

# Package ‘GeneTonic’

April 10, 2023

**Title** Enjoy Analyzing And Integrating The Results From Differential Expression Analysis And Functional Enrichment Analysis

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**Description** This package provides a Shiny application that aims to combine at different levels the existing pieces of the transcriptome data and results, in a way that makes it easier to generate insightful observations and hypothesis - combining the benefits of interactivity and reproducibility, e.g. by capturing the features and gene sets of interest highlighted during the live session, and creating an HTML report as an artifact where text, code, and output coexist.

**Depends** R (>= 4.0.0)

**Imports** AnnotationDbi, backbone, bs4Dash (>= 2.0.0), circlize, colorspace, colourpicker, ComplexHeatmap, ComplexUpset, dendextend, DESeq2, dplyr, DT, dynamicTreeCut, expm, ggforce, ggplot2, ggrepel, GO.db, graphics, grDevices, grid, igraph, matrixStats, methods, plotly, RColorBrewer, rintrojs, rlang, rmarkdown, S4Vectors, scales, shiny, shinyAce, shinycssloaders, shinyWidgets, stats, SummarizedExperiment, tidyr, tippy, tools, utils, viridis, visNetwork

**Suggests** knitr, BiocStyle, htmltools, clusterProfiler, macrophage, org.Hs.eg.db, magrittr, testthat (>= 2.1.0)

**License** MIT + file LICENSE

**Encoding** UTF-8

**VignetteBuilder** knitr

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**BugReports** <https://github.com/federicomarini/GeneTonic/issues>

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|                            |   |
|----------------------------|---|
| <code>.check_pandoc</code> | <i>Check whether pandoc and pandoc-citeproc are available</i> |
|----------------------------|---|

---

**Description**

Check whether pandoc and pandoc-citeproc are available

**Usage**

```
.check_pandoc(ignore_pandoc)
```

**Arguments**

`ignore_pandoc` Logical. If TRUE, just give a warning if one of pandoc or pandoc-citeproc is not available. If FALSE, an error is thrown.

**Details**

Credits to the original implementation proposed by Charlotte Sonesson, upon which this function is **heavily** inspired.

**Value**

No value is returned. If pandoc or pandoc-citeproc are missing, either warning or error messages are triggered.

---

|                   |   |
|-------------------|---|
| checkup_GeneTonic | <i>Checking the input objects for GeneTonic</i> |
|-------------------|---|

---

**Description**

Checking the input objects for GeneTonic, whether these are all set for running the app

**Usage**

```
checkup_GeneTonic(dds, res_de, res_enrich, annotation_obj, verbose = FALSE)
```

**Arguments**

|                |  |
|----------------|--|
| dds            | A DESeqDataSet object, normally obtained after running your data through the DESeq2 framework.   |
| res_de         | A DESeqResults object. As for the dds parameter, this is also commonly used in the DESeq2 framework.   |
| res_enrich     | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present). |
| annotation_obj | A data.frame object, containing two columns, gene_id with a set of unambiguous identifiers (e.g. ENSEMBL ids) and gene_name, containing e.g. HGNC-based gene symbols.  |
| verbose        | Logical, to control level of verbosity of the messages generated   |

**Details**

Some suggestions on the requirements for each parameter are returned in the error messages.

**Value**

Invisible NULL

**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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

checkup_GeneTonic(
  dds = dds_macrophage,
  res_de = res_de,
  res_enrich = res_enrich,
  annotation_obj = anno_df
)
# if all is fine, it should return an invisible NULL and a simple message

```

---

checkup\_gtl

*Checking the gtl input object for GeneTonic*


---

## Description

Checking the gtl ("GeneTonic list") input object for GeneTonic, with the correct content and format expected

## Usage

```
checkup_gtl(gtl, verbose = FALSE)
```

## Arguments

gtl                    A DESeqDataSet object, normally obtained after running your data through the DESeq2 framework. This list should contain

- in the dds slot: A DESeqDataSet object

- in the `res_de`: A `DESeqResults` object
- in the `res_enrich`: A `data.frame` object, storing the result of the functional enrichment analysis
- in the `annotation_obj`: A `data.frame` object, containing two columns, `gene_id` with a set of unambiguous identifiers (e.g. ENSEMBL ids) and `gene_name`, containing e.g. HGNC-based gene symbols.

`verbose` Logical, to control level of verbosity of the messages generated

## Details

Some suggestions on the requirements for the `gtl` are returned in the error messages.

## Value

Invisible `NULL`

## 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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

gtl <- list(
```

```
    dds = dds_macrophage,  
    res_de = res_de,  
    res_enrich = res_enrich,  
    annotation_obj = anno_df  
  )  
  
  checkup_gtl(gtl)  
  # if all is fine, it should return an invisible NULL and a simple message
```

---

check\_colors

*Check colors*

---

## Description

Check correct specification of colors

## Usage

```
check_colors(x)
```

## Arguments

x                    A vector of strings specifying colors

## Details

This is a vectorized version of [grDevices::col2rgb\(\)](#)

## Value

A vector of logical values, one for each specified color - TRUE if the color is specified correctly

## Examples

```
# simple case  
mypal <- c("steelblue", "#FF1100")  
check_colors(mypal)  
mypal2 <- rev(  
  scales::alpha(  
    colorRampPalette(RColorBrewer::brewer.pal(name = "RdYlBu", 11))(50), 0.4  
  )  
)  
check_colors(mypal2)  
# useful with long vectors to check at once if all cols are fine  
all(check_colors(mypal2))
```

---

cluster\_markov

*Markov Clustering (MCL) for community detection*


---

### Description

This function implements the Markov Clustering (MCL) algorithm for finding community structure, in an analogous way to other existing algorithms in igraph.

### Usage

```
cluster_markov(
  g,
  add_self_loops = TRUE,
  loop_value = 1,
  mcl_expansion = 2,
  mcl_inflation = 2,
  allow_singletons = TRUE,
  max_iter = 100,
  return_node_names = TRUE,
  return_esm = FALSE
)
```

### Arguments

|                                |   |
|--------------------------------|---|
| <code>g</code>                 | The input graph object  |
| <code>add_self_loops</code>    | Logical, whether to add self-loops to the matrix by setting the diagonal to <code>loop_value</code>   |
| <code>loop_value</code>        | Numeric, the value to use for self-loops  |
| <code>mcl_expansion</code>     | Numeric, cluster expansion factor for the Markov clustering iteration - defaults to 2   |
| <code>mcl_inflation</code>     | Numeric, cluster inflation factor for the Markov clustering iteration - defaults to 2   |
| <code>allow_singletons</code>  | Logical; if TRUE, single isolated vertices are allowed to form their own cluster. If set to FALSE, all clusters of size = 1 are grouped in one cluster (to be interpreted as background noise). |
| <code>max_iter</code>          | Numeric value for the maximum number of iterations for the Markov clustering  |
| <code>return_node_names</code> | Logical, if the graph is named and set to TRUE, returns the node names.   |
| <code>return_esm</code>        | Logical, controlling whether the equilibrium state matrix should be returned  |

### Details

This implementation has been driven by the nice explanations provided in

- [https://sites.cs.ucsb.edu/~xian/classes/CS595D-2009winter/MCL\\_Presentation2.pdf](https://sites.cs.ucsb.edu/~xian/classes/CS595D-2009winter/MCL_Presentation2.pdf)



- <https://medium.com/analytics-vidhya/demystifying-markov-clustering-aeb6cdabfc7>
- [https://github.com/GuyAllard/markov\\_clustering](https://github.com/GuyAllard/markov_clustering) (python implementation)

More info on the MCL: <https://micans.org/mcl/index.html>, and [https://micans.org/mcl/sec\\_description1.html](https://micans.org/mcl/sec_description1.html)

## Value

This function returns a `communities` object, containing the numbers of the assigned membership (in the slot `membership`). Please see the `igraph::communities()` manual page for additional details

## References

van Dongen, S.M., Graph clustering by flow simulation (2000) PhD thesis, Utrecht University Repository - <https://dspace.library.uu.nl/handle/1874/848>

Enright AJ, van Dongen SM, Ouzounis CA, An efficient algorithm for large-scale detection of protein families (2002) *Nucleic Acids Research*, Volume 30, Issue 7, 1 April 2002, Pages 1575–1584, <https://doi.org/10.1093/nar/30.7.1575>

## Examples

```
library("igraph")
g <- make_full_graph(5) %du% make_full_graph(5) %du% make_full_graph(5)
g <- add_edges(g, c(1, 6, 1, 11, 6, 11))
cluster_markov(g)
V(g)$color <- cluster_markov(g)$membership
plot(g)
```

---

`create_jaccard_matrix` *Compute the overlap matrix for enrichment results*

---

## Description

Compute the overlap matrix for enrichment results, based on the Jaccard Index between each pair of sets

## Usage

```
create_jaccard_matrix(
  res_enrich,
  gtl = NULL,
  n_gs = nrow(res_enrich),
  gs_ids = NULL,
  return_sym = FALSE
)
```

**Arguments**

|            |   |
|------------|---|
| res_enrich | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to see the formatting requirements.                                       |
| gtl        | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting |
| n_gs       | Integer value, corresponding to the maximal number of gene sets to be included (from the top ranked ones). Defaults to the number of rows of res_enrich   |
| gs_ids     | Character vector, containing a subset of gs_id as they are available in res_enrich. Lists the gene sets to be included, additionally to the ones specified via n_gs. Defaults to NULL.                                |
| return_sym | Logical, whether to return the symmetrical matrix or just the upper triangular - as needed by <a href="#">enrichment_map()</a> , for example.   |

**Value**

A matrix with the kappa scores between gene sets

**See Also**

[gs\\_mds\(\)](#), [enrichment\\_map\(\)](#)

**Examples**

```
# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOTableResult(topgoDE_macrophage_IFNg_vs_naive)

jmat <- create_jaccard_matrix(res_enrich[1:200, ])
dim(jmat)
```

---

create\_kappa\_matrix     *Compute the kappa matrix for enrichment results*

---

**Description**

Compute the kappa matrix for enrichment results, as a measure of overlap

**Usage**

```
create_kappa_matrix(
  res_enrich,
  gtl = NULL,
  n_gs = nrow(res_enrich),
  gs_ids = NULL
)
```

**Arguments**

|            |   |
|------------|---|
| res_enrich | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to see the formatting requirements.                                       |
| gtl        | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting |
| n_gs       | Integer value, corresponding to the maximal number of gene sets to be included (from the top ranked ones). Defaults to the number of rows of res_enrich   |
| gs_ids     | Character vector, containing a subset of gs_id as they are available in res_enrich. Lists the gene sets to be included, additionally to the ones specified via n_gs. Defaults to NULL.                                |

**Value**

A matrix with the kappa scores between gene sets

**See Also**

[gs\\_mds\(\)](#)

**Examples**

```
# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)

kmat <- create_kappa_matrix(res_enrich[1:200, ])
dim(kmat)
```

---

|                  |                                       |
|------------------|---------------------------------------|
| create_upsetdata | <i>Create a geneset upset dataset</i> |
|------------------|---------------------------------------|

---

**Description**

Create a data frame that can be fed to the upset function

**Usage**

```
create_upsetdata(res_enrich, use_ids = FALSE)
```

**Arguments**

|            |   |
|------------|---|
| res_enrich | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to see the formatting requirements. |
| use_ids    | Logical - whether to use the gs_identifiers as names, or the values provided as gs_description. Defaults to FALSE, using the full descriptions                                  |

**Value**

A data.frame to be used in ComplexUpset::upset()

**Examples**

```
# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)

create_upsetdata(res_enrich[1:20, ])
dim(create_upsetdata(res_enrich[1:20, ]))

create_upsetdata(res_enrich[1:5, ], use_ids = TRUE)
```

---

describe\_gtl

*Describe a GeneTonic list*

---

**Description**

Obtain a quick textual overview of the essential features of the components of the GeneTonic list object

**Usage**

```
describe_gtl(gtl)
```

**Arguments**

gtl            A GeneTonic-list object, containing in its named slots the required dds, res\_de, res\_enrich, and annotation\_obj

**Value**

A character string, that can further be processed (e.g. by message() or cat()), or easily rendered inside Shiny's renderText elements)

---

|                |   |
|----------------|---|
| deseqresult2df | <i>Generate a table from the DESeq2 results</i> |
|----------------|---|

---

**Description**

Generate a tidy table with the results of DESeq2

**Usage**

```
deseqresult2df(res_de, FDR = NULL)
```

**Arguments**

|        |   |
|--------|---|
| res_de | A DESeqResults object.  |
| 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**

```
data(res_de_macrophage, package = "GeneTonic")
head(res_macrophage_IFNg_vs_naive)
res_df <- deseqresult2df(res_macrophage_IFNg_vs_naive)
head(res_df)
```

---

|                    |                                   |
|--------------------|-----------------------------------|
| distill_enrichment | <i>Distill enrichment results</i> |
|--------------------|-----------------------------------|

---

**Description**

Distill the main topics from the enrichment results, based on the graph derived from constructing an enrichment map

**Usage**

```
distill_enrichment(
  res_enrich,
  res_de,
  annotation_obj,
  gtl = NULL,
  n_gs = nrow(res_enrich),
  cluster_fun = "cluster_markov"
)
```

**Arguments**

|                             |   |
|-----------------------------|---|
| <code>res_enrich</code>     | A <code>data.frame</code> object, storing the result of the functional enrichment analysis.   |
| <code>res_de</code>         | A <code>DESeqResults</code> object. As for the <code>dds</code> parameter, this is also commonly used in the <code>DESeq2</code> framework.   |
| <code>annotation_obj</code> | A <code>data.frame</code> object, containing two columns, <code>gene_id</code> with a set of unambiguous identifiers (e.g. ENSEMBL ids) and <code>gene_name</code> , containing e.g. HGNC-based gene symbols.   |
| <code>gtl</code>            | A <code>GeneTonic-list</code> object, containing in its slots the arguments specified above: <code>dds</code> , <code>res_de</code> , <code>res_enrich</code> , and <code>annotation_obj</code> - the names of the list <i>must</i> be specified following the content they are expecting |
| <code>n_gs</code>           | Integer value, corresponding to the maximal number of gene sets to be used.   |
| <code>cluster_fun</code>    | Character, referring to the name of the function used for the community detection in the enrichment map graph. Could be one of "cluster_markov", "cluster_louvain", or "cluster_walktrap", as they all return a <code>communities</code> object.  |

**Value**

A list containing three objects:

- the distilled table of enrichment, `distilled_table`, where the new meta-genesets are identified and defined, specifying e.g. the names of each component, and the genes associated to these.
- the distilled graph for the enrichment map, `distilled_em`, with the information on the membership
- the original `res_enrich`, augmented with the information of the membership related to the meta-genesets

**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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

distilled <- distill_enrichment(res_enrich,
  res_de,
  annotation_obj,
  n_gs = 100,
  cluster_fun = "cluster_markov"
)
colnames(distilled$distilled_table)
distilled$distilled_em
```

---

editor\_to\_vector\_sanitized

*Extract vectors from editor content*

---

## Description

Extract vectors from the shinyAce editor content, also removing comments and whitespaces from text.

## Usage

```
editor_to_vector_sanitized(txt)
```

## Arguments

txt                    A single character text input.

## Value

A character vector representing valid lines in the text input of the editor.

---

|               |   |
|---------------|---|
| enhance_table | <i>Visually enhances a functional enrichment result table</i> |
|---------------|---|

---

### Description

Creates a visual summary for the results of a functional enrichment analysis, by displaying also the components of each gene set and their expression change in the contrast of interest

### Usage

```
enhance_table(
  res_enrich,
  res_de,
  annotation_obj,
  gtl = NULL,
  n_gs = 50,
  gs_ids = NULL,
  chars_limit = 70,
  plot_title = NULL
)
```

### Arguments

|                |   |
|----------------|---|
| res_enrich     | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <code>GeneTonic()</code> , to check the formatting requirements (a minimal set of columns should be present). |
| res_de         | A DESeqResults object.  |
| annotation_obj | A data.frame object with the feature annotation. information, with at least two columns, gene_id and gene_name.   |
| gtl            | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting       |
| n_gs           | Integer value, corresponding to the maximal number of gene sets to be displayed.  |
| gs_ids         | Character vector, containing a subset of gs_id as they are available in res_enrich. Lists the gene sets to be displayed.  |
| chars_limit    | Integer, number of characters to be displayed for each geneset name.  |
| plot_title     | Character string, used as title for the plot. If left NULL, it defaults to a general description of the plot and of the DE contrast   |

### Value

A ggplot object



## 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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)
enhance_table(res_enrich,
  res_de,
  anno_df,
  n_gs = 10
)
```

---

enrichment\_map

*Creates an enrichment map for the results of functional enrichment*

---

## Description

Generates a graph for the enrichment map, combining information from `res_enrich` and `res_de`. This object can be further plotted, e.g. statically via `igraph::plot.igraph()`, or dynamically via `visNetwork::visIgraph()`

**Usage**

```
enrichment_map(
  res_enrich,
  res_de,
  annotation_obj,
  gtl = NULL,
  n_gs = 50,
  gs_ids = NULL,
  overlap_threshold = 0.1,
  scale_edges_width = 200,
  scale_nodes_size = 5,
  color_by = "gs_pvalue"
)
```

**Arguments**

|                   |   |
|-------------------|---|
| res_enrich        | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <code>GeneTonic()</code> , to check the formatting requirements (a minimal set of columns should be present). |
| res_de            | A DESeqResults object.  |
| annotation_obj    | A data.frame object with the feature annotation information, with at least two columns, gene_id and gene_name.  |
| gtl               | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting       |
| n_gs              | Integer value, corresponding to the maximal number of gene sets to be displayed   |
| gs_ids            | Character vector, containing a subset of gs_id as they are available in res_enrich. Lists the gene sets to be displayed.  |
| overlap_threshold | Numeric value, between 0 and 1. Defines the threshold to be used for removing edges in the enrichment map - edges below this value will be excluded from the final graph. Defaults to 0.1.                                  |
| scale_edges_width | A numeric value, to define the scaling factor for the edges between nodes. Defaults to 200 (works well chained to visNetwork functions).  |
| scale_nodes_size  | A numeric value, to define the scaling factor for the node sizes. Defaults to 5 - works well chained to visNetwork functions.   |
| color_by          | Character, specifying the column of res_enrich to be used for coloring the plotted gene sets. Defaults to gs_pvalue.  |

**Value**

An igraph object to be further manipulated or processed/plotted

**See Also**

[GeneTonic\(\)](#) embeds an interactive visualization for the enrichment map

**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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

em <- enrichment_map(res_enrich,
  res_de,
  anno_df,
  n_gs = 20
)

em

# could be viewed interactively with
# library("visNetwork")
# library("magrittr")
# em %>%
#   visIgraph() %>%
#   visOptions(highlightNearest = list(enabled = TRUE,
#                                       degree = 1,

```

```
#                                     hover = TRUE),
#                                     nodesIdSelection = TRUE)
```

---

```
enrichr_output_macrophage
```

*A sample output from Enrichr*

---

## Description

A sample output object as created from a call to Enrichr, with the interface provided by enrichR - using the enrichr() function

## Details

This object has been created on the data from the macrophage package by analyzing downstream the differentially expressed genes when comparing IFNg treated samples vs naive samples, accounting for the different cell lines included.

Details on how this object has been created are included in the create\_gt\_data.R script, included in the scripts folder of the GeneTonic package.

## 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

Other pathway-analysis-results: [gostres\\_macrophage](#), [topgoDE\\_macrophage\\_IFNg\\_vs\\_naive](#)

---

```
export_for_iSEE      export_for_iSEE
```

---

## Description

Combine data from a typical DESeq2 run

## Usage

```
export_for_iSEE(dds, res_de, gtl = NULL)
```

## Arguments

|        |   |
|--------|---|
| dds    | A <a href="#">DESeqDataSet</a> object.  |
| res_de | A <a href="#">DESeqResults</a> object.  |
| gtl    | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting |

**Details**

Combines the DESeqDataSet input and DESeqResults into a SummarizedExperiment object, which can be readily explored with iSEE.

A typical usage would be after running the DESeq2 pipeline and/or after exploring the functional enrichment results with [GeneTonic\(\)](#)

**Value**

A SummarizedExperiment object, with raw counts, normalized counts, and variance-stabilizing transformed counts in the assay slots; and with colData and rowData extracted from the corresponding input parameters - mainly the results for differential expression analysis.

**Examples**

```
library("macrophage")
library("DESeq2")

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

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# now everything is in place to launch the app
# dds_macrophage <- DESeq2::DESeq(dds_macrophage)
se_macrophage <- export_for_iSEE(dds_macrophage, res_de)
# iSEE(se_macrophage)
```

---

export\_to\_sif

*Export to sif*

---

**Description**

Export a graph to a Simple Interaction Format file

**Usage**

```
export_to_sif(g, sif_file = "", edge_label = "relates_to")
```

**Arguments**

|            |  |
|------------|--|
| g          | An igraph object   |
| sif_file   | Character string, the path to the file where to save the exported graph as .sif file       |
| edge_label | Character string, defining the name of the interaction type. Defaults here to "relates_to" |

**Value**

Returns the path to the exported file, invisibly

**Examples**

```
library("igraph")
g <- make_full_graph(5) %du% make_full_graph(5) %du% make_full_graph(5)
g <- add_edges(g, c(1, 6, 1, 11, 6, 11))
export_to_sif(g, tempfile())
```

---

fgseaRes

*A sample output from fgsea*


---

**Description**

A sample output object as created from a call to the fgsea() function, in the fgsea package, as a practical framework for performing GSEA

**Details**

This object has been created on the data from the macrophage package by analyzing downstream the differentially expressed genes when comparing IFNg treated samples vs naive samples, accounting for the different cell lines included.

Details on how this object has been created are included in the create\_gt\_data.R script, included in the scripts folder of the GeneTonic package.

**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.

---

geneinfo\_2\_html

*Information on a gene*


---

**Description**

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

**Usage**

```
geneinfo_2_html(gene_id, res_de = NULL)
```

**Arguments**

|         |   |
|---------|---|
| gene_id | Character specifying the gene identifier for which to retrieve information  |
| res_de  | A DESeqResults object, storing the result of the differential expression analysis. 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. |

**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_2_html("ACTB")
geneinfo_2_html("Pf4")
```

---

GeneTonic

*GeneTonic*


---

**Description**

GeneTonic, main function for the Shiny app

**Usage**

```
GeneTonic(
  dds = NULL,
  res_de = NULL,
  res_enrich = NULL,
  annotation_obj = NULL,
  gtl = NULL,
  project_id = "",
  size_gtl = 50
)
```

**Arguments**

|        |  |
|--------|--|
| dds    | A DESeqDataSet object, normally obtained after running your data through the DESeq2 framework.       |
| res_de | A DESeqResults object. As for the dds parameter, this is also commonly used in the DESeq2 framework. |

|                             |   |
|-----------------------------|---|
| <code>res_enrich</code>     | <p>A <code>data.frame</code> object, storing the result of the functional enrichment analysis. Required columns for enjoying the full functionality of <code>GeneTonic()</code> include:</p> <ul style="list-style-type: none"> <li>• a gene set identifier (e.g. GeneOntology id, <code>gs_id</code>) and its term description (<code>gs_description</code>)</li> <li>• a numeric value for the significance of the enrichment (<code>gs_pvalue</code>)</li> <li>• a column named <code>gs_genes</code> containing a comma separated vector of the gene names associated to the term, one for each term</li> <li>• the number of genes in the geneset of interest detected as differentially expressed (<code>gs_de_count</code>), or in the background set of genes (<code>gs_bg_count</code>)</li> </ul> <p>See <code>shake_topG0tableResult()</code> or <code>shake_enrichResult()</code> for examples of such formatting helpers</p> |
| <code>annotation_obj</code> | <p>A <code>data.frame</code> object, containing two columns, <code>gene_id</code> with a set of unambiguous identifiers (e.g. ENSEMBL ids) and <code>gene_name</code>, containing e.g. HGNC-based gene symbols. This object can be constructed via the <code>org.eg.XX.db</code> packages, e.g. with convenience functions such as <code>pcaExplorer::get_annotation_orgdb()</code>.</p>  |
| <code>gtl</code>            | <p>A GeneTonic-list object, containing in its slots the arguments specified above: <code>dds</code>, <code>res_de</code>, <code>res_enrich</code>, and <code>annotation_obj</code> - the names of the list <i>must</i> be specified following the content they are expecting</p>  |
| <code>project_id</code>     | <p>A character string, which can be considered as an identifier for the set/session, and will be e.g. used in the title of the report created via <code>happy_hour()</code></p>   |
| <code>size_gtl</code>       | <p>Numeric value, specifying the maximal size in MB for the accepted GeneTonicList object - this applies when uploading the dataset at runtime</p>  |

### Value

A Shiny app object is returned, for interactive data exploration

### Author(s)

Federico Marini

### 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)
dds_macrophage <- estimateSizeFactors(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)
  )

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOTableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

# now everything is in place to launch the app
if (interactive()) {
  GeneTonic(
    dds = dds_macrophage,
    res_de = res_de,
    res_enrich = res_enrich,
    annotation_obj = anno_df,
    project_id = "myexample"
  )
}
# alternatively...
gtl_macrophage <- GeneTonicList(
  dds = dds_macrophage,
  res_de = res_de,
  res_enrich = res_enrich,
  annotation_obj = anno_df
)
# GeneTonic(gtl = gtl_macrophage)

# if running it "as a server", without input data specified:
if (interactive()) {
  GeneTonic(size_gtl = 300) # for fairly large gtl objects
}

```

---

GeneTonic-pkg

*GeneTonic*


---

## Description

GeneTonic is a Bioconductor package that provides an interactive Shiny-based graphical user interface for streamlining the interpretation of RNA-seq data

## Details

GeneTonic simplifies and optimizes the integration of all components of Differential Expression analysis, with functional enrichment analysis and the original expression quantifications. It does so in a way that makes it easier to generate insightful observations and hypothesis - combining the benefits of interactivity and reproducibility, e.g. by capturing the features and gene sets of interest highlighted during the live session, and creating an HTML report as an artifact where text, code, and output coexist.

## Author(s)

Federico Marini <marinif@uni-mainz.de>

---

|               |                                      |
|---------------|--------------------------------------|
| GeneTonicList | <i>Create a GeneTonicList object</i> |
|---------------|--------------------------------------|

---

## Description

Create a list for GeneTonic from the single required components.

## Usage

```
GeneTonicList(dds, res_de, res_enrich, annotation_obj)
```

```
GeneTonic_list(dds, res_de, res_enrich, annotation_obj)
```

## Arguments

|                |  |
|----------------|--|
| dds            | A DESeqDataSet object, normally obtained after running your data through the DESeq2 framework.   |
| res_de         | A DESeqResults object. As for the dds parameter, this is also commonly used in the DESeq2 framework.   |
| res_enrich     | <p>A data.frame object, storing the result of the functional enrichment analysis. Required columns for enjoying the full functionality of <code>GeneTonic()</code> include:</p> <ul style="list-style-type: none"> <li>• a gene set identifier (e.g. GeneOntology id, <code>gs_id</code>) and its term description (<code>gs_description</code>)</li> <li>• a numeric value for the significance of the enrichment (<code>gs_pvalue</code>)</li> <li>• a column named <code>gs_genes</code> containing a comma separated vector of the gene names associated to the term, one for each term</li> <li>• the number of genes in the geneset of interest detected as differentially expressed (<code>gs_de_count</code>), or in the background set of genes (<code>gs_bg_count</code>)</li> </ul> <p>See <code>shake_topG0tableResult()</code> or <code>shake_enrichResult()</code> for examples of such formatting helpers</p> |
| annotation_obj | A data.frame object, containing two columns, <code>gene_id</code> with a set of unambiguous identifiers (e.g. ENSEMBL ids) and <code>gene_name</code> , containing e.g. HGNC-based gene symbols. This object can be constructed via the <code>org.eg.XX.db</code> packages, e.g. with convenience functions such as <code>pcaExplorer::get_annotation_organismdb()</code> .  |

## Details

Having this dedicated function saves the pain of remembering which names the components of the list should have. For backwards compatibility, the `GeneTonic_list` function is still provided as a synonym, and will likely be deprecated in the upcoming release cycles.

## Value

A `GeneTonic-list` object, containing in its named slots the arguments specified above: `dds`, `res_de`, `res_enrich`, and `annotation_obj` - the names of the list are specified following the requirements for using it as single input to `GeneTonic()`

## Author(s)

Federico Marini

## 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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

gtl_macrophage <- GeneTonicList(
```

```

dds = dds_macrophage,
res_de = res_de,
res_enrich = res_enrich,
annotation_obj = anno_df
)

# now everything is in place to launch the app
if (interactive()) {
  GeneTonic(gtl = gtl_macrophage)
}

```

---

gene\_plot

*Plot expression values for a gene*


---

### Description

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

### Usage

```

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

```

### Arguments

|                |   |
|----------------|---|
| dds            | 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(dds) to use for grouping. Note: the vector components should be categorical variables. |
| assay          | Character, specifies with assay of the dds 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.  |
| gtl            | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting  |

## 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)
dds_macrophage <- estimateSizeFactors(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(dds_macrophage,
  gene = "ENSG00000125347",
  intgroup = "condition",
  annotation_obj = anno_df
)
```

---

|                |  |
|----------------|--|
| get_aggrscores | <i>Compute aggregated scores for gene sets</i> |
|----------------|--|

---

### Description

Computes for each gene set in the `res_enrich` object a Z score and an aggregated score (using the `log2FoldChange` values, provided in the `res_de`)

### Usage

```
get_aggrscores(res_enrich, res_de, annotation_obj, gtl = NULL, aggrfun = mean)
```

### Arguments

|                             |   |
|-----------------------------|---|
| <code>res_enrich</code>     | A <code>data.frame</code> object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present).   |
| <code>res_de</code>         | A <code>DESeqResults</code> object.   |
| <code>annotation_obj</code> | A <code>data.frame</code> object with the feature annotation information, with at least two columns, <code>gene_id</code> and <code>gene_name</code> .  |
| <code>gtl</code>            | A <code>GeneTonic-list</code> object, containing in its slots the arguments specified above: <code>dds</code> , <code>res_de</code> , <code>res_enrich</code> , and <code>annotation_obj</code> - the names of the list <i>must</i> be specified following the content they are expecting |
| <code>aggrfun</code>        | Specifies the function to use for aggregating the scores for each term. Common values could be <code>mean</code> or <code>median</code> .   |

### Value

A `data.frame` with the same columns as provided in the input, with additional information on the `z_score` and the `aggr_score` for each gene set. This information is used by other functions such as [gs\\_volcano\(\)](#) or [enrichment\\_map\(\)](#)

### See Also

[gs\\_volcano\(\)](#) and [enrichment\\_map\(\)](#) make efficient use of the computed aggregated scores

**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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)

res_enrich <- get_aggrscores(
  res_enrich,
  res_de,
  anno_df
)
```

---

get\_expression\_values *Get expression values*

---

**Description**

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

**Usage**

```
get_expression_values(
  dds,
```

```

    gene,
    intgroup,
    assay = "counts",
    normalized = TRUE,
    gtl = NULL
  )

```

### Arguments

|            |   |
|------------|---|
| dds        | 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(dds) to use for grouping.  |
| assay      | Character, specifies with assay of the dds 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"  |
| gtl        | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting |

### 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)
dds_macrophage <- estimateSizeFactors(dds_macrophage)

df_exp <- get_expression_values(dds_macrophage,
  gene = "ENSG00000125347",
  intgroup = "condition"
)
head(df_exp)

```



ggs\_backbone

*Extract the backbone for the gene-geneset graph***Description**

Extract the backbone for the gene-geneset graph, either for the genes or for the genesets

**Usage**

```
ggs_backbone(
  res_enrich,
  res_de,
  annotation_obj = NULL,
  gtl = NULL,
  n_gs = 15,
  gs_ids = NULL,
  bb_on = c("genesets", "features"),
  bb_method = c("sdsm", "fdsm", "fixedrow"),
  bb_extract_alpha = 0.05,
  bb_extract_fwer = c("none", "bonferroni", "holm"),
  bb_fullinfo = FALSE,
  bb_remove_singletons = TRUE,
  color_graph = TRUE,
  color_by_geneset = "z_score",
  color_by_feature = "log2FoldChange",
  ...
)
```

**Arguments**

|                |  |
|----------------|--|
| res_enrich     | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present). |
| res_de         | A DESeqResults object.   |
| annotation_obj | A data.frame object with the feature annotation information, with at least two columns, gene_id and gene_name.   |
| gtl            | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting          |
| n_gs           | Integer value, corresponding to the maximal number of gene sets to be included   |
| gs_ids         | Character vector, containing a subset of gs_id as they are available in res_enrich. Lists the gene sets to be included in addition to the top ones (via n_gs)  |
| bb_on          | A character string, either "genesets" or "features", to specify which entity should be based the backbone graph on.  |

|                                   |   |
|-----------------------------------|---|
| <code>bb_method</code>            | A character string, referring to the function to be called ( from the backbone package) for computing the backbone of the specified bipartite graph. Defaults to "sdsm", as recommended in the backbone package.  |
| <code>bb_extract_alpha</code>     | A numeric value, specifying the significance level to use when detecting the backbone of the network  |
| <code>bb_extract_fwer</code>      | A character string, defaulting to "none", specifying which method to use for the multiple testing correction for controlling the family-wise error rate   |
| <code>bb_fullinfo</code>          | Logical value, determining what will be returned as output: either a simple <code>igraph</code> object with the graph backbone (if set to FALSE), or a list object containing also the backbone object, and the gene-geneset graph used for the computation (if TRUE) |
| <code>bb_remove_singletons</code> | Logical value, defines whether to remove or leave in the returned graph the nodes that are not connected to other vertices  |
| <code>color_graph</code>          | Logical value, specifies whether to use information about genesets or features to colorize the nodes, e.g. for this info to be used in interactive versions of the graph  |
| <code>color_by_geneset</code>     | Character string, corresponding to the column in <code>res_enrich</code> to be used for coloring the nodes if <code>bb_on</code> is set to "genesets". Defaults to the "z_score", which can be obtained via <code>get_aggrscores()</code>                             |
| <code>color_by_feature</code>     | Character string, corresponding to the column in <code>res_de</code> to be used for coloring the nodes if <code>bb_on</code> is set to "features". Defaults to the "log2FoldChange", which should be normally included in a <code>DESeqResults</code> object.         |
| <code>...</code>                  | Additional parameters to be passed internally   |

## Value

According to the `bb_fullinfo`, either a simple `igraph` object with the graph backbone, or a named list object containing:

- the `igraph` of the extracted backbone
- the backbone object itself
- the gene-geneset graph used for the computation

## 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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

ggs_bbg <- ggs_backbone(res_enrich,
  res_de,
  anno_df,
  n_gs = 50,
  bb_on = "genesets",
  color_graph = TRUE,
  color_by_geneset = "z_score"
)
plot(ggs_bbg)

# if desired, one can also plot the interactive version
visNetwork::visIgraph(ggs_bbg)

```

---

ggs\_graph

*Construct a gene-geneset-graph*


---

## Description

Construct a gene-geneset-graph from the results of a functional enrichment analysis

## Usage

```

ggs_graph(
  res_enrich,
  res_de,
  annotation_obj = NULL,

```

```

gtl = NULL,
n_gs = 15,
gs_ids = NULL,
prettify = TRUE,
geneset_graph_color = "gold",
genes_graph_colpal = NULL
)

```

### Arguments

|                                  |  |
|----------------------------------|--|
| <code>res_enrich</code>          | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <code>GeneTonic()</code> , to check the formatting requirements (a minimal set of columns should be present).  |
| <code>res_de</code>              | A DESeqResults object.   |
| <code>annotation_obj</code>      | A data.frame object with the feature annotation information, with at least two columns, <code>gene_id</code> and <code>gene_name</code> .  |
| <code>gtl</code>                 | A GeneTonic-list object, containing in its slots the arguments specified above: <code>dds</code> , <code>res_de</code> , <code>res_enrich</code> , and <code>annotation_obj</code> - the names of the list <i>must</i> be specified following the content they are expecting |
| <code>n_gs</code>                | Integer value, corresponding to the maximal number of gene sets to be included   |
| <code>gs_ids</code>              | Character vector, containing a subset of <code>gs_id</code> as they are available in <code>res_enrich</code> . Lists the gene sets to be included in addition to the top ones (via <code>n_gs</code> )   |
| <code>prettify</code>            | Logical, controlling the aspect of the returned graph object. If TRUE (default value), different shapes of the nodes are returned, based on the node type  |
| <code>geneset_graph_color</code> | Character value, specifying which color should be used for the fill of the shapes related to the gene sets.  |
| <code>genes_graph_colpal</code>  | A vector of colors, also provided with their hex string, to be used as a palette for coloring the gene nodes. If unspecified, defaults to a color ramp palette interpolating from blue through yellow to red.  |

### Value

An `igraph` object to be further manipulated or processed/plotted (e.g. via `igraph::plot.igraph()` or `visNetwork::visIgraph()`)

### 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)

```

```

dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

ggs <- ggs_graph(
  res_enrich,
  res_de,
  anno_df
)

ggs

#' # could be viewed interactively with
# library(visNetwork)
# library(magrittr)
# ggs %>%
#   visIgraph() %>%
#   visOptions(highlightNearest = list(enabled = TRUE,
#                                       degree = 1,
#                                       hover = TRUE),
#             nodesIdSelection = TRUE)

```

---

gostres\_macrophage      *A sample output from g:Profiler*

---

## Description

A sample output object as created from a call to `g:Profiler`, with the interface provided by `gprofiler2` - using the `gost()` function

**Details**

This object has been created on the data from the macrophage package by analyzing downstream the differentially expressed genes when comparing IFNg treated samples vs naive samples, accounting for the different cell lines included.

Details on how this object has been created are included in the `create_gt_data.R` script, included in the `scripts` folder of the `GeneTonic` package.

**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**

Other pathway-analysis-results: [enrichr\\_output\\_macrophage](#), [topgoDE\\_macrophage\\_IFNg\\_vs\\_naive](#)

---

go\_2\_html

*Information on a GeneOntology identifier*

---

**Description**

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

**Usage**

```
go_2_html(go_id, res_enrich = NULL)
```

**Arguments**

|                         |  |
|-------------------------|--|
| <code>go_id</code>      | Character, specifying the GeneOntology identifier for which to retrieve information  |
| <code>res_enrich</code> | A <code>data.frame</code> 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. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present). |

**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_2_html("GO:0002250")
go_2_html("GO:0043368")
```

---

gs\_alluvial

*Alluvial (sankey) plot for a set of genesets and the associated genes*


---

**Description**

Generate an interactive alluvial plot linking genesets to their associated genes

**Usage**

```
gs_alluvial(
  res_enrich,
  res_de,
  annotation_obj,
  gtl = NULL,
  n_gs = 5,
  gs_ids = NULL
)
```

```
gs_sankey(
  res_enrich,
  res_de,
  annotation_obj,
  gtl = NULL,
  n_gs = 5,
  gs_ids = NULL
)
```

**Arguments**

|                |  |
|----------------|--|
| res_enrich     | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present). |
| res_de         | A DESeqResults object.   |
| annotation_obj | A data.frame object with the feature annotation information, with at least two columns, gene_id and gene_name.   |
| gtl            | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting          |
| n_gs           | Integer value, corresponding to the maximal number of gene sets to be displayed  |
| gs_ids         | Character vector, containing a subset of gs_id as they are available in res_enrich. Lists the gene sets to be displayed.   |

**Value**

A plotly object

**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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

gs_alluvial(
  res_enrich = res_enrich,
  res_de = res_de,
  annotation_obj = anno_df,
  n_gs = 4
)
# or using the alias...
gs_sankey(
  res_enrich = res_enrich,
  res_de = res_de,
  annotation_obj = anno_df,
  n_gs = 4
)
```



---

|           |  |
|-----------|--|
| gs_dendro | <i>Dendrogram of the gene set enrichment results</i> |
|-----------|--|

---

## Description

Calculate (and plot) the dendrogram of the gene set enrichment results

## Usage

```
gs_dendro(
  res_enrich,
  gtl = NULL,
  n_gs = nrow(res_enrich),
  gs_ids = NULL,
  gs_dist_type = "kappa",
  clust_method = "ward.D2",
  color_leaves_by = "z_score",
  size_leaves_by = "gs_pvalue",
  color_branches_by = "clusters",
  create_plot = TRUE
)
```

## Arguments

|                 |   |
|-----------------|---|
| res_enrich      | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to see the formatting requirements.                                       |
| gtl             | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting |
| n_gs            | Integer value, corresponding to the maximal number of gene sets to be included (from the top ranked ones). Defaults to the number of rows of res_enrich   |
| gs_ids          | Character vector, containing a subset of gs_id as they are available in res_enrich. Lists the gene sets to be included, additionally to the ones specified via n_gs. Defaults to NULL.                                |
| gs_dist_type    | Character string, specifying which type of similarity (and therefore distance measure) will be used. Defaults to kappa, which uses <a href="#">create_kappa_matrix()</a>  |
| clust_method    | Character string defining the agglomeration method to be used for the hierarchical clustering. See <a href="#">stats::hclust()</a> for details, defaults to ward.D2   |
| color_leaves_by | Character string, which columns of res_enrich will define the color of the leaves. Defaults to z_score  |
| size_leaves_by  | Character string, which columns of res_enrich will define the size of the leaves. Defaults to the gs_pvalue   |

`color_branches_by` Character string, which columns of `res_enrich` will define the color of the branches. Defaults to `clusters`, which calls `dynamicTreeCut::cutreeDynamic()` to define the clusters

`create_plot` Logical, whether to create the plot as well.

## Value

A dendrogram object is returned invisibly, and a plot can be generated as well on that object.

## 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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

gs_dendro(res_enrich,
  n_gs = 100
)
```

---

gs\_fuzzyclustering      *Compute fuzzy clusters of gene sets*

---

### Description

Compute fuzzy clusters of different gene sets, aiming to identify grouped categories that can better represent the distinct biological themes in the enrichment results

### Usage

```
gs_fuzzyclustering(
  res_enrich,
  gtl = NULL,
  n_gs = nrow(res_enrich),
  gs_ids = NULL,
  similarity_matrix = NULL,
  similarity_threshold = 0.35,
  fuzzy_seeding_initial_neighbors = 3,
  fuzzy_multilinkage_rule = 0.5
)
```

### Arguments

|                                 |   |
|---------------------------------|---|
| res_enrich                      | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present).  |
| gtl                             | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting   |
| n_gs                            | Integer value, corresponding to the maximal number of gene sets to be displayed   |
| gs_ids                          | Character vector, containing a subset of gs_id as they are available in res_enrich. Lists the gene sets to be displayed.  |
| similarity_matrix               | A similarity matrix between gene sets. Can be e.g. computed with <a href="#">create_kappa_matrix()</a> or <a href="#">create_jaccard_matrix()</a> or a similar function, returning a symmetric matrix with numeric values (max = 1). If not provided, this will be computed on the fly with <a href="#">create_kappa_matrix()</a>                           |
| similarity_threshold            | A numeric value for the similarity matrix, used to determine the initial seeds as in the implementation of DAVID. Higher values will lead to more genesets being initially unclustered, leading to a functional classification result with fewer groups and fewer geneset members. Defaults to 0.35, recommended to not go below 0.3 (see DAVID help pages) |
| fuzzy_seeding_initial_neighbors | Integer value, corresponding to the minimum geneset number in a seeding group. Lower values will lead to the inclusion of more genesets in the functional groups, and may generate a lot of small size groups. Defaults to 3  |

**fuzzy\_multilinkage\_rule**

Numeric value, comprised between 0 and 1. This parameter will determine how the seeding groups merge with each other, by specifying the percentage of shared genesets required to merge the two subsets into one group. Higher values will give sharper separation between the groups of genesets. Defaults to 0.5 (50%)

**Value**

A data frame, shaped in a similar way as the originally provided `res_enrich` object, containing two extra columns: `gs_fuzzycluster`, to specify the identifier of the fuzzy cluster of genesets, and `gs_cluster_status`, which can specify whether the geneset is the "Representative" for that cluster or a simple "Member". Notably, the number of rows in the returned object can be higher than the original number of rows in `res_enrich`.

**References**

See [https://david.ncicrf.gov/helps/functional\\_classification.html#clustering](https://david.ncicrf.gov/helps/functional_classification.html#clustering) for details on the original implementation

**Examples**

```
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
# taking a smaller subset
res_enrich_subset <- res_enrich[1:100, ]

fuzzy_subset <- gs_fuzzyclustering(
  res_enrich = res_enrich_subset,
  n_gs = nrow(res_enrich_subset),
  gs_ids = NULL,
  similarity_matrix = NULL,
  similarity_threshold = 0.35,
  fuzzy_seeding_initial_neighbors = 3,
  fuzzy_multilinkage_rule = 0.5
)

# show all genesets members of the first cluster
fuzzy_subset[fuzzy_subset$gs_fuzzycluster == "1", ]

# list only the representative clusters
head(fuzzy_subset[fuzzy_subset$gs_cluster_status == "Representative", ], 10)
```

---

 gs\_heatmap

*Plot a heatmap of the gene signature on the data*


---

**Description**

Plot a heatmap for the selected gene signature on the provided data, with the possibility to compactly display also DE only genes

**Usage**

```
gs_heatmap(
  se,
  res_de,
  res_enrich,
  annotation_obj = NULL,
  gtl = NULL,
  geneset_id = NULL,
  genelist = NULL,
  FDR = 0.05,
  de_only = FALSE,
  cluster_rows = TRUE,
  cluster_columns = FALSE,
  center_mean = TRUE,
  scale_row = FALSE,
  winsorize_threshold = NULL,
  anno_col_info = NULL,
  plot_title = NULL,
  ...
)
```

**Arguments**

|                 |  |
|-----------------|--|
| se              | A SummarizedExperiment object, or an object derived from this class, such as a DESeqTransform object (variance stabilized transformed data, or regularized logarithm transformed), in where the transformation has been applied to make the data more homoscedastic and thus a better fit for visualization. |
| res_de          | A DESeqResults object.   |
| res_enrich      | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present).   |
| annotation_obj  | A data.frame object with the feature annotation information, with at least two columns, gene_id and gene_name.   |
| gtl             | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting  |
| geneset_id      | Character specifying the gene set identifier to be plotted   |
| genelist        | A vector of character strings, specifying the identifiers contained in the row names of the se input object.   |
| FDR             | Numeric value, specifying the significance level for thresholding adjusted p-values. Defaults to 0.05.   |
| de_only         | Logical, whether to include only differentially expressed genes in the plot  |
| cluster_rows    | Logical, determining if rows should be clustered, as specified by <a href="#">ComplexHeatmap::Heatmap()</a>  |
| cluster_columns | Logical, determining if columns should be clustered, as specified by <a href="#">ComplexHeatmap::Heatmap()</a>   |

|                     |  |
|---------------------|--|
| center_mean         | Logical, whether to perform mean centering on the row-wise   |
| scale_row           | Logical, whether to standardize by row the expression values   |
| winsorize_threshold | Numeric value, to be applied as value to winsorize the extreme values of the heatmap. Should be a positive number. Defaults to NULL, which corresponds to not applying any winsorization. Suggested values: enter 2 or 3 if using row-standardized values (scale_row is TRUE), or visually inspect the range of the values if using simply mean centered values. |
| anno_col_info       | A character vector of names in colData(dds) to use for decorating the heatmap as annotation.   |
| plot_title          | Character string, to specify the title of the plot, displayed over the heatmap. If left to NULL as by default, it tries to use the information on the geneset identifier provided  |
| ...                 | Additional arguments passed to other methods, e.g. in the call to <a href="#">ComplexHeatmap::Heatmap()</a>  |

### Value

A plot returned by the [ComplexHeatmap::Heatmap\(\)](#) function

### 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)
dds_macrophage <- estimateSizeFactors(dds_macrophage)

vst_macrophage <- vst(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive
```

```

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

gs_heatmap(vst_macrophage,
  res_de,
  res_enrich,
  anno_df,
  geneset_id = res_enrich$gs_id[1],
  cluster_columns = TRUE,
  anno_col_info = "condition"
)

```

---

gs\_horizon

*Plots a summary of enrichment results*


---

## Description

Plots a summary of enrichment results - horizon plot to compare one or more sets of results

## Usage

```

gs_horizon(
  res_enrich,
  compared_res_enrich_list,
  n_gs = 20,
  p_value_column = "gs_pvalue",
  color_by = "z_score",
  ref_name = "ref_scenario",
  sort_by = c("clustered", "first_set")
)

```

## Arguments

|                          |   |
|--------------------------|---|
| res_enrich               | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present).            |
| compared_res_enrich_list | A named list, where each element is a data.frame formatted like the standard res_enrich objects used by GeneTonic. The names of the list are the names of the scenarios.  |
| n_gs                     | Integer value, corresponding to the maximal number of gene sets to be displayed   |
| p_value_column           | Character string, specifying the column of res_enrich where the p-value to be represented is specified. Defaults to gs_pvalue (it could have other values, in case more than one p-value - or an adjusted p-value - have been specified). |

|          |  |
|----------|--|
| color_by | Character, specifying the column of res_enrich to be used for coloring the plotted gene sets. Defaults sensibly to z_score.  |
| ref_name | Character, defining the name of the scenario to compare against (the one in res_enrich) - defaults to "ref_scenario".  |
| sort_by  | Character string, either "clustered", or "first_set". This controls the sorting order of the included terms in the final plot. "clustered" presents the terms grouped by the scenario where they assume the highest values. "first_set" sorts the terms by the significance value in the reference scenario. |

### Details

It makes sense to have the results in res\_enrich sorted by increasing gs\_pvalue, to make sure the top results are first sorted by the significance (when selecting the common gene sets across the res\_enrich elements provided in compared\_res\_enrich\_list)

The gene sets included are a subset of the ones in common to all different scenarios included in res\_enrich and the elements of compared\_res\_enrich\_list.

### Value

A ggplot object

### See Also

[gs\\_summary\\_overview\(\)](#), [gs\\_summary\\_overview\\_pair\(\)](#)

### 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)
dds_macrophage <- estimateSizeFactors(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)
)
```



```

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOTableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

res_enrich2 <- res_enrich[1:42, ]
res_enrich3 <- res_enrich[1:42, ]
res_enrich4 <- res_enrich[1:42, ]

set.seed(2 * 42)
shuffled_ones_2 <- sample(seq_len(42)) # to generate permuted p-values
res_enrich2$gs_pvalue <- res_enrich2$gs_pvalue[shuffled_ones_2]
res_enrich2$z_score <- res_enrich2$z_score[shuffled_ones_2]
res_enrich2$aggr_score <- res_enrich2$aggr_score[shuffled_ones_2]

set.seed(3 * 42)
shuffled_ones_3 <- sample(seq_len(42)) # to generate permuted p-values
res_enrich3$gs_pvalue <- res_enrich3$gs_pvalue[shuffled_ones_3]
res_enrich3$z_score <- res_enrich3$z_score[shuffled_ones_3]
res_enrich3$aggr_score <- res_enrich3$aggr_score[shuffled_ones_3]

set.seed(4 * 42)
shuffled_ones_4 <- sample(seq_len(42)) # to generate permuted p-values
res_enrich4$gs_pvalue <- res_enrich4$gs_pvalue[shuffled_ones_4]
res_enrich4$z_score <- res_enrich4$z_score[shuffled_ones_4]
res_enrich4$aggr_score <- res_enrich4$aggr_score[shuffled_ones_4]

compa_list <- list(
  scenario2 = res_enrich2,
  scenario3 = res_enrich3,
  scenario4 = res_enrich4
)

gs_horizon(res_enrich,
  compared_res_enrich_list = compa_list,
  n_gs = 50,
  sort_by = "clustered"
)
gs_horizon(res_enrich,
  compared_res_enrich_list = compa_list,
  n_gs = 20,
  sort_by = "first_set"
)

```

**Description**

Multi Dimensional Scaling plot for gene sets, extracted from a `res_enrich` object

**Usage**

```
gs_mds(
  res_enrich,
  res_de,
  annotation_obj,
  gtl = NULL,
  n_gs = nrow(res_enrich),
  gs_ids = NULL,
  similarity_measure = "kappa_matrix",
  mds_k = 2,
  mds_labels = 0,
  mds_colorby = "z_score",
  gs_labels = NULL,
  plot_title = NULL,
  return_data = FALSE
)
```

**Arguments**

|                                 |   |
|---------------------------------|---|
| <code>res_enrich</code>         | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <code>GeneTonic()</code> , to check the formatting requirements (a minimal set of columns should be present).   |
| <code>res_de</code>             | A <code>DESeqResults</code> object.   |
| <code>annotation_obj</code>     | A data.frame object with the feature annotation information, with at least two columns, <code>gene_id</code> and <code>gene_name</code> .   |
| <code>gtl</code>                | A <code>GeneTonic-list</code> object, containing in its slots the arguments specified above: <code>dds</code> , <code>res_de</code> , <code>res_enrich</code> , and <code>annotation_obj</code> - the names of the list <i>must</i> be specified following the content they are expecting |
| <code>n_gs</code>               | Integer value, corresponding to the maximal number of gene sets to be included (from the top ranked ones). Defaults to the number of rows of <code>res_enrich</code>  |
| <code>gs_ids</code>             | Character vector, containing a subset of <code>gs_id</code> as they are available in <code>res_enrich</code> . Lists the gene sets to be included, additionally to the ones specified via <code>n_gs</code> . Defaults to <code>NULL</code> .   |
| <code>similarity_measure</code> | Character, currently defaults to <code>kappa_matrix</code> , to specify how to compute the similarity measure between gene sets   |
| <code>mds_k</code>              | Integer value, number of dimensions to compute in the multi dimensional scaling procedure   |
| <code>mds_labels</code>         | Integer, defines the number of labels to be plotted on top of the scatter plot for the provided gene sets.  |
| <code>mds_colorby</code>        | Character specifying the column of <code>res_enrich</code> to be used for coloring the plotted gene sets. Defaults sensibly to <code>z_score</code> .   |

|             |   |
|-------------|---|
| gs_labels   | Character vector, containing a subset of gs_id as they are available in res_enrich. Lists the gene sets to be labeled.                                |
| plot_title  | Character string, used as title for the plot. If left NULL, it defaults to a general description of the plot and of the DE contrast                   |
| return_data | Logical, whether the function should just return the data.frame of the MDS coordinates, related to the original res_enrich object. Defaults to FALSE. |

**Value**

A ggplot object

**See Also**

[create\\_kappa\\_matrix\(\)](#) is used to calculate the similarity between gene sets

**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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

gs_mds(res_enrich,
  res_de,
```

```

anno_df,
n_gs = 200,
mds_labels = 10
)

```

---

gs\_radar

*Radar (spider) plot for gene sets*


---

### Description

Radar (spider) plot for gene sets, either for one or more results from functional enrichment analysis.

### Usage

```

gs_radar(
  res_enrich,
  res_enrich2 = NULL,
  n_gs = 20,
  p_value_column = "gs_pvalue"
)

```

```

gs_spider(
  res_enrich,
  res_enrich2 = NULL,
  n_gs = 20,
  p_value_column = "gs_pvalue"
)

```

### Arguments

|                |   |
|----------------|---|
| res_enrich     | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present).                      |
| res_enrich2    | Analogous to res_enrich1, another data.frame object, storing the result of the functional enrichment analysis, but for a different setting (e.g. another contrast). Defaults to NULL (in this case, a single set of enrichment results is plotted). |
| n_gs           | Integer value, corresponding to the maximal number of gene sets to be displayed   |
| p_value_column | Character string, specifying the column of res_enrich where the p-value to be represented is specified. Defaults to gs_pvalue (it could have other values, in case more than one p-value - or an adjusted p-value - have been specified).           |

### Value

A plotly object

**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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)
gs_radar(res_enrich = res_enrich)
# or using the alias...
gs_spider(res_enrich = res_enrich)

# with more than one set
res_enrich2 <- res_enrich[1:60, ]
set.seed(42)
shuffled_ones <- sample(seq_len(60)) # to generate permuted p-values
res_enrich2$gs_pvalue <- res_enrich2$gs_pvalue[shuffled_ones]
# ideally, I would also permute the z scores and aggregated scores
gs_radar(
  res_enrich = res_enrich,
  res_enrich2 = res_enrich2
)

```

**Description**

Compute gene set scores for each sample, by transforming the gene-wise change to a geneset-wise change

**Usage**

```
gs_scores(se, res_de, res_enrich, annotation_obj = NULL, gtl = NULL)
```

**Arguments**

|                |  |
|----------------|--|
| se             | A SummarizedExperiment object, or an object derived from this class, such as a DESeqTransform object (variance stabilized transformed data, or regularized logarithm transformed), in where the transformation has been applied to make the data more homoscedastic and thus a better fit for visualization. |
| res_de         | A DESeqResults object.   |
| res_enrich     | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present).   |
| annotation_obj | A data.frame object with the feature annotation information, with at least two columns, gene_id and gene_name.   |
| gtl            | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting  |

**Value**

A matrix with the geneset Z scores, e.g. to be plotted with [gs\\_scoresheat\(\)](#)

**See Also**

[gs\\_scoresheat\(\)](#) plots these scores

**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)
dds_macrophage <- estimateSizeFactors(dds_macrophage)

vst_macrophage <- vst(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

scores_mat <- gs_scores(
  vst_macrophage,
  res_de,
  res_enrich[1:50, ],
  anno_df
)
```

---

gs\_scoresheat

*Plots a matrix of geneset scores*

---

## Description

Plots a matrix of geneset Z scores, across all samples

## Usage

```
gs_scoresheat(
  mat,
  n_gs = nrow(mat),
  gs_ids = NULL,
  clustering_distance_rows = "euclidean",
  clustering_distance_cols = "euclidean",
  cluster_rows = TRUE,
  cluster_cols = TRUE
)
```

## Arguments

mat                    A matrix, e.g. returned by the `gs_scores()` function

|                          |  |
|--------------------------|--|
| n_gs                     | Integer value, corresponding to the maximal number of gene sets to be displayed.   |
| gs_ids                   | Character vector, containing a subset of gs_id as they are available in res_enrich. Lists the gene sets to be displayed. |
| clustering_distance_rows | Character, a distance measure used in clustering rows  |
| clustering_distance_cols | Character, a distance measure used in clustering columns   |
| cluster_rows             | Logical, determining if rows should be clustered   |
| cluster_cols             | Logical, determining if columns should be clustered  |

**Value**

A ggplot object

**See Also**

[gs\\_scores\(\)](#) computes the scores plotted by this function

**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)
dds_macrophage <- estimateSizeFactors(dds_macrophage)

vst_macrophage <- vst(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive
```



```
# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

scores_mat <- gs_scores(
  vst_macrophage,
  res_de,
  res_enrich[1:30, ],
  anno_df
)
gs_scoresheat(scores_mat,
  n_gs = 30
)
```

---

gs\_simplify

*Simplify results from functional enrichment analysis*

---

## Description

Simplify results from functional enrichment analysis, removing genesets that are redundant to enhance interpretation of the results

## Usage

```
gs_simplify(res_enrich, gs_overlap = 0.75)
```

## Arguments

|            |  |
|------------|--|
| res_enrich | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present).   |
| gs_overlap | Numeric value, which defines the threshold for removing terms that present an overlap greater than the specified value. Changing its value can control the granularity of how redundant terms are removed from the original res_enrich for the next steps, e.g. plotting this via <a href="#">gs_volcano()</a> |

## Value

A data.frame with a subset of the original gene sets

## See Also

[gs\\_volcano\(\)](#) and [ggs\\_graph\(\)](#) can e.g. show an overview on the simplified table of gene sets

**Examples**

```
# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)

dim(res_enrich)
res_enrich_simplified <- gs_simplify(res_enrich)
dim(res_enrich_simplified)
# and then use this further for all other functions expecting a res_enrich
```

---

|                              |   |
|------------------------------|---|
| <code>gs_summary_heat</code> | <i>Plots a heatmap for genes and genesets</i> |
|------------------------------|---|

---

**Description**

Plots a heatmap for genes and genesets, useful to spot out intersections across genesets and an overview of them

**Usage**

```
gs_summary_heat(res_enrich, res_de, annotation_obj, gtl = NULL, n_gs = 80)
```

**Arguments**

|                             |  |
|-----------------------------|--|
| <code>res_enrich</code>     | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present).   |
| <code>res_de</code>         | A DESeqResults object.   |
| <code>annotation_obj</code> | A data.frame object with the feature annotation information, with at least two columns, <code>gene_id</code> and <code>gene_name</code> .  |
| <code>gtl</code>            | A GeneTonic-list object, containing in its slots the arguments specified above: <code>dds</code> , <code>res_de</code> , <code>res_enrich</code> , and <code>annotation_obj</code> - the names of the list <i>must</i> be specified following the content they are expecting |
| <code>n_gs</code>           | Integer value, corresponding to the maximal number of gene sets to be displayed  |

**Value**

A ggplot object

**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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

gs_summary_heat(
  res_enrich = res_enrich,
  res_de = res_de,
  annotation_obj = anno_df,
  n_gs = 20
)
```

---

gs\_summary\_overview *Plots a summary of enrichment results*

---

## Description

Plots a summary of enrichment results for one set

## Usage

```
gs_summary_overview(
  res_enrich,
  gtl = NULL,
  n_gs = 20,
  p_value_column = "gs_pvalue",
  color_by = "z_score",
  return_barchart = FALSE
)
```

**Arguments**

|                              |  |
|------------------------------|--|
| <code>res_enrich</code>      | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <code>GeneTonic()</code> , to check the formatting requirements (a minimal set of columns should be present).  |
| <code>gtl</code>             | A GeneTonic-list object, containing in its slots the arguments specified above: <code>dds</code> , <code>res_de</code> , <code>res_enrich</code> , and <code>annotation_obj</code> - the names of the list <i>must</i> be specified following the content they are expecting |
| <code>n_gs</code>            | Integer value, corresponding to the maximal number of gene sets to be displayed  |
| <code>p_value_column</code>  | Character string, specifying the column of <code>res_enrich</code> where the p-value to be represented is specified. Defaults to <code>gs_pvalue</code> (it could have other values, in case more than one p-value - or an adjusted p-value - have been specified).          |
| <code>color_by</code>        | Character, specifying the column of <code>res_enrich</code> to be used for coloring the plotted gene sets. Defaults sensibly to <code>z_score</code> .   |
| <code>return_barchart</code> | Logical, whether to return a barchart (instead of the default dot-segment plot); defaults to FALSE.  |

**Value**

A ggplot object

**See Also**

[gs\\_summary\\_overview\\_pair\(\)](#), [gs\\_horizon\(\)](#)

**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)
dds_macrophage <- estimateSizeFactors(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)
)
```

```

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

gs_summary_overview(res_enrich)

# if desired, it can also be shown as a barplot
gs_summary_overview(res_enrich, n_gs = 30, return_barchart = TRUE)

```

---

```
gs_summary_overview_pair
```

*Plots a summary of enrichment results*

---

## Description

Plots a summary of enrichment results - for two sets of results

## Usage

```

gs_summary_overview_pair(
  res_enrich,
  res_enrich2,
  n_gs = 20,
  p_value_column = "gs_pvalue",
  color_by = "z_score",
  alpha_set2 = 1
)

```

## Arguments

|                |   |
|----------------|---|
| res_enrich     | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <code>GeneTonic()</code> , to check the formatting requirements (a minimal set of columns should be present).               |
| res_enrich2    | As res_enrich, the result of functional enrichment analysis, in a scenario/contrast different than the first set.   |
| n_gs           | Integer value, corresponding to the maximal number of gene sets to be displayed   |
| p_value_column | Character string, specifying the column of res_enrich where the p-value to be represented is specified. Defaults to gs_pvalue (it could have other values, in case more than one p-value - or an adjusted p-value - have been specified). |

|            |   |
|------------|---|
| color_by   | Character, specifying the column of res_enrich to be used for coloring the plotted gene sets. Defaults sensibly to z_score. |
| alpha_set2 | Numeric value, between 0 and 1, which specified the alpha transparency used for plotting the points for gene set 2.         |

**Value**

A ggplot object

**See Also**

[gs\\_summary\\_overview\(\)](#), [gs\\_horizon\(\)](#)

**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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

res_enrich2 <- res_enrich[1:42, ]
set.seed(42)
shuffled_ones <- sample(seq_len(42)) # to generate permuted p-values
res_enrich2$gs_pvalue <- res_enrich2$gs_pvalue[shuffled_ones]
res_enrich2$z_score <- res_enrich2$z_score[shuffled_ones]
```

```

res_enrich2$aggr_score <- res_enrich2$aggr_score[shuffled_ones]
# ideally, I would also permute the z scores and aggregated scores
gs_summary_overview_pair(
  res_enrich = res_enrich,
  res_enrich2 = res_enrich2
)

```

---

gs\_upset

*Upset plot for genesets*


---

## Description

Create an upset plot for genesets

## Usage

```

gs_upset(
  res_enrich,
  res_de = NULL,
  annotation_obj = NULL,
  n_gs = 10,
  gtl = NULL,
  gs_ids = NULL,
  add_de_direction = FALSE,
  add_de_gsgenes = FALSE,
  col_upDE = "#E41A1C",
  col_downDE = "#377EB8",
  upset_geom = geom_point(size = 2),
  return_upsetgsg = FALSE
)

```

## Arguments

|                |  |
|----------------|--|
| res_enrich     | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <a href="#">GeneTonic()</a> , to check the formatting requirements (a minimal set of columns should be present). |
| res_de         | A DESeqResults object.   |
| annotation_obj | A data.frame object with the feature annotation information, with at least two columns, gene_id and gene_name.   |
| n_gs           | Integer value, corresponding to the maximal number of gene sets to be included   |
| gtl            | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting          |
| gs_ids         | Character vector, containing a subset of gs_id as they are available in res_enrich. Lists the gene sets to be included in addition to the top ones (via n_gs)  |

**add\_de\_direction** Logical, whether to add an annotation with info on the DE direction of single genes  
**add\_de\_gsgenes** Logical, if set to TRUE adds an annotation with detail on the single components of each defined subset  
**col\_upDE** Character, specifying the color value to be used to mark upregulated genes  
**col\_downDE** Character, specifying the color value to be used to mark downregulated genes  
**upset\_geom** A geom specification to be used in the upset chart. Defaults sensibly to `geom_point(size = 2)`  
**return\_upsetgsg** Logical, controlling the returned value. If set to TRUE, this function will not generate the plot but only create the corresponding data.frame, in case the user wants to proceed with a custom call to create an upset plot.

### Value

A ggplot object (if plotting), or alternatively a data.frame

### 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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
  
```



```
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)
gs_upset(res_enrich,
  n_gs = 10
)

gs_upset(res_enrich,
  res_de = res_de, annotation_obj = anno_df,
  n_gs = 8,
  add_de_direction = TRUE, add_de_gsgenes = TRUE
)

# or using the practical gtl (GeneTonicList)
gtl_macrophage <- GeneTonic_list(
  dds = dds_macrophage,
  res_de = res_de,
  res_enrich = res_enrich,
  annotation_obj = anno_df
)

gs_upset(
  gtl = gtl_macrophage,
  n_gs = 15,
  add_de_direction = TRUE, add_de_gsgenes = TRUE
)
```

---

gs\_volcano

*Volcano plot for gene sets*

---

## Description

Volcano plot for gene sets, to summarize visually the functional enrichment results

## Usage

```
gs_volcano(
  res_enrich,
  gtl = NULL,
  p_threshold = 0.05,
  color_by = "aggr_score",
  volcano_labels = 10,
  scale_circles = 1,
  gs_ids = NULL,
  plot_title = NULL
)
```

## Arguments

**res\_enrich** A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, [GeneTonic\(\)](#), to check the formatting requirements (a minimal set of columns should be present). This object needs to be

|                             |  |
|-----------------------------|--|
|                             | processed first by a function such as <code>get_aggrscores()</code> to compute the term-wise <code>z_score</code> or <code>aggr_score</code> , which will be used for plotting   |
| <code>gtl</code>            | A GeneTonic-list object, containing in its slots the arguments specified above: <code>dds</code> , <code>res_de</code> , <code>res_enrich</code> , and <code>annotation_obj</code> - the names of the list <i>must</i> be specified following the content they are expecting |
| <code>p_threshold</code>    | Numeric, defines the threshold to be used for filtering the gene sets to display. Defaults to 0.05   |
| <code>color_by</code>       | Character specifying the column of <code>res_enrich</code> to be used for coloring the plotted gene sets. Defaults to <code>aggr_score</code> .  |
| <code>volcano_labels</code> | Integer, maximum number of labels for the gene sets to be plotted as labels on the volcano scatter plot.   |
| <code>scale_circles</code>  | A numeric value, to define the scaling factor for the circle sizes. Defaults to 1.   |
| <code>gs_ids</code>         | Character vector, containing a subset of <code>gs_id</code> as they are available in <code>res_enrich</code> . Lists the gene sets to be labeled.  |
| <code>plot_title</code>     | Character string, used as title for the plot. If left NULL, it defaults to a general description of the plot and of the DE contrast  |

### Details

It is also possible to reduce the redundancy of the input `res_enrich` object, if it is passed in advance to the `gs_simplify()` function.

### Value

A ggplot object

### See Also

`gs_simplify()` can be applied in advance to `res_enrich` to reduce the redundancy of the displayed gene sets

### 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)
dds_macrophage <- estimateSizeFactors(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)
  )

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

gs_volcano(res_enrich)
```

---

happy\_hour

*Happy hour!*

---

## Description

Start the happy hour, creating a report containing a document full of goodies derived from the provided objects.

## Usage

```
happy_hour(
  dds,
  res_de,
  res_enrich,
  annotation_obj,
  gtl = NULL,
  project_id,
  mygenesets,
  mygenes,
  mygroup = NULL,
  usage_mode = "batch_mode",
  input_rmd = NULL,
  output_file = "my_first_GeneTonic_happyhour.html",
  output_dir = tempdir(),
  output_format = NULL,
  force_overwrite = FALSE,
  knitr_show_progress = FALSE,
  ignore_pandoc = FALSE,
  open_after_creating = TRUE,
  ...
)
```

**Arguments**

|                     |   |
|---------------------|---|
| dds                 | A DESeqDataSet object, normally obtained after running your data through the DESeq2 framework.  |
| res_de              | A DESeqResults object. As for the dds parameter, this is also commonly used in the DESeq2 framework.  |
| res_enrich          | A data.frame object, storing the result of the functional enrichment analysis. See <a href="#">GeneTonic()</a> for the formatting requirements.   |
| annotation_obj      | A data.frame object with the feature annotation information, with at least two columns, gene_id and gene_name. See <a href="#">GeneTonic()</a> for the formatting requirements.                                       |
| gtl                 | A GeneTonic-list object, containing in its slots the arguments specified above: dds, res_de, res_enrich, and annotation_obj - the names of the list <i>must</i> be specified following the content they are expecting |
| project_id          | A character string, which can be considered as an identifier for the set/session, and will be e.g. used in the title of the report created via <a href="#">happy_hour()</a>   |
| mygenesets          | A vector of character strings, containing the genesets to focus on in the report - for each geneset, e.g. a signature heatmap can be created.   |
| mygenes             | A vector of character strings, containing the genes to focus on in the report - for each gene, the plot of the expression values is included.   |
| mygroup             | A character string, or a vector thereof. Contains the experimental variables to be used to split into groups the expression data, and color accordingly.  |
| usage_mode          | A character string, which controls the behavior of the Rmd document, based on whether the rendering is triggered while using the app ("shiny_mode"), or offline, in batch mode. Defaults to "batch_mode".             |
| input_rmd           | Character string with the path to the RMarkdown (.Rmd) file that will be used as the template for generating the report. Defaults to NULL, which will then use the one provided with the GeneTonic package.           |
| output_file         | Character string, specifying the file name of the output report. The file name extension must be either .html or .pdf, and consistent with the value of output_format.  |
| output_dir          | Character, defining the path to the output directory where the report will be generated. Defaults to the temp directory (tempdir()).  |
| output_format       | The format of the output report. Either html_document or pdf_document. The file name extension of output_file must be consistent with this choice. Can also be left empty and determined accordingly.                 |
| force_overwrite     | Logical, whether to force overwrite an existing report with the same name in the output directory. Defaults to FALSE.   |
| knitr_show_progress | Logical, whether to display the progress of knitr while generating the report. Defaults to FALSE.   |
| ignore_pandoc       | Logical, controlling how the report generation function will behave if pandoc or pandoc-citeproc are missing.   |

open\_after\_creating  
 Logical, whether to open the report in the default browser after being generated.  
 Defaults to TRUE.

... Other arguments that will be passed to `rmarkdown::render()`.

### Details

When `happy_hour` is called, a RMarkdown template file will be copied into the output directory, and `rmarkdown::render()` will be called to generate the final report.

As a default template, `happy_hour` uses the one delivered together with the `GeneTonic` package, which provides a comprehensive overview of what the user can extract. Experienced users can take that as a starting point to further edit and customize.

If there is already a `.Rmd` file with the same name in the output directory, the function will raise an error and stop, to avoid overwriting the existing file. The reason for this behaviour is that the copied template in the output directory will be deleted once the report is generated.

Credits to the original implementation proposed by Charlotte Sonesson, upon which this function is **heavily** inspired.

### Value

Generates a fully fledged report in the `output_dir` directory, called `output_file` and returns (invisibly) the name of the generated report.

### See Also

[GeneTonic\(\)](#), [shake\\_topGOtableResult\(\)](#), [shake\\_enrichResult\(\)](#)

### 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)
dds_macrophage <- estimateSizeFactors(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)
```

```

)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)
## Not run:
happy_hour(
  dds = dds_macrophage,
  res_de = res_de,
  res_enrich = res_enrich,
  annotation_obj = anno_df,
  project_id = "examplerun",
  mygroup = "condition",
  # mygroup = "line", # alternatively
  mygenesets = res_enrich$gs_id[c(1:5, 11, 31)],
  mygenes = c(
    "ENSG00000125347",
    "ENSG00000172399",
    "ENSG00000137496"
  )
)
)

## End(Not run)

```

---

map2color

*Maps numeric values to color values*


---

## Description

Maps numeric continuous values to values in a color palette

## Usage

```
map2color(x, pal, symmetric = TRUE, limits = NULL)
```

## Arguments

|           |   |
|-----------|---|
| x         | A character vector of numeric values (e.g. log2FoldChange values) to be converted to a vector of colors   |
| pal       | A vector of characters specifying the definition of colors for the palette, e.g. obtained via <a href="#">brewer.pal</a>                                  |
| symmetric | Logical value, whether to return a palette which is symmetrical with respect to the minimum and maximum values - "respecting" the zero. Defaults to TRUE. |
| limits    | A vector containing the limits of the values to be mapped. If not specified, defaults to the range of values in the x 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")
map2color(a, pal)
plot(a, col = map2color(a, pal), pch = 20, cex = 4)

b <- 1:50
pal2 <- grDevices::colorRampPalette(
  RColorBrewer::brewer.pal(name = "RdYlBu", 11)
)(50)
plot(b, col = map2color(b, pal2), pch = 20, cex = 3)
```

---

overlap\_coefficient    *Calculate overlap coefficient*

---

**Description**

Calculate similarity coefficient between two sets, based on the overlap

**Usage**

```
overlap_coefficient(x, y)
```

**Arguments**

|   |  |
|---|--|
| x | Character vector, corresponding to set 1 |
| y | Character vector, set 2                  |

**Value**

A numeric value between 0 and 1

**See Also**

[https://en.wikipedia.org/wiki/Overlap\\_coefficient](https://en.wikipedia.org/wiki/Overlap_coefficient)

**Examples**

```
a <- seq(1, 21, 2)
b <- seq(1, 11, 2)
overlap_coefficient(a, b)
```

---

overlap\_jaccard\_index *Calculate Jaccard Index between two sets*

---

**Description**

Calculate similarity coefficient with the Jaccard Index

**Usage**

```
overlap_jaccard_index(x, y)
```

**Arguments**

x                    Character vector, corresponding to set 1  
y                    Character vector, corresponding to set 2

**Value**

A numeric value between 0 and 1

**Examples**

```
a <- seq(1, 21, 2)  
b <- seq(1, 11, 2)  
overlap_jaccard_index(a, b)
```

---

res\_macrophage\_IFNg\_vs\_naive  
*A sample DESeqResults object*

---

**Description**

A sample DESeqResults object, generated in the DESeq2 framework

**Details**

This DESeqResults object on the data from the macrophage package has been created comparing IFNg treated samples vs naive samples, accounting for the different cell lines included.

Details on how this object has been created are included in the create\_gt\_data.R script, included in the scripts folder of the GeneTonic package.

**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.



---

shake\_davidResult      *Convert the output of DAVID*

---

**Description**

Convert the output of DAVID for straightforward use in [GeneTonic\(\)](#)

**Usage**

```
shake_davidResult(david_output_file)
```

**Arguments**

david\_output\_file  
The location of the text file output, as exported from DAVID

**Value**

A data.frame compatible for use in [GeneTonic\(\)](#) as res\_enrich

**See Also**

Other shakers: [shake\\_enrichResult\(\)](#), [shake\\_enrichrResult\(\)](#), [shake\\_fgseaResult\(\)](#), [shake\\_gprofilerResult\(\)](#), [shake\\_gsenrichResult\(\)](#), [shake\\_topG0tableResult\(\)](#)

**Examples**

```
david_output_file <- system.file("extdata",  
  "david_output_chart_BPonly_ifng_vs_naive.txt",  
  package = "GeneTonic"  
)  
res_enrich <- shake_davidResult(david_output_file)
```

---

shake\_enrichResult      *Convert an enrichResult object*

---

**Description**

Convert an enrichResult object for straightforward use in [GeneTonic\(\)](#)

**Usage**

```
shake_enrichResult(obj)
```

**Arguments**

obj      An enrichResult object, obtained via clusterProfiler (or also via reactomePA)

## Details

This function is able to handle the output of clusterProfiler and reactomePA, as they both return an object of class `enrichResult` - and this in turn contains the information required to create correctly a `res_enrich` object.

## Value

A data.frame compatible for use in `GeneTonic()` as `res_enrich`

## See Also

Other shakers: `shake_davidResult()`, `shake_enrichrResult()`, `shake_fgseaResult()`, `shake_gprofilerResult()`, `shake_gsenrichResult()`, `shake_topGOtableResult()`

## Examples

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

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive
de_symbols_IFNg_vs_naive <- res_macrophage_IFNg_vs_naive[
  (!(is.na(res_macrophage_IFNg_vs_naive$padj))) &
  (res_macrophage_IFNg_vs_naive$padj <= 0.05), "SYMBOL"
]
bg_ids <- rowData(dds_macrophage)$SYMBOL[rowSums(counts(dds_macrophage)) > 0]
## Not run:
library("clusterProfiler")
library("org.Hs.eg.db")
ego_IFNg_vs_naive <- enrichGO(
  gene = de_symbols_IFNg_vs_naive,
  universe = bg_ids,
  keyType = "SYMBOL",
  OrgDb = org.Hs.eg.db,
  ont = "BP",
  pAdjustMethod = "BH",
  pvalueCutoff = 0.01,
  qvalueCutoff = 0.05,
  readable = FALSE
)

res_enrich <- shake_enrichResult(ego_IFNg_vs_naive)
head(res_enrich)

## End(Not run)
```

---

shake\_enrichrResult     *Convert the output of Enrichr*

---

### Description

Convert the output of Enrichr for straightforward use in [GeneTonic\(\)](#)

### Usage

```
shake_enrichrResult(enrichr_output_file, enrichr_output = NULL)
```

### Arguments

enrichr\_output\_file  
The location of the text file output, as exported from Enrichr

enrichr\_output A data.frame with the output of enrichr, related to a specific set of genesets. Usually it is one of the members of the list returned by the initial call to enrichr.

### Value

A data.frame compatible for use in [GeneTonic\(\)](#) as res\_enrich

### See Also

Other shakers: [shake\\_davidResult\(\)](#), [shake\\_enrichResult\(\)](#), [shake\\_fgseaResult\(\)](#), [shake\\_gprofilerResult\(\)](#), [shake\\_gsenrichResult\(\)](#), [shake\\_topGOtableResult\(\)](#)

### Examples

```
# library("enrichR")
# dbs <- c("GO_Molecular_Function_2018",
#         "GO_Cellular_Component_2018",
#         "GO_Biological_Process_2018",
#         "KEGG_2019_Human",
#         "Reactome_2016",
#         "WikiPathways_2019_Human")
# degenes <- (deseqresult2df(res_macrophage_IFNg_vs_naive, FDR = 0.01)$SYMBOL)
# if called directly within R...
# enrichr_output_macrophage <- enrichr(degenes, dbs)
# or alternatively, if downloaded from the website in tabular format
enrichr_output_file <- system.file("extdata",
  "enrichr_tblexport_IFNg_vs_naive.txt",
  package = "GeneTonic"
)
res_from_enrichr <- shake_enrichrResult(enrichr_output_file = enrichr_output_file)
# res_from_enrichr2 <- shake_enrichrResult(
#   enrichr_output = enrichr_output_macrophage[["GO_Biological_Process_2018"]])
```

---

shake\_fgseaResult      *Convert the output of fgsea*

---

**Description**

Convert the output of fgsea for straightforward use in [GeneTonic\(\)](#)

**Usage**

```
shake_fgseaResult(fgsea_output)
```

**Arguments**

fgsea\_output      A data.frame with the output of fgsea() in fgsea.

**Value**

A data.frame compatible for use in [GeneTonic\(\)](#) as res\_enrich

**See Also**

Other shakers: [shake\\_davidResult\(\)](#), [shake\\_enrichResult\(\)](#), [shake\\_enrichrResult\(\)](#), [shake\\_gprofilerResult\(\)](#), [shake\\_gsenrichResult\(\)](#), [shake\\_topG0tableResult\(\)](#)

**Examples**

```
data(fgseaRes, package = "GeneTonic")
res_from_fgsea <- shake_fgseaResult(fgseaRes)
```

---

shake\_gprofilerResult      *Convert the output of g:Profiler*

---

**Description**

Convert the output of g:Profiler for straightforward use in [GeneTonic\(\)](#)

**Usage**

```
shake_gprofilerResult(gprofiler_output_file, gprofiler_output = NULL)
```

**Arguments**

gprofiler\_output\_file

The location of the text file output, as exported from g:Profiler

gprofiler\_output

A data.frame with the output of gost() in gprofiler2. Usually it is one of the members of the list returned by the initial call to gost.

**Value**

A data.frame compatible for use in [GeneTonic\(\)](#) as res\_enrich

**See Also**

Other shakers: [shake\\_davidResult\(\)](#), [shake\\_enrichResult\(\)](#), [shake\\_enrichrResult\(\)](#), [shake\\_fgseaResult\(\)](#), [shake\\_gsenrichResult\(\)](#), [shake\\_topG0tableResult\(\)](#)

**Examples**

```
# degenes <- (deseqresult2df(res_macrophage_IFNg_vs_naive, FDR = 0.01)$SYMBOL)
# if called directly within R...
# enrichr_output_macrophage <- enrichr(degenes, dbs)
# or alternatively, if downloaded from the website in tabular format
gprofiler_output_file <- system.file(
  "extdata",
  "gProfiler_hsapiens_5-25-2020_tblexport_IFNg_vs_naive.csv",
  package = "GeneTonic"
)
res_from_gprofiler <- shake_gprofilerResult(gprofiler_output_file = gprofiler_output_file)

data(gostres_macrophage, package = "GeneTonic")
res_from_gprofiler_2 <- shake_gprofilerResult(
  gprofiler_output = gostres_macrophage$result
)
```

---

shake\_gsenrichResult *Convert a gseaResult object*

---

**Description**

Convert a gseaResult object for straightforward use in [GeneTonic\(\)](#)

**Usage**

```
shake_gsenrichResult(obj)
```

**Arguments**

obj                    A gseaResult object, obtained via clusterProfiler

**Details**

This function is able to handle the output of clusterProfiler's gseG0 and GSEA, as they both return an object of class gseaResult - and this in turn contains the information required to create correctly a res\_enrich object.

**Value**

A data.frame compatible for use in `GeneTonic()` as `res_enrich`

**See Also**

Other shakers: `shake_davidResult()`, `shake_enrichResult()`, `shake_enrichrResult()`, `shake_fgseaResult()`, `shake_gprofilerResult()`, `shake_topGtableResult()`

**Examples**

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

# res object
data(res_de_macrophage, package = "GeneTonic")
sorted_genes <- sort(
  setNames(res_macrophage_IFNg_vs_naive$log2FoldChange,
           res_macrophage_IFNg_vs_naive$SYMBOL),
  decreasing = TRUE
)
## Not run:
library("clusterProfiler")
library("org.Hs.eg.db")
gsego_IFNg_vs_naive <- gseGO(
  geneList = sorted_genes,
  ont = "BP",
  OrgDb = org.Hs.eg.db,
  keyType = "SYMBOL",
  minGSSize = 10,
  maxGSSize = 500,
  pvalueCutoff = 0.05,
  verbose = TRUE
)

res_enrich <- shake_gsenrichResult(gsego_IFNg_vs_naive)
head(res_enrich)
gtl_macrophage <- GeneTonicList(
  dds = dds_macrophage,
  res_de = res_macrophage_IFNg_vs_naive,
  res_enrich = res_enrich,
  annotation_obj = anno_df
)

## End(Not run)
```

---

`shake_topGOTableResult`*Convert a topGOTableResult object*

---

**Description**

Convert a topGOTableResult object for straightforward use in [GeneTonic\(\)](#)

**Usage**

```
shake_topGOTableResult(obj, p_value_column = "p.value_elim")
```

**Arguments**

`obj` A topGOTableResult object

`p_value_column` Character, specifying which column the p value for enrichment has to be used. Example values are "p.value\_elim" or "p.value\_classic"

**Value**

A data.frame compatible for use in [GeneTonic\(\)](#) as `res_enrich`

**See Also**

Other shakers: [shake\\_davidResult\(\)](#), [shake\\_enrichResult\(\)](#), [shake\\_enrichrResult\(\)](#), [shake\\_fgseaResult\(\)](#), [shake\\_gprofilerResult\(\)](#), [shake\\_gsenrichResult\(\)](#)

**Examples**

```
# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")

res_enrich <- shake_topGOTableResult(topgoDE_macrophage_IFNg_vs_naive)
```

---

`signature_volcano`*Plot a volcano plot of a geneset*

---

**Description**

Plot a volcano plot for the geneset of the provided data, with the remaining genes as shaded dots in the background of the plot.

**Usage**

```
signature_volcano(
  res_de,
  res_enrich,
  annotation_obj = NULL,
  gtl = NULL,
  geneset_id = NULL,
  genelist = NULL,
  FDR = 0.05,
  color = "#1a81c2",
  volcano_labels = 25,
  plot_title = NULL
)
```

**Arguments**

|                             |   |
|-----------------------------|---|
| <code>res_de</code>         | A DESeqResults object.  |
| <code>res_enrich</code>     | A data.frame object, storing the result of the functional enrichment analysis. See more in the main function, <code>GeneTonic()</code> , to check the formatting requirements (a minimal set of columns should be present).   |
| <code>annotation_obj</code> | A data.frame object with the feature annotation information, with at least two columns, <code>gene_id</code> and <code>gene_name</code> .   |
| <code>gtl</code>            | A GeneTonic-list object, containing in its slots the arguments specified above: <code>dds</code> , <code>res_de</code> , <code>res_enrich</code> , and <code>annotation_obj</code> - the names of the list <i>must</i> be specified following the content they are expecting. |
| <code>geneset_id</code>     | Character specifying the gene set identifier to be plotted.   |
| <code>genelist</code>       | A vector of character strings, specifying the identifiers contained in the rownames of the <code>res_de</code> input object.  |
| <code>FDR</code>            | Numeric value, specifying the significance level for thresholding adjusted p-values. Defaults to 0.05.  |
| <code>color</code>          | Character string to specify color of filtered points in the plot. Defaults to #1a81c2 (shade of blue).  |
| <code>volcano_labels</code> | Integer, maximum number of labels for the gene sets to be plotted as labels on the volcano scatter plot. Defaults to 25.  |
| <code>plot_title</code>     | Character string, to specify the title of the plot, displayed over the volcano plot. If left to NULL as by default, it tries to use the information on the geneset identifier provided.   |

**Value**

A plot returned by the `ggplot()` function

**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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

signature_volcano(res_de,
  res_enrich,
  anno_df,
  geneset_id = res_enrich$gs_id[1]
)

# alternatively

chemokine_list <- c(
  "ENSG00000108702",
  "ENSG00000172156",
  "ENSG00000181374",
  "ENSG00000276409"
)

signature_volcano(res_de,
  res_enrich,
  anno_df,
  genelists = chemokine_list
)
```

---

 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

```
data(res_de_macrophage, package = "GeneTonic")
res_df <- deseqresult2df(res_macrophage_IFNg_vs_naive)
library("magrittr")
library("DT")
DT::datatable(res_df[1:50, ],
  options = list(
    pageLength = 25,
    columnDefs = list(
```

```

        list(className = "dt-center", targets = "_all")
      )
    )
  ) %>%
  formatRound(columns = c("log2FoldChange"), digits = 3) %>%
  formatStyle(
    "log2FoldChange",
    background = styleColorBar_divergent(
      res_df$log2FoldChange,
      scales::alpha("navyblue", 0.4),
      scales::alpha("darkred", 0.4)
    ),
    backgroundSize = "100% 90%",
    backgroundRepeat = "no-repeat",
    backgroundPosition = "center"
  )

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

# or with a very simple data frame
DT::datatable(simplest_df) %>%
  formatStyle(
    "value",
    background = styleColorBar_divergent(
      simplest_df$value,
      scales::alpha("forestgreen", 0.4),
      scales::alpha("gold", 0.4)
    ),
    backgroundSize = "100% 90%",
    backgroundRepeat = "no-repeat",
    backgroundPosition = "center"
  )

```

---

summarize\_ggs\_hubgenes

*Summarize information on the hub genes*

---

## Description

Summarize information on the hub genes in the Gene-Geneset graph

## Usage

```
summarize_ggs_hubgenes(g)
```

**Arguments**

`g` An igraph object, as generated by the `ggs_graph()` function

**Value**

A data.frame object, formatted for use in `DT::datatable()`

**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)
dds_macrophage <- estimateSizeFactors(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)
)

# res object
data(res_de_macrophage, package = "GeneTonic")
res_de <- res_macrophage_IFNg_vs_naive

# res_enrich object
data(res_enrich_macrophage, package = "GeneTonic")
res_enrich <- shake_topGOtableResult(topgoDE_macrophage_IFNg_vs_naive)
res_enrich <- get_aggrscores(res_enrich, res_de, anno_df)

ggs <- ggs_graph(
  res_enrich,
  res_de,
  anno_df
)
dt_df <- summarize_ggs_hubgenes(ggs)
DT::datatable(dt_df, escape = FALSE)
```

---

topgoDE\_macrophage\_IFNg\_vs\_naive  
*A sample res\_enrich object*

---

**Description**

A sample res\_enrich object, generated with the topGOtable function (from the pcaExplorer package).

**Details**

This res\_enrich object on the data from the macrophage package has been created by analyzing downstream the differentially expressed genes when comparing IFNg treated samples vs naive samples, accounting for the different cell lines included.

Details on how this object has been created are included in the create\_gt\_data.R script, included in the scripts folder of the GeneTonic package.

**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**

Other pathway-analysis-results: [enrichr\\_output\\_macrophage](#), [gostres\\_macrophage](#)

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