

Arguments

de_container An object containing the data for a Differential Expression workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqDataSet object, normally obtained after running your data through the DESeq2 framework.

verbose Logical, whether to add messages telling the user which steps were taken.

Value

An invisible NULL after performing the checks

Examples

```
library("macrophage")
library("DESeq2")
data(gse, package = "macrophage")

dds_macrophage <- DESeqDataSet(gse, design = ~ line + condition)
rownames(dds_macrophage) <- substr(rownames(dds_macrophage), 1, 15)
keep <- rowSums(counts(dds_macrophage) >= 10) >= 6
dds_macrophage <- dds_macrophage[keep, ]
# dds_macrophage <- DESeq(dds_macrophage)

mosdef_de_container_check(dds_macrophage)
```

| | |
|------------------|------------------------------------------------------------------------|
| mosdef_res_check | <i>A function checking if your res_de contains everything you need</i> |
|------------------|------------------------------------------------------------------------|

Description

A function checking if your res_de contains everything you need

Usage

```
mosdef_res_check(res_de, verbose = FALSE)
```

Arguments

| | |
|---------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| res_de | An object containing the results of the Differential Expression analysis workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqResults object created using the DESeq2 framework. |
| verbose | Logical, whether to add messages telling the user which steps were taken |

Value

An invisible NULL after performing the checks

Examples

```
data(res_de_macrophage, package = "mosdef")
mosdef_res_check(res_macrophage_IFNg_vs_naive)
```

| | |
|-----------|--------------------------------------------------------------------|
| pair_corr | <i>Pairwise scatter plot matrix and correlation plot of counts</i> |
|-----------|--------------------------------------------------------------------|

Description

Pairwise scatter plot matrix and correlation plot of counts

Usage

```
pair_corr(df, log = TRUE, method = "pearson", use_subset = TRUE)
```

Arguments

| | |
|------------|-----------------------------------------------------------------------------------------------------------|
| df | A data frame, containing the (raw/normalized/transformed) counts |
| log | Logical, whether to convert the input values to log2 (with addition of a pseudo-count). Defaults to TRUE. |
| method | Character string, one of pearson (default), kendall, or spearman as in cor |
| use_subset | Logical value. If TRUE, only 1000 values per sample will be used to speed up the plotting operations. |

Value

A plot with pairwise scatter plots and correlation coefficients

Examples

```
library("macrophage")
library("DESeq2")
data(gse, package = "macrophage")
## dds object
dds_macrophage <- DESeqDataSet(gse, design = ~ line + condition)
rownames(dds_macrophage) <- substr(rownames(dds_macrophage), 1, 15)
dds_macrophage <- estimateSizeFactors(dds_macrophage)

## Using just a subset for the example
pair_corr(counts(dds_macrophage, normalized = TRUE)[1:100, 1:8])
```

plot_ma

MA-plot from base means and log fold changes

Description

MA-plot from base means and log fold changes, in the ggplot2 framework, with additional support to annotate genes if provided.

Usage

```
plot_ma(
  res_de,
  FDR = 0.05,
  point_alpha = 0.2,
  sig_color = "red",
  annotation_obj = NULL,
  draw_y0 = TRUE,
  hlines = NULL,
  title = NULL,
  xlab = "mean of normalized counts - log10 scale",
  ylim = NULL,
  add_rug = TRUE,
  intgenes = NULL,
  intgenes_color = "steelblue",
  labels_intgenes = TRUE,
  labels_repel = TRUE
)
```

Arguments

| | |
|-----------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| res_de | An object containing the results of the Differential Expression analysis workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqResults object created using the DESeq2 framework. |
| FDR | Numeric value, the significance level for thresholding adjusted p-values |
| point_alpha | Alpha transparency value for the points (0 = transparent, 1 = opaque) |
| sig_color | Color to use to mark differentially expressed genes. Defaults to red |
| annotation_obj | A data.frame object, with row.names as gene identifiers (e.g. ENSEMBL ids) and a column, gene_name, containing e.g. HGNC-based gene symbols. Optional |
| draw_y0 | Logical, whether to draw the horizontal line at y=0. Defaults to TRUE. |
| hlines | The y coordinate (in absolute value) where to draw horizontal lines, optional |
| title | A title for the plot, optional |
| xlab | X axis label, defaults to "mean of normalized counts - log10 scale" |
| ylim | Vector of two numeric values, Y axis limits to restrict the view |
| add_rug | Logical, whether to add rug plots in the margins |
| intgenes | Vector of genes of interest. Gene symbols if a symbol column is provided in res_de, or else the identifiers specified in the row names |
| intgenes_color | The color to use to mark the genes on the main plot. |
| labels_intgenes | Logical, whether to add the gene identifiers/names close to the marked plots |
| labels_repel | Logical, whether to use ggrepel::geom_text_repel for placing the labels on the features to mark |

Details

The genes of interest are to be provided as gene symbols if a symbol column is provided in res_de, or else by using the identifiers specified in the row names

Value

An object created by ggplot

Examples

```
data(res_de_macrophage, package = "mosdef")

plot_ma(res_macrophage_IFNg_vs_naive, FDR = 0.05, hlines = 1)

plot_ma(res_macrophage_IFNg_vs_naive,
        FDR = 0.1,
        intgenes = c(
          "ENSG00000103196", # CRISPLD2
          "ENSG00000120129", # DUSP1
          "ENSG00000163884", # KLF15
        ))
```

```
      "ENSG00000179094" # PER1
    )
  )
```

res_enrich_macrophage_cluPro
A sample enrichment object

Description

A sample enrichment object, generated in the `mosdef` and `clusterProfiler` framework

Format

An `enrichResult` object

Details

This enrichment object is on the data from the `macrophage` package

Specifically, this set of enrichment results was created using the Biological Process ontology, mapping the gene identifiers through the `org.Hs.eg.db` package.

Source

Details on how this object has been created are included in the `create_mosdef_data.R` script, included in the (installed) `inst/scripts` folder of the `mosdef` package. This is also available at https://github.com/imbeimainz/mosdef/blob/devel/inst/scripts/create_mosdef_data.R

References

Alasoo, et al. "Shared genetic effects on chromatin and gene expression indicate a role for enhancer priming in immune response", *Nature Genetics*, January 2018 doi: 10.1038/s41588-018-0046-7.

See Also

[res_macrophage_IFNg_vs_naive](#)

res_enrich_macrophage_goseq
A sample enrichment object

Description

A sample enrichment object, generated in the mosdef and goseq framework

Format

A data.frame object

Details

This enrichment object is on the data from the macrophage package

Specifically, this set of enrichment results was created using the Biological Process ontology, mapping the gene symbol identifiers through the org.Hs.eg.db package - the gene length information is retrieved by the internal routines of goseq.

Source

Details on how this object has been created are included in the create_mosdef_data.R script, included in the (installed) inst/scripts folder of the mosdef package. This is also available at https://github.com/imbeimainz/mosdef/blob/devel/inst/scripts/create_mosdef_data.R

References

Alasoo, et al. "Shared genetic effects on chromatin and gene expression indicate a role for enhancer priming in immune response", Nature Genetics, January 2018 doi: 10.1038/s41588-018-0046-7.

See Also

[res_macrophage_IFNg_vs_naive](#)

res_enrich_macrophage_topGO
A sample enrichment object

Description

A sample enrichment object, generated in the mosdef and topGO framework

Format

A data.frame object

Details

This enrichment object is on the data from the macrophage package.

Specifically, this set of enrichment results was created using the Biological Process ontology, mapping the gene symbol identifiers through the org.Hs.eg.db package.

Source

Details on how this object has been created are included in the create_mosdef_data.R script, included in the (installed) inst/scripts folder of the mosdef package. This is also available at https://github.com/imbeimainz/mosdef/blob/devel/inst/scripts/create_mosdef_data.R

References

Alasoo, et al. "Shared genetic effects on chromatin and gene expression indicate a role for enhancer priming in immune response", Nature Genetics, January 2018 doi: 10.1038/s41588-018-0046-7.

See Also

[res_macrophage_IFNg_vs_naive](#)

res_macrophage_IFNg_vs_naive

A sample DESeqResults object

Description

A sample DESeqResults object, generated in the DESeq2 framework

Format

A DESeqResults object

Details

This DESeqResults object is on the data from the macrophage package. This result set has been created by setting the design to ~line + condition to detect the effect of the condition while accounting for the different cell lines included.

Specifically, this object contains the differences between the IFNg vs naive samples, testing against a logFC threshold of 1 for robustness.

Source

Details on how this object has been created are included in the create_mosdef_data.R script, included in the (installed) inst/scripts folder of the mosdef package. This is also available at https://github.com/imbeimainz/mosdef/blob/devel/inst/scripts/create_mosdef_data.R

References

Alasoo, et al. "Shared genetic effects on chromatin and gene expression indicate a role for enhancer priming in immune response", *Nature Genetics*, January 2018 doi: 10.1038/s41588-018-0046-7.

| | |
|------------|------------------------------------------------------------------------------------|
| run_cluPro | <i>Extract functional terms enriched in the DE genes, based on clusterProfiler</i> |
|------------|------------------------------------------------------------------------------------|

Description

A wrapper for extracting functional GO terms enriched in a list of (DE) genes, based on the algorithm and the implementation in the clusterProfiler package

Usage

```
run_cluPro(
  de_container = NULL,
  res_de = NULL,
  de_genes = NULL,
  bg_genes = NULL,
  top_de = NULL,
  FDR_threshold = 0.05,
  min_counts = 0,
  mapping = "org.Hs.eg.db",
  de_type = "up_and_down",
  keyType = "SYMBOL",
  verbose = TRUE,
  ...
)
```

Arguments

| | |
|---------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| de_container | An object containing the data for a Differential Expression workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqDataSet object, normally obtained after running your data through the DESeq2 framework. |
| res_de | An object containing the results of the Differential Expression analysis workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqResults object created using the DESeq2 framework. |
| de_genes | A vector of (differentially expressed) genes |
| bg_genes | A vector of background genes, e.g. all (expressed) genes in the assays |
| top_de | numeric, how many of the top differentially expressed genes to use for the enrichment analysis. Attempts to reduce redundancy. Assumes the data is sorted by padj (default in DESeq2). |
| FDR_threshold | The pvalue threshold to use for counting genes as de. Default is 0.05 |

| | |
|------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| min_counts | numeric, min number of counts a gene needs to have to be included in the gene-set that the de genes are compared to. Default is 0, recommended only for advanced users. |
| mapping | Which org.XX.eg.db package to use for annotation - select according to the species |
| de_type | One of: 'up', 'down', or 'up_and_down' Which genes to use for GOterm calculations |
| keyType | Gene format to input into enrichGO from clusterProfiler. If res_de and de_container are used use "SYMBOL" for more information check the enrichGO documentation |
| verbose | Logical, whether to add messages telling the user which steps were taken |
| ... | Further parameters to use for the <code>clusterProfiler::enrichGO()</code> function from clusterProfiler. |

Value

A table containing the computed GO Terms and related enrichment scores.

See Also

`clusterProfiler::enrichGO()` for the underlying method

Other Enrichment functions: `run_goseq()`, `run_topGO()`

Examples

```
library("macrophage")
library("DESeq2")
data(gse, package = "macrophage")

dds_macrophage <- DESeqDataSet(gse, design = ~ line + condition)
rownames(dds_macrophage) <- substr(rownames(dds_macrophage), 1, 15)
keep <- rowSums(counts(dds_macrophage) >= 10) >= 6
dds_macrophage <- dds_macrophage[keep, ]
dds_macrophage <- DESeq(dds_macrophage)
data(res_de_macrophage, package = "mosdef")

library("AnnotationDbi")
library("org.Hs.eg.db")
library("clusterProfiler")
CluProde_macrophage <- run_cluPro(
  res_de = res_macrophage_IFNg_vs_naive,
  de_container = dds_macrophage,
  mapping = "org.Hs.eg.db"
)
```

run_goseq

*Extract functional terms enriched in the DE genes, based on goseq***Description**

A wrapper for extracting functional GO terms enriched in a list of (DE) genes, based on the algorithm and the implementation in the goseq package

Usage

```
run_goseq(
  de_container = NULL,
  res_de = NULL,
  de_genes = NULL,
  bg_genes = NULL,
  top_de = NULL,
  FDR_threshold = 0.05,
  min_counts = 0,
  genome = "hg38",
  id = "ensGene",
  de_type = "up_and_down",
  testCats = c("GO:BP", "GO:MF", "GO:CC"),
  mapping = "org.Hs.eg.db",
  add_gene_to_terms = TRUE,
  verbose = TRUE
)
```

Arguments

| | |
|---------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| de_container | An object containing the data for a Differential Expression workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqDataSet object, normally obtained after running your data through the DESeq2 framework. |
| res_de | An object containing the results of the Differential Expression analysis workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqResults object created using the DESeq2 framework. |
| de_genes | A vector of (differentially expressed) genes |
| bg_genes | A vector of background genes, e.g. all (expressed) genes in the assays |
| top_de | numeric, how many of the top differentially expressed genes to use for the enrichment analysis. Attempts to reduce redundancy. Assumes the data is sorted by padj (default in DESeq2). |
| FDR_threshold | The pvalue threshold to use for counting genes as de. Default is 0.05 |
| min_counts | numeric, min number of counts a gene needs to have to be included in the gene-set that the de genes are compared to. Default is 0, recommended only for advanced users. |

| | |
|-------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------|
| genome | A string identifying the genome that genes refer to, as in the <code>goseq::goseq()</code> function |
| id | A string identifying the gene identifier used by genes, as in the <code>goseq::goseq()</code> function |
| de_type | One of: 'up', 'down', or 'up_and_down' Which genes to use for GOterm calculations: upregulated, downregulated or both |
| testCats | A vector specifying which categories to test for overrepresentation amongst DE genes - can be any combination of "GO:CC", "GO:BP", "GO:MF" & "KEGG" |
| mapping | Character string, named as the org.XX.eg.db package which should be available in Bioconductor |
| add_gene_to_terms | Logical, whether to add a column with all genes annotated to each GO term |
| verbose | Logical, whether to add messages telling the user which steps were taken |

Details

Note: the feature length retrieval is based on the `goseq::goseq()` function, and requires that the corresponding TxDb packages are installed and available

Value

A table containing the computed GO Terms and related enrichment scores

See Also

`goseq::goseq()` for the underlying method

Other Enrichment functions: `run_cluPro()`, `run_topGO()`

Examples

```
library("macrophage")
library("DESeq2")
data(gse, package = "macrophage")

dds_macrophage <- DESeqDataSet(gse, design = ~ line + condition)
rownames(dds_macrophage) <- substr(rownames(dds_macrophage), 1, 15)
keep <- rowSums(counts(dds_macrophage) >= 10) >= 6
dds_macrophage <- dds_macrophage[keep, ]
dds_macrophage <- DESeq(dds_macrophage)

data(res_de_macrophage, package = "mosdef")
res_de <- res_macrophage_IFNg_vs_naive
mygo <- run_goseq(
  res_de = res_macrophage_IFNg_vs_naive,
  de_container = dds_macrophage,
  mapping = "org.Hs.eg.db",
  testCats = "GO:BP",
  add_gene_to_terms = TRUE
)
```

```
head(mygo)
```

```
run_topGO
```

Extract functional terms enriched in the DE genes, based on topGO

Description

A wrapper for extracting functional GO terms enriched in the DE genes, based on the algorithm and the implementation in the topGO package

Usage

```
run_topGO(
  de_container = NULL,
  res_de = NULL,
  de_genes = NULL,
  bg_genes = NULL,
  top_de = NULL,
  FDR_threshold = 0.05,
  min_counts = 0,
  ontology = "BP",
  annot = annFUN.org,
  mapping = "org.Mm.eg.db",
  gene_id = "symbol",
  full_names_in_rows = TRUE,
  add_gene_to_terms = TRUE,
  de_type = "up_and_down",
  topGO_method2 = "elim",
  do_padj = FALSE,
  verbose = TRUE
)
```

Arguments

| | |
|--------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| de_container | An object containing the data for a Differential Expression workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqDataSet object, normally obtained after running your data through the DESeq2 framework. |
| res_de | An object containing the results of the Differential Expression analysis workflow (e.g. DESeq2, edgeR or limma). Currently, this can be a DESeqResults object created using the DESeq2 framework. |
| de_genes | A vector of (differentially expressed) genes |
| bg_genes | A vector of background genes, e.g. all (expressed) genes in the assays |
| top_de | numeric, how many of the top differentially expressed genes to use for the enrichment analysis. Attempts to reduce redundancy. Assumes the data is sorted by padj (default in DESeq2). |

| | |
|--------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| FDR_threshold | The pvalue threshold to us for counting genes as de. Default is 0.05 |
| min_counts | numeric, min number of counts a gene needs to have to be included in the gene-set that the de genes are compared to. Default is 0, recommended only for advanced users. |
| ontology | Which Gene Ontology domain to analyze: BP (Biological Process), MF (Molecular Function), or CC (Cellular Component) |
| annot | Which function to use for annotating genes to GO terms. Defaults to annFUN.org |
| mapping | Which org.XX.eg.db package to use for annotation - select according to the species |
| gene_id | Which format the genes are provided. Defaults to symbol, could also be entrez or ENSEMBL |
| full_names_in_rows | Logical, whether to display or not the full names for the GO terms |
| add_gene_to_terms | Logical, whether to add a column with all genes annotated to each GO term |
| de_type | One of: 'up', 'down', or 'up_and_down' Which genes to use for GOterm calculations: upregulated, downregulated or both |
| topGO_method2 | Character, specifying which of the methods implemented by topGO should be used, in addition to the classic algorithm. Defaults to elim. |
| do_padj | Logical, whether to perform the adjustment on the p-values from the specific topGO method, based on the FDR correction. Defaults to FALSE, since the assumption of independent hypotheses is somewhat violated by the intrinsic DAG-structure of the Gene Ontology Terms |
| verbose | Logical, whether to add messages telling the user which steps were taken |

Details

Allowed values assumed by the topGO_method2 parameter are one of the following: elim, weight, weight01, lea, parentchild. For more details on this, please refer to the original documentation of the topGO package itself

Value

A table containing the computed GO Terms and related enrichment scores

See Also

[topGO::topGOdata-class\(\)](#) and [topGO::runTest\(\)](#) for the class objects and underlying methods
Other Enrichment functions: [run_cluPro\(\)](#), [run_goseq\(\)](#)

Examples

```
library("macrophage")
library("DESeq2")
data(gse, package = "macrophage")
```

```

dds_macrophage <- DESeqDataSet(gse, design = ~ line + condition)
rownames(dds_macrophage) <- substr(rownames(dds_macrophage), 1, 15)
keep <- rowSums(counts(dds_macrophage) >= 10) >= 6
dds_macrophage <- dds_macrophage[keep, ]
dds_macrophage <- DESeq(dds_macrophage)

data(res_de_macrophage, package = "mosdef")

library("AnnotationDbi")
library("org.Hs.eg.db")
library("topGO")
topgoDE_macrophage <- run_topGO(
  de_container = dds_macrophage,
  res_de = res_macrophage_IFNg_vs_naive,
  ontology = "BP",
  mapping = "org.Hs.eg.db",
  gene_id = "symbol",
)

```

styleColorBar_divergent

Style DT color bars

Description

Style DT color bars for values that diverge from 0.

Usage

```
styleColorBar_divergent(data, color_pos, color_neg)
```

Arguments

| | |
|-----------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| data | The numeric vector whose range will be used for scaling the table data from 0-100 before being represented as color bars. A vector of length 2 is acceptable here for specifying a range possibly wider or narrower than the range of the table data itself. |
| color_pos | The color of the bars for the positive values |
| color_neg | The color of the bars for the negative values |

Details

This function draws background color bars behind table cells in a column, with the width of bars being proportional to the column values *and* the color dependent on the sign of the value.

A typical usage is for values such as log₂FoldChange for tables resulting from differential expression analysis. Still, the functionality of this can be quickly generalized to other cases - see in the examples.

The code of this function is heavily inspired from styleColorBar, and borrows at full hands from an excellent post on StackOverflow - <https://stackoverflow.com/questions/33521828/stylecolorbar-center-and-shift-left-right-dependent-on-sign/33524422#33524422>

Value

This function generates JavaScript and CSS code from the values specified in R, to be used in DT tables formatting.

Examples

```
# With a very simple data frame

simplest_df <- data.frame(
  a = c(rep("a", 9)),
  value = c(-4, -3, -2, -1, 0, 1, 2, 3, 4)
)

library("DT")
DT::datatable(simplest_df) |>
  formatStyle(
    "value",
    background = styleColorBar_divergent(
      simplest_df$value,
      scales::alpha("forestgreen", 0.4),
      scales::alpha("gold", 0.4)
    ),
    backgroundColor = "100% 90%",
    backgroundRepeat = "no-repeat",
    backgroundPosition = "center"
  )
```

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