# Package 'Giotto'

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addCellIntMetadata

add Cell Int Metadata

# Description

Creates an additional metadata column with information about interacting and non-interacting cell types of the selected cell-cell interaction.

# Usage

```
addCellIntMetadata(
  gobject,
  spatial_network = "spatial_network",
  cluster_column,
  cell_interaction,
  name = "select_int",
  return_gobject = TRUE
)
```

# Arguments

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#### **Details**

This function will create an additional metadata column which selects interacting cell types for a specific cell-cell interaction. For example, if you want to color interacting astrocytes and oligodendrocytes it will create a new metadata column with the values "select\_astrocytes", "select\_oligodendrocytes", "other\_astrocytes", "other\_oligodendrocytes" and "other". Where "other" is all other cell types found within the selected cell type column.

#### Value

Giotto object

addCellMetadata a

addCellMetadata

#### **Description**

adds cell metadata to the giotto object

### Usage

```
addCellMetadata(
  gobject,
  new_metadata,
  vector_name = NULL,
  by_column = FALSE,
  column_cell_ID = NULL
)
```

# **Arguments**

```
gobject giotto object

new_metadata new cell metadata to use (data.table, data.frame, ...)

vector_name (optional) custom name if you provide a single vector

by_column merge metadata based on cell_ID column in pDataDT (default = FALSE)

column_cell_ID column name of new metadata to use if by_column = TRUE
```

# **Details**

You can add additional cell metadata in two manners:

- 1. Provide a data.table or data.frame with cell annotations in the same order as the cell\_ID column in pDataDT(gobject)
- 2. Provide a data.table or data.frame with cell annotations and specificy which column contains the cell IDs, these cell IDs need to match with the cell\_ID column in pDataDT(gobject)

addCellStatistics 9

#### Value

giotto object

addCellStatistics

addCellStatistics

# Description

adds cells statistics to the giotto object

# Usage

```
addCellStatistics(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  detection_threshold = 0,
  return_gobject = TRUE
)
```

# **Arguments**

# **Details**

This function will add the following statistics to cell metadata:

- nr\_genes: Denotes in how many genes are detected per cell
- perc\_genes: Denotes what percentage of genes is detected per cell
- total\_expr: Shows the total sum of gene expression per cell

#### Value

```
giotto object if return_gobject = TRUE
```

# **Examples**

```
data(mini_giotto_single_cell)
updated_giotto_object = addCellStatistics(mini_giotto_single_cell)
```

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addGeneMetadata

addGeneMetadata

# **Description**

adds gene metadata to the giotto object

#### Usage

```
addGeneMetadata(gobject, new_metadata, by_column = F, column_gene_ID = NULL)
```

# **Arguments**

```
gobject giotto object
```

new\_metadata new metadata to use

by\_column merge metadata based on gene\_ID column in fDataDT

column\_gene\_ID column name of new metadata to use if by\_column = TRUE

# **Details**

You can add additional gene metadata in two manners: 1. Provide a data.table or data.frame with gene annotations in the same order as the gene\_ID column in fDataDT(gobject) 2. Provide a data.table or data.frame with gene annotations and specificy which column contains the gene IDs, these gene IDs need to match with the gene\_ID column in fDataDT(gobject)

### Value

giotto object

addGenesPerc

addGenesPerc

# Description

calculates the total percentage of (normalized) counts for a subset of selected genes

```
addGenesPerc(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  genes = NULL,
  vector_name = "gene_perc",
  return_gobject = TRUE
)
```

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#### **Arguments**

```
gobject giotto object
expression_values
expression values to use
genes vector of selected genes
vector_name column name as seen in pDataDT()
return_gobject boolean: return giotto object (default = TRUE)
```

#### Value

giotto object if return\_gobject = TRUE, else a vector with

#### **Examples**

addGeneStatistics

addGeneStatistics

#### **Description**

adds gene statistics to the giotto object

### Usage

```
addGeneStatistics(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  detection_threshold = 0,
  return_gobject = TRUE
)
```

# **Arguments**

12 addGiottoImage

#### **Details**

This function will add the following statistics to gene metadata:

- nr\_cells: Denotes in how many cells the gene is detected
- per\_cells: Denotes in what percentage of cells the gene is detected
- total\_expr: Shows the total sum of gene expression in all cells
- mean\_expr: Average gene expression in all cells
- mean\_expr\_det: Average gene expression in cells with detectable levels of the gene

# Value

```
giotto object if return_gobject = TRUE
```

# **Examples**

```
data(mini_giotto_single_cell)
updated_giotto_object = addGeneStatistics(mini_giotto_single_cell)
```

addGiottoImage

addGiottoImage

# **Description**

Adds giotto image objects to your giotto object

# Usage

```
addGiottoImage(gobject, images)
```

# **Arguments**

gobject giotto object

images list of giotto image objects, see createGiottoImage

#### Value

an updated Giotto object with access to the list of images

```
add {\tt GiottoImageToSpatPlot}
```

add Giot to Image To Spat Plot

# Description

Add a giotto image to a spatial ggplot object post creation

# Usage

```
addGiottoImageToSpatPlot(spatpl = NULL, gimage = NULL)
```

# Arguments

spatpl a spatial ggplot object

gimage a giotto image, see createGiottoImage

#### Value

an updated spatial ggplot object

addHMRF addHMRF

# **Description**

Add selected results from doHMRF to the giotto object

# Usage

```
addHMRF(gobject, HMRFoutput, k = NULL, betas_to_add = NULL, hmrf_name = NULL)
```

# Arguments

gobject giotto object

 $\label{eq:hmrf} HMRF \ output \ from \ do HMRF()$ 

k number of domains

hmrf\_name specify a custom name

# Value

giotto object

14 addNetworkLayout

addNetworkLayout

addNetworkLayout

# **Description**

Add a network layout for a selected nearest neighbor network

# Usage

```
addNetworkLayout(
  gobject,
  nn_network_to_use = "sNN",
  network_name = "sNN.pca",
  layout_type = c("drl"),
  options_list = NULL,
  layout_name = "layout",
  return_gobject = TRUE
)
```

# **Arguments**

# Details

This function creates layout coordinates based on the provided kNN or sNN. Currently only the force-directed graph layout "drl", see layout\_with\_drl, is implemented. This provides an alternative to tSNE or UMAP based visualizations.

#### Value

giotto object with updated layout for selected NN network

addStatistics 15

 ${\tt addStatistics}$ 

addStatistics

# **Description**

adds genes and cells statistics to the giotto object

# Usage

```
addStatistics(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  detection_threshold = 0,
  return_gobject = TRUE
)
```

# **Arguments**

```
gobject giotto object

expression_values

expression values to use

detection_threshold

detection threshold to consider a gene detected

return_gobject boolean: return giotto object (default = TRUE)
```

#### **Details**

See addGeneStatistics and addCellStatistics

#### Value

```
giotto object if return_gobject = TRUE, else a list with results
```

# **Examples**

```
data(mini_giotto_single_cell)
updated_giotto_object = addStatistics(mini_giotto_single_cell)
```

adjustGiottoMatrix

adjust Giot to Matrix

# **Description**

Adjust expression values to account for known batch effects or technological covariates.

16 anndataToGiotto

#### Usage

```
adjustGiottoMatrix(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  batch_columns = NULL,
  covariate_columns = NULL,
  return_gobject = TRUE,
  update_slot = c("custom")
)
```

# Arguments

#### **Details**

This function implements the removeBatchEffect function to remove known batch effects and to adjust expression values according to provided covariates.

# Value

giotto object

# **Examples**

```
data(mini_giotto_single_cell)
adjust_gobject = adjustGiottoMatrix(mini_giotto_single_cell)
```

 $ann data To {\tt Giotto}$ 

anndata To Giotto

#### **Description**

Converts a spatial anndata (e.g. scanpy) .h5ad file into a Giotto object

```
anndataToGiotto(
  anndata_path,
  metadata_cols = c("total_counts", "pct_counts_mt"),
  instructions = NULL,
  ...
)
```

annotateGiotto 17

#### **Arguments**

```
anndata_path path to the .h5ad file
metadata_cols metadata columns to include
instructions giotto instructions
... additional parameters to createGiottoObject
```

#### **Details**

Function in beta. Converts a .h5ad file into a Giotto object.

#### Value

Giotto object

annotateGiotto

annotateGiotto

# Description

Converts cluster results into a user provided annotation.

# Usage

```
annotateGiotto(
  gobject,
  annotation_vector = NULL,
  cluster_column = NULL,
  name = "cell_types"
)
```

# **Arguments**

### **Details**

You need to specifify which (cluster) column you want to annotate and you need to provide an annotation vector like this:

- 1. identify the cell type of each cluster
- 2. create a vector of these cell types, e.g. cell\_types = c('T-cell', 'B-cell', 'Stromal')
- 3. provide original cluster names to previous vector, e.g. names(cell\_types) = c(2, 1, 3)

# Value

giotto object

18 annotateSpatialGrid

### **Examples**

 $annotate Spatial Grid \qquad annotate Spatial Grid$ 

#### **Description**

annotate spatial grid with cell ID and cell metadata (optional)

### Usage

```
annotateSpatialGrid(
  gobject,
  spatial_grid_name = "spatial_grid",
  cluster_columns = NULL
)
```

# **Arguments**

#### Value

annotated spatial grid data.table

```
annotate Spatial Network
```

annotate Spatial Network

#### **Description**

Annotate spatial network with cell metadata information.

#### Usage

```
annotateSpatialNetwork(
  gobject,
  spatial_network_name = "Delaunay_network",
  cluster_column,
  create_full_network = F
)
```

# Arguments

#### Value

annotated network in data.table format

binSpect

binSpect

# Description

Previously: binGetSpatialGenes. BinSpect (Binary Spatial Extraction of genes) is a fast computational method that identifies genes with a spatially coherent expression pattern.

```
binSpect(
  gobject,
  bin_method = c("kmeans", "rank"),
  expression_values = c("normalized", "scaled", "custom"),
  subset_genes = NULL,
  spatial_network_name = "Delaunay_network",
  spatial_network_k = NULL,
  reduce_network = FALSE,
  kmeans_algo = c("kmeans", "kmeans_arma", "kmeans_arma_subset"),
```

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```
nstart = 3,
      iter_max = 10,
      extreme_nr = 50,
      sample_nr = 50,
      percentage_rank = 30,
      do_fisher_test = TRUE,
      adjust_method = "fdr",
      calc_hub = FALSE,
      hub_min_int = 3,
      get_av_expr = TRUE,
      get_high_expr = TRUE,
      implementation = c("data.table", "simple", "matrix"),
      group_size = "automatic",
      do_parallel = TRUE,
      cores = NA,
      verbose = T,
      knn_params = NULL,
      set.seed = NULL,
      bin_matrix = NULL,
      summarize = c("p.value", "adj.p.value")
    )
Arguments
                     giotto object
    gobject
   bin_method
                     method to binarize gene expression
    expression_values
                     expression values to use
                     only select a subset of genes to test
    subset_genes
    spatial_network_name
                     name of spatial network to use (default = 'spatial_network')
    spatial_network_k
                     different k's for a spatial kNN to evaluate
    reduce_network default uses the full network
                     kmeans algorithm to use (kmeans, kmeans_arma, kmeans_arma_subset)
   kmeans_algo
                     kmeans: nstart parameter
    nstart
    iter_max
                     kmeans: iter.max parameter
    extreme_nr
                     number of top and bottom cells (see details)
                     total number of cells to sample (see details)
    sample_nr
    percentage_rank
                     percentage of top cells for binarization
    do_fisher_test perform fisher test
                     p-value adjusted method to use (see p.adjust)
    adjust_method
    calc_hub
                     calculate the number of hub cells
   hub_min_int
                     minimum number of cell-cell interactions for a hub cell
    get_av_expr
                     calculate the average expression per gene of the high expressing cells
                     calculate the number of high expressing cells per gene
    get_high_expr
```

binSpect 21

implementation enrichment implementation (data.table, simple, matrix) group\_size number of genes to process together with data.table implementation (default = automatic) run calculations in parallel with mclapply do\_parallel cores number of cores to use if do\_parallel = TRUE be verbose verbose knn\_params list of parameters to create spatial kNN network set.seed set a seed before kmeans binarization bin\_matrix a binarized matrix, when provided it will skip the binarization process summarize summarize the p-values or adjusted p-values

#### Details

We provide two ways to identify spatial genes based on gene expression binarization. Both methods are identicial except for how binarization is performed.

- 1. binarize: Each gene is binarized (0 or 1) in each cell with **kmeans** (k = 2) or based on **rank** percentile
- 2. network: Alll cells are connected through a spatial network based on the physical coordinates
- 3. contingency table: A contingency table is calculated based on all edges of neighboring cells and the binarized expression (0-0, 0-1, 1-0 or 1-1)
- 4. For each gene an odds-ratio (OR) and fisher.test (optional) is calculated

Three different kmeans algorithmes have been implemented:

- 1. kmeans: default, see kmeans
- 2. kmeans\_arma: from ClusterR, see KMeans\_arma
- 3. kmeans\_arma\_subst: from ClusterR, see KMeans\_arma, but random subsetting the vector for each gene to increase speed. Change extreme\_nr and sample\_nr for control.

Other statistics are provided (optional):

- Number of cells with high expression (binary = 1)
- Average expression of each gene within high expressing cells
- Number of hub cells, these are high expressing cells that have a user defined number of high expressing neighbors

By selecting a subset of likely spatial genes (e.g. soft thresholding highly variable genes) can accelerate the speed. The simple implementation is usually faster, but lacks the possibility to run in parallel and to calculate hub cells. The data.table implementation might be more appropriate for large datasets by setting the group\_size (number of genes) parameter to divide the workload.

#### Value

data.table with results (see details)

22 binSpectMulti

binSpectMulti

binSpectMulti

### **Description**

binSpect for multiple spatial kNN networks

### Usage

```
binSpectMulti(
  gobject,
  bin_method = c("kmeans", "rank"),
  expression_values = c("normalized", "scaled", "custom"),
  subset_genes = NULL,
  spatial_network_k = c(5, 10, 20),
  reduce_network = FALSE,
  kmeans_algo = c("kmeans", "kmeans_arma", "kmeans_arma_subset"),
  nstart = 3,
  iter_max = 10,
  extreme_nr = 50,
  sample_nr = 50,
  percentage_rank = c(10, 30),
  do_fisher_test = TRUE,
  adjust_method = "fdr",
  calc_hub = FALSE,
  hub_min_int = 3,
  get_av_expr = TRUE,
  get_high_expr = TRUE,
  implementation = c("data.table", "simple", "matrix"),
  group_size = "automatic",
  do_parallel = TRUE,
  cores = NA,
  verbose = T,
  knn_params = NULL,
  set.seed = NULL,
  summarize = c("adj.p.value", "p.value")
)
```

#### **Arguments**

```
gobject giotto object

bin_method method to binarize gene expression

expression_values

expression values to use

subset_genes only select a subset of genes to test

spatial_network_k

different k's for a spatial kNN to evaluate

reduce_network default uses the full network

kmeans_algo kmeans algorithm to use (kmeans, kmeans_arma, kmeans_arma_subset)
```

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nstart kmeans: nstart parameter iter\_max kmeans: iter.max parameter

extreme\_nr number of top and bottom cells (see details)
sample\_nr total number of cells to sample (see details)

percentage\_rank

percentage of top cells for binarization

do\_fisher\_test perform fisher test

adjust\_method p-value adjusted method to use (see p.adjust)

calc\_hub calculate the number of hub cells

hub\_min\_int minimum number of cell-cell interactions for a hub cell

get\_av\_expr calculate the average expression per gene of the high expressing cells

get\_high\_expr calculate the number of high expressing cells per gene implementation enrichment implementation (data.table, simple, matrix)

group\_size number of genes to process together with data.table implementation (default =

automatic)

do\_parallel run calculations in parallel with mclapply cores number of cores to use if do\_parallel = TRUE

verbose be verbose

knn\_params list of parameters to create spatial kNN network

set. seed set a seed before kmeans binarization

summarize summarize the p-values or adjusted p-values

### **Details**

We provide two ways to identify spatial genes based on gene expression binarization. Both methods are identicial except for how binarization is performed.

- 1. binarize: Each gene is binarized (0 or 1) in each cell with **kmeans** (k = 2) or based on **rank** percentile
- 2. network: Alll cells are connected through a spatial network based on the physical coordinates
- 3. contingency table: A contingency table is calculated based on all edges of neighboring cells and the binarized expression (0-0, 0-1, 1-0 or 1-1)
- 4. For each gene an odds-ratio (OR) and fisher.test (optional) is calculated

Three different kmeans algorithmes have been implemented:

- 1. kmeans: default, see kmeans
- 2. kmeans\_arma: from ClusterR, see KMeans\_arma
- 3. kmeans\_arma\_subst: from ClusterR, see KMeans\_arma, but random subsetting the vector for each gene to increase speed. Change extreme\_nr and sample\_nr for control.

Other statistics are provided (optional):

- Number of cells with high expression (binary = 1)
- Average expression of each gene within high expressing cells

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• Number of hub cells, these are high expressing cells that have a user defined number of high expressing neighbors

By selecting a subset of likely spatial genes (e.g. soft thresholding highly variable genes) can accelerate the speed. The simple implementation is usually faster, but lacks the possibility to run in parallel and to calculate hub cells. The data.table implementation might be more appropriate for large datasets by setting the group\_size (number of genes) parameter to divide the workload.

#### Value

data.table with results (see details)

binSpectSingle

binSpectSingle

#### **Description**

binSpect for a single spatial network

```
binSpectSingle(
  gobject,
  bin_method = c("kmeans", "rank"),
  expression_values = c("normalized", "scaled", "custom"),
  subset_genes = NULL,
  spatial_network_name = "Delaunay_network",
  reduce_network = FALSE,
  kmeans_algo = c("kmeans", "kmeans_arma", "kmeans_arma_subset"),
  nstart = 3,
  iter_max = 10,
  extreme_nr = 50,
  sample_nr = 50,
  percentage_rank = 30,
  do_fisher_test = TRUE,
  adjust\_method = "fdr",
  calc_hub = FALSE,
  hub_min_int = 3,
  get_av_expr = TRUE,
  get_high_expr = TRUE,
  implementation = c("data.table", "simple", "matrix"),
  group_size = "automatic",
  do_parallel = TRUE,
  cores = NA,
  verbose = T,
  set.seed = NULL,
  bin_matrix = NULL
)
```

binSpectSingle 25

#### **Arguments**

gobject giotto object

bin\_method method to binarize gene expression

expression\_values

expression values to use

subset\_genes only select a subset of genes to test

spatial\_network\_name

name of spatial network to use (default = 'spatial\_network')

reduce\_network default uses the full network

kmeans\_algo kmeans algorithm to use (kmeans, kmeans\_arma, kmeans\_arma\_subset)

nstart kmeans: nstart parameter iter\_max kmeans: iter.max parameter

extreme\_nr number of top and bottom cells (see details)
sample\_nr total number of cells to sample (see details)

percentage\_rank

percentage of top cells for binarization

do\_fisher\_test perform fisher test

adjust\_method p-value adjusted method to use (see p.adjust)

calc\_hub calculate the number of hub cells

hub\_min\_int minimum number of cell-cell interactions for a hub cell

get\_av\_expr calculate the average expression per gene of the high expressing cells

get\_high\_expr calculate the number of high expressing cells per gene implementation enrichment implementation (data.table, simple, matrix)

group\_size number of genes to process together with data.table implementation (default =

automatic)

do\_parallel run calculations in parallel with mclapply cores number of cores to use if do\_parallel = TRUE

verbose be verbose

set.seed set a seed before kmeans binarization

bin\_matrix a binarized matrix, when provided it will skip the binarization process

#### **Details**

We provide two ways to identify spatial genes based on gene expression binarization. Both methods are identicial except for how binarization is performed.

- 1. binarize: Each gene is binarized (0 or 1) in each cell with **kmeans** (k = 2) or based on **rank** percentile
- 2. network: Alll cells are connected through a spatial network based on the physical coordinates
- 3. contingency table: A contingency table is calculated based on all edges of neighboring cells and the binarized expression (0-0, 0-1, 1-0 or 1-1)
- 4. For each gene an odds-ratio (OR) and fisher.test (optional) is calculated

Three different kmeans algorithmes have been implemented:

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- 1. kmeans: default, see kmeans
- 2. kmeans\_arma: from ClusterR, see KMeans\_arma
- 3. kmeans\_arma\_subst: from ClusterR, see KMeans\_arma, but random subsetting the vector for each gene to increase speed. Change extreme\_nr and sample\_nr for control.

Other statistics are provided (optional):

- Number of cells with high expression (binary = 1)
- Average expression of each gene within high expressing cells
- Number of hub cells, these are high expressing cells that have a user defined number of high expressing neighbors

By selecting a subset of likely spatial genes (e.g. soft thresholding highly variable genes) can accelerate the speed. The simple implementation is usually faster, but lacks the possibility to run in parallel and to calculate hub cells. The data.table implementation might be more appropriate for large datasets by setting the group\_size (number of genes) parameter to divide the workload.

#### Value

data.table with results (see details)

calculateHVG

calculateHVG

# **Description**

compute highly variable genes

```
calculateHVG(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
 method = c("cov_groups", "cov_loess"),
 reverse_log_scale = FALSE,
  logbase = 2,
  expression_threshold = 0,
 nr_expression_groups = 20,
 zscore_threshold = 1.5,
 HVGname = "hvg",
  difference_in_cov = 0.1,
  show_plot = NA,
 return_plot = NA,
  save_plot = NA,
  save_param = list(),
 default_save_name = "HVGplot",
  return_gobject = TRUE
)
```

calculateHVG 27

#### **Arguments**

```
gobject
                  giotto object
expression_values
                  expression values to use
method
                  method to calculate highly variable genes
reverse_log_scale
                  reverse log-scale of expression values (default = FALSE)
logbase
                  if reverse_log_scale is TRUE, which log base was used?
expression_threshold
                  expression threshold to consider a gene detected
nr_expression_groups
                  number of expression groups for cov_groups
zscore_threshold
                  zscore to select hvg for cov_groups
HVGname
                  name for highly variable genes in cell metadata
difference_in_cov
                  minimum difference in coefficient of variance required
show_plot
                  show plot
                  return ggplot object
return_plot
save_plot
                  directly save the plot [boolean]
save_param
                  list of saving parameters from all_plots_save_function
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

#### **Details**

Currently we provide 2 ways to calculate highly variable genes:

return\_gobject boolean: return giotto object (default = TRUE)

#### 1. high coeff of variance (COV) within groups:

First genes are binned (*nr\_expression\_groups*) into average expression groups and the COV for each gene is converted into a z-score within each bin. Genes with a z-score higher than the threshold (*zscore\_threshold*) are considered highly variable.

# 2. high COV based on loess regression prediction:

A predicted COV is calculated for each gene using loess regression (COV~log(mean expression)) Genes that show a higher than predicted COV (difference\_in\_cov) are considered highly variable.

#### Value

giotto object highly variable genes appended to gene metadata (fDataDT)

# Examples

```
data(mini_giotto_single_cell) # loads existing Giotto object
# update a giotto object
mini_giotto_single_cell <- calculateHVG(gobject = mini_giotto_single_cell,</pre>
```

28 calculateMetaTable

calculateMetaTable

calculateMetaTable

# **Description**

calculates the average gene expression for one or more (combined) annotation columns.

# Usage

```
calculateMetaTable(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  metadata_cols = NULL,
  selected_genes = NULL
)
```

#### **Arguments**

```
gobject giotto object
expression_values
expression values to use
metadata_cols annotation columns found in pDataDT(gobject)
selected_genes subset of genes to use
```

#### Value

data.table with average expression values for each gene per (combined) annotation

# **Examples**

calculateMetaTableCells 29

```
calculateMetaTableCells
```

calculateMetaTableCells

# **Description**

calculates the average metadata values for one or more (combined) annotation columns.

# Usage

```
calculateMetaTableCells(
  gobject,
  value_cols = NULL,
  metadata_cols = NULL,
  spat_enr_names = NULL
)
```

# **Arguments**

```
gobject giotto object
value_cols metadata or enrichment value columns to use
metadata_cols annotation columns found in pDataDT(gobject)
spat_enr_names which spatial enrichment results to include
```

#### Value

data.table with average metadata values per (combined) annotation

```
cellProximityBarplot cellProximityBarplot
```

# **Description**

Create barplot from cell-cell proximity scores

```
cellProximityBarplot(
  gobject,
  CPscore,
  min_orig_ints = 5,
  min_sim_ints = 5,
  p_val = 0.05,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "cellProximityBarplot"
)
```

#### **Arguments**

gobject giotto object CPscore, output from cellProximityEnrichment() **CPscore** min\_orig\_ints filter on minimum original cell-cell interactions filter on minimum simulated cell-cell interactions min\_sim\_ints p\_val p-value show\_plot show plot return\_plot return ggplot object save\_plot directly save the plot [boolean] list of saving parameters from all\_plots\_save\_function save\_param default\_save\_name default save name for saving, don't change, change save\_name in save\_param

#### **Details**

This function creates a barplot that shows the spatial proximity enrichment or depletion of cell type pairs.

#### Value

ggplot barplot

```
cellProximityEnrichment
```

cellProximityEnrichment

# Description

Compute cell-cell interaction enrichment (observed vs expected)

cellProximityHeatmap 31

#### **Arguments**

# **Details**

Spatial proximity enrichment or depletion between pairs of cell types is calculated by calculating the observed over the expected frequency of cell-cell proximity interactions. The expected frequency is the average frequency calculated from a number of spatial network simulations. Each individual simulation is obtained by reshuffling the cell type labels of each node (cell) in the spatial network.

#### Value

List of cell Proximity scores (CPscores) in data.table format. The first data.table (raw\_sim\_table) shows the raw observations of both the original and simulated networks. The second data.table (enrichm\_res) shows the enrichment results.

```
cellProximityHeatmap cellProximityHeatmap
```

### **Description**

Create heatmap from cell-cell proximity scores

```
cellProximityHeatmap(
  gobject,
  CPscore,
  scale = T,
  order_cell_types = T,
  color_breaks = NULL,
  color_names = NULL,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "cellProximityHeatmap")
```

#### **Arguments**

```
gobject
                  giotto object
                  CPscore, output from cellProximityEnrichment()
CPscore
scale
                  scale cell-cell proximity interaction scores
order_cell_types
                  order cell types based on enrichment correlation
                  numerical vector of length 3 to represent min, mean and maximum
color_breaks
                  character color vector of length 3
color_names
show_plot
                  show plot
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
                  list of saving parameters from all_plots_save_function
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

#### **Details**

This function creates a heatmap that shows the spatial proximity enrichment or depletion of cell type pairs.

#### Value

ggplot heatmap

```
cell Proximity Network \qquad cell Proximity Network
```

# **Description**

Create network from cell-cell proximity scores

```
cellProximityNetwork(
  gobject,
  CPscore,
  remove_self_edges = FALSE,
  self_loop_strength = 0.1,
  color_depletion = "lightgreen",
  color_enrichment = "red",
  rescale_edge_weights = TRUE,
  edge_weight_range_depletion = c(0.1, 1),
  edge_weight_range_enrichment = c(1, 5),
  layout = c("Fruchterman", "DrL", "Kamada-Kawai"),
  only_show_enrichment_edges = F,
  edge_width_range = c(0.1, 2),
  node_size = 4,
  node_text_size = 6,
```

cellProximityNetwork 33

```
show_plot = NA,
return_plot = NA,
save_plot = NA,
save_param = list(),
default_save_name = "cellProximityNetwork"
)
```

### **Arguments**

```
giotto object
gobject
CPscore
                  CPscore, output from cellProximityEnrichment()
remove_self_edges
                  remove enrichment/depletion edges with itself
self_loop_strength
                  size of self-loops
color_depletion
                  color for depleted cell-cell interactions
color_enrichment
                  color for enriched cell-cell interactions
rescale_edge_weights
                  rescale edge weights (boolean)
edge_weight_range_depletion
                  numerical vector of length 2 to rescale depleted edge weights
edge_weight_range_enrichment
                  numerical vector of length 2 to rescale enriched edge weights
                  layout algorithm to use to draw nodes and edges
layout
only_show_enrichment_edges
                  show only the enriched pairwise scores
edge_width_range
                  range of edge width
                  size of nodes
node_size
node_text_size size of node labels
show_plot
                  show plot
return_plot
                  return ggplot object
                  directly save the plot [boolean]
save_plot
                  list of saving parameters from all_plots_save_function
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

#### **Details**

This function creates a network that shows the spatial proximity enrichment or depletion of cell type pairs.

# Value

```
igraph plot
```

cellProximitySpatPlot cellProximitySpatPlot

#### **Description**

Visualize 2D cell-cell interactions according to spatial coordinates in ggplot mode

#### Usage

```
cellProximitySpatPlot(gobject, ...)
```

# **Arguments**

```
gobject
                 giotto object
                  Arguments passed on to cellProximitySpatPlot2D
                  interaction_name cell-cell interaction name
                 cluster_column cluster column with cell clusters
                  sdimx x-axis dimension name (default = 'sdimx')
                 sdimy y-axis dimension name (default = 'sdimy')
                 cell_color color for cells (see details)
                 cell_color_code named vector with colors
                 color_as_factor convert color column to factor
                 show_other_cells decide if show cells not in network
                 show_network show spatial network of selected cells
                 show_other_network show spatial network of not selected cells
                 network_color color of spatial network
                 spatial_network_name name of spatial network to use
                 show_grid show spatial grid
                 grid_color color of spatial grid
                 spatial_grid_name name of spatial grid to use
                 coord_fix_ratio fix ratio between x and y-axis
                 show_legend show legend
                 point_size_select size of selected points
                 point_select_border_col border color of selected points
                 point_select_border_stroke stroke size of selected points
                 point_size_other size of other points
                 point_alpha_other opacity of other points
                 point_other_border_col border color of other points
                 point_other_border_stroke stroke size of other points
                 show_plot show plots
                 return_plot return ggplot object
                 save_plot directly save the plot [boolean]
                 save_param list of saving parameters from all_plots_save_function
                 default_save_name default save name for saving, don't change, change save_name
                      in save_param
```

#### **Details**

Description of parameters.

#### Value

ggplot

#### See Also

cellProximitySpatPlot2D and cellProximitySpatPlot3D for 3D

```
cellProximitySpatPlot2D
```

cellProximitySpatPlot2D

# **Description**

Visualize 2D cell-cell interactions according to spatial coordinates in ggplot mode

```
cellProximitySpatPlot2D(
  gobject,
  interaction_name = NULL,
  cluster_column = NULL,
  sdimx = "sdimx",
  sdimy = "sdimy",
  cell_color = NULL,
  cell_color_code = NULL,
  color_as_factor = T,
  show_other_cells = F,
  show_network = F,
  show_other_network = F,
  network_color = NULL,
  spatial_network_name = "Delaunay_network",
  show\_grid = F,
  grid_color = NULL,
  spatial_grid_name = "spatial_grid",
  coord_fix_ratio = 1,
  show_legend = T,
  point_size_select = 2,
  point_select_border_col = "black",
  point_select_border_stroke = 0.05,
  point_size_other = 1,
  point_alpha_other = 0.3,
  point_other_border_col = "lightgrey",
  point_other_border_stroke = 0.01,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
```

```
default_save_name = "cellProximitySpatPlot2D"
)
```

# **Arguments**

show\_plot

return\_plot

show plots

return ggplot object

giotto object gobject interaction\_name cell-cell interaction name cluster\_column cluster column with cell clusters sdimx x-axis dimension name (default = 'sdimx') sdimy y-axis dimension name (default = 'sdimy') color for cells (see details) cell\_color cell\_color\_code named vector with colors color\_as\_factor convert color column to factor show\_other\_cells decide if show cells not in network show\_network show spatial network of selected cells show\_other\_network show spatial network of not selected cells network\_color color of spatial network spatial\_network\_name name of spatial network to use show\_grid show spatial grid grid\_color color of spatial grid spatial\_grid\_name name of spatial grid to use coord\_fix\_ratio fix ratio between x and y-axis show\_legend show legend point\_size\_select size of selected points point\_select\_border\_col border color of selected points point\_select\_border\_stroke stroke size of selected points point\_size\_other size of other points point\_alpha\_other opacity of other points point\_other\_border\_col border color of other points point\_other\_border\_stroke stroke size of other points

#### **Details**

Description of parameters.

#### Value

ggplot

```
cell Proximity SpatPlot 3D \\ cell Proximity SpatPlot 2D
```

#### **Description**

Visualize 3D cell-cell interactions according to spatial coordinates in plotly mode

```
cellProximitySpatPlot3D(
  gobject,
  interaction_name = NULL,
  cluster_column = NULL,
  sdimx = "sdimx",
  sdimy = "sdimy",
  sdimz = "sdimz",
  cell_color = NULL,
  cell_color_code = NULL,
  color_as_factor = T,
  show_other_cells = T,
  show_network = T,
  show_other_network = F,
  network_color = NULL,
  spatial_network_name = "Delaunay_network",
  show\_grid = F,
  grid_color = NULL,
  spatial_grid_name = "spatial_grid",
  show_legend = T,
  point_size_select = 4,
  point_size_other = 2,
  point_alpha_other = 0.5,
  axis_scale = c("cube", "real", "custom"),
  custom_ratio = NULL,
  x_ticks = NULL,
  y_ticks = NULL,
  z_{ticks} = NULL,
  show_plot = NA,
```

```
return_plot = NA,
  save_plot = NA,
  save_param = list(),
 default_save_name = "cellProximitySpatPlot3D",
)
```

## **Arguments**

show\_plot

show plots

```
giotto object
gobject
interaction_name
                  cell-cell interaction name
cluster_column cluster column with cell clusters
                  x-axis dimension name (default = 'sdimx')
sdimx
                  y-axis dimension name (default = 'sdimy')
sdimy
sdimz
                  z-axis dimension name (default = 'sdimz')
cell_color
                  color for cells (see details)
cell_color_code
                  named vector with colors
color_as_factor
                  convert color column to factor
show_other_cells
                  decide if show cells not in network
show_network
                  show spatial network of selected cells
show_other_network
                  show spatial network of not selected cells
                  color of spatial network
network_color
spatial_network_name
                  name of spatial network to use
show_grid
                  show spatial grid
grid_color
                  color of spatial grid
spatial_grid_name
                  name of spatial grid to use
show_legend
                  show legend
point_size_select
                  size of selected points
point_size_other
                  size of other points
point_alpha_other
                  opacity of other points
                  scale of axis
axis_scale
custom_ratio
                  custom ratio of axes
                  ticks on x-axis
x_ticks
y_ticks
                  ticks on y-axis
z_ticks
                  ticks on z-axis
```

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#### **Details**

Description of parameters.

#### Value

plotly

```
cell Proximity VisPlot \quad \textit{cellProximityVisPlot}
```

## **Description**

Visualize cell-cell interactions according to spatial coordinates

```
cellProximityVisPlot(
  gobject,
  interaction_name = NULL,
  cluster_column = NULL,
  sdimx = NULL,
  sdimy = NULL,
  sdimz = NULL,
  cell_color = NULL,
  cell_color_code = NULL,
  color_as_factor = T,
  show_other_cells = F,
  show_network = F,
  show_other_network = F,
  network_color = NULL,
  spatial_network_name = "Delaunay_network",
  show\_grid = F,
  grid_color = NULL,
  spatial_grid_name = "spatial_grid",
  coord_fix_ratio = 1,
  show_legend = T,
  point_size_select = 2,
  point_select_border_col = "black",
  point_select_border_stroke = 0.05,
  point_size_other = 1,
  point_alpha_other = 0.3,
  point_other_border_col = "lightgrey",
```

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```
point_other_border_stroke = 0.01,
      axis_scale = c("cube", "real", "custom"),
      custom_ratio = NULL,
      x_ticks = NULL,
      y_ticks = NULL,
      z_ticks = NULL,
      plot_method = c("ggplot", "plotly"),
    )
Arguments
    gobject
                     giotto object
    interaction_name
                     cell-cell interaction name
    cluster_column cluster column with cell clusters
                     x-axis dimension name (default = 'sdimx')
    sdimx
    sdimy
                     y-axis dimension name (default = 'sdimy')
    sdimz
                     z-axis dimension name (default = 'sdimz')
    cell_color
                     color for cells (see details)
    cell_color_code
                     named vector with colors
    color_as_factor
                     convert color column to factor
    show_other_cells
                     show not selected cells
    show_network
                     show underlying spatial network
    show_other_network
                     show underlying spatial network of other cells
                     color of spatial network
    network_color
    spatial_network_name
                     name of spatial network to use
                     show spatial grid
    show_grid
                     color of spatial grid
    grid_color
    spatial_grid_name
                     name of spatial grid to use
    coord_fix_ratio
                     fix ratio between x and y-axis
    show_legend
                     show legend
   point_size_select
                     size of selected points
   point_select_border_col
                     border color of selected points
    point_select_border_stroke
                     stroke size of selected points
   point_size_other
```

size of other points

```
point_alpha_other
                 alpha of other points
point_other_border_col
                 border color of other points
point_other_border_stroke
                 stroke size of other points
axis_scale
                 scale of axis
                 custom ratio of scales
custom_ratio
x_ticks
                 x ticks
y_ticks
                 y ticks
z_ticks
                 z ticks
plot_method
                 method to plot
                 additional parameters
```

## **Details**

Description of parameters.

## Value

ggplot or plotly

 ${\tt change} {\tt GiottoInstructions}$ 

*changeGiottoInstructions* 

## **Description**

Function to change one or more instructions from giotto object

# Usage

```
changeGiottoInstructions(
  gobject,
  params = NULL,
  new_values = NULL,
  return_gobject = TRUE
)
```

# Arguments

```
gobject giotto object
params parameter(s) to change
new_values new value(s) for parameter(s)
```

return\_gobject (boolean) return giotto object

# Value

giotto object with one or more changed instructions

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changeImageBg changeImageBg

## **Description**

Function to change the background color of a magick image plot to another color

# Usage

```
changeImageBg(
  mg_object,
  bg_color,
  perc_range = 10,
  new_color = "#FFFFFF",
  new_name = NULL
)
```

# Arguments

mg\_object magick image or giotto image object bg\_color estimated current background color

perc\_range range around estimated background color to include (percentage)

new\_color new background color

new\_name change name of Giotto image

#### Value

magick image or giotto image object with updated background color

 ${\tt checkGiottoEnvironment}$ 

checkGiottoEnvironment

## **Description**

checkGiottoEnvironment

## Usage

```
checkGiottoEnvironment(verbose = TRUE)
```

#### **Arguments**

verbose be verbose

#### **Details**

Checks if a miniconda giotto environment can be found. Can be installed with installGiottoEnvironment.

clusterCells 43

clusterCells

clusterCells

#### **Description**

cluster cells using a variety of different methods

```
clusterCells(
  gobject,
 cluster_method = c("leiden", "louvain_community", "louvain_multinet", "randomwalk",
    "sNNclust", "kmeans", "hierarchical"),
  name = "cluster_name",
  nn_network_to_use = "sNN",
  network_name = "sNN.pca",
  pyth_leid_resolution = 1,
  pyth_leid_weight_col = "weight",
  pyth_leid_part_type = c("RBConfigurationVertexPartition",
    "ModularityVertexPartition"),
  pyth_leid_init_memb = NULL,
  pyth_leid_iterations = 1000,
  pyth_louv_resolution = 1,
  pyth_louv_weight_col = NULL,
  python_louv_random = F,
  python_path = NULL,
  louvain_gamma = 1,
  louvain\_omega = 1,
  walk_steps = 4,
  walk_clusters = 10,
  walk_weights = NA,
  sNNclust_k = 20,
  sNNclust_eps = 4,
  sNNclust_minPts = 16,
  borderPoints = TRUE,
  expression_values = c("normalized", "scaled", "custom"),
  genes_to_use = NULL,
  dim_reduction_to_use = c("cells", "pca", "umap", "tsne"),
  dim_reduction_name = "pca",
  dimensions_to_use = 1:10,
  distance_method = c("original", "pearson", "spearman", "euclidean", "maximum",
    "manhattan", "canberra", "binary", "minkowski"),
  km_{centers} = 10,
  km_iter_max = 100,
  km_nstart = 1000,
  km_algorithm = "Hartigan-Wong",
 hc_agglomeration_method = c("ward.D2", "ward.D", "single", "complete", "average",
    "mcquitty", "median", "centroid"),
  hc_k = 10,
  hc_h = NULL
  return_gobject = TRUE,
```

44 clusterCells

```
set_seed = T,
seed_number = 1234
)
```

#### **Arguments**

giotto object gobject cluster\_method community cluster method to use name for new clustering result name nn\_network\_to\_use type of NN network to use (kNN vs sNN) name of NN network to use network\_name pyth\_leid\_resolution resolution for leiden pyth\_leid\_weight\_col column to use for weights pyth\_leid\_part\_type partition type to use pyth\_leid\_init\_memb initial membership pyth\_leid\_iterations number of iterations pyth\_louv\_resolution resolution for louvain pyth\_louv\_weight\_col python louvain param: weight column python\_louv\_random python louvain param: random python\_path specify specific path to python if required louvain param: gamma or resolution louvain\_gamma louvain\_omega louvain param: omega walk\_steps randomwalk: number of steps walk\_clusters randomwalk: number of clusters randomwalk: weight column walk\_weights SNNclust: k neighbors to use sNNclust\_k SNNclust: epsilon sNNclust\_eps sNNclust\_minPts SNNclust: min points borderPoints SNNclust: border points expression\_values expression values to use genes\_to\_use = NULL. dim\_reduction\_to\_use dimension reduction to use dim\_reduction\_name

name of reduction 'pca',

clusterSpatialCorGenes

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```
dimensions_to_use
```

dimensions to use

distance\_method

distance method

km\_centers kmeans centers km\_iter\_max kmeans iterations

km\_nstart kmeans random starting points

km\_algorithm kmeans algorithm

hc\_agglomeration\_method

hierarchical clustering method

hc\_k hierachical number of clusters

hc\_h hierarchical tree cutoff

return\_gobject boolean: return giotto object (default = TRUE)

set\_seed set seed

seed\_number number for seed

## **Details**

Wrapper for the different clustering methods.

#### Value

giotto object with new clusters appended to cell metadata

## See Also

 $\label{lem:cluster_doLouvainCluster_multinet} do Louvain Cluster\_community, do Louvain Cluster\_multinet, do Louvain Cluster, do Random Walk Cluster, do SNN Cluster, do Kmeans, do H clust Cluster, do Louvain Cluster, do Louva$ 

```
clusterSpatialCorGenes
```

cluster Spatial Cor Genes

# Description

Cluster based on spatially correlated genes

```
clusterSpatialCorGenes(
   spatCorObject,
   name = "spat_clus",
   hclust_method = "ward.D",
   k = 10,
   return_obj = TRUE
)
```

46 colSums\_giotto

## **Arguments**

spatCorObject spatial correlation object

name name for spatial clustering results
hclust\_method method for hierarchical clustering
k number of clusters to extract

return\_obj return spatial correlation object (spatCorObject)

## Value

spatCorObject or cluster results

colMeans\_giotto colMeans\_giotto

# Description

colMeans function that works with multiple matrix representations

# Usage

```
colMeans_giotto(mymatrix)
```

# Arguments

mymatrix matrix object

# Value

numeric vector

 $colSums\_giotto$   $colSums\_giotto$ 

## **Description**

colSums function that works with multiple matrix representations

# Usage

```
colSums_giotto(mymatrix)
```

# Arguments

mymatrix matrix object

## Value

numeric vector

combCCcom 47

combCCcom combCCcom

## **Description**

Combine spatial and expression based cell-cell communication data.tables

## Usage

```
combCCcom(
  spatialCC,
  exprCC,
  min_lig_nr = 3,
  min_rec_nr = 3,
  min_padj_value = 1,
  min_log2fc = 0,
  min_av_diff = 0,
  detailed = FALSE
)
```

# Arguments

```
spatialCC spatial cell-cell communication scores

exprCC expression cell-cell communication scores

min_lig_nr minimum number of ligand cells

min_rec_nr minimum number of receptor cells

min_padj_value minimum adjusted p-value

min_log2fc minimum log2 fold-change

min_av_diff minimum average expression difference

detailed detailed option used with spatCellCellcom (default = FALSE)
```

## Value

combined data.table with spatial and expression communication data

```
combine {\tt CellProximity Genes} \\ combine {\tt CellProximity Genes}
```

# Description

Combine ICG scores in a pairwise manner.

```
combineCellProximityGenes(...)
```

48 combineCPG

#### **Arguments**

... Arguments passed on to combineInteractionChangedGenes

cpg0bject ICG (interaction changed gene) score object

selected\_ints subset of selected cell-cell interactions (optional)

selected\_genes subset of selected genes (optional)

specific\_genes\_1 specific geneset combo (need to position match specific genes 2)

specific\_genes\_2 specific geneset combo (need to position match specific\_genes\_1)

min\_cells minimum number of target cell type

min\_int\_cells minimum number of interacting cell type

min\_fdr minimum adjusted p-value

min\_spat\_diff minimum absolute spatial expression difference

min\_log2\_fc minimum absolute log2 fold-change

do\_parallel run calculations in parallel with mclapply

cores number of cores to use if do\_parallel = TRUE

verbose verbose

#### See Also

combineInteractionChangedGenes

combineCPG

combineCPG

## **Description**

Combine ICG scores in a pairwise manner.

#### Usage

combineCPG(...)

## **Arguments**

... Arguments passed on to combineICG

cpg0bject ICG (interaction changed gene) score object

selected\_ints subset of selected cell-cell interactions (optional)

selected\_genes subset of selected genes (optional)

specific\_genes\_1 specific geneset combo (need to position match specific\_genes\_2)

specific\_genes\_2 specific geneset combo (need to position match specific\_genes\_1)

min\_cells minimum number of target cell type

min\_int\_cells minimum number of interacting cell type

min\_fdr minimum adjusted p-value

min\_spat\_diff minimum absolute spatial expression difference

min\_log2\_fc minimum absolute log2 fold-change

do\_parallel run calculations in parallel with mclapply

cores number of cores to use if do\_parallel = TRUE

verbose verbose

combineICG 49

#### See Also

combineICG

combineICG

combineICG

## Description

Combine ICG scores in a pairwise manner.

## Usage

```
combineICG(
  cpgObject,
  selected_ints = NULL,
  selected_genes = NULL,
  specific_genes_1 = NULL,
  specific_genes_2 = NULL,
  min_cells = 5,
  min_int_cells = 3,
  min_fdr = 0.05,
  min_spat_diff = 0,
  min_log2_fc = 0.5,
  do_parallel = TRUE,
  cores = NA,
  verbose = T
)
```

#### **Arguments**

```
cpgObject
                  ICG (interaction changed gene) score object
                  subset of selected cell-cell interactions (optional)
selected_ints
selected_genes subset of selected genes (optional)
specific_genes_1
                  specific geneset combo (need to position match specific_genes_2)
specific_genes_2
                  specific geneset combo (need to position match specific_genes_1)
min_cells
                  minimum number of target cell type
min_int_cells
                  minimum number of interacting cell type
                  minimum adjusted p-value
min_fdr
min_spat_diff
                  minimum absolute spatial expression difference
                  minimum absolute log2 fold-change
min_log2_fc
                  run calculations in parallel with mclapply
do_parallel
                  number of cores to use if do_parallel = TRUE
cores
verbose
                  verbose
```

#### Value

cpgObject that contains the filtered differential gene scores

```
{\tt combineInteractionChangedGenes}
```

combine Interaction Changed Genes

## **Description**

Combine ICG scores in a pairwise manner.

# Usage

```
combineInteractionChangedGenes(
  cpgObject,
  selected_ints = NULL,
  selected_genes = NULL,
  specific_genes_1 = NULL,
  specific_genes_2 = NULL,
  min_cells = 5,
  min_int_cells = 3,
  min_fdr = 0.05,
  min_spat_diff = 0,
  min_log2_fc = 0.5,
  do_parallel = TRUE,
  cores = NA,
  verbose = T
)
```

# **Arguments**

```
cpgObject
                  ICG (interaction changed gene) score object
selected_ints
                  subset of selected cell-cell interactions (optional)
selected_genes subset of selected genes (optional)
specific_genes_1
                  specific geneset combo (need to position match specific_genes_2)
specific_genes_2
                  specific geneset combo (need to position match specific_genes_1)
                  minimum number of target cell type
min_cells
min_int_cells
                  minimum number of interacting cell type
min_fdr
                  minimum adjusted p-value
min_spat_diff
                  minimum absolute spatial expression difference
                  minimum absolute log2 fold-change
min_log2_fc
                  run calculations in parallel with mclapply
do_parallel
cores
                  number of cores to use if do_parallel = TRUE
verbose
                  verbose
```

#### Value

cpgObject that contains the filtered differential gene scores

combineMetadata 51

combine Metadata
------------------

## **Description**

This function combines the cell metadata with spatial locations and enrichment results from runSpatialEnrich

# Usage

```
combineMetadata(gobject, spat_enr_names = NULL)
```

## **Arguments**

```
gobject Giotto object
spat_enr_names names of spatial enrichment results to include
```

#### Value

Extended cell metadata in data.table format.

```
convertEnsemblToGeneSymbol
```

convert Ensembl To Gene Symbol

# Description

This function convert ensembl gene IDs from a matrix to official gene symbols

# Usage

```
convertEnsemblToGeneSymbol(matrix, species = c("mouse", "human"))
```

## **Arguments**

matrix an expression matrix with ensembl gene IDs as rownames

species species to use for gene symbol conversion

## **Details**

This function requires that the biomaRt library is installed

# Value

expression matrix with gene symbols as rownames

52 createCrossSection

createCrossSection

createCrossSection

#### **Description**

Create a virtual 2D cross section.

#### Usage

```
createCrossSection(
  gobject.
 name = "cross_section",
  spatial_network_name = "Delaunay_network",
  thickness_unit = c("cell", "natural"),
  slice_thickness = 2,
  cell_distance_estimate_method = "mean",
  extend_ratio = 0.2,
 method = c("equation", "3 points", "point and norm vector",
    "point and two plane vectors"),
  equation = NULL,
  point1 = NULL,
  point2 = NULL,
  point3 = NULL,
 normVector = NULL,
 planeVector1 = NULL,
 planeVector2 = NULL,
 mesh\_grid\_n = 20,
  return_gobject = TRUE
)
```

## **Arguments**

gobject giotto object name name of cress section object. (default = cross\_section) spatial\_network\_name name of spatial network object. (default = Delaunay\_network) thickness\_unit unit of the virtual section thickness. If "cell", average size of the observed cells is used as length unit. If "natural", the unit of cell location coordinates is used.(default = cell) slice\_thickness thickness of slice. default = 2 ${\tt cell\_distance\_estimate\_method}$ method to estimate average distance between neighboring cells. (default = mean) deciding the span of the cross section meshgrid, as a ratio of extension compared extend\_ratio to the borders of the vitural tissue section. (default = 0.2) method method to define the cross section plane. If equation, the plane is defined by a four element numerical vector (equation) in the form of c(A,B,C,D), corresponding to a plane with equation Ax+By+Cz=D. If 3 points, the plane is define

by the coordinates of 3 points, as given by point1, point2, and point3. If point

createGiottoImage 53

	and norm vector, the plane is defined by the coordinates of one point (point1) in the plane and the coordinates of one norm vector (normVector) to the plane. If point and two plane vector, the plane is defined by the coordinates of one point (point1) in the plane and the coordinates of two vectors (planeVector1, planeVector2) in the plane. (default = equation)
equation	equation required by method "equation".equations needs to be a numerical vector of length 4, in the form of $c(A,B,C,D)$ , which defines plane $Ax+By+Cz=D$ .
point1	coordinates of the first point required by method "3 points", "point and norm vector", and "point and two plane vectors".
point2	coordinates of the second point required by method "3 points"
point3	coordinates of the third point required by method "3 points"
normVector	coordinates of the norm vector required by method "point and norm vector"
planeVector1	coordinates of the first plane vector required by method "point and two plane vectors"
planeVector2	coordinates of the second plane vector required by method "point and two plane vectors"
mesh_grid_n	numer of meshgrid lines to generate along both directions for the cross section plane.
return_gobject	boolean: return giotto object (default = TRUE)

## **Details**

Creates a virtual 2D cross section object for a given spatial network object. The users need to provide the definition of the cross section plane (see method).

#### Value

giotto object with updated spatial network slot

createGiottoImage

# Description

Creates a giotto image that can be added to a Giotto object and/or used to add an image to the spatial plotting functions

```
createGiottoImage(
  gobject = NULL,
  spatial_locs = NULL,
  mg_object,
  name = "image",
  xmax_adj = 0,
  xmin_adj = 0,
  ymax_adj = 0,
  ymin_adj = 0
)
```

54 createGiottoInstructions

## **Arguments**

```
gobject
                  giotto object
spatial_locs
                  spatial locations (alternative if giobject = NULL)
                  magick image object
mg_object
name
                  name for the image
                  adjustment of the maximum x-value to align the image
xmax_adj
xmin_adj
                  adjustment of the minimum x-value to align the image
                  adjustment of the maximum y-value to align the image
ymax_adj
                  adjustment of the minimum y-value to align the image
ymin_adj
```

#### Value

a giotto image object

createGiottoInstructions

createGiottoInstructions

## **Description**

Function to set global instructions for giotto functions

## Usage

```
createGiottoInstructions(
  python_path = NULL,
  show_plot = NULL,
  return_plot = NULL,
  save_plot = NULL,
  save_dir = NULL,
  plot_format = NULL,
  dpi = NULL,
  units = NULL,
  height = NULL,
  width = NULL,
  is_docker = FALSE
)
```

# Arguments

```
path to python binary to use
show_plot print plot to console, default = TRUE
return_plot return plot as object, default = TRUE
save_plot automatically save plot, dafault = FALSE
save_dir path to directory where to save plots
plot_format format of plots (defaults to png)
dpi resolution for raster images
```

createGiottoObject 55

```
units units of format (defaults to in)
height height of plots
width width of plots
is_docker using docker implementation of Giotto (defaults to FALSE)
```

## Value

named vector with giotto instructions

#### See Also

More online information can be found here https://rubd.github.io/Giotto\_site/articles/instructions\_and\_plotting.html

## **Description**

Function to create a giotto object

## Usage

```
createGiottoObject(
  raw_exprs,
  spatial_locs = NULL,
  norm_expr = NULL,
  norm_scaled_expr = NULL,
  custom_expr = NULL,
  cell_metadata = NULL,
  gene_metadata = NULL,
  spatial_network = NULL,
  spatial_network_name = NULL,
  spatial_grid = NULL,
  spatial_grid_name = NULL,
  spatial_enrichment = NULL,
  spatial_enrichment_name = NULL,
  dimension_reduction = NULL,
  nn_network = NULL,
  images = NULL,
  offset_file = NULL,
  instructions = NULL,
  cores = NA
)
```

# Arguments

```
raw_exprs matrix with raw expression counts [required]
spatial_locs data.table or data.frame with coordinates for cell centroids
norm_expr normalized expression values
```

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```
norm_scaled_expr
                  scaled expression values
                  custom expression values
custom_expr
                  cell annotation metadata
cell_metadata
gene_metadata
                  gene annotation metadata
spatial_network
                  list of spatial network(s)
spatial_network_name
                  list of spatial network name(s)
                  list of spatial grid(s)
spatial_grid
spatial_grid_name
                  list of spatial grid name(s)
spatial_enrichment
                  list of spatial enrichment score(s) for each spatial region
spatial_enrichment_name
                  list of spatial enrichment name(s)
dimension_reduction
                  list of dimension reduction(s)
                  list of nearest neighbor network(s)
nn_network
                  list of images
images
offset_file
                  file used to stitch fields together (optional)
instructions
                  list of instructions or output result from createGiottoInstructions
                  how many cores or threads to use to read data if paths are provided
cores
```

#### **Details**

[**Requirements**] To create a giotto object you need to provide at least a matrix with genes as row names and cells as column names. This matrix can be provided as a base matrix, sparse Matrix, data.frame, data.table or as a path to any of those. To include spatial information about cells (or regions) you need to provide a matrix, data.table or data.frame (or path to them) with coordinates for all spatial dimensions. This can be 2D (x and y) or 3D (x, y, x). The row order for the cell coordinates should be the same as the column order for the provided expression data.

[Instructions] Additionally an instruction file, generated manually or with createGiottoInstructions can be provided to instructions, if not a default instruction file will be created for the Giotto object.

[Multiple fields] In case a dataset consists of multiple fields, like seqFISH+ for example, an offset file can be provided to stitch the different fields together. stitchFieldCoordinates can be used to generate such an offset file.

[**Processed data**] Processed count data, such as normalized data, can be provided using one of the different expression slots (norm\_expr, norm\_scaled\_expr, custom\_expr).

[Metadata] Cell and gene metadata can be provided using the cell and gene metadata slots. This data can also be added afterwards using the addGeneMetadata or addCellMetadata functions.

[Other information] Additional information can be provided through the appropriate slots:

- · spatial networks
- · spatial girds
- spatial enrichments
- · dimensions reductions
- nearest neighbours networks
- · images

#### Value

```
giotto object
```

```
createGiottoVisiumObject
```

createGiottoVisiumObject

## **Description**

creates Giotto object directly from a 10X visium folder

## Usage

```
createGiottoVisiumObject(
  visium_dir = NULL,
  expr_data = c("raw", "filter"),
  gene_column_index = 1,
  png_name = NULL,
  xmax_adj = 0,
  xmin_adj = 0,
  ymax_adj = 0,
  ymin_adj = 0,
  instructions = NULL,
  cores = NA
)
```

#### **Arguments**

```
visium_dir
                  path to the 10X visium directory [required]
expr_data
                  raw or filtered data (see details)
gene_column_index
                  which column index to select (see details)
png_name
                  select name of png to use (see details)
xmax_adj
                  adjustment of the maximum x-value to align the image
                  adjustment of the minimum x-value to align the image
xmin_adj
                  adjustment of the maximum y-value to align the image
ymax_adj
                  adjustment of the minimum y-value to align the image
ymin_adj
instructions
                  list of instructions or output result from createGiottoInstructions
cores
                  how many cores or threads to use to read data if paths are provided
```

## **Details**

- expr\_data: raw will take expression data from raw\_feature\_bc\_matrix and filter from filtered\_feature\_bc\_matrix
- gene\_column\_index: which gene identifiers (names) to use if there are multiple columns (e.g. ensemble and gene symbol)
- png\_name: by default the first png will be selected, provide the png name to override this (e.g. myimage.png)

58 createMetagenes

#### Value

giotto object

createMetagenes

createMetagenes

## **Description**

This function creates an average metagene for gene clusters.

## Usage

```
createMetagenes(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  gene_clusters,
  name = "metagene",
  return_gobject = TRUE
)
```

## **Arguments**

```
gobject Giotto object
expression_values
expression values to use
gene_clusters numerical vector with genes as names
name name of the metagene results
return_gobject return giotto object
```

#### **Details**

```
An example for the 'gene_clusters' could be like this: cluster_vector = c(1, 1, 2, 2); names(cluster_vector) = c('geneA', 'geneB', 'geneC', 'geneD')
```

#### Value

giotto object

## **Examples**

createNearestNetwork 59

createNearestNetwork createNearestNetwork

## **Description**

create a nearest neighbour (NN) network

## Usage

```
createNearestNetwork(
  gobject,
  type = c("sNN", "kNN"),
  dim_reduction_to_use = "pca",
  dim_reduction_name = "pca",
  dimensions_to_use = 1:10,
  genes_to_use = NULL,
  expression_values = c("normalized", "scaled", "custom"),
  name = "sNN.pca",
  return_gobject = TRUE,
  k = 30,
  minimum_shared = 5,
  top_shared = 3,
  verbose = T,
  ...
)
```

## **Arguments**

```
giotto object
gobject
                  sNN or kNN
tvpe
dim_reduction_to_use
                  dimension reduction method to use
dim_reduction_name
                  name of dimension reduction set to use
{\tt dimensions\_to\_use}
                  number of dimensions to use as input
                  if dim_reduction_to_use = NULL, which genes to use
genes_to_use
expression_values
                  expression values to use
                  arbitrary name for NN network
return_gobject boolean: return giotto object (default = TRUE)
```

60 createNearestNetwork

```
k number of k neighbors to use
minimum_shared minimum shared neighbors
top_shared keep at ...
verbose be verbose
additional parameters for kNN and sNN functions from dbscan
```

#### **Details**

This function creates a k-nearest neighbour (kNN) or shared nearest neighbour (sNN) network based on the provided dimension reduction space. To run it directly on the gene expression matrix set  $dim\_reduction\_to\_use = NULL$ .

See also kNN and sNN for more information about how the networks are created.

#### Output for kNN:

• from: cell\_ID for source cell

• to: cell\_ID for target cell

• distance: distance between cells

• weight: weight = 1/(1 + distance)

#### Output for sNN:

• from: cell ID for source cell

• to: cell\_ID for target cell

• distance: distance between cells

• weight: 1/(1 + distance)

• shared: number of shared neighbours

• rank: ranking of pairwise cell neighbours

For sNN networks two additional parameters can be set:

- minimum\_shared: minimum number of shared neighbours needed
- top\_shared: keep this number of the top shared neighbours, irrespective of minimum\_shared setting

#### Value

giotto object with updated NN network

## **Examples**

```
create {\tt Spatial Default Grid} \\ {\it create Spatial Default Grid}
```

## **Description**

Create a spatial grid using the default method

# Usage

```
createSpatialDefaultGrid(
  gobject,
  sdimx_stepsize = NULL,
  sdimy_stepsize = NULL,
  sdimz_stepsize = NULL,
  minimum_padding = 1,
  name = NULL,
  return_gobject = TRUE
)
```

## **Arguments**

```
gobject giotto object

sdimx_stepsize stepsize along the x-axis

sdimy_stepsize stepsize along the y-axis

sdimz_stepsize stepsize along the z-axis

minimum_padding

minimum padding on the edges

name name for spatial grid (default = 'spatial_grid')

return_gobject boolean: return giotto object (default = TRUE)
```

## **Details**

Creates a spatial grid with defined x, y (and z) dimensions. The dimension units are based on the provided spatial location units.

## Value

giotto object with updated spatial grid slot

```
createSpatialDelaunayNetwork
```

createSpatialDelaunayNetwork

#### **Description**

Create a spatial Delaunay network based on cell centroid physical distances.

#### Usage

```
createSpatialDelaunayNetwork(
  gobject,
  method = c("deldir", "delaunayn_geometry", "RTriangle"),
  dimensions = "all",
  name = "Delaunay_network",
  maximum_distance = "auto",
  minimum_k = 0,
  options = "Pp",
  Y = TRUE,
  j = TRUE,
  S = 0,
  verbose = T,
  return_gobject = TRUE,
  ...
)
```

# Arguments

gobject giotto object

method package to use to create a Delaunay network

dimensions which spatial dimensions to use. Use "sdimx" (spatial dimension x), "sdimy",

"sdimz" respectively to refer to X (or the 1st), Y (or the 2nd) and Z(or the 3rd)

dimension, see details. (default = all)

name for spatial network (default = 'delaunay\_network')

maximum\_distance

distance cuttof for Delaunay neighbors to consider. If "auto", "upper wisker" value of the distance vector between neighbors is used; see the boxplotgraphics

documentation for more details.(default = "auto")

minimum\_k minimum number of neighbours if maximum\_distance != NULL

options (geometry) String containing extra control options for the underlying Qhull

command; see the Qhull documentation (../doc/qhull/html/qdelaun.html) for the

available options. (default = 'Pp', do not report precision problems)

Y (RTriangle) If TRUE prohibits the insertion of Steiner points on the mesh bound-

ary.

j (RTriangle) If TRUE jettisons vertices that are not part of the final triangulation

from the output.

S (RTriangle) Specifies the maximum number of added Steiner points.

verbose verbose

return\_gobject boolean: return giotto object (default = TRUE)

... Other additional parameters

createSpatialEnrich 63

#### **Details**

Creates a spatial Delaunay network as explained in delaunayn (default), deldir, or triangulate.

#### Value

giotto object with updated spatial network slot

createSpatialEnrich createSpatialEnrich

#### **Description**

Function to calculate gene signature enrichment scores per spatial position using an enrichment test.

#### Usage

```
createSpatialEnrich(...)
```

#### **Arguments**

```
Arguments passed on to runSpatialEnrich
. . .
                  gobject Giotto object
                  enrich_method method for gene signature enrichment calculation
                  sign_matrix Matrix of signature genes for each cell type / process
                  expression_values expression values to use
                  reverse_log_scale reverse expression values from log scale
                  min_overlap_genes minimum number of overlapping genes in sign_matrix
                      required to calculate enrichment (PAGE)
                  logbase log base to use if reverse_log_scale = TRUE
                  p_value calculate p-value (default = FALSE)
                  n_times (page/rank) number of permutation iterations to calculate p-value
                  rbp_p (rank) fractional binarization threshold (default = 0.99)
                  num_agg (rank) number of top genes to aggregate (default = 100)
                  max\_block number of lines to process together (default = 20e6)
                  top_percentage (hyper) percentage of cells that will be considered to have
                      gene expression with matrix binarization
                  output_enrichment how to return enrichment output
                  name to give to spatial enrichment results, default = PAGE
                  verbose be verbose
                  return_gobject return giotto object
```

#### See Also

runSpatialEnrich

64 createSpatialGrid

# Description

Create a spatial grid using the default method

## Usage

```
createSpatialGrid(
  gobject,
  name = NULL,
  method = c("default"),
  sdimx_stepsize = NULL,
  sdimy_stepsize = NULL,
  sdimz_stepsize = NULL,
  minimum_padding = 1,
  return_gobject = TRUE
)
```

## **Arguments**

```
gobject giotto object

name name for spatial grid

method method to create a spatial grid

sdimx_stepsize stepsize along the x-axis

sdimy_stepsize stepsize along the y-axis

sdimz_stepsize stepsize along the z-axis

minimum_padding

minimum padding on the edges

return_gobject boolean: return giotto object (default = TRUE)
```

#### **Details**

Creates a spatial grid with defined x, y (and z) dimensions. The dimension units are based on the provided spatial location units.

• default method: createSpatialDefaultGrid

## Value

giotto object with updated spatial grid slot

```
createSpatialKNNnetwork
```

createSpatialKNNnetwork

#### **Description**

Create a spatial knn network.

## Usage

```
createSpatialKNNnetwork(
  gobject,
  method = "dbscan",
  dimensions = "all",
  name = "knn_network",
  k = 4,
  maximum_distance = NULL,
  minimum_k = 0,
  verbose = F,
  return_gobject = TRUE,
  ...
)
```

## **Arguments**

gobject

```
method method to create kNN network

dimensions which spatial dimensions to use (default = all)

name name for spatial network (default = 'spatial_network')

k number of nearest neighbors based on physical distance

maximum_distance

distance cuttof for nearest neighbors to consider for kNN network

minimum_k minimum nearest neighbours if maximum_distance != NULL

verbose verbose

return_gobject boolean: return giotto object (default = TRUE)
```

## Value

giotto object with updated spatial network slot

giotto object

**dimensions:** default = 'all' which takes all possible dimensions. Alternatively you can provide a character vector that specififies the spatial dimensions to use, e.g. c("sdimx', "sdimy") or a numerical vector, e.g. 2:3

additional arguments to the selected method function

**maximum\_distance:** to create a network based on maximum distance only, you also need to set k to a very high value, e.g. k = 100

66 createSpatialNetwork

```
create Spatial Network \\ create Spatial Network
```

giotto object

# **Description**

Create a spatial network based on cell centroid physical distances.

# Usage

```
createSpatialNetwork(
  gobject,
  name = NULL,
  dimensions = "all",
  method = c("Delaunay", "kNN"),
delaunay_method = c("deldir", "delaunayn_geometry", "RTriangle"),
maximum_distance_delaunay = "auto",
  options = "Pp",
  Y = TRUE,
  j = TRUE,
  S = 0,
  minimum_k = 0,
  knn_method = "dbscan",
  k = 4,
  maximum_distance_knn = NULL,
  verbose = F,
  return_gobject = TRUE,
)
```

# Arguments

gobject

name name for spatial network (default = 'spatial_network')		
dimensions which spatial dimensions to use (default = all)		
method which method to use to create a spatial network. (default = Delaunay)		
delaunay_method		
Delaunay method to use		
maximum_distance_delaunay		
distance cuttof for nearest neighbors to consider for Delaunay network		
options (geometry) String containing extra control options for the underlying Qhu command; see the Qhull documentation (/doc/qhull/html/qdelaun.html) for the available options. (default = 'Pp', do not report precision problems)		
Y (RTriangle) If TRUE prohibits the insertion of Steiner points on the mesh bound ary.		
j (RTriangle) If TRUE jettisons vertices that are not part of the final triangulation from the output.		
S (RTriangle) Specifies the maximum number of added Steiner points.		
minimum_k minimum nearest neigbhours if maximum_distance != NULL		

#### **Details**

Creates a spatial network connecting single-cells based on their physical distance to each other. For Delaunay method, neighbors will be decided by delaunay triangulation and a maximum distance criteria. For kNN method, number of neighbors can be determined by k, or maximum distance from each cell with or without setting a minimum k for each cell.

**dimensions:** default = 'all' which takes all possible dimensions. Alternatively you can provide a character vector that specififies the spatial dimensions to use, e.g. c("sdimx', "sdimy") or a numerical vector, e.g. 2:3

#### Value

giotto object with updated spatial network slot

#### **Description**

create a crossSection object

```
create_crossSection_object(
 name = NULL,
 method = NULL,
  thickness_unit = NULL,
  slice_thickness = NULL,
  cell_distance_estimate_method = NULL,
  extend_ratio = NULL,
 plane_equation = NULL,
 mesh_grid_n = NULL,
 mesh_obj = NULL,
 cell_subset = NULL,
  cell_subset_spatial_locations = NULL,
  cell_subset_projection_locations = NULL,
  cell_subset_projection_PCA = NULL,
  cell_subset_projection_coords = NULL
)
```

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#### **Arguments**

```
name
                  name of cress section object. (default = cross_sectino)
method
                  method to define the cross section plane.
thickness_unit unit of the virtual section thickness. If "cell", average size of the observed
                  cells is used as length unit. If "natural", the unit of cell location coordinates
                  is used.(default = cell)
slice_thickness
                  thickness of slice
cell_distance_estimate_method
                  method to estimate average distance between neighboring cells. (default = mean)
extend_ratio
                  deciding the span of the cross section meshgrid, as a ratio of extension compared
                  to the borders of the vitural tissue section. (default = 0.2)
plane_equation a numerical vector of length 4, in the form of c(A,B,C,D), which defines plane
                  Ax+By+Cz=D.
                  numer of meshgrid lines to generate along both directions for the cross section
mesh_grid_n
mesh_obj
                  object that stores the cross section meshgrid information.
cell_subset
                  cells selected by the cross section
cell_subset_spatial_locations
                  locations of cells selected by the cross section
cell_subset_projection_locations
                  3D projection coordinates of selected cells onto the cross section plane
cell_subset_projection_PCA
                  pca of projection coordinates
cell_subset_projection_coords
                  2D PCA coordinates of selected cells in the cross section plane
```

 ${\tt crossSectionGenePlot} \quad {\it crossSectionGenePlot}$ 

#### **Description**

Visualize cells and gene expression in a virtual cross section according to spatial coordinates

```
crossSectionGenePlot(
  gobject = NULL,
  crossSection_obj = NULL,
  name = NULL,
  spatial_network_name = "Delaunay_network",
  default_save_name = "crossSectionGenePlot",
  ...
)
```

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## **Arguments**

```
gobject giotto object

crossSection_obj

crossSection object

name name of virtual cross section to use

spatial_network_name

name of spatial network to use

default_save_name

default save name for saving, don't change, change save_name in save_param

parameters for spatGenePlot2D
```

#### **Details**

Description of parameters.

#### Value

ggplot

# See Also

```
spatGenePlot3D and spatGenePlot2D
```

```
crossSectionGenePlot3D
```

crossSectionGenePlot3D

## **Description**

Visualize cells and gene expression in a virtual cross section according to spatial coordinates

## Usage

```
crossSectionGenePlot3D(
  gobject,
  crossSection_obj = NULL,
  name = NULL,
  spatial_network_name = "Delaunay_network",
  other_cell_color = alpha("lightgrey", 0),
  default_save_name = "crossSectionGenePlot3D",
   ...
)
```

## **Arguments**

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#### **Details**

Description of parameters.

#### Value

ggplot

crossSectionPlot

cross Section Plot

# Description

Visualize cells in a virtual cross section according to spatial coordinates

## Usage

```
crossSectionPlot(
  gobject,
  crossSection_obj = NULL,
  name = NULL,
  spatial_network_name = "Delaunay_network",
  default_save_name = "crossSectionPlot",
   ...
)
```

# Arguments

## Details

Description of parameters.

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#### Value

ggplot

## See Also

crossSectionPlot

 ${\tt crossSectionPlot3D}$ 

cross Section Plot 3D

# Description

Visualize cells in a virtual cross section according to spatial coordinates

# Usage

```
crossSectionPlot3D(
  gobject,
  crossSection_obj = NULL,
  name = NULL,
  spatial_network_name = "Delaunay_network",
  show_other_cells = T,
  other_cell_color = alpha("lightgrey", 0),
  default_save_name = "crossSection3D",
  ...
)
```

## **Arguments**

## **Details**

Description of parameters.

# Value

ggplot

detectSpatialCorGenes detectSpatialCorGenes

## **Description**

Detect genes that are spatially correlated

## Usage

```
detectSpatialCorGenes(
  gobject,
  method = c("grid", "network"),
  expression_values = c("normalized", "scaled", "custom"),
  subset_genes = NULL,
  spatial_network_name = "Delaunay_network",
  network_smoothing = NULL,
  spatial_grid_name = "spatial_grid",
  min_cells_per_grid = 4,
  cor_method = c("pearson", "kendall", "spearman")
)
```

## **Arguments**

```
gobject
                  giotto object
method
                  method to use for spatial averaging
expression_values
                  gene expression values to use
subset_genes
                  subset of genes to use
spatial_network_name
                  name of spatial network to use
network_smoothing
                  smoothing factor beteen 0 and 1 (default: automatic)
spatial_grid_name
                  name of spatial grid to use
min_cells_per_grid
                  minimum number of cells to consider a grid
cor_method
                  correlation method
```

## **Details**

For method = network, it expects a fully connected spatial network. You can make sure to create a fully connected network by setting  $minimal_k > 0$  in the createSpatialNetwork function.

- 1. grid-averaging: average gene expression values within a predefined spatial grid
- 2. network-averaging: smoothens the gene expression matrix by averaging the expression within one cell by using the neighbours within the predefined spatial network. b is a smoothening factor that defaults to 1 1/k, where k is the median number of k-neighbors in the selected spatial network. Setting b = 0 means no smoothing and b = 1 means no contribution from its own expression.

The spatCorObject can be further explored with showSpatialCorGenes()

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#### Value

```
returns a spatial correlation object: "spatCorObject"
```

#### See Also

```
showSpatialCorGenes
```

```
detectSpatialPatterns detectSpatialPatterns
```

## **Description**

Identify spatial patterns through PCA on average expression in a spatial grid.

#### Usage

```
detectSpatialPatterns(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  spatial_grid_name = "spatial_grid",
  min_cells_per_grid = 4,
  scale_unit = F,
  ncp = 100,
  show_plot = T,
  PC_zscore = 1.5
)
```

# Arguments

```
gobject
                  giotto object
expression_values
                  expression values to use
spatial_grid_name
                  name of spatial grid to use (default = 'spatial_grid')
min_cells_per_grid
                  minimum number of cells in a grid to be considered
scale_unit
                  scale features
                  number of principal components to calculate
ncp
                  show plots
show_plot
PC_zscore
                  minimum z-score of variance explained by a PC
```

### Details

Steps to identify spatial patterns:

- 1. average gene expression for cells within a grid, see createSpatialGrid
- 2. perform PCA on the average grid expression profiles
- 3. convert variance of principlal components (PCs) to z-scores and select PCs based on a z-score threshold

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#### Value

spatial pattern object 'spatPatObj'

dimCellPlot

dimCellPlot

### **Description**

Visualize cells according to dimension reduction coordinates

# Usage

```
dimCellPlot(gobject, ...)
```

# Arguments

```
gobject
                 giotto object
                  Arguments passed on to dimCellPlot2D
. . .
                 dim_reduction_to_use dimension reduction to use
                 dim_reduction_name dimension reduction name
                 dim1_to_use dimension to use on x-axis
                 dim2_to_use dimension to use on y-axis
                 spat_enr_names names of spatial enrichment results to include
                 cell_annotation_values numeric cell annotation columns
                 show_NN_network show underlying NN network
                 nn_network_to_use type of NN network to use (kNN vs sNN)
                 network_name name of NN network to use, if show_NN_network = TRUE
                 cell color code named vector with colors for cell annotation values
                 cell_color_gradient vector with 3 colors for numeric data
                 gradient_midpoint midpoint for color gradient
                 gradient_limits vector with lower and upper limits
                  select_cell_groups select subset of cells/clusters based on cell_color param-
                      eter
                 select_cells select subset of cells based on cell IDs
                 show_other_cells display not selected cells
                 other_cell_color color of not selected cells
                 other_point_size size of not selected cells
                 show_cluster_center plot center of selected clusters
                  show_center_label plot label of selected clusters
                 center_point_size size of center points
                 center_point_border_col border color of center points
                 center_point_border_stroke border stroke size of center points
                 label_size size of labels
                 label_fontface font of labels
                 edge_alpha column to use for alpha of the edges
```

point\_shape point with border or not (border or no\_border)

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```
point_size size of point (cell)
point_alpha transparancy of dim. reduction points
point_border_col color of border around points
point_border_stroke stroke size of border around points
show_legend show legend
legend_text size of legend text
legend_symbol_size size of legend symbols
background_color color of plot background
axis_text size of axis text
axis_title size of axis title
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_plot show plot
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
default_save_name default save name for saving, don't change, change save_name
    in save param
```

#### **Details**

Description of parameters. For 3D plots see dimCellPlot2D

# Value

ggplot

### See Also

Other dimension reduction cell annotation visualizations: dimCellPlot2D()

# **Examples**

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dimCellPlot2D

dimCellPlot2D

#### **Description**

Visualize cells according to dimension reduction coordinates

### Usage

```
dimCellPlot2D(
  gobject,
  dim_reduction_to_use = "umap",
  dim_reduction_name = "umap",
  dim1_to_use = 1,
  dim2\_to\_use = 2,
  spat_enr_names = NULL,
  cell_annotation_values = NULL,
  show_NN_network = F,
  nn_network_to_use = "sNN",
  network_name = "sNN.pca",
  cell_color_code = NULL,
  cell_color_gradient = c("blue", "white", "red"),
  gradient_midpoint = NULL,
  gradient_limits = NULL,
  select_cell_groups = NULL,
  select_cells = NULL,
  show_other_cells = T,
  other_cell_color = "lightgrey",
  other_point_size = 0.5,
  show_cluster_center = F,
  show_center_label = T,
  center_point_size = 4,
  center_point_border_col = "black",
  center_point_border_stroke = 0.1,
  label_size = 4,
  label_fontface = "bold",
  edge_alpha = NULL,
  point_shape = c("border", "no_border"),
  point_size = 1,
  point_alpha = 1,
  point_border_col = "black",
  point_border_stroke = 0.1,
  show_legend = T,
  legend_text = 8,
  legend_symbol_size = 1,
  background_color = "white",
  axis_text = 8,
  axis_title = 8,
  cow_n_col = 2,
  cow_rel_h = 1,
  cow_rel_w = 1,
```

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```
cow_align = "h",
      show_plot = NA,
      return_plot = NA,
      save_plot = NA,
      save_param = list(),
      default_save_name = "dimCellPlot2D"
    )
Arguments
    gobject
                     giotto object
    dim_reduction_to_use
                     dimension reduction to use
    dim_reduction_name
                     dimension reduction name
                     dimension to use on x-axis
    dim1_to_use
                     dimension to use on y-axis
    dim2_to_use
    spat_enr_names names of spatial enrichment results to include
    cell_annotation_values
                     numeric cell annotation columns
    show_NN_network
                     show underlying NN network
    nn_network_to_use
                     type of NN network to use (kNN vs sNN)
    network_name
                     name of NN network to use, if show_NN_network = TRUE
    cell_color_code
                     named vector with colors for cell annotation values
    cell_color_gradient
                     vector with 3 colors for numeric data
    gradient_midpoint
                     midpoint for color gradient
    gradient_limits
                     vector with lower and upper limits
    select_cell_groups
                     select subset of cells/clusters based on cell_color parameter
    select_cells
                     select subset of cells based on cell IDs
    show_other_cells
                     display not selected cells
    other_cell_color
                     color of not selected cells
    other_point_size
                     size of not selected cells
    show_cluster_center
                     plot center of selected clusters
    show_center_label
                     plot label of selected clusters
    center_point_size
                     size of center points
```

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```
center_point_border_col
                  border color of center points
center_point_border_stroke
                  border stroke size of center points
label_size
                  size of labels
label_fontface font of labels
                  column to use for alpha of the edges
edge_alpha
                  point with border or not (border or no_border)
point_shape
point_size
                  size of point (cell)
point_alpha
                  transparancy of dim. reduction points
point_border_col
                  color of border around points
point_border_stroke
                  stroke size of border around points
show_legend
                  show legend
legend_text
                  size of legend text
legend_symbol_size
                  size of legend symbols
background_color
                  color of plot background
                  size of axis text
axis_text
axis_title
                  size of axis title
cow_n_col
                  cowplot param: how many columns
cow_rel_h
                  cowplot param: relative height
                  cowplot param: relative width
cow_rel_w
cow_align
                  cowplot param: how to align
show_plot
                  show plot
return_plot
                  return ggplot object
                  directly save the plot [boolean]
save_plot
                  list of saving parameters, see showSaveParameters
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

### **Details**

Description of parameters. For 3D plots see dimPlot3D

### Value

ggplot

## See Also

Other dimension reduction cell annotation visualizations: dimCellPlot()

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#### **Examples**

dimGenePlot

dimGenePlot

#### **Description**

Visualize gene expression according to dimension reduction coordinates

#### Usage

```
dimGenePlot(...)
```

# **Arguments**

```
Arguments passed on to dimGenePlot2D
. . .
                 gobject giotto object
                 expression_values gene expression values to use
                 genes genes to show
                 dim_reduction_to_use dimension reduction to use
                 dim_reduction_name dimension reduction name
                 dim1_to_use dimension to use on x-axis
                 dim2_to_use dimension to use on y-axis
                 show_NN_network show underlying NN network
                 nn_network_to_use type of NN network to use (kNN vs sNN)
                 network_name name of NN network to use, if show_NN_network = TRUE
                 network_color color of NN network
                 edge_alpha column to use for alpha of the edges
                 scale_alpha_with_expression scale expression with ggplot alpha parameter
                 point_shape point with border or not (border or no_border)
                 point_size size of point (cell)
                 point_alpha transparancy of points
                 cell_color_gradient vector with 3 colors for numeric data
                 gradient_midpoint midpoint for color gradient
                 gradient_limits vector with lower and upper limits
                 point_border_col color of border around points
```

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```
point_border_stroke stroke size of border around points
show_legend show legend
legend_text size of legend text
background_color color of plot background
axis_text size of axis text
axis_title size of axis title
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_plot show plots
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
default_save_name default save name for saving, don't change, change save_name
    in save_param
```

### **Details**

Description of parameters.

#### Value

ggplot

## See Also

```
dimGenePlot3D
```

Other dimension reduction gene expression visualizations: dimGenePlot2D(), dimGenePlot3D()

# **Examples**

```
data(mini_giotto_single_cell)
all_genes = slot(mini_giotto_single_cell, 'gene_ID')
selected_genes = all_genes[1:2]
dimGenePlot(mini_giotto_single_cell, genes = selected_genes, point_size = 3)
```

dimGenePlot2D

dimGenePlot2D

## **Description**

Visualize gene expression according to dimension reduction coordinates

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#### Usage

```
dimGenePlot2D(
     gobject,
     expression_values = c("normalized", "scaled", "custom"),
     genes = NULL,
     dim_reduction_to_use = "umap",
     dim_reduction_name = "umap",
     dim1_to_use = 1,
     dim2\_to\_use = 2,
     show_NN_network = F,
     nn_network_to_use = "sNN",
     network_name = "sNN.pca",
     network_color = "lightgray",
     edge_alpha = NULL,
     scale_alpha_with_expression = FALSE,
     point_shape = c("border", "no_border"),
     point_size = 1,
     point_alpha = 1,
     cell_color_gradient = c("blue", "white", "red"),
     gradient_midpoint = NULL,
     gradient_limits = NULL,
     point_border_col = "black",
     point_border_stroke = 0.1,
     show_legend = T,
     legend_text = 8,
     background_color = "white",
     axis_text = 8,
     axis_title = 8,
     cow_n_col = 2,
     cow_rel_h = 1,
     cow_rel_w = 1,
     cow_align = "h",
     show_plot = NA,
     return_plot = NA,
     save_plot = NA,
     save_param = list(),
     default_save_name = "dimGenePlot2D"
Arguments
   gobject
                   giotto object
   expression_values
                   gene expression values to use
   genes
                   genes to show
   dim_reduction_to_use
```

dimension reduction to use

dimension reduction name

dimension to use on x-axis

dimension to use on y-axis

dim\_reduction\_name

dim1\_to\_use

dim2\_to\_use

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```
show_NN_network
                 show underlying NN network
nn_network_to_use
                 type of NN network to use (kNN vs sNN)
network_name
                 name of NN network to use, if show_NN_network = TRUE
network_color
                 color of NN network
edge_alpha
                 column to use for alpha of the edges
scale_alpha_with_expression
                 scale expression with ggplot alpha parameter
point_shape
                 point with border or not (border or no_border)
point_size
                 size of point (cell)
point_alpha
                 transparancy of points
cell_color_gradient
                 vector with 3 colors for numeric data
gradient_midpoint
                 midpoint for color gradient
gradient_limits
                  vector with lower and upper limits
point_border_col
                 color of border around points
point_border_stroke
                 stroke size of border around points
                 show legend
show_legend
legend_text
                 size of legend text
background_color
                 color of plot background
                 size of axis text
axis_text
axis_title
                 size of axis title
cow_n_col
                 cowplot param: how many columns
cow_rel_h
                 cowplot param: relative height
cow_rel_w
                 cowplot param: relative width
                 cowplot param: how to align
cow_align
show_plot
                 show plots
return_plot
                 return ggplot object
                 directly save the plot [boolean]
save_plot
save_param
                 list of saving parameters, see showSaveParameters
default_save_name
                 default save name for saving, don't change, change save_name in save_param
```

### **Details**

Description of parameters.

#### Value

ggplot

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#### See Also

```
dimGenePlot3D
```

Other dimension reduction gene expression visualizations: dimGenePlot3D(), dimGenePlot()

#### **Examples**

```
data(mini_giotto_single_cell)
all_genes = slot(mini_giotto_single_cell, 'gene_ID')
selected_genes = all_genes[1:2]
dimGenePlot2D(mini_giotto_single_cell, genes = selected_genes, point_size = 3)
```

dimGenePlot3D

dimGenePlot3D

## Description

Visualize cells and gene expression according to dimension reduction coordinates

### Usage

```
dimGenePlot3D(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  genes = NULL,
  dim_reduction_to_use = "umap",
  dim_reduction_name = "umap",
  dim1_to_use = 1,
  dim2_to_use = 2,
  dim3_to_use = 3,
  show_NN_network = F,
  nn_network_to_use = "sNN",
  network_name = "sNN.pca",
  network_color = "lightgray",
  cluster_column = NULL,
  select_cell_groups = NULL,
  select_cells = NULL,
  show_other_cells = T,
  other_cell_color = "lightgrey",
  other_point_size = 1,
  edge_alpha = NULL,
  point_size = 2,
  genes_high_color = NULL,
  genes_mid_color = "white",
  genes_low_color = "blue",
  show_legend = T,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
```

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```
default_save_name = "dimGenePlot3D"
Arguments
                     giotto object
    gobject
    expression_values
                     gene expression values to use
    genes
                     genes to show
    dim_reduction_to_use
                     dimension reduction to use
    dim_reduction_name
                     dimension reduction name
    dim1_to_use
                     dimension to use on x-axis
                     dimension to use on y-axis
    dim2_to_use
    dim3_to_use
                     dimension to use on z-axis
    show_NN_network
                     show underlying NN network
    nn_network_to_use
                     type of NN network to use (kNN vs sNN)
    network_name
                     name of NN network to use, if show_NN_network = TRUE
    network_color
                     color of NN network
    cluster_column cluster column to select groups
    select_cell_groups
                     select subset of cells/clusters based on cell_color parameter
                     select subset of cells based on cell IDs
    select_cells
    show_other_cells
                     display not selected cells
    other_cell_color
                     color of not selected cells
    other_point_size
                     size of not selected cells
    edge_alpha
                     column to use for alpha of the edges
    point_size
                     size of point (cell)
    genes_high_color
                      color for high expression levels
    genes_mid_color
                     color for medium expression levels
    genes_low_color
                     color for low expression levels
    show_legend
                     show legend
    show_plot
                     show plots
    return_plot
                     return ggplot object
                     directly save the plot [boolean]
    save_plot
                     list of saving parameters, see showSaveParameters
    save_param
    default_save_name
```

default save name for saving, don't change, change save\_name in save\_param

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#### **Details**

Description of parameters.

#### Value

ggplot

#### See Also

Other dimension reduction gene expression visualizations: dimGenePlot2D(), dimGenePlot()

dimPlot

dimPlot

### Description

Visualize cells according to dimension reduction coordinates

### Usage

```
dimPlot(...)
```

### **Arguments**

Arguments passed on to dimPlot2D gobject giotto object group\_by create multiple plots based on cell annotation column group\_by\_subset subset the group\_by factor column dim\_reduction\_to\_use dimension reduction to use dim\_reduction\_name dimension reduction name dim1\_to\_use dimension to use on x-axis dim2\_to\_use dimension to use on y-axis spat\_enr\_names names of spatial enrichment results to include show\_NN\_network show underlying NN network nn\_network\_to\_use type of NN network to use (kNN vs sNN) network\_name name of NN network to use, if show\_NN\_network = TRUE cell\_color color for cells (see details) color\_as\_factor convert color column to factor cell\_color\_code named vector with colors cell\_color\_gradient vector with 3 colors for numeric data gradient\_midpoint midpoint for color gradient gradient\_limits vector with lower and upper limits select\_cell\_groups select subset of cells/clusters based on cell\_color paramselect\_cells select subset of cells based on cell IDs show\_other\_cells display not selected cells other\_cell\_color color of not selected cells other\_point\_size size of not selected cells

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```
show_cluster_center plot center of selected clusters
show_center_label plot label of selected clusters
center_point_size size of center points
center_point_border_col border color of center points
center_point_border_stroke border stroke size of center points
label_size size of labels
label_fontface font of labels
edge_alpha column to use for alpha of the edges
point_shape point with border or not (border or no_border)
point_size size of point (cell)
point_alpha transparancy of point
point_border_col color of border around points
point_border_stroke stroke size of border around points
title title for plot, defaults to cell_color parameter
show_legend show legend
legend_text size of legend text
legend_symbol_size size of legend symbols
background_color color of plot background
axis_text size of axis text
axis_title size of axis title
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_plot show plot
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
default_save_name default save name for saving, don't change, change save_name
    in save_param
```

### **Details**

Description of parameters, see dimPlot2D. For 3D plots see dimPlot3D

#### Value

ggplot

### See Also

```
Other reduced dimension visualizations: dimPlot2D(), dimPlot3D(), plotPCA_2D(), plotPCA_3D(), plotPCA(), plotTSNE_2D(), plotTSNE_3D(), plotUMAP_2D(), plotUMAP_3D(), plot
```

#### **Examples**

```
data(mini_giotto_single_cell)
dimPlot(mini_giotto_single_cell)
dimPlot(mini_giotto_single_cell, cell_color = 'cell_types', point_size = 3)
```

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dimPlot2D

dimPlot2D

## **Description**

Visualize cells according to dimension reduction coordinates

### Usage

```
dimPlot2D(
  gobject,
  group_by = NULL,
  group_by_subset = NULL,
  dim_reduction_to_use = "umap",
  dim_reduction_name = "umap",
  dim1_to_use = 1,
  dim2\_to\_use = 2,
  spat_enr_names = NULL,
  show_NN_network = F,
  nn_network_to_use = "sNN",
  network_name = "sNN.pca",
  cell_color = NULL,
  color_as_factor = T,
  cell_color_code = NULL,
  cell_color_gradient = c("blue", "white", "red"),
  gradient_midpoint = NULL,
  gradient_limits = NULL,
  select_cell_groups = NULL,
  select_cells = NULL,
  show_other_cells = T,
  other_cell_color = "lightgrey",
  other_point_size = 0.5,
  show_cluster_center = F,
  show_center_label = T,
  center_point_size = 4,
  center_point_border_col = "black",
  center_point_border_stroke = 0.1,
  label_size = 4,
  label_fontface = "bold",
  edge_alpha = NULL,
  point_shape = c("border", "no_border"),
  point_size = 1,
  point_alpha = 1,
  point_border_col = "black",
  point_border_stroke = 0.1,
  title = NULL,
  show_legend = T,
  legend_text = 8,
  legend_symbol_size = 1,
  background_color = "white",
  axis_text = 8,
```

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```
axis_title = 8,
      cow_n_col = 2,
      cow_rel_h = 1,
      cow_rel_w = 1,
      cow_align = "h",
      show_plot = NA,
      return_plot = NA,
      save_plot = NA,
      save_param = list(),
      default_save_name = "dimPlot2D"
    )
Arguments
    gobject
                     giotto object
    group_by
                     create multiple plots based on cell annotation column
    group_by_subset
                     subset the group_by factor column
    dim_reduction_to_use
                     dimension reduction to use
    dim_reduction_name
                     dimension reduction name
    dim1_to_use
                     dimension to use on x-axis
    dim2_to_use
                     dimension to use on y-axis
    spat_enr_names names of spatial enrichment results to include
    show_NN_network
                     show underlying NN network
    nn_network_to_use
                     type of NN network to use (kNN vs sNN)
    network_name
                     name of NN network to use, if show_NN_network = TRUE
    cell_color
                     color for cells (see details)
    color_as_factor
                     convert color column to factor
    cell_color_code
                     named vector with colors
    cell_color_gradient
                     vector with 3 colors for numeric data
    gradient_midpoint
                     midpoint for color gradient
    gradient_limits
                     vector with lower and upper limits
    select_cell_groups
                     select subset of cells/clusters based on cell_color parameter
    select_cells
                     select subset of cells based on cell IDs
    show_other_cells
                     display not selected cells
    other_cell_color
```

color of not selected cells

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```
other_point_size
                  size of not selected cells
show_cluster_center
                  plot center of selected clusters
show_center_label
                  plot label of selected clusters
center_point_size
                  size of center points
center_point_border_col
                  border color of center points
{\tt center\_point\_border\_stroke}
                  border stroke size of center points
label_size
                  size of labels
label_fontface font of labels
                  column to use for alpha of the edges
edge_alpha
                  point with border or not (border or no_border)
point_shape
point_size
                  size of point (cell)
point_alpha
                  transparancy of point
point_border_col
                  color of border around points
point_border_stroke
                  stroke size of border around points
title
                  title for plot, defaults to cell_color parameter
show_legend
                  show legend
legend_text
                  size of legend text
legend_symbol_size
                  size of legend symbols
background_color
                  color of plot background
axis_text
                  size of axis text
axis_title
                  size of axis title
cow_n_col
                  cowplot param: how many columns
cow_rel_h
                  cowplot param: relative height
                  cowplot param: relative width
cow_rel_w
                  cowplot param: how to align
cow_align
show_plot
                  show plot
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
save_param
                  list of saving parameters, see showSaveParameters
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

## **Details**

Description of parameters. For 3D plots see dimPlot3D

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#### Value

ggplot

#### See Also

```
Other reduced dimension visualizations: dimPlot3D(), dimPlot(), plotPCA_2D(), plotPCA_3D(), plotPCA(), plotTSNE_2D(), plotTSNE_3D(), plotTSNE(), plotUMAP_2D(), plotUMAP_3D(), plotUMAP()
```

### **Examples**

```
data(mini_giotto_single_cell)
dimPlot2D(mini_giotto_single_cell)
dimPlot2D(mini_giotto_single_cell, cell_color = 'cell_types', point_size = 3)
```

dimPlot3D

dimPlot3D

#### **Description**

Visualize cells according to dimension reduction coordinates

### Usage

```
dimPlot3D(
  gobject,
  dim_reduction_to_use = "umap",
  dim_reduction_name = "umap",
  dim1_to_use = 1,
  dim2\_to\_use = 2,
  dim3_to_use = 3,
  spat_enr_names = NULL,
  select_cell_groups = NULL,
  select_cells = NULL,
  show_other_cells = T,
  other_cell_color = "lightgrey",
  other_point_size = 2,
  show_NN_network = F,
  nn_network_to_use = "sNN",
  network_name = "sNN.pca",
  color_as_factor = T,
  cell_color = NULL,
  cell_color_code = NULL,
  show_cluster_center = F,
  show_center_label = T,
  center_point_size = 4,
  label_size = 4,
  edge_alpha = NULL,
  point_size = 3,
  show_plot = NA,
```

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```
return_plot = NA,
      save_plot = NA,
      save_param = list(),
      default_save_name = "dim3D"
Arguments
    gobject
                      giotto object
    dim_reduction_to_use
                      dimension reduction to use
    dim_reduction_name
                      dimension reduction name
    dim1_to_use
                      dimension to use on x-axis
    dim2_to_use
                      dimension to use on y-axis
    dim3_to_use
                      dimension to use on z-axis
    spat_enr_names names of spatial enrichment results to include
    select_cell_groups
                      select subset of cells/clusters based on cell_color parameter
    select_cells
                      select subset of cells based on cell IDs
    show_other_cells
                      display not selected cells
    other_cell_color
                      color of not selected cells
    other_point_size
                      size of not selected cells
    show_NN_network
                      show underlying NN network
    nn_network_to_use
                      type of NN network to use (kNN vs sNN)
                      name of NN network to use, if show_NN_network = TRUE
    network_name
    color_as_factor
                      convert color column to factor
                      color for cells (see details)
    cell_color
    cell_color_code
                      named vector with colors
    show_cluster_center
                      plot center of selected clusters
    show_center_label
                      plot label of selected clusters
    center_point_size
                      size of center points
    label_size
                      size of labels
    edge_alpha
                      column to use for alpha of the edges
    point_size
                      size of point (cell)
    show_plot
                      show plot
```

return\_plot

return ggplot object

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#### **Details**

Description of parameters.

#### Value

plotly

#### See Also

```
Other reduced dimension visualizations: dimPlot2D(), dimPlot(), plotPCA_2D(), plotPCA_3D(), plotPCA(), plotTSNE_2D(), plotTSNE_3D(), plotUMAP_2D(), plotUMAP_3D(), plotUMAP()
```

doHclust

doHclust

### **Description**

cluster cells using hierarchical clustering algorithm

# Usage

```
doHclust(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  genes_to_use = NULL,
  dim_reduction_to_use = c("cells", "pca", "umap", "tsne"),
  dim_reduction_name = "pca",
  dimensions_to_use = 1:10,
  distance_method = c("pearson", "spearman", "original", "euclidean", "maximum",
    "manhattan", "canberra", "binary", "minkowski"),
  agglomeration_method = c("ward.D2", "ward.D", "single", "complete", "average",
    "mcquitty", "median", "centroid"),
  k = 10,
  h = NULL,
  name = "hclust",
  return_gobject = TRUE,
  set_seed = T,
  seed_number = 1234
)
```

doHclust 93

### **Arguments**

```
gobject
                 giotto object
expression_values
                 expression values to use
genes_to_use
                 subset of genes to use
dim_reduction_to_use
                 dimension reduction to use
dim_reduction_name
                 dimensions reduction name
dimensions_to_use
                 dimensions to use
distance_method
                 distance method
agglomeration_method
                 agglomeration method for hclust
                 number of final clusters
                 cut hierarchical tree at height = h
h
name
                 name for hierarchical clustering
return_gobject boolean: return giotto object (default = TRUE)
set_seed
                 set seed
seed_number
                 number for seed
```

### **Details**

Description on how to use Kmeans clustering method.

### Value

giotto object with new clusters appended to cell metadata

#### See Also

hclust

### **Examples**

```
data(mini_giotto_single_cell)
mini_giotto_single_cell = doHclust(mini_giotto_single_cell, k = 4, name = 'hier_clus')
plotUMAP_2D(mini_giotto_single_cell, cell_color = 'hier_clus', point_size = 3)
```

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doHMRF

doHMRF

### **Description**

Run HMRF

### Usage

```
doHMRF(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  spatial_network_name = "Delaunay_network",
  spatial_genes = NULL,
  spatial_dimensions = c("sdimx", "sdimy", "sdimz"),
  dim_reduction_to_use = NULL,
  dim_reduction_name = "pca",
  dimensions_to_use = 1:10,
  name = "test",
  k = 10,
  betas = c(0, 2, 50),
  tolerance = 1e-10,
  zscore = c("none", "rowcol", "colrow"),
  numinit = 100,
  python_path = NULL,
  output_folder = NULL,
  overwrite_output = TRUE
)
```

### **Arguments**

```
giotto object
gobject
expression_values
                 expression values to use
spatial_network_name
                 name of spatial network to use for HMRF
                 spatial genes to use for HMRF
spatial_genes
spatial_dimensions
                 select spatial dimensions to use, default is all possible dimensions
dim_reduction_to_use
                 use another dimension reduction set as input
dim_reduction_name
                 name of dimension reduction set to use
dimensions_to_use
                 number of dimensions to use as input
                 name of HMRF run
name
                 number of HMRF domains
k
                 betas to test for
betas
tolerance
                 tolerance
```

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```
zscore zscore
numinit number of initializations

python_path python path to use

output_folder output folder to save results

overwrite_output

overwrite output folder
```

#### **Details**

Description of HMRF parameters ...

#### Value

Creates a directory with results that can be viewed with viewHMRFresults

doKmeans

doKmeans

### **Description**

cluster cells using kmeans algorithm

# Usage

```
doKmeans(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  genes_to_use = NULL,
  dim_reduction_to_use = c("cells", "pca", "umap", "tsne"),
  dim_reduction_name = "pca",
  dimensions_to_use = 1:10,
  distance_method = c("original", "pearson", "spearman", "euclidean", "maximum",
    "manhattan", "canberra", "binary", "minkowski"),
  centers = 10,
  iter_max = 100,
  nstart = 1000,
  algorithm = "Hartigan-Wong",
  name = "kmeans",
  return_gobject = TRUE,
  set\_seed = T,
  seed_number = 1234
)
```

# **Arguments**

```
gobject giotto object
expression_values
expression values to use
genes_to_use subset of genes to use
```

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dim\_reduction\_to\_use

dimension reduction to use

dim\_reduction\_name

dimensions reduction name

dimensions\_to\_use

dimensions to use

distance\_method

distance method

centers number of final clusters

iter\_max kmeans maximum iterations

nstart kmeans nstart

algorithm kmeans algorithm

name name for kmeans clustering

return\_gobject boolean: return giotto object (default = TRUE)

set\_seed set seed

seed\_number number for seed

### **Details**

Description on how to use Kmeans clustering method.

# Value

giotto object with new clusters appended to cell metadata

# See Also

kmeans

### **Examples**

```
data(mini_giotto_single_cell)
mini_giotto_single_cell = doKmeans(mini_giotto_single_cell, centers = 4, name = 'kmeans_clus')
plotUMAP_2D(mini_giotto_single_cell, cell_color = 'kmeans_clus', point_size = 3)
```

doLeidenCluster

doLeidenCluster

# **Description**

cluster cells using a NN-network and the Leiden community detection algorithm

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#### **Usage**

```
doLeidenCluster(
  gobject,
  name = "leiden_clus",
  nn_network_to_use = "sNN",
  network_name = "sNN.pca",
  python_path = NULL,
  resolution = 1,
  weight_col = "weight",
  partition_type = c("RBConfigurationVertexPartition", "ModularityVertexPartition"),
  init_membership = NULL,
  n_iterations = 1000,
  return_gobject = TRUE,
  set_seed = T,
  seed_number = 1234
)
```

### **Arguments**

gobject giotto object name for cluster name nn\_network\_to\_use type of NN network to use (kNN vs sNN) name of NN network to use network\_name specify specific path to python if required python\_path resolution resolution weight\_col weight column to use for edges partition\_type The type of partition to use for optimisation. init\_membership initial membership of cells for the partition number of interations to run the Leiden algorithm. If the number of iterations n\_iterations is negative, the Leiden algorithm is run until an iteration in which there was no improvement. return\_gobject boolean: return giotto object (default = TRUE) set\_seed set seed number for seed seed\_number

#### **Details**

This function is a wrapper for the Leiden algorithm implemented in python, which can detect communities in graphs of millions of nodes (cells), as long as they can fit in memory. See the <a href="https://github.com/vtraag/leidenalgleidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalgleidenalggithub.com/vtraag/leidenalggithub.com/vtra

Partition types available and information:

• RBConfigurationVertexPartition: Implements Reichardt and Bornholdt's Potts model with a configuration null model. This quality function is well-defined only for positive edge weights. This quality function uses a linear resolution parameter.

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• Modularity Vertex Partition: Implements modularity. This quality function is well-defined only for positive edge weights. It does *not* use the resolution parameter

Set  $weight\_col = NULL$  to give equal weight (=1) to each edge.

#### Value

giotto object with new clusters appended to cell metadata

doLeidenSubCluster doLeidenSubCluster

## **Description**

Further subcluster cells using a NN-network and the Leiden algorithm

### Usage

```
doLeidenSubCluster(
  gobject,
  name = "sub_pleiden_clus",
  cluster_column = NULL,
  selected_clusters = NULL,
 hvg_param = list(reverse_log_scale = T, difference_in_cov = 1, expression_values =
    "normalized"),
  hvg_min_perc_cells = 5,
  hvg_mean_expr_det = 1,
  use_all_genes_as_hvg = FALSE,
  min_nr_of_hvg = 5,
  pca_param = list(expression_values = "normalized", scale_unit = T),
  nn_param = list(dimensions_to_use = 1:20),
  k_neighbors = 10,
  resolution = 0.5,
  n_{iterations} = 500,
  python_path = NULL,
  nn_network_to_use = "sNN",
  network_name = "sNN.pca",
  return_gobject = TRUE,
  verbose = T
)
```

#### **Arguments**

```
gobject giotto object

name name for new clustering result

cluster_column cluster column to subcluster

selected_clusters

only do subclustering on these clusters

hvg_param parameters for calculateHVG

hvg_min_perc_cells

threshold for detection in min percentage of cells
```

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hvg\_mean\_expr\_det

threshold for mean expression level in cells with detection

use\_all\_genes\_as\_hvg

forces all genes to be HVG and to be used as input for PCA

min\_nr\_of\_hvg minimum number of HVG, or all genes will be used as input for PCA

pca\_param parameters for runPCA

nn\_param parameters for parameters for createNearestNetwork

k\_neighbors number of k for createNearestNetwork

resolution resolution of Leiden clustering

n\_iterations number of interations to run the Leiden algorithm.

python\_path specify specific path to python if required

nn\_network\_to\_use

type of NN network to use (kNN vs sNN)

 ${\tt network\_name} \quad \quad name \ of \ NN \ network \ to \ use$ 

return\_gobject boolean: return giotto object (default = TRUE)

verbose verbose

#### **Details**

This function performs subclustering using the Leiden algorithm on selected clusters. The systematic steps are:

- 1. subset Giotto object
- 2. identify highly variable genes
- 3. run PCA
- 4. create nearest neighbouring network
- 5. do Leiden clustering

#### Value

giotto object with new subclusters appended to cell metadata

### See Also

doLeidenCluster

doLouvainCluster doLouvainCluster

# Description

cluster cells using a NN-network and the Louvain algorithm.

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#### Usage

```
doLouvainCluster(
 gobject,
 version = c("community", "multinet"),
 name = "louvain_clus",
 nn_network_to_use = "sNN",
 network_name = "sNN.pca",
 python_path = NULL,
  resolution = 1,
 weight_col = NULL,
 gamma = 1,
 omega = 1,
  louv_random = F,
  return_gobject = TRUE,
  set_seed = F,
 seed_number = 1234,
)
```

### **Arguments**

gobject giotto object version implemented version of Louvain clustering to use name name for cluster nn\_network\_to\_use type of NN network to use (kNN vs sNN) name of NN network to use network\_name [community] specify specific path to python if required python\_path resolution [community] resolution  $weight\_col$ weight column name gamma [multinet] Resolution parameter for modularity in the generalized louvain method. [multinet] Inter-layer weight parameter in the generalized louvain method omega louv\_random [community] Will randomize the node evaluation order and the community evaluation order to get different partitions at each call return\_gobject boolean: return giotto object (default = TRUE) set\_seed set seed seed\_number number for seed additional parameters

## **Details**

Louvain clustering using the community or multinet implementation of the louvain clustering algorithm.

## Value

giotto object with new clusters appended to cell metadata

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#### See Also

doLouvainCluster\_community and doLouvainCluster\_multinet

doLouvainSubCluster doLouvainSubCluster

#### **Description**

subcluster cells using a NN-network and the Louvain algorithm

### Usage

```
doLouvainSubCluster(
  gobject,
  name = "sub_louvain_clus",
  version = c("community", "multinet"),
  cluster_column = NULL,
  selected_clusters = NULL,
 hvg_param = list(reverse_log_scale = T, difference_in_cov = 1, expression_values =
    "normalized"),
  hvg_min_perc_cells = 5,
  hvg_mean_expr_det = 1,
  use_all_genes_as_hvg = FALSE,
  min_nr_of_hvg = 5,
  pca_param = list(expression_values = "normalized", scale_unit = T),
  nn_param = list(dimensions_to_use = 1:20),
  k_neighbors = 10,
  resolution = 0.5,
  gamma = 1,
  omega = 1,
  python_path = NULL,
  nn_network_to_use = "sNN",
  network_name = "sNN.pca",
  return_gobject = TRUE,
  verbose = T
)
```

## **Arguments**

```
gobject giotto object

name name for new clustering result

version version of Louvain algorithm to use

cluster_column cluster column to subcluster

selected_clusters

only do subclustering on these clusters

hvg_param parameters for calculateHVG

hvg_min_perc_cells

threshold for detection in min percentage of cells
```

102 doLouvainSubCluster

hvg\_mean\_expr\_det

threshold for mean expression level in cells with detection

use\_all\_genes\_as\_hvg

forces all genes to be HVG and to be used as input for PCA

min\_nr\_of\_hvg minimum number of HVG, or all genes will be used as input for PCA

pca\_param parameters for runPCA

nn\_param parameters for parameters for createNearestNetwork

 $k\_neighbors \qquad number \ of \ k \ for \ createNearestNetwork$ 

resolution resolution for community algorithm

gamma gamma omega

python\_path specify specific path to python if required

nn\_network\_to\_use

type of NN network to use (kNN vs sNN)

network\_name name of NN network to use

return\_gobject boolean: return giotto object (default = TRUE)

verbose verbose

#### **Details**

This function performs subclustering using the Louvain algorithm on selected clusters. The systematic steps are:

- 1. subset Giotto object
- 2. identify highly variable genes
- 3. run PCA
- 4. create nearest neighbouring network
- 5. do Louvain clustering

#### Value

giotto object with new subclusters appended to cell metadata

## See Also

doLouvainCluster\_multinet and doLouvainCluster\_community

doRandomWalkCluster 103

 $do Random Walk Cluster \qquad do Random Walk Cluster$ 

### **Description**

Cluster cells using a random walk approach.

### Usage

```
doRandomWalkCluster(
  gobject,
  name = "random_walk_clus",
  nn_network_to_use = "sNN",
  network_name = "sNN.pca",
  walk_steps = 4,
  walk_clusters = 10,
  walk_weights = NA,
  return_gobject = TRUE,
  set_seed = F,
  seed_number = 1234
)
```

### **Arguments**

```
giotto object
gobject
                 name for cluster
name
nn_network_to_use
                 type of NN network to use (kNN vs sNN)
                 name of NN network to use
network_name
walk_steps
                 number of walking steps
walk_clusters
                 number of final clusters
                 cluster column defining the walk weights
walk_weights
return_gobject boolean: return giotto object (default = TRUE)
set\_seed
                 set seed
seed_number
                 number for seed
```

#### **Details**

See cluster\_walktrap function from the igraph package in R for more information.

# Value

giotto object with new clusters appended to cell metadata

104 doSNNCluster

doSNNCluster doSNNCluster

# **Description**

Cluster cells using a SNN cluster approach.

### Usage

```
doSNNCluster(
  gobject,
  name = "sNN_clus",
  nn_network_to_use = "kNN",
  network_name = "kNN.pca",
  k = 20,
  eps = 4,
  minPts = 16,
  borderPoints = TRUE,
  return_gobject = TRUE,
  set_seed = F,
  seed_number = 1234
)
```

### **Arguments**

gobject giotto object name name for cluster

nn\_network\_to\_use

type of NN network to use (only works on kNN)

 ${\tt network\_name} \qquad {\tt name} \ of \ kNN \ network \ to \ use$ 

k Neighborhood size for nearest neighbor sparsification to create the shared NN

graph.

eps Two objects are only reachable from each other if they share at least eps nearest

neighbors.

minPts minimum number of points that share at least eps nearest neighbors for a point

to be considered a core points.

borderPoints should borderPoints be assigned to clusters like in DBSCAN?

return\_gobject boolean: return giotto object (default = TRUE)

set\_seed set seed

seed\_number number for seed

## **Details**

See sNNclust from dbscan package

### Value

giotto object with new clusters appended to cell metadata

estimateImageBg 105

estimateImageBg

estimateImageBg

# **Description**

helps to estimate which color is the background color of your plot

# Usage

```
estimateImageBg(mg_object, top_color_range = 1:50)
```

# **Arguments**

```
mg_object magick image or Giotto image object top_color_range top possible background colors to return
```

### Value

vector of pixel color frequencies and an associated barplot

exportGiottoViewer

exportGiottoViewer

### **Description**

compute highly variable genes

## Usage

```
exportGiottoViewer(
  gobject,
  output_directory = NULL,
  spat_enr_names = NULL,
  factor_annotations = NULL,
  numeric_annotations = NULL,
  dim_reductions,
  dim_reduction_names,
  expression_values = c("scaled", "normalized", "custom"),
  dim_red_rounding = NULL,
  dim_red_rescale = c(-20, 20),
  expression_rounding = 2,
  overwrite_dir = T,
  verbose = T
```

106 exportGiottoViewer

### **Arguments**

```
gobject
                  giotto object
output_directory
                  directory where to save the files
spat_enr_names spatial enrichment results to include for annotations
factor_annotations
                  giotto cell annotations to view as factor
numeric_annotations
                  giotto cell annotations to view as numeric
dim_reductions high level dimension reductions to view
dim_reduction_names
                  specific dimension reduction names
expression_values
                  expression values to use in Viewer
dim_red_rounding
                  numerical indicating how to round the coordinates
dim_red_rescale
                  numericals to rescale the coordinates
expression_rounding
                  numerical indicating how to round the expression data
                  overwrite files in the directory if it already existed
overwrite_dir
verbose
                  be verbose
```

## **Details**

Giotto Viewer expects the results from Giotto Analyzer in a specific format, which is provided by this function. To include enrichment results from createSpatialEnrich include the provided spatial enrichment name (default PAGE or rank) and add the gene signature names (.e.g cell types) to the numeric annotations parameter.

# Value

writes the necessary output to use in Giotto Viewer

### **Examples**

```
## Not run:
data(mini_giotto_single_cell)
exportGiottoViewer(mini_giotto_single_cell)
## End(Not run)
```

exprCellCellcom 107

exprCellCellcom exprCellCellcom

## Description

Cell-Cell communication scores based on expression only

#### Usage

### **Arguments**

```
giotto object to use
gobject
cluster_column cluster column with cell type information
random_iter
                  number of iterations
                  first specific gene set from gene pairs
gene_set_1
gene_set_2
                  second specific gene set from gene pairs
log2FC_addendum
                  addendum to add when calculating log2FC
detailed
                  provide more detailed information (random variance and z-score)
                  which method to adjust p-values
adjust_method
                  adjust multiple hypotheses at the cell or gene level
adjust_target
                  set seed for random simulations (default = TRUE)
set_seed
seed_number
                  seed number
verbose
                  verbose
```

### **Details**

Statistical framework to identify if pairs of genes (such as ligand-receptor combinations) are expressed at higher levels than expected based on a reshuffled null distribution of gene expression values, without considering the spatial position of cells. More details will follow soon.

# Value

Cell-Cell communication scores for gene pairs based on expression only

fDataDT

fDataDT

# Description

show gene metadata

### Usage

fDataDT(gobject)

### **Arguments**

gobject

giotto object

#### Value

data.table with gene metadata

#### **Examples**

```
data(mini_giotto_single_cell) # loads existing Giotto object
fDataDT(mini_giotto_single_cell)
```

filterCellProximityGenes

filter Cell Proximity Genes

### **Description**

Filter Interaction Changed Gene scores.

#### Usage

```
filterCellProximityGenes(...)
```

## **Arguments**

... Arguments passed on to findICG

gobject giotto object
expression\_values expression values to use
selected\_genes subset of selected genes (optional)
cluster\_column name of column to use for cell types
spatial\_network\_name name of spatial network to use
minimum\_unique\_cells minimum number of target cells required
minimum\_unique\_int\_cells minimum number of interacting cells required
diff\_test which differential expression test
mean\_method method to use to calculate the mean

filterCombinations 109

offset offset value to use when calculating log2 ratio
adjust\_method which method to adjust p-values
nr\_permutations number of permutations if diff\_test = permutation
exclude\_selected\_cells\_from\_test exclude interacting cells other cells
do\_parallel run calculations in parallel with mclapply
cores number of cores to use if do\_parallel = TRUE
set\_seed set a seed for reproducibility
seed\_number seed number

# See Also

findICG

filterCombinations

*filterCombinations* 

# **Description**

Shows how many genes and cells are lost with combinations of thresholds.

# Usage

```
filterCombinations(
 gobject,
  expression_values = c("raw", "normalized", "scaled", "custom"),
 expression_thresholds = c(1, 2),
 gene_det_in_min_cells = c(5, 50),
 min_det_genes_per_cell = c(200, 400),
  scale_x_axis = "identity",
 x_axis_offset = 0,
  scale_y_axis = "identity",
 y_axis_offset = 0,
  show_plot = TRUE,
 return_plot = FALSE,
  save_plot = NA,
 save_param = list(),
 default_save_name = "filterCombinations"
)
```

### **Arguments**

```
gobject giotto object

expression_values

expression values to use

expression_thresholds

all thresholds to consider a gene expressed

gene_det_in_min_cells

minimum number of cells that should express a gene to consider that gene further
```

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```
min_det_genes_per_cell
                  minimum number of expressed genes per cell to consider that cell further
                  ggplot transformation for x-axis (e.g. log2)
scale_x_axis
x_axis_offset
                  x-axis offset to be used together with the scaling transformation
scale_y_axis
                  ggplot transformation for y-axis (e.g. log2)
y_axis_offset
                  y-axis offset to be used together with the scaling transformation
show_plot
                  show plot
return_plot
                  return only ggplot object
                  directly save the plot [boolean]
save_plot
                  list of saving parameters from all_plots_save_function
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

### **Details**

Creates a scatterplot that visualizes the number of genes and cells that are lost with a specific combination of a gene and cell threshold given an arbitrary cutoff to call a gene expressed. This function can be used to make an informed decision at the filtering step with filterGiotto.

### Value

list of data.table and ggplot object

### **Examples**

```
data(mini_giotto_single_cell)
# assess the effect of multiple filter criteria
filterCombinations(mini_giotto_single_cell,
gene_det_in_min_cells = c(2, 4, 8),
min_det_genes_per_cell = c(5, 10, 20))
```

filterCPG filterCPG

# **Description**

Filter Interaction Changed Gene scores.

```
filterCPG(...)
```

filterDistributions 111

#### **Arguments**

```
... Arguments passed on to filterICG

cpgObject ICG (interaction changed gene) score object

min_cells minimum number of source cell type

min_cells_expr minimum expression level for source cell type

min_int_cells minimum number of interacting neighbor cell type

min_int_cells_expr minimum expression level for interacting neighbor cell

type

min_fdr minimum adjusted p-value

min_spat_diff minimum absolute spatial expression difference

min_log2_fc minimum log2 fold-change

min_zscore minimum z-score change

zscores_column calculate z-scores over cell types or genes

direction differential expression directions to keep
```

### See Also

filterICG

filterDistributions filterDistributions

# **Description**

show gene or cell distribution after filtering on expression threshold

```
filterDistributions(
 gobject,
 expression_values = c("raw", "normalized", "scaled", "custom"),
 expression_threshold = 1,
 detection = c("genes", "cells"),
 plot_type = c("histogram", "violin"),
 nr_bins = 30,
 fill_color = "lightblue",
  scale_axis = "identity",
 axis_offset = 0,
  show_plot = NA,
 return_plot = NA,
  save_plot = NA,
 save_param = list(),
 default_save_name = "filterDistributions"
)
```

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### **Arguments**

```
gobject
                  giotto object
expression_values
                  expression values to use
expression_threshold
                  threshold to consider a gene expressed
                  consider genes or cells
detection
plot_type
                  type of plot
nr_bins
                  number of bins for histogram plot
fill_color
                  fill color for plots
scale_axis
                  ggplot transformation for axis (e.g. log2)
axis_offset
                  offset to be used together with the scaling transformation
show_plot
                  show plot
return_plot
                  return ggplot object
                  directly save the plot [boolean]
save_plot
                  list of saving parameters from all_plots_save_function
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

### Value

ggplot object

# **Examples**

```
data(mini_giotto_single_cell)

# distribution plot of genes
filterDistributions(mini_giotto_single_cell, detection = 'genes')

# distribution plot of cells
filterDistributions(mini_giotto_single_cell, detection = 'cells')
```

filterGiotto

filterGiotto

# **Description**

filter Giotto object based on expression threshold

```
filterGiotto(
  gobject,
  expression_values = c("raw", "normalized", "scaled", "custom"),
  expression_threshold = 1,
   gene_det_in_min_cells = 100,
   min_det_genes_per_cell = 100,
  verbose = F
)
```

filterICG 113

# **Arguments**

```
gobject giotto object
expression_values
expression values to use
expression_threshold
threshold to consider a gene expressed
gene_det_in_min_cells
minimum # of cells that need to express a gene
min_det_genes_per_cell
minimum # of genes that need to be detected in a cell
verbose verbose
```

# **Details**

The function filterCombinations can be used to explore the effect of different parameter values.

# Value

giotto object

# **Examples**

filterICG

filterICG

# Description

Filter Interaction Changed Gene scores.

```
filterICG(
  cpgObject,
  min_cells = 4,
  min_cells_expr = 1,
  min_int_cells = 4,
  min_int_cells_expr = 1,
  min_fdr = 0.1,
  min_spat_diff = 0.2,
  min_log2_fc = 0.2,
  min_zscore = 2,
  zscores_column = c("cell_type", "genes"),
  direction = c("both", "up", "down")
)
```

# **Arguments**

```
cpgObject
                 ICG (interaction changed gene) score object
min_cells
                 minimum number of source cell type
min_cells_expr minimum expression level for source cell type
min_int_cells
                 minimum number of interacting neighbor cell type
min_int_cells_expr
                 minimum expression level for interacting neighbor cell type
min_fdr
                 minimum adjusted p-value
min_spat_diff
                 minimum absolute spatial expression difference
                 minimum log2 fold-change
min_log2_fc
min_zscore
                 minimum z-score change
zscores_column calculate z-scores over cell types or genes
direction
                 differential expression directions to keep
```

#### Value

cpgObject that contains the filtered differential gene scores

```
filterInteractionChangedGenes
```

filterInteractionChangedGenes

# **Description**

Filter Interaction Changed Gene scores.

```
filterInteractionChangedGenes(
  cpgObject,
  min_cells = 4,
  min_cells_expr = 1,
  min_int_cells = 4,
  min_int_cells_expr = 1,
  min_fdr = 0.1,
  min_spat_diff = 0.2,
  min_log2_fc = 0.2,
  min_zscore = 2,
  zscores_column = c("cell_type", "genes"),
  direction = c("both", "up", "down")
)
```

### **Arguments**

cpgObject ICG (interaction changed gene) score object min\_cells minimum number of source cell type min\_cells\_expr minimum expression level for source cell type min\_int\_cells minimum number of interacting neighbor cell type min\_int\_cells\_expr minimum expression level for interacting neighbor cell type min\_fdr minimum adjusted p-value minimum absolute spatial expression difference min\_spat\_diff minimum log2 fold-change min\_log2\_fc minimum z-score change min\_zscore zscores\_column calculate z-scores over cell types or genes direction differential expression directions to keep

### Value

cpgObject that contains the filtered differential gene scores

findCellProximityGenes

findCellProximityGenes

### **Description**

Identifies cell-to-cell Interaction Changed Genes (ICG), i.e. genes that are differentially expressed due to proximity to other cell types.

# Usage

```
findCellProximityGenes(...)
```

# **Arguments**

Arguments passed on to findInteractionChangedGenes
gobject giotto object
expression\_values expression values to use
selected\_genes subset of selected genes (optional)
cluster\_column name of column to use for cell types
spatial\_network\_name name of spatial network to use
minimum\_unique\_cells minimum number of target cells required
minimum\_unique\_int\_cells minimum number of interacting cells required
diff\_test which differential expression test
mean\_method method to use to calculate the mean
offset offset value to use when calculating log2 ratio
adjust\_method which method to adjust p-values
nr\_permutations number of permutations if diff\_test = permutation

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exclude\_selected\_cells\_from\_test exclude interacting cells other cells do\_parallel run calculations in parallel with mclapply cores number of cores to use if do\_parallel = TRUE set\_seed set a seed for reproducibility seed\_number seed number

### See Also

 ${\tt findInteractionChangedGenes}$ 

findCPG

findCPG

# Description

Identifies cell-to-cell Interaction Changed Genes (ICG), i.e. genes that are differentially expressed due to proximity to other cell types.

# Usage

findCPG(...)

### **Arguments**

... Arguments passed on to findICG

gobject giotto object expression\_values expression values to use selected\_genes subset of selected genes (optional) cluster\_column name of column to use for cell types spatial\_network\_name name of spatial network to use minimum\_unique\_cells minimum number of target cells required minimum\_unique\_int\_cells minimum number of interacting cells required diff\_test which differential expression test mean\_method method to use to calculate the mean offset offset value to use when calculating log2 ratio adjust\_method which method to adjust p-values nr\_permutations number of permutations if diff\_test = permutation exclude\_selected\_cells\_from\_test exclude interacting cells other cells do\_parallel run calculations in parallel with mclapply cores number of cores to use if do\_parallel = TRUE set\_seed set a seed for reproducibility seed\_number seed number

### See Also

findICG

findGiniMarkers 117

findGiniMarkers findGiniMarkers

**Description** 

Identify marker genes for selected clusters based on gini detection and expression scores.

# Usage

```
findGiniMarkers(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column,
  subset_clusters = NULL,
  group_1 = NULL,
  group_2 = NULL,
  min_expr_gini_score = 0.2,
  min_det_gini_score = 0.2,
  detection_threshold = 0,
  rank_score = 1,
  min_genes = 5
)
```

# **Arguments**

```
gobject
                  giotto object
expression_values
                  gene expression values to use
cluster_column clusters to use
subset_clusters
                  selection of clusters to compare
                  group 1 cluster IDs from cluster_column for pairwise comparison
group_1
group_2
                  group 2 cluster IDs from cluster_column for pairwise comparison
min_expr_gini_score
                  filter on minimum gini coefficient for expression
min_det_gini_score
                  filter on minimum gini coefficient for detection
detection_threshold
                  detection threshold for gene expression
                  rank scores for both detection and expression to include
rank_score
                  minimum number of top genes to return
min_genes
```

### **Details**

Detection of marker genes using the <a href="https://en.wikipedia.org/wiki/Gini\_coefficientgini">https://en.wikipedia.org/wiki/Gini\_coefficientgini</a> coefficient is based on the following steps/principles per gene:

- 1. calculate average expression per cluster
- 2. calculate detection fraction per cluster

- 3. calculate gini-coefficient for av. expression values over all clusters
- 4. calculate gini-coefficient for detection fractions over all clusters
- 5. convert gini-scores to rank scores
- 6. for each gene create combined score = detection rank x expression rank x expr gini-coefficient x detection gini-coefficient
- 7. for each gene sort on expression and detection rank and combined score

As a results "top gini" genes are genes that are very selectivily expressed in a specific cluster, however not always expressed in all cells of that cluster. In other words highly specific, but not necessarily sensitive at the single-cell level.

To perform differential expression between cluster groups you need to specificy cluster IDs to the parameters *group\_1* and *group\_2*.

### Value

data.table with marker genes

### **Examples**

```
find {\it GiniMarkers\_one\_vs\_all} \\ {\it find GiniMarkers\_one\_vs\_all}
```

# **Description**

Identify marker genes for all clusters in a one vs all manner based on gini detection and expression scores.

```
findGiniMarkers_one_vs_all(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column,
  subset_clusters = NULL,
  min_expr_gini_score = 0.5,
  min_det_gini_score = 0.5,
  detection_threshold = 0,
  rank_score = 1,
  min_genes = 4,
  verbose = TRUE
)
```

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### **Arguments**

```
gobject
                  giotto object
expression_values
                  gene expression values to use
cluster_column clusters to use
subset_clusters
                  selection of clusters to compare
min_expr_gini_score
                  filter on minimum gini coefficient on expression
min_det_gini_score
                  filter on minimum gini coefficient on detection
detection_threshold
                  detection threshold for gene expression
                  rank scores for both detection and expression to include
rank_score
                  minimum number of top genes to return
min_genes
verbose
                  be verbose
```

#### Value

data.table with marker genes

#### See Also

findGiniMarkers

### **Examples**

 ${\tt findICG} \hspace{1cm} \textit{findICG}$ 

# Description

Identifies cell-to-cell Interaction Changed Genes (ICG), i.e. genes that are differentially expressed due to proximity to other cell types.

```
findICG(
  gobject,
  expression_values = "normalized",
  selected_genes = NULL,
  cluster_column,
  spatial_network_name = "Delaunay_network",
```

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# Arguments

```
gobject
                  giotto object
expression_values
                  expression values to use
selected_genes subset of selected genes (optional)
cluster_column name of column to use for cell types
spatial_network_name
                  name of spatial network to use
minimum_unique_cells
                  minimum number of target cells required
minimum_unique_int_cells
                  minimum number of interacting cells required
diff_test
                  which differential expression test
mean_method
                  method to use to calculate the mean
offset
                  offset value to use when calculating log2 ratio
                  which method to adjust p-values
adjust_method
nr_permutations
                  number of permutations if diff_test = permutation
exclude_selected_cells_from_test
                  exclude interacting cells other cells
                  run calculations in parallel with mclapply
do_parallel
                  number of cores to use if do_parallel = TRUE
cores
                  set a seed for reproducibility
set_seed
seed_number
                  seed number
```

#### **Details**

Function to calculate if genes are differentially expressed in cell types when they interact (approximated by physical proximity) with other cell types. The results data.table in the cpgObject contains - at least - the following columns:

- genes: All or selected list of tested genes
- sel: average gene expression in the interacting cells from the target cell type

- other: average gene expression in the NOT-interacting cells from the target cell type
- log2fc: log2 fold-change between sel and other
- diff: spatial expression difference between sel and other
- p.value: associated p-value
- p.adj: adjusted p-value
- cell\_type: target cell type
- int\_cell\_type: interacting cell type
- nr select: number of cells for selected target cell type
- int\_nr\_select: number of cells for interacting cell type
- nr other: number of other cells of selected target cell type
- int\_nr\_other: number of other cells for interacting cell type
- unif\_int: cell-cell interaction

#### Value

cpgObject that contains the differential gene scores

findInteractionChangedGenes

findInteractionChangedGenes

# Description

Identifies cell-to-cell Interaction Changed Genes (ICG), i.e. genes that are differentially expressed due to proximity to other cell types.#'

```
findInteractionChangedGenes(
  gobject.
  expression_values = "normalized",
  selected_genes = NULL,
  cluster_column,
  spatial_network_name = "Delaunay_network",
 minimum_unique_cells = 1,
 minimum_unique_int_cells = 1,
 diff_test = c("permutation", "limma", "t.test", "wilcox"),
 mean_method = c("arithmic", "geometric"),
 offset = 0.1,
 adjust_method = c("bonferroni", "BH", "holm", "hochberg", "hommel", "BY", "fdr",
    "none"),
 nr_permutations = 1000,
 exclude_selected_cells_from_test = T,
 do_parallel = TRUE,
 cores = NA,
  set_seed = TRUE,
  seed_number = 1234
)
```

#### **Arguments**

gobject giotto object expression\_values expression values to use selected\_genes subset of selected genes (optional) cluster\_column name of column to use for cell types spatial\_network\_name name of spatial network to use minimum\_unique\_cells minimum number of target cells required minimum\_unique\_int\_cells minimum number of interacting cells required diff\_test which differential expression test mean\_method method to use to calculate the mean offset offset value to use when calculating log2 ratio adjust\_method which method to adjust p-values nr\_permutations number of permutations if diff\_test = permutation exclude\_selected\_cells\_from\_test exclude interacting cells other cells run calculations in parallel with mclapply do\_parallel number of cores to use if do\_parallel = TRUE cores set a seed for reproducibility set\_seed seed number seed\_number

# **Details**

Function to calculate if genes are differentially expressed in cell types when they interact (approximated by physical proximity) with other cell types. The results data.table in the cpgObject contains - at least - the following columns:

- genes: All or selected list of tested genes
- sel: average gene expression in the interacting cells from the target cell type
- other: average gene expression in the NOT-interacting cells from the target cell type
- log2fc: log2 fold-change between sel and other
- diff: spatial expression difference between sel and other
- p.value: associated p-value
- p.adj: adjusted p-value
- cell\_type: target cell type
- int\_cell\_type: interacting cell type
- nr\_select: number of cells for selected target cell type
- int\_nr\_select: number of cells for interacting cell type
- nr\_other: number of other cells of selected target cell type
- int\_nr\_other: number of other cells for interacting cell type
- unif\_int: cell-cell interaction

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#### Value

cpgObject that contains the Interaction Changed differential gene scores

findMarkers

findMarkers

# **Description**

Identify marker genes for selected clusters.

### Usage

```
findMarkers(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column = NULL,
  method = c("scran", "gini", "mast"),
  subset_clusters = NULL,
  group_1 = NULL,
  group_2 = NULL,
  min_expr_gini_score = 0.5,
  min_det_gini_score = 0.5,
  detection_threshold = 0,
  rank_score = 1,
  min\_genes = 4,
  group_1_name = NULL,
  group_2_name = NULL,
  adjust_columns = NULL,
)
```

# **Arguments**

```
gobject
                  giotto object
expression_values
                  gene expression values to use
cluster_column clusters to use
method
                  method to use to detect differentially expressed genes
subset_clusters
                  selection of clusters to compare
group_1
                  group 1 cluster IDs from cluster_column for pairwise comparison
                  group 2 cluster IDs from cluster_column for pairwise comparison
group_2
min_expr_gini_score
                  gini: filter on minimum gini coefficient for expression
min_det_gini_score
                  gini: filter minimum gini coefficient for detection
detection_threshold
                  gini: detection threshold for gene expression
```

```
rank_score gini: rank scores to include

min_genes minimum number of top genes to return (for gini)

group_1_name mast: custom name for group_1 clusters

group_2_name mast: custom name for group_2 clusters

adjust_columns mast: column in pDataDT to adjust for (e.g. detection rate)

... additional parameters for the findMarkers function in scran or zlm function in MAST
```

# **Details**

Wrapper for all individual functions to detect marker genes for clusters.

### Value

data.table with marker genes

### See Also

findScranMarkers, findGiniMarkers and findMastMarkers

```
findMarkers_one_vs_all findMarkers_one_vs_all
```

# Description

Identify marker genes for all clusters in a one vs all manner.

```
findMarkers_one_vs_all(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column,
  subset_clusters = NULL,
  method = c("scran", "gini", "mast"),
  pval = 0.01,
  logFC = 0.5,
  min_genes = 10,
  min_expr_gini_score = 0.5,
  min_det_gini_score = 0.5,
  detection_threshold = 0,
  rank_score = 1,
  adjust_columns = NULL,
  verbose = TRUE,
)
```

findMastMarkers 125

### **Arguments**

gobject giotto object

expression\_values

gene expression values to use

cluster\_column clusters to use

subset\_clusters

selection of clusters to compare

method method to use to detect differentially expressed genes

pval scran & mast: filter on minimal p-value

logFC scan & mast: filter on logFC

min\_genes minimum genes to keep per cluster, overrides pval and logFC

min\_expr\_gini\_score

gini: filter on minimum gini coefficient for expression

min\_det\_gini\_score

gini: filter minimum gini coefficient for detection

detection\_threshold

gini: detection threshold for gene expression

rank\_score gini: rank scores to include

 $adjust\_columns \quad mast: \ column \ in \ pDataDT \ to \ adjust \ for \ (e.g. \ detection \ rate)$ 

verbose be verbose

... additional parameters for the findMarkers function in scran or zlm function in

**MAST** 

# Details

Wrapper for all one vs all functions to detect marker genes for clusters.

# Value

data.table with marker genes

# See Also

 $find Scran Markers\_one\_vs\_all, find Gini Markers\_one\_vs\_all \ and \ find Mast Marke$ 

findMastMarkers findMastMarkers

# **Description**

Identify marker genes for selected clusters based on the MAST package.

126 findMastMarkers

#### Usage

```
findMastMarkers(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column,
  group_1 = NULL,
  group_1_name = NULL,
  group_2 = NULL,
  group_2_name = NULL,
  adjust_columns = NULL,
  verbose = FALSE,
  ...
)
```

### **Arguments**

```
gobject
                  giotto object
expression_values
                  gene expression values to use
cluster_column clusters to use
group_1
                  group 1 cluster IDs from cluster_column for pairwise comparison
group_1_name
                  custom name for group_1 clusters
                  group 2 cluster IDs from cluster_column for pairwise comparison
group_2
                  custom name for group_2 clusters
group_2_name
adjust_columns column in pDataDT to adjust for (e.g. detection rate)
                  be verbose
verbose
                  additional parameters for the zlm function in MAST
. . .
```

### **Details**

This is a minimal convenience wrapper around the zlm from the MAST package to detect differentially expressed genes. Caution: with large datasets MAST might take a long time to run and finish

#### Value

data.table with marker genes

# **Examples**

```
\label{lem:cone_vs_all} find \textit{MastMarkers\_one\_vs\_all} \\ find \textit{MastMarkers\_one\_vs\_all}
```

# Description

Identify marker genes for all clusters in a one vs all manner based on the MAST package.

# Usage

```
findMastMarkers_one_vs_all(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column,
  subset_clusters = NULL,
  adjust_columns = NULL,
  pval = 0.001,
  logFC = 1,
  min_genes = 10,
  verbose = TRUE,
  ...
)
```

# **Arguments**

```
gobject
                  giotto object
expression_values
                  gene expression values to use
cluster_column clusters to use
subset_clusters
                  selection of clusters to compare
adjust_columns column in pDataDT to adjust for (e.g. detection rate)
                  filter on minimal p-value
pval
logFC
                  filter on logFC
                  minimum genes to keep per cluster, overrides pval and logFC
min_genes
verbose
                  be verbose
                  additional parameters for the zlm function in MAST
```

# Value

data.table with marker genes

# See Also

findMastMarkers

### **Examples**

findNetworkNeighbors findNetworkNeighbors

# **Description**

Find the spatial neighbors for a selected group of cells within the selected spatial network.

# Usage

```
findNetworkNeighbors(
  gobject,
  spatial_network_name,
  source_cell_ids = NULL,
  name = "nb_cells"
)
```

# **Arguments**

#### Value

data.table

# **Examples**

findScranMarkers 129

findScranMarkers findScranMarkers

# **Description**

Identify marker genes for all or selected clusters based on scran's implementation of findMarkers.

### Usage

```
findScranMarkers(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column,
  subset_clusters = NULL,
  group_1 = NULL,
  group_2 = NULL,
  verbose = FALSE,
  ...
)
```

### **Arguments**

```
gobject giotto object
expression_values
gene expression values to use

cluster_column clusters to use
subset_clusters
selection of clusters to compare
group_1 group 1 cluster IDs from cluster_column for pairwise comparison
group_2 group 2 cluster IDs from cluster_column for pairwise comparison
verbose be verbose (default = FALSE)
... additional parameters for the findMarkers function in scran
```

### **Details**

This is a minimal convenience wrapper around the findMarkers function from the scran package. To perform differential expression between cluster groups you need to specificy cluster IDs to the

#### Value

data.table with marker genes

parameters group\_1 and group\_2.

### **Examples**

```
group_2 = 2)
```

```
\label{lem:findScranMarkers_one_vs_all} findScranMarkers\_one\_vs\_all
```

# Description

Identify marker genes for all clusters in a one vs all manner based on scran's implementation of findMarkers.

# Usage

```
findScranMarkers_one_vs_all(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column,
  subset_clusters = NULL,
  pval = 0.01,
  logFC = 0.5,
  min_genes = 10,
  verbose = TRUE,
  ...
)
```

# **Arguments**

```
gobject
                  giotto object
expression_values
                  gene expression values to use
cluster_column clusters to use
subset_clusters
                  subset of clusters to use
                  filter on minimal p-value
pval
logFC
                  filter on logFC
min_genes
                  minimum genes to keep per cluster, overrides pval and logFC
verbose
                  be verbose
                  additional parameters for the findMarkers function in scran
```

# Value

data.table with marker genes

# See Also

findScranMarkers

get10Xmatrix 131

# **Examples**

get10Xmatrix

get10Xmatrix

# **Description**

This function creates an expression matrix from a 10X structured folder

# Usage

```
get10Xmatrix(path_to_data, gene_column_index = 1)
```

# **Arguments**

```
path_to_data path to the 10X folder gene_column_index which column from the features or genes .tsv file to use for row ids
```

# **Details**

A typical 10X folder is named raw\_feature\_bc\_matrix or raw\_feature\_bc\_matrix and tt has 3 files:

- barcodes.tsv(.gz)
- features.tsv(.gz) or genes.tsv(.gz)
- matrix.mtx(.gz)

By default the first column of the features or genes .tsv file will be used, however if multiple annotations are provided (e.g. ensembl gene ids and gene symbols) the user can select another column.

### Value

sparse expression matrix from 10X

132 getClusterSimilarity

```
getClusterSimilarity
getClusterSimilarity
```

# Description

Creates data.table with pairwise correlation scores between each cluster.

# Usage

```
getClusterSimilarity(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column,
  cor = c("pearson", "spearman")
)
```

# **Arguments**

```
gobject giotto object
expression_values
expression values to use

cluster_column name of column to use for clusters

cor correlation score to calculate distance
```

### **Details**

Creates data.table with pairwise correlation scores between each cluster and the group size (# of cells) for each cluster. This information can be used together with mergeClusters to combine very similar or small clusters into bigger clusters.

### Value

data.table

# **Examples**

getDendrogramSplits 133

```
{\tt getDendrogramSplits} \qquad {\tt getDendrogramSplits}
```

# Description

Split dendrogram at each node and keep the leave (label) information..

# Usage

```
getDendrogramSplits(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column,
  cor = c("pearson", "spearman"),
  distance = "ward.D",
  h = NULL,
  h_color = "red",
  show_dend = TRUE,
  verbose = TRUE
)
```

### **Arguments**

```
gobject
                  giotto object
expression_values
                  expression values to use
cluster_column name of column to use for clusters
                  correlation score to calculate distance
cor
distance
                  distance method to use for hierarchical clustering
                  height of horizontal lines to plot
h_color
                  color of horizontal lines
show_dend
                  show dendrogram
verbose
                  be verbose
```

#### Details

Creates a data.table with three columns and each row represents a node in the dendrogram. For each node the height of the node is given together with the two subdendrograms. This information can be used to determine in a hierarchical manner differentially expressed marker genes at each node.

### Value

data.table object

### **Examples**

```
data("mini_giotto_single_cell")
splits = getDendrogramSplits(mini_giotto_single_cell, cluster_column = 'leiden_clus')
```

134 getGiottoImage

 ${\tt getDistinctColors}$ 

 ${\it getDistinctColors}$ 

# Description

Returns a number of distint colors based on the RGB scale

# Usage

```
getDistinctColors(n)
```

# Arguments

n

number of colors wanted

# Value

number of distinct colors

getGiottoImage

getGiottoImage

# Description

```
get get a giotto image from a giotto object
```

# Usage

```
getGiottoImage(gobject, image_name)
```

# Arguments

gobject giotto object

# Value

a giotto image

getSpatialDataset 135

getSpatialDataset getSpatialDataset

### **Description**

This package will automatically download the spatial locations and expression matrix for the chosen dataset. These files are already in the right format to create a Giotto object. If wget is installed on your machine, you can add 'method = wget' to the parameters to download files faster.

### Usage

```
getSpatialDataset(
  dataset = c("ST_OB1", "ST_OB2", "codex_spleen", "cycif_PDAC", "starmap_3D_cortex",
        "osmfish_SS_cortex", "merfish_preoptic", "seqfish_SS_cortex", "seqfish_OB",
        "slideseq_cerebellum"),
        directory = getwd(),
        ...
)
```

# Arguments

dataset dataset to download
directory directory to save the data to
... additional parameters to download.file

giotto-class

S4 giotto Class

# **Description**

Framework of giotto object to store and work with spatial expression data

### **Slots**

```
raw_exprs raw expression counts

norm_expr normalized expression counts

norm_scaled_expr normalized and scaled expression counts

custom_expr custom normalized counts

spatial_locs spatial location coordinates for cells

cell_metadata metadata for cells

gene_metadata metadata for genes

cell_ID unique cell IDs

gene_ID unique gene IDs

spatial_network spatial network in data.table/data.frame format

spatial_grid spatial grid in data.table/data.frame format
```

```
spatial_enrichment slot to save spatial enrichment-like results dimension_reduction slot to save dimension reduction coordinates nn_network nearest neighbor network in igraph format images slot to store giotto images parameters slot to save parameters that have been used instructions slot for global function instructions offset_file offset file used to stitch together image fields OS_platform Operating System to run Giotto analysis on
```

heatmSpatialCorGenes heatmSpatialCorGenes

# **Description**

Create heatmap of spatially correlated genes

# Usage

```
heatmSpatialCorGenes(
  gobject,
  spatCorObject,
  use_clus_name = NULL,
  show_cluster_annot = TRUE,
  show_row_dend = T,
  show_column_dend = F,
  show_row_names = F,
  show_column_names = F,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "heatmSpatialCorGenes",
  ...
)
```

# **Arguments**

hyperGeometricEnrich 137

show\_plot show plot

return\_plot return ggplot object

save\_plot directly save the plot [boolean]

save\_param list of saving parameters, see showSaveParameters

default\_save\_name

default save name for saving, don't change, change save\_name in save\_param

... additional parameters to the Heatmap function from ComplexHeatmap

#### Value

Heatmap generated by ComplexHeatmap

hyperGeometricEnrich hyperGeometricEnrich

### **Description**

Function to calculate gene signature enrichment scores per spatial position using a hypergeometric test.

### Usage

```
hyperGeometricEnrich(...)
```

### **Arguments**

... Arguments passed on to runHyperGeometricEnrich

gobject Giotto object

sign\_matrix Matrix of signature genes for each cell type / process

expression\_values expression values to use

reverse\_log\_scale reverse expression values from log scale

logbase log base to use if reverse\_log\_scale = TRUE

top\_percentage percentage of cells that will be considered to have gene ex-

pression with matrix binarization

output\_enrichment how to return enrichment output

p\_value calculate p-values (boolean, default = FALSE)

name to give to spatial enrichment results, default = rank

return\_gobject return giotto object

#### See Also

runHyperGeometricEnrich

```
insert {\tt CrossSectionGenePlot3D} \\ insert {\tt CrossSectionGenePlot3D}
```

# **Description**

Visualize cells and gene expression in a virtual cross section according to spatial coordinates

# Usage

```
insertCrossSectionGenePlot3D(
 gobject,
 crossSection_obj = NULL,
 name = NULL,
 spatial_network_name = "Delaunay_network",
 mesh_grid_color = "#1f77b4",
 mesh_grid_width = 3,
 mesh_grid_style = "dot",
 sdimx = "sdimx",
  sdimy = "sdimy",
  sdimz = "sdimz",
  show_other_cells = F,
 axis_scale = c("cube", "real", "custom"),
  custom_ratio = NULL,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
 default_save_name = "spatGenePlot3D_with_cross_section",
)
```

### **Arguments**

```
gobject
                  giotto object
crossSection_obj
                  cross section object as alternative input. default = NULL.
                  name of virtual cross section to use
name
spatial_network_name
                  name of spatial network to use
mesh_grid_color
                  color for the meshgrid lines
mesh_grid_width
                  width for the meshgrid lines
mesh_grid_style
                  style for the meshgrid lines
                  x-axis dimension name (default = 'sdimx')
sdimx
sdimy
                  y-axis dimension name (default = 'sdimy')
sdimz
                  z-axis dimension name (default = 'sdimy')
```

```
show_other_cells
                  display not selected cells
axis_scale
                  axis_scale
custom_ratio
                  custom_ratio
show_plot
                  show plots
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
                  list of saving parameters from all_plots_save_function
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
                  parameters for spatGenePlot3D
. . .
```

### **Details**

Description of parameters.

#### Value

ggplot

```
insert {\tt CrossSectionSpatPlot3D} \\ insert {\tt CrossSectionSpatPlot3D}
```

# **Description**

Visualize the meshgrid lines of cross section together with cells

```
insertCrossSectionSpatPlot3D(
  gobject,
  crossSection_obj = NULL,
  name = NULL,
  spatial_network_name = "Delaunay_network",
  mesh_grid_color = "#1f77b4",
  mesh_grid_width = 3,
  mesh_grid_style = "dot",
  sdimx = "sdimx",
  sdimy = "sdimy",
  sdimz = "sdimz",
  show_other_cells = F,
  axis_scale = c("cube", "real", "custom"),
  custom_ratio = NULL,
  default_save_name = "spat3D_with_cross_section",
)
```

140 installGiottoEnvironment

### **Arguments**

```
gobject
                  giotto object
crossSection_obj
                  cross section object as alternative input. default = NULL.
                  name of virtual cross section to use
name
spatial_network_name
                  name of spatial network to use
mesh_grid_color
                  color for the meshgrid lines
mesh_grid_width
                  width for the meshgrid lines
mesh_grid_style
                  style for the meshgrid lines
sdimx
                  x-axis dimension name (default = 'sdimx')
                  y-axis dimension name (default = 'sdimy')
sdimy
sdimz
                  z-axis dimension name (default = 'sdimy')
show_other_cells
                  display not selected cells
axis_scale
                  axis_scale
custom_ratio
                  custom ratio
default_save_name
                  default save name for saving, don't change, change save_name in save_param
                  parameters for spatPlot3D
. . .
```

#### **Details**

Description of parameters.

# Value

ggplot

installGiottoEnvironment

install Giot to Environment

# Description

Installs a giotto environment

jackstrawPlot 141

#### **Arguments**

#### **Details**

This function will install a local giotto environment using the miniconda system as implemented by reticulate. Once this giotto environment is installed it will be automatically detected when you run the Giotto toolbox. If you want to use your own python path then you can set the python\_path in the createGiottoInstructions and provide the instructions to the createGiottoObject function.

### Value

installs a giotto environment using the reticulate miniconda system

# **Examples**

```
## Not run:

# this command will install r-miniconda
# and a giotto environment with all necessary python modules
installGiottoEnvironment()

## End(Not run)
```

jackstrawPlot

jackstrawPlot

# Description

identify significant prinicipal components (PCs)

```
jackstrawPlot(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  reduction = c("cells", "genes"),
  genes_to_use = NULL,
  center = FALSE,
  scale_unit = FALSE,
  ncp = 20,
  ylim = c(0, 1),
  iter = 10,
  threshold = 0.01,
```

142 jackstrawPlot

```
verbose = TRUE,
show_plot = NA,
return_plot = NA,
save_plot = NA,
save_param = list(),
default_save_name = "jackstrawPlot"
)
```

# **Arguments**

gobject giotto object
expression\_values

expression values to use

reduction cells or genes

genes\_to\_use subset of genes to use for PCA

center center data before PCA scale\_unit scale features before PCA

ncp number of principal components to calculate

ylim y-axis limits on jackstraw plot
iter number of interations for jackstraw
threshold p-value threshold to call a PC significant
verbose show progress of jackstraw method

show\_plot show plot

return\_plot return ggplot object

save\_plot directly save the plot [boolean]

save\_param list of saving parameters from all\_plots\_save\_function()

default\_save\_name

default save name for saving, don't change, change save\_name in save\_param

### **Details**

The Jackstraw method uses the permutationPA function. By systematically permuting genes it identifies robust, and thus significant, PCs.

#### Value

ggplot object for jackstraw method

# **Examples**

```
data(mini_giotto_single_cell)
# jackstraw package is required to run
jackstrawPlot(mini_giotto_single_cell, ncp = 10)
```

loadHMRF 143

loadHMRF

loadHMRF

# **Description**

load previous HMRF

# Usage

```
loadHMRF(
  name_used = "test",
  output_folder_used,
  k_used = 10,
  betas_used,
  python_path_used
)
```

# **Arguments**

```
name_used name of HMRF that was run
output_folder_used
output folder that was used
k_used number of HMRF domains that was tested
betas_used betas that were tested
python_path_used
python path that was used
```

# **Details**

Description of HMRF parameters ...

# Value

reloads a previous ran HMRF from doHRMF

makeSignMatrixPAGE

makeSignMatrixPAGE

# Description

Function to convert a list of signature genes (e.g. for cell types or processes) into a binary matrix format that can be used with the PAGE enrichment option. Each cell type or process should have a vector of cell-type or process specific genes. These vectors need to be combined into a list (sign\_list). The names of the cell types or processes that are provided in the list need to be given (sign\_names).

```
makeSignMatrixPAGE(sign_names, sign_list)
```

144 makeSignMatrixRank

# **Arguments**

```
sign_names vector with names for each provided gene signature sign_list list of genes (signature)
```

### Value

matrix

### See Also

**PAGEEnrich** 

 ${\it make Sign Matrix Rank}$ 

make Sign Matrix Rank

# Description

Function to convert a single-cell count matrix and a corresponding single-cell cluster vector into a rank matrix that can be used with the Rank enrichment option.

# Usage

```
makeSignMatrixRank(
   sc_matrix,
   sc_cluster_ids,
   ties_method = c("random", "max"),
   gobject = NULL
)
```

# **Arguments**

sc\_matrix matrix of single-cell RNAseq expression data

sc\_cluster\_ids vector of cluster ids
ties\_method how to handle rank ties

gobject if giotto object is given then only genes present in both datasets will be consid-

ered

# Value

matrix

# See Also

rankEnrich

mean\_giotto 145

 $mean\_giotto$ 

 $mean\_giotto$ 

# Description

mean function that works with multiple matrix representations

# Usage

```
mean_giotto(x, ...)
```

# **Arguments**

x vector

... additional parameters

## Value

numeric

mergeClusters

mergeClusters

# Description

Merge selected clusters based on pairwise correlation scores and size of cluster.

```
mergeClusters(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column,
  cor = c("pearson", "spearman"),
  new_cluster_name = "merged_cluster",
  min_cor_score = 0.8,
  max_group_size = 20,
  force_min_group_size = 10,
  max_sim_clusters = 10,
  return_gobject = TRUE,
  verbose = TRUE
)
```

146 mergeClusters

#### **Arguments**

```
gobject
                  giotto object
expression_values
                  expression values to use
cluster_column name of column to use for clusters
                  correlation score to calculate distance
new_cluster_name
                  new name for merged clusters
                  min correlation score to merge pairwise clusters
min_cor_score
max_group_size max cluster size that can be merged
force_min_group_size
                  size of clusters that will be merged with their most similar neighbor(s)
max_sim_clusters
                  maximum number of clusters to potentially merge to reach force_min_group_size
return_gobject return giotto object
                  be verbose
verbose
```

### **Details**

Merge selected clusters based on pairwise correlation scores and size of cluster. To avoid large clusters to merge the max\_group\_size can be lowered. Small clusters can be forcibly merged with their most similar pairwise cluster by adjusting the force\_min\_group\_size parameter. Clusters smaller than this value will be merged independent on the provided min\_cor\_score value. The force\_min\_group\_size might not always be reached if clusters have already been merged before A giotto object is returned by default, if FALSE then the merging vector will be returned.

### Value

Giotto object

# Examples

mini\_giotto\_3D 147

mini\_giotto\_3D

mini Giotto object for spatial single-cell 3D data

## **Description**

Mini Giotto object created from the STARmap data.

# Usage

```
data(mini_giotto_3D)
```

### **Format**

An object of class "giotto"; see createGiottoObject.

## References

```
Wang et al. (2018) Science (PubMed)
```

## **Examples**

```
data(mini_giotto_3D)
## Not run: spatPlot3D(mini_giotto_3D, cell_color = 'cell_types', point_size = 5)
```

```
mini_giotto_multi_cell
```

mini Giotto object for spatial multi-cell resolution data

### **Description**

Mini Giotto object created from the Brain Visium 10X data.

## Usage

```
data(mini_giotto_multi_cell)
```

## **Format**

An object of class "giotto"; see createGiottoObject.

### References

10 Genomics Visium technology (10xgenomics)

# **Examples**

```
data(mini_giotto_multi_cell)
## Not run: spatPlot(mini_giotto_multi_cell, cell_color = 'cell_types', point_size = 5)
```

148 normalizeGiotto

```
mini_giotto_single_cell
```

mini Giotto object for spatial single-cell resolution data

# Description

Mini Giotto object created from the seqFISH+ data.

## Usage

```
data(mini_giotto_single_cell)
```

### **Format**

An object of class "giotto"; see createGiottoObject.

### References

```
Eng et al. (2019) Nature (PubMed)
```

# **Examples**

```
data(mini_giotto_single_cell)
## Not run: spatPlot2D(mini_giotto_single_cell,cell_color = 'cell_types', point_size = 5)
```

normalizeGiotto

normalizeGiotto

### **Description**

fast normalize and/or scale expresion values of Giotto object

```
normalizeGiotto(
  gobject,
  norm_methods = c("standard", "osmFISH"),
  library_size_norm = TRUE,
  scalefactor = 6000,
  log_norm = TRUE,
  log_offset = 1,
  logbase = 2,
  scale_genes = T,
  scale_cells = T,
  scale_order = c("first_genes", "first_cells"),
  verbose = F
)
```

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### **Arguments**

gobject giotto object

norm\_methods normalization method to use
library\_size\_norm

normalize cells by library size

scalefactor scale factor to use after library size normalization
log\_norm transform values to log-scale
log\_offset offset value to add to expression matrix, default = 1

logbase log base to use to log normalize expression values scale\_genes z-score genes over all cells scale\_cells z-score cells over all genes scale\_order order to scale genes and cells

verbose be verbose

#### **Details**

Currently there are two 'methods' to normalize your raw counts data.

A. The standard method follows the standard protocol which can be adjusted using the provided parameters and follows the following order:

- 1. Data normalization for total library size and scaling by a custom scale-factor.
- 2. Log transformation of data.
- 3. Z-scoring of data by genes and/or cells.
- B. The normalization method as provided by the osmFISH paper is also implemented:
  - 1. First normalize genes, for each gene divide the counts by the total gene count and multiply by the total number of genes.
  - 2. Next normalize cells, for each cell divide the normalized gene counts by the total counts per cell and multiply by the total number of cells.

This data will be saved in the Giotto slot for custom expression.

#### Value

giotto object

### **Examples**

```
data(mini_giotto_single_cell)
norm_gobject = normalizeGiotto(mini_giotto_single_cell)
```

pDataDT

**PAGEEnrich** 

**PAGEEnrich** 

## Description

Function to calculate gene signature enrichment scores per spatial position using PAGE.

## Usage

```
PAGEEnrich(...)
```

### **Arguments**

... Arguments passed on to runPAGEEnrich

gobject Giotto object

sign\_matrix Matrix of signature genes for each cell type / process

expression\_values expression values to use

min\_overlap\_genes minimum number of overlapping genes in sign\_matrix required to calculate enrichment

reverse\_log\_scale reverse expression values from log scale

logbase log base to use if reverse\_log\_scale = TRUE

output\_enrichment how to return enrichment output

p\_value calculate p-values (boolean, default = FALSE)

include\_depletion calculate both enrichment and depletion

n\_times number of permutations to calculate for p\_value

 $max\_block$  number of lines to process together (default = 20e6)

name to give to spatial enrichment results, default = PAGE

verbose be verbose

return\_gobject return giotto object

## See Also

runPAGEEnrich

pDataDT

pDataDT

# Description

show cell metadata

### Usage

pDataDT(gobject)

### **Arguments**

gobject

giotto object

plotCCcomDotplot 151

#### Value

data.table with cell metadata

#### **Examples**

```
data(mini_giotto_single_cell) # loads existing Giotto object
pDataDT(mini_giotto_single_cell)
```

 $\verb|plotCCcomDotplot|$ 

plot CC com Dot plot

# Description

Plots dotplot for ligand-receptor communication scores in cell-cell interactions

# Usage

```
plotCCcomDotplot(
  gobject,
  comScores,
  selected_LR = NULL,
  selected_cell_LR = NULL,
  show_LR_names = TRUE,
  show_cell_LR_names = TRUE,
  cluster_on = c("PI", "LR_expr", "log2fc"),
  cor_method = c("pearson", "kendall", "spearman"),
 aggl_method = c("ward.D", "ward.D2", "single", "complete", "average", "mcquitty",
    "median", "centroid"),
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "plotCCcomDotplot"
```

```
gobject
                  giotto object
comScores
                  communinication scores from exprCellCellcom or spatCellCellcom
selected_LR
                  selected ligand-receptor combinations
selected_cell_LR
                  selected cell-cell combinations for ligand-receptor combinations
show_LR_names
                  show ligand-receptor names
show_cell_LR_names
                  show cell-cell names
                  values to use for clustering of cell-cell and ligand-receptor pairs
cluster_on
cor_method
                  correlation method used for clustering
aggl_method
                  agglomeration method used by hclust
```

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```
show_plot show plots

return_plot return plotting object

save_plot directly save the plot [boolean]

save_param list of saving parameters from all_plots_save_function

default_save_name

default save name for saving, don't change, change save_name in save_param
```

#### Value

ggplot

plotCCcomHeatmap plotCCcomHeatmap

### **Description**

Plots heatmap for ligand-receptor communication scores in cell-cell interactions

## Usage

```
plotCCcomHeatmap(
  gobject,
  comScores,
  selected_LR = NULL,
  selected_cell_LR = NULL,
  show_LR_names = TRUE,
  show_cell_LR_names = TRUE,
  show = c("PI", "LR_expr", "log2fc"),
  cor_method = c("pearson", "kendall", "spearman"),
  aggl_method = c("ward.D", "ward.D2", "single", "complete", "average", "mcquitty",
        "median", "centroid"),
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "plotCCcomHeatmap"
)
```

```
gobject giotto object

comScores communinication scores from exprCellCellcom or spatCellCellcom

selected_LR selected ligand-receptor combinations

selected_cell_LR selected cell-cell combinations for ligand-receptor combinations

show_LR_names show ligand-receptor names

show_cell_LR_names show cell-cell names

show values to show on heatmap
```

```
cor_method correlation method used for clustering

aggl_method agglomeration method used by hclust

show_plot show plots

return_plot return plotting object

save_plot directly save the plot [boolean]

save_param list of saving parameters from all_plots_save_function

default_save_name

default save name for saving, don't change, change save_name in save_param
```

#### Value

ggplot

```
plotCellProximityGenes
```

plotCellProximityGenes

## **Description**

Create visualization for cell proximity gene scores

```
plotCellProximityGenes(
  gobject,
  cpgObject,
  method = c("volcano", "cell_barplot", "cell-cell", "cell_sankey", "heatmap",
    "dotplot"),
  min_cells = 4,
  min_cells_expr = 1,
  min_int_cells = 4,
  min_int_cells_expr = 1,
  min_fdr = 0.1,
  min_spat_diff = 0.2,
  min_log2_fc = 0.2,
  min_zscore = 2,
  zscores_column = c("cell_type", "genes"),
  direction = c("both", "up", "down"),
  cell_color_code = NULL,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "plotCellProximityGenes"
```

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### **Arguments**

gobject giotto object cpgObject ICG (interaction changed gene) score object method plotting method to use min\_cells minimum number of source cell type min\_cells\_expr minimum expression level for source cell type minimum number of interacting neighbor cell type min\_int\_cells min\_int\_cells\_expr minimum expression level for interacting neighbor cell type min\_fdr minimum adjusted p-value minimum absolute spatial expression difference min\_spat\_diff minimum log2 fold-change min\_log2\_fc minimum z-score change min\_zscore zscores\_column calculate z-scores over cell types or genes differential expression directions to keep direction cell\_color\_code vector of colors with cell types as names show\_plot show plots return\_plot return plotting object save\_plot directly save the plot [boolean] list of saving parameters from all\_plots\_save\_function save\_param default\_save\_name

default save name for saving, don't change, change save\_name in save\_param

# Value

plot

plotCombineCCcom plotCombineCCcom

## **Description**

Create visualization for combined (pairwise) cell proximity gene scores

```
plotCombineCCcom(
  gobject,
  combCCcom,
  selected_LR = NULL,
  selected_cell_LR = NULL,
  detail_plot = T,
  simple_plot = F,
  simple_plot_facet = c("interaction", "genes"),
```

```
facet_scales = "fixed",
facet_ncol = length(selected_LR),
facet_nrow = length(selected_cell_LR),
colors = c("#9932CC", "#FF8C00"),
show_plot = NA,
return_plot = NA,
save_plot = NA,
save_param = list(),
default_save_name = "plotCombineCCcom")
```

### **Arguments**

```
gobject
                  giotto object
{\sf combCCcom}
                  combined communcation scores, output from combCCcom()
selected_LR
                  selected ligand-receptor pair
selected_cell_LR
                  selected cell-cell interaction pair for ligand-receptor pair
detail_plot
                  show detailed info in both interacting cell types
simple_plot
                  show a simplified plot
simple_plot_facet
                  facet on interactions or genes with simple plot
facet_scales
                  ggplot facet scales paramter
facet_ncol
                  ggplot facet ncol parameter
facet_nrow
                  ggplot facet nrow parameter
colors
                  vector with two colors to use
show_plot
                  show plots
                  return plotting object
return_plot
save_plot
                  directly save the plot [boolean]
                  list of saving parameters from all_plots_save_function
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

## Value

ggplot

```
plot {\tt Combine Cell Cell Communication} \\ plot {\tt Combine Cell Cell Communication} \\
```

# Description

Create visualization for combined (pairwise) cell proximity gene scores

#### Usage

```
plotCombineCellCellCommunication(
  gobject,
  combCCcom,
  selected_LR = NULL,
  selected_cell_LR = NULL,
  detail_plot = T,
  simple_plot = F,
  simple_plot_facet = c("interaction", "genes"),
  facet_scales = "fixed",
  facet_ncol = length(selected_LR),
  facet_nrow = length(selected_cell_LR),
  colors = c("#9932CC", "#FF8C00"),
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "plotCombineCellCellCommunication"
)
```

## **Arguments**

```
gobject
                  giotto object
combCCcom
                  combined communcation scores, output from combCCcom()
selected_LR
                  selected ligand-receptor pair
selected_cell_LR
                  selected cell-cell interaction pair for ligand-receptor pair
{\tt detail\_plot}
                  show detailed info in both interacting cell types
                  show a simplified plot
simple_plot
simple_plot_facet
                  facet on interactions or genes with simple plot
facet_scales
                  ggplot facet scales paramter
facet_ncol
                  ggplot facet ncol parameter
facet_nrow
                  ggplot facet nrow parameter
                  vector with two colors to use
colors
show_plot
                  show plots
return_plot
                  return plotting object
                  directly save the plot [boolean]
save_plot
                  list of saving parameters from all_plots_save_function
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

#### Value

ggplot

```
plot {\tt Combine Cell Proximity Genes} \\ plot {\tt Combine Cell Proximity Genes}
```

## **Description**

Create visualization for combined (pairwise) ICG scores

### Usage

```
plotCombineCellProximityGenes(...)
```

## **Arguments**

Arguments passed on to plotCombineInteractionChangedGenes . . . gobject giotto object combCpgObject ICGscores, output from combineInteractionChangedGenes() selected\_interactions interactions to show selected\_gene\_to\_gene pairwise gene combinations to show detail\_plot show detailed info in both interacting cell types simple\_plot show a simplified plot simple\_plot\_facet facet on interactions or genes with simple plot facet\_scales ggplot facet scales paramter facet\_ncol ggplot facet ncol parameter facet\_nrow ggplot facet nrow parameter colors vector with two colors to use show\_plot show plots return\_plot return plotting object save\_plot directly save the plot [boolean] save\_param list of saving parameters from all\_plots\_save\_function default\_save\_name default save name for saving, don't change, change save\_name in save\_param

### See Also

 $\verb|plotCombineInteractionChangedGenes||$ 

plotCombineCPG plotCombineCPG

### **Description**

Create visualization for combined (pairwise) ICG scores

```
plotCombineCPG(...)
```

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#### **Arguments**

```
Arguments passed on to plotCombineICG
gobject giotto object
combCpgObject ICGscores, output from combineInteractionChangedGenes()
selected_interactions interactions to show
selected_gene_to_gene pairwise gene combinations to show
detail_plot show detailed info in both interacting cell types
simple_plot show a simplified plot
simple_plot_facet facet on interactions or genes with simple plot
facet_scales ggplot facet scales paramter
facet_ncol ggplot facet ncol parameter
facet_nrow ggplot facet nrow parameter
colors vector with two colors to use
show_plot show plots
return_plot return plotting object
save_plot directly save the plot [boolean]
save_param list of saving parameters from all_plots_save_function
default_save_name default save name for saving, don't change, change save_name
    in save_param
```

### See Also

plotCombineICG

plotCombineICG

plotCombineICG

## **Description**

Create visualization for combined (pairwise) ICG scores

```
plotCombineICG(
  gobject,
  combCpgObject,
  selected_interactions = NULL,
  selected_gene_to_gene = NULL,
  detail_plot = T,
  simple_plot = F,
  simple_plot_facet = c("interaction", "genes"),
  facet_scales = "fixed",
  facet_ncol = length(selected_gene_to_gene),
  facet_nrow = length(selected_interactions),
  colors = c("#9932CC", "#FF8C00"),
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
```

```
save_param = list(),
  default_save_name = "plotCombineICG"
)
```

#### **Arguments**

```
gobject
                  giotto object
combCpgObject
                  ICGscores, output from combineInteractionChangedGenes()
selected_interactions
                  interactions to show
selected_gene_to_gene
                  pairwise gene combinations to show
detail_plot
                  show detailed info in both interacting cell types
simple_plot
                  show a simplified plot
simple_plot_facet
                  facet on interactions or genes with simple plot
facet_scales
                  ggplot facet scales paramter
facet_ncol
                  ggplot facet ncol parameter
facet_nrow
                  ggplot facet nrow parameter
colors
                  vector with two colors to use
                  show plots
show_plot
return_plot
                  return plotting object
save_plot
                  directly save the plot [boolean]
                  list of saving parameters from all_plots_save_function
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

## Value

ggplot

```
plot {\tt Combine Interaction Changed Genes} \\ plot {\tt Combine Interaction Changed Genes}
```

### **Description**

Create visualization for combined (pairwise) ICG scores

```
plotCombineInteractionChangedGenes(
  gobject,
  combCpgObject,
  selected_interactions = NULL,
  selected_gene_to_gene = NULL,
  detail_plot = T,
```

```
simple_plot = F,
simple_plot_facet = c("interaction", "genes"),
facet_scales = "fixed",
facet_ncol = length(selected_gene_to_gene),
facet_nrow = length(selected_interactions),
colors = c("#9932CC", "#FF8C00"),
show_plot = NA,
return_plot = NA,
save_plot = NA,
save_param = list(),
default_save_name = "plotCombineICG"
)
```

## **Arguments**

```
gobject
                 giotto object
                 ICGscores, output from combineInteractionChangedGenes()
combCpgObject
selected_interactions
                 interactions to show
selected_gene_to_gene
                 pairwise gene combinations to show
detail_plot
                 show detailed info in both interacting cell types
simple_plot
                 show a simplified plot
simple_plot_facet
                 facet on interactions or genes with simple plot
facet_scales
                 ggplot facet scales paramter
                 ggplot facet ncol parameter
facet_ncol
                 ggplot facet nrow parameter
facet_nrow
colors
                 vector with two colors to use
show_plot
                 show plots
return_plot
                 return plotting object
                 directly save the plot [boolean]
save_plot
                 list of saving parameters from all_plots_save_function
save_param
default_save_name
                 default save name for saving, don't change, change save_name in save_param
```

#### Value

ggplot

plotCPG 161

plotCPG plotCPG

## **Description**

Create visualization for cell proximity gene scores

### Usage

```
plotCPG(
  gobject,
  cpgObject,
  method = c("volcano", "cell_barplot", "cell-cell", "cell_sankey", "heatmap",
    "dotplot"),
  min_cells = 5,
  min_cells_expr = 1,
  min_int_cells = 3,
  min_int_cells_expr = 1,
  min_fdr = 0.05,
  min_spat_diff = 0.2,
  min_log2_fc = 0.2,
  min_zscore = 2,
  zscores_column = c("cell_type", "genes"),
  direction = c("both", "up", "down"),
  cell_color_code = NULL,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "plotCPG"
```

```
giotto object
gobject
cpgObject
                 ICG (interaction changed gene) score object
method
                 plotting method to use
min_cells
                 minimum number of source cell type
min_cells_expr minimum expression level for source cell type
                 minimum number of interacting neighbor cell type
min_int_cells
min_int_cells_expr
                 minimum expression level for interacting neighbor cell type
                 minimum adjusted p-value
min_fdr
min_spat_diff
                 minimum absolute spatial expression difference
min_log2_fc
                 minimum log2 fold-change
min_zscore
                 minimum z-score change
zscores_column calculate z-scores over cell types or genes
```

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direction differential expression directions to keep

cell\_color\_code

vector of colors with cell types as names

show\_plot show plots

return\_plot return plotting object

save\_plot directly save the plot [boolean]

save\_param list of saving parameters from all\_plots\_save\_function

default\_save\_name

default save name for saving, don't change, change save\_name in save\_param

## Value

plot

plotGiottoImage

plotGiottoImage

# Description

get plot a giotto image from a giotto object

# Usage

```
plotGiottoImage(gobject, image_name)
```

# Arguments

gobject giotto object

image\_name name of giotto image showGiottoImageNames

### Value

plot

plotHeatmap

plotHeatmap

# Description

Creates heatmap for genes and clusters.

plotHeatmap 163

## Usage

```
plotHeatmap(
     gobject,
     expression_values = c("normalized", "scaled", "custom"),
     genes,
     cluster_column = NULL,
     cluster_order = c("size", "correlation", "custom"),
     cluster_custom_order = NULL,
     cluster_color_code = NULL,
     cluster_cor_method = "pearson",
     cluster_hclust_method = "ward.D",
     gene_order = c("correlation", "custom"),
     gene_custom_order = NULL,
     gene_cor_method = "pearson",
     gene_hclust_method = "complete",
     show_values = c("rescaled", "z-scaled", "original"),
     size_vertical_lines = 1.1,
     gradient_colors = c("blue", "yellow", "red"),
     gene_label_selection = NULL,
     axis_text_y_size = NULL,
     legend_nrows = 1,
     show_plot = NA,
     return_plot = NA,
     save_plot = NA,
     save_param = list(),
     default_save_name = "plotHeatmap"
   )
Arguments
   gobject
                    giotto object
   expression_values
                    expression values to use
   genes
                    genes to use
   cluster_column name of column to use for clusters
   cluster_order
                    method to determine cluster order
   cluster_custom_order
                    custom order for clusters
   cluster_color_code
                    color code for clusters
   cluster_cor_method
                    method for cluster correlation
   cluster_hclust_method
                    method for hierarchical clustering of clusters
   gene_order
                    method to determine gene order
   gene_custom_order
                    custom order for genes
   gene_cor_method
```

method for gene correlation

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```
gene_hclust_method
                  method for hierarchical clustering of genes
show_values
                  which values to show on heatmap
size_vertical_lines
                  sizes for vertical lines
gradient_colors
                  colors for heatmap gradient
gene_label_selection
                  subset of genes to show on y-axis
axis_text_y_size
                  size for y-axis text
legend_nrows
                  number of rows for the cluster legend
show_plot
                  show plot
return_plot
                  return ggplot object
                  directly save the plot [boolean]
save_plot
                  list of saving parameters, see showSaveParameters
save_param
default_save_name
                  default save name
```

### **Details**

If you want to display many genes there are 2 ways to proceed:

- 1. set axis\_text\_y\_size to a really small value and show all genes
- 2. provide a subset of genes to display to gene\_label\_selection

#### Value

ggplot

### **Examples**

plotICG 165

plotICG plotICG

## **Description**

Create barplot to visualize interaction changed genes

# Usage

```
plotICG(
  gobject,
  cpgObject,
  source_type,
  source_markers,
  ICG_genes,
  cell_color_code = NULL,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "plotICG"
)
```

# Arguments

```
gobject
                  giotto object
cpgObject
                  ICG (interaction changed gene) score object
                  cell type of the source cell
source_type
source_markers markers for the source cell type
                  named character vector of ICG genes
ICG_genes
cell_color_code
                  cell color code for the interacting cell types
show_plot
                  show plots
return_plot
                  return plotting object
                  directly save the plot [boolean]
save_plot
save_param
                  list of saving parameters from all_plots_save_function
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

### Value

plot

```
plotInteraction {\tt Changed Genes} \\ plotInteraction {\tt Changed Genes} \\
```

## **Description**

Create barplot to visualize interaction changed genes

### Usage

```
plotInteractionChangedGenes(
   gobject,
   cpgObject,
   source_type,
   source_markers,
   ICG_genes,
   cell_color_code = NULL,
   show_plot = NA,
   return_plot = NA,
   save_plot = NA,
   save_param = list(),
   default_save_name = "plotInteractionChangedGenes")
```

### **Arguments**

```
gobject
                  giotto object
                  ICG (interaction changed gene) score object
cpgObject
                  cell type of the source cell
source_type
source_markers markers for the source cell type
ICG_genes
                  named character vector of ICG genes
cell_color_code
                  cell color code for the interacting cell types
show_plot
                  show plots
return_plot
                  return plotting object
save_plot
                  directly save the plot [boolean]
save_param
                  list of saving parameters from all_plots_save_function
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

## Value

plot

## **Description**

Creates heatmap for numeric cell metadata within aggregated clusters.

# Usage

```
plotMetaDataCellsHeatmap(
  gobject,
  metadata_cols = NULL,
  spat_enr_names = NULL,
  value_cols = NULL,
  first_meta_col = NULL,
  second_meta_col = NULL,
  show_values = c("zscores", "original", "zscores_rescaled"),
  custom_cluster_order = NULL,
  clus_cor_method = "pearson",
  clus_cluster_method = "complete",
  custom_values_order = NULL,
  values_cor_method = "pearson",
  values_cluster_method = "complete",
  midpoint = 0,
  x_{text_size} = 8,
  x_{text_angle} = 45,
  y_{text_size} = 8,
  strip_text_size = 8,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "plotMetaDataCellsHeatmap"
)
```

```
gobject giotto object

metadata_cols annotation columns found in pDataDT(gobject)

spat_enr_names spatial enrichment results to include

value_cols value columns to use

first_meta_col if more than 1 metadata column, select the x-axis factor

second_meta_col

if more than 1 metadata column, select the facetting factor

show_values which values to show on heatmap

custom_cluster_order

custom cluster order (default = NULL)
```

```
clus_cor_method
                  correlation method for clusters
clus_cluster_method
                  hierarchical cluster method for the clusters
custom_values_order
                  custom values order (default = NULL)
values_cor_method
                  correlation method for values
values\_cluster\_method
                  hierarchical cluster method for the values
midpoint
                  midpoint of show_values
x_text_size
                  size of x-axis text
x_text_angle
                  angle of x-axis text
                  size of y-axis text
y_text_size
strip_text_size
                  size of strip text
show_plot
                  show plot
return_plot
                  return ggplot object
                  directly save the plot [boolean]
save_plot
save_param
                  list of saving parameters, see showSaveParameters
default_save_name
```

## **Details**

Creates heatmap for the average values of selected value columns in the different annotation groups.

default save name for saving, don't change, change save\_name in save\_param

#### Value

ggplot or data.table

#### See Also

plotMetaDataHeatmap for gene expression instead of numeric cell annotation data.

plotMetaDataHeatmap

# Description

Creates heatmap for genes within aggregated clusters.

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### Usage

clus\_cor\_method

clus\_cluster\_method

custom\_gene\_order

gene\_cor\_method

```
plotMetaDataHeatmap(
     gobject,
      expression_values = c("normalized", "scaled", "custom"),
     metadata_cols = NULL,
      selected_genes = NULL,
      first_meta_col = NULL,
      second_meta_col = NULL,
      show_values = c("zscores", "original", "zscores_rescaled"),
      custom_cluster_order = NULL,
      clus_cor_method = "pearson",
      clus_cluster_method = "complete",
     custom_gene_order = NULL,
      gene_cor_method = "pearson",
      gene_cluster_method = "complete",
      gradient_color = c("blue", "white", "red"),
     gradient_midpoint = 0,
      gradient_limits = NULL,
      x_{text_size} = 10,
     x_{text_angle} = 45,
     y_{text_size} = 10,
      strip_text_size = 8,
      show_plot = NA,
     return_plot = NA,
      save_plot = NA,
     save_param = list(),
     default_save_name = "plotMetaDataHeatmap"
   )
Arguments
   gobject
                    giotto object
   expression_values
                    expression values to use
   metadata_cols annotation columns found in pDataDT(gobject)
   selected_genes subset of genes to use
   first_meta_col if more than 1 metadata column, select the x-axis factor
    second_meta_col
                    if more than 1 metadata column, select the facetting factor
   show_values
                    which values to show on heatmap
   custom_cluster_order
                    custom cluster order (default = NULL)
```

correlation method for clusters

correlation method for genes

hierarchical cluster method for the clusters

custom gene order (default = NULL)

```
gene_cluster_method
                  hierarchical cluster method for the genes
gradient_color vector with 3 colors for numeric data
gradient_midpoint
                  midpoint for color gradient
gradient_limits
                  vector with lower and upper limits
x_text_size
                  size of x-axis text
x_text_angle
                  angle of x-axis text
y_text_size
                  size of y-axis text
strip_text_size
                  size of strip text
show_plot
                  show plot
return_plot
                  return ggplot object
                  directly save the plot [boolean]
save_plot
save_param
                  list of saving parameters, see showSaveParameters
default_save_name
                  default save name
```

### **Details**

Creates heatmap for the average expression of selected genes in the different annotation/cluster groups. Calculation of cluster or gene order is done on the provided expression values, but visualization is by default on the z-scores. Other options are the original values or z-scores rescaled per gene (-1 to 1).

## Value

ggplot or data.table

#### See Also

plotMetaDataCellsHeatmap for numeric cell annotation instead of gene expression.

# **Examples**

plotPCA 171

plotPCA plotPCA

### **Description**

Short wrapper for PCA visualization

#### Usage

```
plotPCA(gobject, dim_reduction_name = "pca", default_save_name = "PCA", ...)
```

```
gobject
                 giotto object
dim_reduction_name
                 name of PCA
default_save_name
                 default save name of PCA plot
                 Arguments passed on to dimPlot2D
. . .
                 group_by create multiple plots based on cell annotation column
                 group_by_subset subset the group_by factor column
                 dim1_to_use dimension to use on x-axis
                 dim2_to_use dimension to use on y-axis
                 spat_enr_names names of spatial enrichment results to include
                 show_NN_network show underlying NN network
                 nn_network_to_use type of NN network to use (kNN vs sNN)
                 network_name name of NN network to use, if show_NN_network = TRUE
                 cell_color color for cells (see details)
                 color_as_factor convert color column to factor
                 cell_color_code named vector with colors
                 cell_color_gradient vector with 3 colors for numeric data
                 gradient_midpoint midpoint for color gradient
                 gradient_limits vector with lower and upper limits
                 select_cell_groups select subset of cells/clusters based on cell_color param-
                 select_cells select subset of cells based on cell IDs
                  show_other_cells display not selected cells
                 other_cell_color color of not selected cells
                 other_point_size size of not selected cells
                 show_cluster_center plot center of selected clusters
                 show_center_label plot label of selected clusters
                 center_point_size size of center points
                  center_point_border_col border color of center points
                 center_point_border_stroke border stroke size of center points
                 label_size size of labels
                 label_fontface font of labels
```

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```
edge_alpha column to use for alpha of the edges
point_shape point with border or not (border or no_border)
point_size size of point (cell)
point_alpha transparancy of point
point_border_col color of border around points
point_border_stroke stroke size of border around points
title title for plot, defaults to cell_color parameter
show_legend show legend
legend_text size of legend text
legend_symbol_size size of legend symbols
background_color color of plot background
axis_text size of axis text
axis_title size of axis title
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_plot show plot
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
```

## **Details**

Description of parameters, see dimPlot2D. For 3D plots see plotPCA\_3D

### Value

ggplot

### See Also

```
Other reduced dimension visualizations: dimPlot2D(), dimPlot3D(), dimPlot(), plotPCA_2D(), plotPCA_3D(), plotTSNE_3D(), plotTSNE(), plotUMAP_2D(), plotUMAP_3D(), plotUMAP()
```

### **Examples**

```
data(mini_giotto_single_cell)
plotPCA(mini_giotto_single_cell)
plotPCA(mini_giotto_single_cell, cell_color = 'cell_types', point_size = 3)
```

plotPCA\_2D 173

plotPCA\_2D

plotPCA\_2D

### **Description**

Short wrapper for PCA visualization

# Usage

```
plotPCA_2D(
  gobject,
  dim_reduction_name = "pca",
  default_save_name = "PCA_2D",
   ...
)
```

```
giotto object
gobject
dim_reduction_name
                 name of PCA
default_save_name
                 default save name of PCA plot
                 Arguments passed on to dimPlot2D
                 group_by create multiple plots based on cell annotation column
                 group_by_subset subset the group_by factor column
                 dim1_to_use dimension to use on x-axis
                 dim2_to_use dimension to use on y-axis
                 spat_enr_names names of spatial enrichment results to include
                 show_NN_network show underlying NN network
                 nn_network_to_use type of NN network to use (kNN vs sNN)
                 network_name name of NN network to use, if show_NN_network = TRUE
                 cell_color color for cells (see details)
                 color_as_factor convert color column to factor
                 cell_color_code named vector with colors
                 cell_color_gradient vector with 3 colors for numeric data
                 gradient_midpoint midpoint for color gradient
                 gradient_limits vector with lower and upper limits
                  select_cell_groups select subset of cells/clusters based on cell_color param-
                      eter
                 select_cells select subset of cells based on cell IDs
                 show_other_cells display not selected cells
                 other_cell_color color of not selected cells
                 other_point_size size of not selected cells
                 show_cluster_center plot center of selected clusters
                 show_center_label plot label of selected clusters
                 center_point_size size of center points
```

plotPCA\_2D

```
center_point_border_col border color of center points
center_point_border_stroke border stroke size of center points
label_size size of labels
label_fontface font of labels
edge_alpha column to use for alpha of the edges
point_shape point with border or not (border or no_border)
point_size size of point (cell)
point_alpha transparancy of point
point_border_col color of border around points
point_border_stroke stroke size of border around points
title title for plot, defaults to cell_color parameter
show_legend show legend
legend_text size of legend text
legend_symbol_size size of legend symbols
background_color color of plot background
axis_text size of axis text
axis_title size of axis title
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_plot show plot
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
```

## Details

Description of parameters, see dimPlot2D. For 3D plots see plotPCA\_3D

### Value

ggplot

## See Also

```
Other reduced dimension visualizations: dimPlot2D(), dimPlot3D(), dimPlot(), plotPCA_3D(), plotPCA(), plotTSNE_2D(), plotTSNE_3D(), plotTSNE(), plotUMAP_2D(), plotUMAP_3D(), plotUMAP()
```

### **Examples**

```
data(mini_giotto_single_cell)
plotPCA_2D(mini_giotto_single_cell)
plotPCA_2D(mini_giotto_single_cell, cell_color = 'cell_types', point_size = 3)
```

plotPCA\_3D 175

plotPCA\_3D plotPCA\_3D

### **Description**

Visualize cells according to 3D PCA dimension reduction

# Usage

```
plotPCA_3D(
  gobject,
  dim_reduction_name = "pca",
  default_save_name = "PCA_3D",
)
```

```
giotto object
gobject
dim_reduction_name
                 name of PCA
default_save_name
                 default save name of PCA plot
                 Arguments passed on to dimPlot3D
                 dim1_to_use dimension to use on x-axis
                 dim2_to_use dimension to use on y-axis
                 dim3_to_use dimension to use on z-axis
                 spat_enr_names names of spatial enrichment results to include
                 show_NN_network show underlying NN network
                 nn_network_to_use type of NN network to use (kNN vs sNN)
                 network_name name of NN network to use, if show_NN_network = TRUE
                 cell_color color for cells (see details)
                 color_as_factor convert color column to factor
                 cell_color_code named vector with colors
                 select_cell_groups select subset of cells/clusters based on cell_color param-
                 select_cells select subset of cells based on cell IDs
                 show_other_cells display not selected cells
                 other_cell_color color of not selected cells
                 other_point_size size of not selected cells
                 show_cluster_center plot center of selected clusters
                 show_center_label plot label of selected clusters
                 center_point_size size of center points
                 label_size size of labels
                 edge_alpha column to use for alpha of the edges
                 point_size size of point (cell)
                 show_plot show plot
```

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```
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
```

#### Details

Description of parameters.

#### Value

plotly

#### See Also

```
Other reduced dimension visualizations: dimPlot2D(), dimPlot3D(), dimPlot(), plotPCA_2D(), plotPCA_2D(), plotTSNE_2D(), plotTSNE_3D(), plotTSNE(), plotUMAP_2D(), plotUMAP_3D(), plotUMAP()
```

```
plotRankSpatvsExpr plotRankSpatvsExpr
```

## **Description**

Plots dotplot to compare ligand-receptor rankings from spatial and expression information

## Usage

```
plotRankSpatvsExpr(
 gobject,
 combCC,
 expr_rnk_column = "LR_expr_rnk",
  spat_rnk_column = "LR_spat_rnk",
 midpoint = 10,
 size\_range = c(0.01, 1.5),
 xlims = NULL,
 ylims = NULL,
 selected_ranks = c(1, 10, 20),
 show_plot = NA,
 return_plot = NA,
 save_plot = NA,
 save_param = list(),
 default_save_name = "plotRankSpatvsExpr"
)
```

```
gobject giotto object

combCC combined communinication scores from combCCcom

expr_rnk_column

column with expression rank information to use

spat_rnk_column

column with spatial rank information to use
```

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```
midpoint
                  midpoint of colors
                  size ranges of dotplot
size_range
xlims
                  x-limits, numerical vector of 2
ylims
                  y-limits, numerical vector of 2
selected_ranks numerical vector, will be used to print out the percentage of top spatial ranks are
                  recovered
show_plot
                  show plots
return_plot
                  return plotting object
save_plot
                  directly save the plot [boolean]
save_param
                  list of saving parameters from all_plots_save_function
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

#### Value

ggplot

plotRecovery plotRecovery

## Description

Plots recovery plot to compare ligand-receptor rankings from spatial and expression information

### Usage

```
plotRecovery(
   gobject,
   combCC,
   expr_rnk_column = "exprPI_rnk",
   spat_rnk_column = "spatPI_rnk",
   ground_truth = c("spatial", "expression"),
   show_plot = NA,
   return_plot = NA,
   save_plot = NA,
   save_param = list(),
   default_save_name = "plotRecovery"
)
```

```
gobject giotto object

combCC combined communinication scores from combCCcom
expr_rnk_column

column with expression rank information to use
spat_rnk_column

column with spatial rank information to use
ground_truth what to consider as ground truth (default: spatial)
```

```
show_plot show plots

return_plot return plotting object

save_plot directly save the plot [boolean]

save_param list of saving parameters from all_plots_save_function

default_save_name

default save name for saving, don't change, change save_name in save_param
```

### Value

ggplot

plotRecovery\_sub plotRecovery\_sub

### **Description**

Plots recovery plot to compare ligand-receptor rankings from spatial and expression information

### Usage

```
plotRecovery_sub(combCC, first_col = "LR_expr_rnk", second_col = "LR_spat_rnk")
```

### **Arguments**

combCC combined communinication scores from combCCcom

first\_col first column to use second\_col second column to use

plotStatDelaunayNetwork

plotStatDelaunayNetwork

# Description

Plots network statistics for a Delaunay network..

```
plotStatDelaunayNetwork(
  gobject,
  method = c("deldir", "delaunayn_geometry", "RTriangle"),
  dimensions = "all",
  maximum_distance = "auto",
  minimum_k = 0,
  options = "Pp",
  Y = TRUE,
  j = TRUE,
  S = 0,
```

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```
show_plot = NA,
return_plot = NA,
save_plot = NA,
save_param = list(),
default_save_name = "plotStatDelaunayNetwork",
...
)
```

## **Arguments**

gobject giotto object

method package to use to create a Delaunay network

dimensions which spatial dimensions to use (maximum 2 dimensions)

maximum\_distance

distance cuttof for Delaunay neighbors to consider

minimum\_k minimum neighbours if maximum\_distance != NULL

options (geometry) String containing extra control options for the underlying Qhull

command; see the Qhull documentation (../doc/qhull/html/qdelaun.html) for the

available options. (default = 'Pp', do not report precision problems)

Y (RTriangle) If TRUE prohibits the insertion of Steiner points on the mesh bound-

ary.

j (RTriangle) If TRUE jettisons vertices that are not part of the final triangulation

from the output.

S (RTriangle) Specifies the maximum number of added Steiner points.

show\_plot show plots

return\_plot return ggplot object

save\_plot directly save the plot [boolean]

save\_param list of saving parameters, see showSaveParameters

default\_save\_name

default save name for saving, don't change, change save\_name in save\_param

... Other parameters

## Value

giotto object with updated spatial network slot

plotTSNE plotTSNE

## **Description**

Short wrapper for tSNE visualization

```
plotTSNE(gobject, dim_reduction_name = "tsne", default_save_name = "tSNE", ...)
```

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```
gobject
                 giotto object
dim_reduction_name
                 name of TSNE
default_save_name
                 default save name of TSNE plot
                  Arguments passed on to dimPlot2D
. . .
                  group_by create multiple plots based on cell annotation column
                 group_by_subset subset the group_by factor column
                 dim1_to_use dimension to use on x-axis
                 dim2_to_use dimension to use on y-axis
                  spat_enr_names names of spatial enrichment results to include
                  show_NN_network show underlying NN network
                 nn_network_to_use type of NN network to use (kNN vs sNN)
                 network_name name of NN network to use, if show_NN_network = TRUE
                 cell_color color for cells (see details)
                 color_as_factor convert color column to factor
                 cell_color_code named vector with colors
                 cell_color_gradient vector with 3 colors for numeric data
                 gradient_midpoint midpoint for color gradient
                 gradient_limits vector with lower and upper limits
                 select_cell_groups select subset of cells/clusters based on cell_color param-
                  select_cells select subset of cells based on cell IDs
                 show_other_cells display not selected cells
                 other_cell_color color of not selected cells
                  other_point_size size of not selected cells
                  show_cluster_center plot center of selected clusters
                 show_center_label plot label of selected clusters
                  center_point_size size of center points
                  center_point_border_col border color of center points
                  center_point_border_stroke border stroke size of center points
                 label_size size of labels
                 label_fontface font of labels
                  edge_alpha column to use for alpha of the edges
                 point_shape point with border or not (border or no_border)
                 point_size size of point (cell)
                 point_alpha transparancy of point
                 point_border_col color of border around points
                 point_border_stroke stroke size of border around points
                 title title for plot, defaults to cell_color parameter
                  show_legend show legend
                 legend_text size of legend text
                 legend_symbol_size size of legend symbols
                 background_color color of plot background
```

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```
axis_text size of axis text

axis_title size of axis title

cow_n_col cowplot param: how many columns

cow_rel_h cowplot param: relative height

cow_rel_w cowplot param: relative width

cow_align cowplot param: how to align

show_plot show plot

return_plot return ggplot object

save_plot directly save the plot [boolean]

save_param list of saving parameters, see showSaveParameters
```

#### **Details**

Description of parameters, see dimPlot2D. For 3D plots see plotTSNE\_3D

#### Value

ggplot

#### See Also

```
Other reduced dimension visualizations: dimPlot2D(), dimPlot3D(), dimPlot(), plotPCA_2D(), plotPCA_3D(), plotPCA_3D(), plotTSNE_2D(), plotTSNE_3D(), plotUMAP_2D(), plotUMAP_3D(), plotUMAP()
```

## **Examples**

```
data(mini_giotto_single_cell)
plotTSNE(mini_giotto_single_cell)
plotTSNE(mini_giotto_single_cell, cell_color = 'cell_types', point_size = 3)
```

plotTSNE\_2D

plotTSNE\_2D

# **Description**

Short wrapper for tSNE visualization

```
plotTSNE_2D(
  gobject,
  dim_reduction_name = "tsne",
  default_save_name = "tSNE_2D",
   ...
)
```

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#### **Arguments**

gobject giotto object dim\_reduction\_name name of TSNE default\_save\_name default save name of TSNE plot Arguments passed on to dimPlot2D . . . group\_by create multiple plots based on cell annotation column group\_by\_subset subset the group\_by factor column dim1\_to\_use dimension to use on x-axis dim2\_to\_use dimension to use on y-axis spat\_enr\_names names of spatial enrichment results to include show\_NN\_network show underlying NN network nn\_network\_to\_use type of NN network to use (kNN vs sNN) network\_name name of NN network to use, if show\_NN\_network = TRUE cell\_color color for cells (see details) color\_as\_factor convert color column to factor cell\_color\_code named vector with colors cell\_color\_gradient vector with 3 colors for numeric data gradient\_midpoint midpoint for color gradient gradient\_limits vector with lower and upper limits select\_cell\_groups select subset of cells/clusters based on cell\_color paramselect\_cells select subset of cells based on cell IDs show\_other\_cells display not selected cells other\_cell\_color color of not selected cells other\_point\_size size of not selected cells show\_cluster\_center plot center of selected clusters show\_center\_label plot label of selected clusters center\_point\_size size of center points center\_point\_border\_col border color of center points center\_point\_border\_stroke border stroke size of center points label\_size size of labels label\_fontface font of labels edge\_alpha column to use for alpha of the edges point\_shape point with border or not (border or no\_border) point\_size size of point (cell) point\_alpha transparancy of point point\_border\_col color of border around points point\_border\_stroke stroke size of border around points title title for plot, defaults to cell\_color parameter show\_legend show legend legend\_text size of legend text legend\_symbol\_size size of legend symbols background\_color color of plot background

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```
axis_text size of axis text

axis_title size of axis title

cow_n_col cowplot param: how many columns

cow_rel_h cowplot param: relative height

cow_rel_w cowplot param: relative width

cow_align cowplot param: how to align

show_plot show plot

return_plot return ggplot object

save_plot directly save the plot [boolean]

save_param list of saving parameters, see showSaveParameters
```

#### **Details**

Description of parameters, see dimPlot2D. For 3D plots see plotTSNE\_3D

#### Value

ggplot

#### See Also

```
Other reduced dimension visualizations: dimPlot2D(), dimPlot3D(), dimPlot(), plotPCA_2D(), plotPCA_3D(), plotPCA(), plotTSNE_3D(), plotTSNE(), plotUMAP_2D(), plotUMAP_3D(), plotUMAP()
```

# Examples

```
data(mini_giotto_single_cell)
plotTSNE_2D(mini_giotto_single_cell)
plotTSNE_2D(mini_giotto_single_cell, cell_color = 'cell_types', point_size = 3)
```

plotTSNE\_3D

plotTSNE\_3D

# Description

Visualize cells according to dimension reduction coordinates

```
plotTSNE_3D(
  gobject,
  dim_reduction_name = "tsne",
  default_save_name = "TSNE_3D",
   ...
)
```

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#### **Arguments**

```
gobject
                  giotto object
dim_reduction_name
                  name of TSNE
default_save_name
                  default save name of TSNE plot
                  Arguments passed on to dimPlot3D
                  dim1_to_use dimension to use on x-axis
                  dim2_to_use dimension to use on y-axis
                  dim3_to_use dimension to use on z-axis
                  spat_enr_names names of spatial enrichment results to include
                  show_NN_network show underlying NN network
                  nn\_network\_to\_use \ type \ of \ NN \ network \ to \ use \ (kNN \ vs \ sNN)
                  network_name name of NN network to use, if show_NN_network = TRUE
                  cell_color color for cells (see details)
                  color_as_factor convert color column to factor
                  cell_color_code named vector with colors
                  select_cell_groups select subset of cells/clusters based on cell_color param-
                  select_cells select subset of cells based on cell IDs
                  show_other_cells display not selected cells
                  other_cell_color color of not selected cells
                  other_point_size size of not selected cells
                  show_cluster_center plot center of selected clusters
                  show_center_label plot label of selected clusters
                  center_point_size size of center points
                  label_size size of labels
                  edge_alpha column to use for alpha of the edges
                  point_size size of point (cell)
                  show_plot show plot
                  return_plot return ggplot object
                  save_plot directly save the plot [boolean]
                  save_param list of saving parameters, see showSaveParameters
```

#### **Details**

Description of parameters.

#### Value

plotly

#### See Also

```
Other reduced dimension visualizations: dimPlot2D(), dimPlot3D(), dimPlot(), plotPCA_2D(), plotPCA_3D(), plotPCA(), plotTSNE_2D(), plotTSNE(), plotUMAP_2D(), plotUMAP_3D(), plotUMAP()
```

plotUMAP 185

plotUMAP plotUMAP

#### **Description**

Short wrapper for UMAP visualization

#### Usage

```
plotUMAP(gobject, dim_reduction_name = "umap", default_save_name = "UMAP", ...)
```

#### **Arguments**

```
gobject
                 giotto object
dim_reduction_name
                 name of UMAP
default_save_name
                 default save name of UMAP plot
                 Arguments passed on to dimPlot2D
                 group_by create multiple plots based on cell annotation column
                 group_by_subset subset the group_by factor column
                 dim1_to_use dimension to use on x-axis
                 dim2_to_use dimension to use on y-axis
                 spat_enr_names names of spatial enrichment results to include
                 show_NN_network show underlying NN network
                 nn_network_to_use type of NN network to use (kNN vs sNN)
                 network_name name of NN network to use, if show_NN_network = TRUE
                 cell_color color for cells (see details)
                 color_as_factor convert color column to factor
                 cell_color_code named vector with colors
                 cell_color_gradient vector with 3 colors for numeric data
                 gradient_midpoint midpoint for color gradient
                 gradient_limits vector with lower and upper limits
                 select_cell_groups select subset of cells/clusters based on cell_color param-
                 select_cells select subset of cells based on cell IDs
                  show_other_cells display not selected cells
                 other_cell_color color of not selected cells
                 other_point_size size of not selected cells
                 show_cluster_center plot center of selected clusters
                 show_center_label plot label of selected clusters
                 center_point_size size of center points
                 center_point_border_col border color of center points
                 center_point_border_stroke border stroke size of center points
                 label_size size of labels
                 label_fontface font of labels
```

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```
edge_alpha column to use for alpha of the edges
point_shape point with border or not (border or no_border)
point_size size of point (cell)
point_alpha transparancy of point
point_border_col color of border around points
point_border_stroke stroke size of border around points
title title for plot, defaults to cell_color parameter
show_legend show legend
legend_text size of legend text
legend_symbol_size size of legend symbols
background_color color of plot background
axis_text size of axis text
axis_title size of axis title
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_plot show plot
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
```

#### **Details**

Description of parameters, see dimPlot2D. For 3D plots see plotUMAP\_3D

#### Value

ggplot

#### See Also

```
Other reduced dimension visualizations: dimPlot2D(), dimPlot3D(), dimPlot(), plotPCA_2D(), plotPCA_3D(), plotPCA(), plotTSNE_2D(), plotTSNE_3D(), plotTSNE(), plotUMAP_2D(), plotUMAP_3D()
```

## **Examples**

```
data(mini_giotto_single_cell)
plotUMAP(mini_giotto_single_cell)
plotUMAP(mini_giotto_single_cell, cell_color = 'cell_types', point_size = 3)
```

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plotUMAP\_2D

plotUMAP\_2D

#### **Description**

Short wrapper for UMAP visualization

# Usage

```
plotUMAP_2D(
  gobject,
  dim_reduction_name = "umap";
  default_save_name = "UMAP_2D",
)
```

## **Arguments**

```
giotto object
gobject
dim_reduction_name
                 name of UMAP
default_save_name
                 default save name of UMAP plot
                 Arguments passed on to dimPlot2D
                 group_by create multiple plots based on cell annotation column
                 group_by_subset subset the group_by factor column
                 dim1_to_use dimension to use on x-axis
                 dim2_to_use dimension to use on y-axis
                 spat_enr_names names of spatial enrichment results to include
                 show_NN_network show underlying NN network
                 nn_network_to_use type of NN network to use (kNN vs sNN)
                 network_name name of NN network to use, if show_NN_network = TRUE
                 cell_color color for cells (see details)
                 color_as_factor convert color column to factor
                 cell_color_code named vector with colors
                 cell_color_gradient vector with 3 colors for numeric data
                 gradient_midpoint midpoint for color gradient
                 gradient_limits vector with lower and upper limits
                  select_cell_groups select subset of cells/clusters based on cell_color param-
                      eter
                 select_cells select subset of cells based on cell IDs
                 show_other_cells display not selected cells
                 other_cell_color color of not selected cells
                 other_point_size size of not selected cells
                 show_cluster_center plot center of selected clusters
                 show_center_label plot label of selected clusters
                 center_point_size size of center points
```

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```
center_point_border_col border color of center points
center_point_border_stroke border stroke size of center points
label_size size of labels
label_fontface font of labels
edge_alpha column to use for alpha of the edges
point_shape point with border or not (border or no_border)
point_size size of point (cell)
point_alpha transparancy of point
point_border_col color of border around points
point_border_stroke stroke size of border around points
title title for plot, defaults to cell_color parameter
show_legend show legend
legend_text size of legend text
legend_symbol_size size of legend symbols
background_color color of plot background
axis_text size of axis text
axis_title size of axis title
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_plot show plot
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
```

# Details

Description of parameters, see dimPlot2D. For 3D plots see plotUMAP\_3D

### Value

ggplot

## See Also

```
Other reduced dimension visualizations: dimPlot2D(), dimPlot3D(), dimPlot(), plotPCA_2D(), plotPCA_3D(), plotPCA(), plotTSNE_2D(), plotTSNE_3D(), plotTSNE(), plotUMAP_3D(), plotUMAP()
```

#### **Examples**

```
data(mini_giotto_single_cell)
plotUMAP_2D(mini_giotto_single_cell)
plotUMAP_2D(mini_giotto_single_cell, cell_color = 'cell_types', point_size = 3)
```

plotUMAP\_3D 189

plotUMAP\_3D plotUMAP\_3D

#### **Description**

Visualize cells according to dimension reduction coordinates

# Usage

```
plotUMAP_3D(
  gobject,
  dim_reduction_name = "umap"
  default_save_name = "UMAP_3D",
)
```

## **Arguments**

```
giotto object
gobject
dim_reduction_name
                 name of UMAP
default_save_name
                 default save name of UMAP plot
                 Arguments passed on to dimPlot3D
                 dim1_to_use dimension to use on x-axis
                 dim2_to_use dimension to use on y-axis
                 dim3_to_use dimension to use on z-axis
                 spat_enr_names names of spatial enrichment results to include
                 show_NN_network show underlying NN network
                 nn_network_to_use type of NN network to use (kNN vs sNN)
                 network_name name of NN network to use, if show_NN_network = TRUE
                 cell_color color for cells (see details)
                 color_as_factor convert color column to factor
                 cell_color_code named vector with colors
                 select_cell_groups select subset of cells/clusters based on cell_color param-
                 select cells select subset of cells based on cell IDs
                 show_other_cells display not selected cells
                 other_cell_color color of not selected cells
                 other_point_size size of not selected cells
                 show_cluster_center plot center of selected clusters
                 show_center_label plot label of selected clusters
                 center_point_size size of center points
                 label_size size of labels
                 edge_alpha column to use for alpha of the edges
                 point_size size of point (cell)
                 show_plot show plot
```

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```
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
```

#### **Details**

Description of parameters.

#### Value

plotly

#### See Also

```
Other reduced dimension visualizations: dimPlot2D(), dimPlot3D(), dimPlot(), plotPCA_2D(), plotPCA_3D(), plotPCA(), plotTSNE_2D(), plotTSNE_3D(), plotTSNE(), plotUMAP_2D(), plotUMAP()
```

processGiotto

processGiotto

#### **Description**

Wrapper for the different Giotto object processing functions

# Usage

```
processGiotto(
  gobject,
  filter_params = list(),
  norm_params = list(),
  stat_params = list(),
  adjust_params = list(),
  verbose = TRUE
)
```

#### **Arguments**

```
gobject giotto object

filter_params additional parameters to filterGiotto

norm_params additional parameters to normalizeGiotto

stat_params additional parameters to addStatistics

adjust_params additional parameters to adjustGiottoMatrix

verbose be verbose (default is TRUE)
```

## **Details**

See filterGiotto, normalizeGiotto, addStatistics and adjustGiottoMatrix for more information about the different parameters in each step. If you do not provide them it will use the default values.

rankEnrich 191

#### Value

```
giotto object
```

## **Examples**

rankEnrich

rankEnrich

#### **Description**

Function to calculate gene signature enrichment scores per spatial position using a rank based approach.

### Usage

```
rankEnrich(...)
```

# **Arguments**

... Arguments passed on to runRankEnrich

gobject Giotto object
sign\_matrix Matrix of signature genes for each cell type / process
expression\_values expression values to use
reverse\_log\_scale reverse expression values from log scale
logbase log base to use if reverse\_log\_scale = TRUE
output\_enrichment how to return enrichment output
ties\_method how to handle rank ties
p\_value calculate p-values (boolean, default = FALSE)
n\_times number of permutations to calculate for p\_value
rbp\_p fractional binarization threshold (default = 0.99)
num\_agg number of top genes to aggregate (default = 100)
name to give to spatial enrichment results, default = rank
return\_gobject return giotto object

# See Also

runRankEnrich

192 readExprMatrix

```
rank Spatial Cor Groups rank Spatial Cor Groups
```

## **Description**

Rank spatial correlated clusters according to correlation structure

#### Usage

```
rankSpatialCorGroups(
  gobject,
  spatCorObject,
  use_clus_name = NULL,
  show_plot = NA,
  return_plot = FALSE,
  save_plot = NA,
  save_param = list(),
  default_save_name = "rankSpatialCorGroups"
)
```

# **Arguments**

```
gobject
                  giotto object
                  spatial correlation object
spatCorObject
use_clus_name
                  name of clusters to visualize (from clusterSpatialCorGenes())
show_plot
                  show plot
                  return ggplot object
return_plot
                  directly save the plot [boolean]
save_plot
                  list of saving parameters, see showSaveParameters
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

#### Value

data.table with positive (within group) and negative (outside group) scores

```
readExprMatrix readExprMatrix
```

# **Description**

Function to read an expression matrix into a sparse matrix.

```
readExprMatrix(path, cores = NA, transpose = FALSE)
```

readGiottoInstructions 193

## **Arguments**

path path to the expression matrix

cores number of cores to use

transpose transpose matrix

#### **Details**

The expression matrix needs to have both unique column names and row names

#### Value

sparse matrix

 ${\tt readGiottoInstructions}$ 

readGiottoInstrunctions

# Description

Retrieves the instruction associated with the provided parameter

# Usage

```
readGiottoInstructions(giotto_instructions, param = NULL)
```

# **Arguments**

giotto\_instructions

 $giot to\ object\ or\ result\ from\ create Giot to Instructions ()$ 

param parameter to retrieve

## Value

specific parameter

remove Cell Annotation remove Cell Annotation

# Description

removes cell annotation of giotto object

```
removeCellAnnotation(gobject, columns = NULL, return_gobject = TRUE)
```

194 removeGeneAnnotation

## **Arguments**

gobject giotto object

columns names of columns to remove

return\_gobject boolean: return giotto object (default = TRUE)

## **Details**

```
if return_gobject = FALSE, it will return the cell metadata
```

#### Value

giotto object

## **Examples**

removeGeneAnnotation removeGeneAnnotation

## **Description**

removes gene annotation of giotto object

## Usage

```
removeGeneAnnotation(gobject, columns = NULL, return_gobject = TRUE)
```

## **Arguments**

gobject giotto object

columns names of columns to remove

return\_gobject boolean: return giotto object (default = TRUE)

# **Details**

```
if return_gobject = FALSE, it will return the gene metadata
```

# Value

giotto object

removeGiottoEnvironment 195

## **Examples**

removeGiottoEnvironment

removeGiottoEnvironment

# **Description**

removeGiottoEnvironment

## Usage

```
removeGiottoEnvironment(verbose = TRUE)
```

# Arguments

verbose be verbose

# **Details**

Removes a previously installed giotto environment. See installGiottoEnvironment.

```
replaceGiottoInstructions
```

replace Giot to Instructions

# Description

Function to replace all instructions from giotto object

# Usage

```
replaceGiottoInstructions(gobject, instructions = NULL)
```

# Arguments

gobject giotto object

instructions new instructions (e.g. result from createGiottoInstructions)

## Value

giotto object with replaces instructions

rowSums\_giotto

rowMeans\_giotto

rowMeans\_giotto

# Description

rowMeans function that works with multiple matrix representations

# Usage

```
rowMeans_giotto(mymatrix)
```

# Arguments

mymatrix

matrix object

## Value

numeric vector

rowSums\_giotto

rowSums\_giotto

# Description

rowSums function that works with multiple matrix representations

# Usage

```
rowSums_giotto(mymatrix)
```

# Arguments

mymatrix

matrix object

## Value

numeric vector

runDWLSDeconv 197

runDWLSDeconv

runDWLSDeconv

## **Description**

Function to perform DWLS deconvolution based on single cell expression data

#### Usage

```
runDWLSDeconv(
  gobject,
  expression_values = c("normalized"),
  logbase = 2,
  cluster_column = "leiden_clus",
  sign_matrix,
  n_cell = 50,
  cutoff = 2,
  name = NULL,
  return_gobject = TRUE
)
```

## **Arguments**

```
gobject giotto object
expression_values
expression values to use

logbase base used for log normalization
cluster_column name of cluster column
sign_matrix sig matrix for deconvolution
n_cell number of cells per spot
cutoff cut off (default = 2)
```

name name to give to spatial deconvolution results, default = DWLS

return\_gobject return giotto object

## Value

giotto object or deconvolution results

```
\verb"runHyperGeometricEnrich"
```

run Hyper Geometric Enrich

## **Description**

Function to calculate gene signature enrichment scores per spatial position using a hypergeometric test.

198 runPAGEEnrich

#### Usage

```
runHyperGeometricEnrich(
  gobject,
  sign_matrix,
  expression_values = c("normalized", "scaled", "custom"),
  reverse_log_scale = TRUE,
  logbase = 2,
  top_percentage = 5,
  output_enrichment = c("original", "zscore"),
  p_value = FALSE,
  name = NULL,
  return_gobject = TRUE
)
```

#### **Arguments**

gobject Giotto object sign\_matrix Matrix of signature genes for each cell type / process expression\_values expression values to use reverse\_log\_scale reverse expression values from log scale log base to use if reverse\_log\_scale = TRUE logbase top\_percentage percentage of cells that will be considered to have gene expression with matrix binarization output\_enrichment how to return enrichment output calculate p-values (boolean, default = FALSE) p\_value to give to spatial enrichment results, default = rank name return\_gobject return giotto object

# **Details**

The enrichment score is calculated based on the p-value from the hypergeometric test, -log10(p-value).

# Value

data.table with enrichment results

runPAGEEnrich runPAGEEnrich

# Description

Function to calculate gene signature enrichment scores per spatial position using PAGE.

runPAGEEnrich 199

#### Usage

```
runPAGEEnrich(
      gobject,
      sign_matrix,
      expression_values = c("normalized", "scaled", "custom"),
      min_overlap_genes = 5,
      reverse_log_scale = TRUE,
      logbase = 2,
      output_enrichment = c("original", "zscore"),
      p_value = FALSE,
      include_depletion = FALSE,
      n_{times} = 1000,
      max_block = 2e+07,
      name = NULL,
      verbose = TRUE,
      return_gobject = TRUE
    )
Arguments
   gobject
                     Giotto object
    sign_matrix
                     Matrix of signature genes for each cell type / process
    expression_values
                     expression values to use
   min_overlap_genes
                     minimum number of overlapping genes in sign matrix required to calculate en-
    reverse_log_scale
                     reverse expression values from log scale
    logbase
                     log base to use if reverse_log_scale = TRUE
    output_enrichment
                     how to return enrichment output
                     calculate p-values (boolean, default = FALSE)
    p_value
    include_depletion
                     calculate both enrichment and depletion
   n_times
                     number of permutations to calculate for p_value
   max_block
                     number of lines to process together (default = 20e6)
```

# **Details**

name

verbose

sign\_matrix: a binary matrix with genes as row names and cell-types as column names. Alternatively a list of signature genes can be provided to makeSignMatrixPAGE, which will create the matrix for you.

to give to spatial enrichment results, default = PAGE

be verbose

return\_gobject return giotto object

The enrichment Z score is calculated by using method (PAGE) from Kim SY et al., BMC bioinformatics, 2005 as  $Z=((Sm\ \ mu)*m^(1/2))/delta$ . For each gene in each spot, mu is the fold change values versus the mean expression and delta is the standard deviation. Sm is the mean fold change value of a specific marker gene set and m is the size of a given marker gene set.

#### Value

data.table with enrichment results

#### See Also

```
make Sign Matrix PAGE
```

runPAGEEnrich\_OLD

runPAGEEnrich\_OLD

# **Description**

Function to calculate gene signature enrichment scores per spatial position using PAGE.

## Usage

```
runPAGEEnrich_OLD(
  gobject,
  sign_matrix,
  expression_values = c("normalized", "scaled", "custom"),
  reverse_log_scale = TRUE,
  logbase = 2,
  output_enrichment = c("original", "zscore"),
  p_value = FALSE,
  n_times = 1000,
  name = NULL,
  return_gobject = TRUE
)
```

## **Arguments**

gobject

```
sign_matrix
                  Matrix of signature genes for each cell type / process
expression_values
                  expression values to use
reverse_log_scale
                  reverse expression values from log scale
                  log base to use if reverse_log_scale = TRUE
logbase
output_enrichment
                  how to return enrichment output
                  calculate p-values (boolean, default = FALSE)
p_value
                  number of permutations to calculate for p_value
n_times
                  to give to spatial enrichment results, default = PAGE
name
return_gobject return giotto object
```

Giotto object

runPatternSimulation 201

#### **Details**

sign\_matrix: a binary matrix with genes as row names and cell-types as column names. Alternatively a list of signature genes can be provided to makeSignMatrixPAGE, which will create the matrix for you.

The enrichment Z score is calculated by using method (PAGE) from Kim SY et al., BMC bioinformatics, 2005 as  $Z=((Sm\ mu)*m^(1/2))/delta$ . For each gene in each spot, mu is the fold change values versus the mean expression and delta is the standard deviation. Sm is the mean fold change value of a specific marker gene set and m is the size of a given marker gene set.

#### Value

data.table with enrichment results

## See Also

makeSignMatrixPAGE

runPatternSimulation runPatternSimulation

## **Description**

Creates a known spatial pattern for selected genes one-by-one and runs the different spatial gene detection tests

```
runPatternSimulation(
 gobject,
 pattern_name = "pattern",
  pattern_colors = c(`in` = "green", out = "red"),
 pattern_cell_ids = NULL,
  gene_names = NULL,
  spatial\_probs = c(0.5, 1),
  reps = 2,
  spatial_network_name = "kNN_network",
  spat_methods = c("binSpect_single", "binSpect_multi", "spatialDE", "spark",
    "silhouetteRank"),
  spat_methods_params = list(NA, NA, NA, NA, NA),
 spat_methods_names = c("binSpect_single", "binSpect_multi", "spatialDE", "spark",
    "silhouetteRank"),
  scalefactor = 6000,
  save_plot = T,
  save_raw = T,
  save_norm = T,
  save_dir = "~"
 max_col = 4,
 height = 7,
 width = 7,
  run_simulations = TRUE,
```

202 runPatternSimulation

```
)
```

#### **Arguments**

giotto object gobject name of spatial pattern pattern\_name pattern\_colors 2 color vector for the spatial pattern pattern\_cell\_ids cell ids that make up the spatial pattern selected genes gene\_names spatial\_probs probabilities to test for a high expressing gene value to be part of the spatial pattern number of random simulation repetitions reps spatial\_network\_name which spatial network to use for binSpectSingle spat\_methods vector of spatial methods to test spat\_methods\_params list of parameters list for each element in the vector of spatial methods to test spat\_methods\_names name for each element in the vector of spatial elements to test scalefactor library size scaling factor when re-normalizing dataset save\_plot save intermediate random simulation plots or not save the raw expression matrix of the simulation save\_raw save the normalized expression matrix of the simulation save\_norm save\_dir directory to save results to maximum number of columns for final plots max\_col height height of final plots width width of final plots run\_simulations run simulations (default = TRUE)

additional parameters for renormalization

## Value

data.table with results

runPCA 203

runPCA runPCA

## **Description**

runs a Principal Component Analysis

# Usage

```
runPCA(
 gobject,
 expression_values = c("normalized", "scaled", "custom"),
 reduction = c("cells", "genes"),
 name = "pca",
 genes_to_use = "hvg";
 return_gobject = TRUE,
 center = TRUE,
  scale_unit = TRUE,
 ncp = 100,
 method = c("irlba", "factominer"),
 rev = FALSE,
  set_seed = TRUE,
  seed_number = 1234,
 verbose = TRUE,
)
```

# Arguments

gobject

verbose

. . .

expression\_values expression values to use cells or genes reduction arbitrary name for PCA run name subset of genes to use for PCA genes\_to\_use return\_gobject boolean: return giotto object (default = TRUE) center center data first (default = TRUE) scale\_unit scale features before PCA (default = TRUE) number of principal components to calculate ncp method which implementation to use rev do a reverse PCA use of seed set\_seed seed\_number seed number to use

verbosity of the function

additional parameters for PCA (see details)

giotto object

204 runRankEnrich

#### **Details**

See prcomp\_irlba and PCA for more information about other parameters.

- genes\_to\_use = NULL: will use all genes from the selected matrix
- genes\_to\_use = <hvg name>: can be used to select a column name of highly variable genes, created by (see calculateHVG)
- genes\_to\_use = c('geneA', 'geneB', ...): will use all manually provided genes

#### Value

giotto object with updated PCA dimension recuction

#### **Examples**

runRankEnrich

runRankEnrich

## **Description**

Function to calculate gene signature enrichment scores per spatial position using a rank based approach.

```
runRankEnrich(
  gobject,
  sign_matrix,
  expression_values = c("normalized", "raw", "scaled", "custom"),
  reverse_log_scale = TRUE,
  logbase = 2,
  output_enrichment = c("original", "zscore"),
  ties_method = c("random", "max"),
  p_value = FALSE,
  n_times = 1000,
  rbp_p = 0.99,
  num_agg = 100,
  name = NULL,
  return_gobject = TRUE
)
```

runSpatialDeconv 205

#### **Arguments**

gobject Giotto object

sign\_matrix Matrix of signature genes for each cell type / process

expression\_values

expression values to use

reverse\_log\_scale

reverse expression values from log scale

logbase log base to use if reverse\_log\_scale = TRUE

output\_enrichment

how to return enrichment output

ties\_method how to handle rank ties

 $\begin{array}{lll} \textbf{p\_value} & \text{calculate p-values (boolean, default = FALSE)} \\ \textbf{n\_times} & \text{number of permutations to calculate for p\_value} \\ \textbf{rbp\_p} & \text{fractional binarization threshold (default = 0.99)} \\ \textbf{num\_agg} & \text{number of top genes to aggregate (default = 100)} \\ \textbf{name} & \text{to give to spatial enrichment results, default = rank} \\ \end{array}$ 

return\_gobject return giotto object

#### **Details**

sign\_matrix: a rank-fold matrix with genes as row names and cell-types as column names. Alternatively a scRNA-seq matrix and vector with clusters can be provided to makeSignMatrixRank, which will create the matrix for you.

First a new rank is calculated as  $R = (R1*R2)^{n}(1/2)$ , where R1 is the rank of fold-change for each gene in each spot and R2 is the rank of each marker in each cell type. The Rank-Biased Precision is then calculated as: RBP =  $(1 - 0.99) * (0.99)^{n}(R - 1)$  and the final enrichment score is then calculated as the sum of top 100 RBPs.

### Value

data.table with enrichment results

# See Also

makeSignMatrixRank

runSpatialDeconv runSpatialDeconv

# Description

Function to perform deconvolution based on single cell expression data

206 runSpatialEnrich

#### Usage

```
runSpatialDeconv(
  gobject,
  deconv_method = c("DWLS"),
  expression_values = c("normalized"),
  logbase = 2,
  cluster_column = "leiden_clus",
  sign_matrix,
  n_cell = 50,
  cutoff = 2,
  name = NULL,
  return_gobject = TRUE
)
```

#### **Arguments**

```
gobject
                  giotto object
                  method to use for deconvolution
deconv_method
expression_values
                  expression values to use
logbase
                  base used for log normalization
cluster_column name of cluster column
                  signature matrix for deconvolution
sign_matrix
n_cell
                  number of cells per spot
cutoff
                  cut off (default = 2)
                  name to give to spatial deconvolution results
name
```

#### Value

giotto object or deconvolution results

return\_gobject return giotto object

 $run Spatial Enrich \\ run Spatial Enrich$ 

# Description

Function to calculate gene signature enrichment scores per spatial position using an enrichment test.

```
runSpatialEnrich(
  gobject,
  enrich_method = c("PAGE", "rank", "hypergeometric"),
  sign_matrix,
  expression_values = c("normalized", "scaled", "custom"),
  min_overlap_genes = 5,
  reverse_log_scale = TRUE,
```

runSpatialEnrich 207

```
logbase = 2,
p_value = FALSE,
n_times = 1000,
rbp_p = 0.99,
num_agg = 100,
max_block = 2e+07,
top_percentage = 5,
output_enrichment = c("original", "zscore"),
name = NULL,
verbose = TRUE,
return_gobject = TRUE
```

Giotto object

# **Arguments**

gobject

method for gene signature enrichment calculation enrich\_method sign\_matrix Matrix of signature genes for each cell type / process expression\_values expression values to use min\_overlap\_genes minimum number of overlapping genes in sign\_matrix required to calculate enrichment (PAGE) reverse\_log\_scale reverse expression values from log scale logbase log base to use if reverse\_log\_scale = TRUE calculate p-value (default = FALSE) p\_value (page/rank) number of permutation iterations to calculate p-value n\_times (rank) fractional binarization threshold (default = 0.99) rbp\_p (rank) number of top genes to aggregate (default = 100) num\_agg  $\max\_block$ number of lines to process together (default = 20e6) (hyper) percentage of cells that will be considered to have gene expression with top\_percentage matrix binarization output\_enrichment how to return enrichment output

to give to spatial enrichment results, default = PAGE

#### **Details**

name

verbose

For details see the individual functions:

return\_gobject return giotto object

be verbose

PAGE: runPAGEEnrichRank: runRankEnrich

• Hypergeometric: runHyperGeometricEnrich

#### Value

Giotto object or enrichment results if return\_gobject = FALSE

208 runtSNE

runtSNE runtSNE

#### **Description**

run tSNE

#### Usage

```
runtSNE(
 gobject,
 expression_values = c("normalized", "scaled", "custom"),
 reduction = c("cells", "genes"),
 dim_reduction_to_use = "pca",
 dim_reduction_name = "pca",
 dimensions_to_use = 1:10,
 name = "tsne",
  genes_to_use = NULL,
  return_gobject = TRUE,
 dims = 2,
 perplexity = 30,
  theta = 0.5,
 do_PCA_first = F,
  set_seed = T,
  seed_number = 1234,
  verbose = TRUE,
)
```

## **Arguments**

do\_PCA\_first

```
gobject
                 giotto object
expression_values
                 expression values to use
                 cells or genes
reduction
dim_reduction_to_use
                 use another dimension reduction set as input
dim_reduction_name
                 name of dimension reduction set to use
dimensions_to_use
                 number of dimensions to use as input
                 arbitrary name for tSNE run
name
                 if dim_reduction_to_use = NULL, which genes to use
genes_to_use
return_gobject boolean: return giotto object (default = TRUE)
dims
                 tSNE param: number of dimensions to return
                 tSNE param: perplexity
perplexity
theta
                 tSNE param: theta
```

tSNE param: do PCA before tSNE (default = FALSE)

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```
set_seed use of seed
seed_number seed number to use
verbose verbosity of the function
... additional tSNE parameters
```

#### **Details**

See Rtsne for more information about these and other parameters.

- Input for tSNE dimension reduction can be another dimension reduction (default = 'pca')
- To use gene expression as input set dim\_reduction\_to\_use = NULL
- If dim\_reduction\_to\_use = NULL, genes\_to\_use can be used to select a column name of highly variable genes (see calculateHVG) or simply provide a vector of genes
- multiple tSNE results can be stored by changing the name of the analysis

#### Value

giotto object with updated tSNE dimension recuction

# **Examples**

runUMAP

runUMAP

## **Description**

run UMAP

```
runUMAP(
   gobject,
   expression_values = c("normalized", "scaled", "custom"),
   reduction = c("cells", "genes"),
   dim_reduction_to_use = "pca",
   dim_reduction_name = "pca",
   dimensions_to_use = 1:10,
   name = "umap",
   genes_to_use = NULL,
```

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```
return_gobject = TRUE,
n_neighbors = 40,
n_components = 2,
n_epochs = 400,
min_dist = 0.01,
n_threads = NA,
spread = 5,
set_seed = TRUE,
seed_number = 1234,
verbose = T,
...
)
```

# **Arguments**

```
gobject
                 giotto object
expression_values
                 expression values to use
                 cells or genes
reduction
dim_reduction_to_use
                 use another dimension reduction set as input
dim_reduction_name
                 name of dimension reduction set to use
dimensions_to_use
                 number of dimensions to use as input
                 arbitrary name for UMAP run
name
                 if dim_reduction_to_use = NULL, which genes to use
genes_to_use
return_gobject boolean: return giotto object (default = TRUE)
n_neighbors
                 UMAP param: number of neighbors
n_components
                 UMAP param: number of components
n_epochs
                 UMAP param: number of epochs
min_dist
                 UMAP param: minimum distance
                 UMAP param: threads/cores to use
n_threads
spread
                 UMAP param: spread
set_seed
                 use of seed
seed_number
                 seed number to use
verbose
                 verbosity of function
                 additional UMAP parameters
. . .
```

#### **Details**

See umap for more information about these and other parameters.

- Input for UMAP dimension reduction can be another dimension reduction (default = 'pca')
- To use gene expression as input set dim\_reduction\_to\_use = NULL
- If dim\_reduction\_to\_use = NULL, genes\_to\_use can be used to select a column name of highly variable genes (see calculateHVG) or simply provide a vector of genes
- multiple UMAP results can be stored by changing the *name* of the analysis

screePlot 211

#### Value

giotto object with updated UMAP dimension recuction

#### **Examples**

screePlot

screePlot

## **Description**

identify significant prinicipal components (PCs) using an screeplot (a.k.a. elbowplot)

# Usage

```
screePlot(
  gobject,
  name = "pca",
  expression_values = c("normalized", "scaled", "custom"),
  reduction = c("cells", "genes"),
  method = c("irlba", "factominer"),
  rev = FALSE,
  genes_to_use = NULL,
  center = F,
  scale_unit = F,
  ncp = 100,
  ylim = c(0, 20),
  verbose = T,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "screePlot",
)
```

# **Arguments**

```
gobject giotto object

name name of PCA object if available

expression_values

expression values to use
```

212 selectPatternGenes

reduction cells or genes

method which implementation to use

rev do a reverse PCA

genes\_to\_use subset of genes to use for PCA

center center data before PCA
scale\_unit scale features before PCA

ncp number of principal components to calculate

ylim y-axis limits on scree plot

verbose verobsity show\_plot show plot

return\_plot return ggplot object

save\_plot directly save the plot [boolean]

save\_param list of saving parameters from all\_plots\_save\_function()

default\_save\_name

default save name for saving, don't change, change save\_name in save\_param

... additional arguments to pca function, see runPCA

#### **Details**

Screeplot works by plotting the explained variance of each individual PC in a barplot allowing you to identify which PC provides a significant contribution (a.k.a 'elbow method'). Screeplot will use an available pca object, based on the parameter 'name', or it will create it if it's

not available (see runPCA)

#### Value

ggplot object for scree method

# **Examples**

```
data(mini_giotto_single_cell)
screePlot(mini_giotto_single_cell, ncp = 10)
```

selectPatternGenes selectPatternGenes

# **Description**

Select genes correlated with spatial patterns

show,giotto-method 213

#### Usage

```
selectPatternGenes(
  spatPatObj,
  dimensions = 1:5,
  top_pos_genes = 10,
  top_neg_genes = 10,
  min_pos_cor = 0.5,
  min_neg_cor = -0.5,
  return_top_selection = FALSE
)
```

## **Arguments**

spatPatObj Output from detectSpatialPatterns
dimensions dimensions to identify correlated genes for.
top\_pos\_genes Top positively correlated genes.
top\_neg\_genes Top negatively correlated genes.
min\_pos\_cor Minimum positive correlation score to include a gene.
min\_neg\_cor Minimum negative correlation score to include a gene.
return\_top\_selection
only return selection based on correlation criteria (boolean)

## **Details**

Description.

### Value

Data.table with genes associated with selected dimension (PC).

show, giotto-method show method for giotto class

## **Description**

show method for giotto class

# Usage

```
## S4 method for signature 'giotto'
show(object)
```

#### **Arguments**

object giotto object

 $show Cluster Dendrogram \quad show Cluster Dendrogram \quad$ 

# **Description**

Creates dendrogram for selected clusters.

#### Usage

```
showClusterDendrogram(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  cluster_column,
  cor = c("pearson", "spearman"),
  distance = "ward.D",
  h = NULL,
  h_color = "red",
  rotate = FALSE,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "showClusterDendrogram",
  ...
)
```

### **Arguments**

```
gobject
                  giotto object
expression_values
                  expression values to use
cluster_column name of column to use for clusters
                  correlation score to calculate distance
cor
distance
                  distance method to use for hierarchical clustering
h
                  height of horizontal lines to plot
h_color
                  color of horizontal lines
                  rotate dendrogram 90 degrees
rotate
show_plot
                  show plot
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
                  list of saving parameters, see showSaveParameters
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
                  additional parameters for ggdendrogram()
```

# **Details**

Expression correlation dendrogram for selected clusters.

showClusterHeatmap 215

#### Value

ggplot

# **Examples**

showClusterHeatmap

showClusterHeatmap

## **Description**

Creates heatmap based on identified clusters

#### Usage

```
showClusterHeatmap(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  genes = "all",
  cluster_column,
  cor = c("pearson", "spearman"),
  distance = "ward.D",
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "showClusterHeatmap",
  ...
)
```

## **Arguments**

```
gobject giotto object
expression_values
expression values to use
genes vector of genes to use, default to 'all'
cluster_column name of column to use for clusters
cor correlation score to calculate distance
distance distance method to use for hierarchical clustering
show_plot show plot
```

# Details

Correlation heatmap of selected clusters.

#### Value

ggplot

# **Examples**

 $show Giot to Image Names \\ show Giot to Image Names \\$ 

# Description

Prints the available giotto images that are attached to the Giotto object

# Usage

```
showGiottoImageNames(gobject, verbose = TRUE)
```

# Arguments

gobject a giotto object verbose verbosity of function

## Value

a vector of giotto image names attached to the giotto object

showGiottoInstructions 217

showGiottoInstructions

showGiottoInstructions

# Description

Function to display all instructions from giotto object

# Usage

```
showGiottoInstructions(gobject)
```

## Arguments

gobject

giotto object

## Value

named vector with giotto instructions

showGrids

showGrids

# Description

Prints the available spatial grids that are attached to the Giotto object

# Usage

```
showGrids(gobject, verbose = TRUE)
```

# Arguments

gobject

a giotto object

verbose

verbosity of function#'

## Value

vector

218 showPattern

showNetworks showNetworks

# Description

Prints the available spatial networks that are attached to the Giotto object

## Usage

```
showNetworks(gobject, verbose = TRUE)
```

## **Arguments**

gobject a giotto object

verbose verbosity of function#'

#### Value

vector

showPattern showPattern

## **Description**

show patterns for 2D spatial data

## Usage

```
showPattern(gobject, spatPatObj, ...)
```

# Arguments

gobject giotto object

spatPatObj Output from detectSpatialPatterns

... Arguments passed on to showPattern2D

dimension dimension to plot trim Trim ends of the PC values.

background\_color background color for plot

grid\_border\_color color for grid
show\_legend show legend of ggplot

point\_size size of points
show\_plot show plot

return\_plot return ggplot object

save\_plot directly save the plot [boolean]

save\_param list of saving parameters, see showSaveParameters

default\_save\_name default save name for saving, don't change, change save\_name in save\_param

showPattern2D 219

#### Value

ggplot

#### See Also

showPattern2D

showPattern2D

showPattern2D

## **Description**

show patterns for 2D spatial data

## Usage

```
showPattern2D(
  gobject,
  spatPatObj,
  dimension = 1,
  trim = c(0.02, 0.98),
  background_color = "white",
  grid_border_color = "grey",
  show_legend = T,
  point_size = 1,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "showPattern2D")
```

# Arguments

```
gobject
                  giotto object
spatPatObj
                  Output from detectSpatialPatterns
                  dimension to plot
dimension
                  Trim ends of the PC values.
background_color
                  background color for plot
grid_border_color
                  color for grid
                  show legend of ggplot
show_legend
                  size of points
point_size
show_plot
                  show plot
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
save_param
                  list of saving parameters, see showSaveParameters
default_save_name
```

default save name for saving, don't change, change save\_name in save\_param

220 showPattern3D

#### Value

ggplot

showPattern3D

showPattern3D

## Description

show patterns for 3D spatial data

## Usage

```
showPattern3D(
 gobject,
  spatPatObj,
 dimension = 1,
  trim = c(0.02, 0.98),
 background_color = "white",
 grid_border_color = "grey",
  show_legend = T,
 point_size = 1,
 axis_scale = c("cube", "real", "custom"),
 custom_ratio = NULL,
 x_ticks = NULL,
 y_ticks = NULL,
 z_ticks = NULL,
  show_plot = NA,
 return_plot = NA,
 save_plot = NA,
 save_param = list(),
 default_save_name = "showPattern3D"
)
```

```
gobject
                  giotto object
spatPatObj
                  Output from detectSpatialPatterns
dimension
                  dimension to plot
trim
                  Trim ends of the PC values.
background_color
                  background color for plot
grid_border_color
                  color for grid
show_legend
                  show legend of plot
point_size
                  adjust the point size
axis_scale
                  scale the axis
custom_ratio
                  cutomize the scale of the axis
x_ticks
                  the tick number of x_axis
```

showPatternGenes 221

```
y_ticks the tick number of y_axis

z_ticks the tick number of z_axis

show_plot show plot

return_plot return plot object

save_plot directly save the plot [boolean]

save_param list of saving parameters, see showSaveParameters

default_save_name

default save name for saving, don't change, change save_name in save_param
```

#### Value

plotly

showPatternGenes showPatternGenes

#### **Description**

show genes correlated with spatial patterns

## Usage

```
showPatternGenes(
  gobject,
  spatPatObj,
  dimension = 1,
  top_pos_genes = 5,
  top_neg_genes = 5,
  point_size = 1,
  return_DT = FALSE,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "showPatternGenes"
)
```

```
gobject
                  giotto object
spatPatObj
                  Output from detectSpatialPatterns
                  dimension to plot genes for.
dimension
                  Top positively correlated genes.
top_pos_genes
                  Top negatively correlated genes.
top_neg_genes
point_size
                  size of points
                  if TRUE, it will return the data.table used to generate the plots
return_DT
show_plot
                  show plot
```

222 showSaveParameters

```
return_plot return ggplot object
```

save\_plot directly save the plot [boolean]

save\_param list of saving parameters, see showSaveParameters

default\_save\_name

default save name for saving, don't change, change save\_name in save\_param

## Value

ggplot

showProcessingSteps showProcessingSteps

## Description

shows the sequential processing steps that were performed on a Giotto object in a summarized format

## Usage

```
showProcessingSteps(gobject)
```

## **Arguments**

gobject giotto object

## Value

list of processing steps and names

## **Examples**

```
data(mini_giotto_single_cell)
showProcessingSteps(mini_giotto_single_cell)
```

 $\verb|showSaveParameters||$ 

showSaveParameters

# Description

 $Description\ of\ Giot to\ saving\ options,\ links\ to\ \verb"all_plots_save_function"$ 

#### Usage

```
showSaveParameters()
```

#### Value

Instruction on how to use the automatic plot saving options within Giotto

showSpatialCorGenes 223

#### **Examples**

```
showSaveParameters()
```

showSpatialCorGenes

showSpatialCorGenes

## Description

Shows and filters spatially correlated genes

## Usage

```
showSpatialCorGenes(
   spatCorObject,
   use_clus_name = NULL,
   selected_clusters = NULL,
   genes = NULL,
   min_spat_cor = 0.5,
   min_expr_cor = NULL,
   min_cor_diff = NULL,
   min_rank_diff = NULL,
   show_top_genes = NULL
)
```

# Arguments

```
spatCorObject
                      spatial correlation object
                      cluster information to show
use_clus_name
selected_clusters
                      subset of clusters to show
genes
                      subset of genes to show
                      filter on minimum spatial correlation
min_spat_cor
min_expr_cor
                      filter on minimum single-cell expression correlation
min_cor_diff
                      filter on minimum correlation difference (spatial vs expression)
min_rank_diff
                      filter on minimum correlation rank difference (spatial vs expression)
show\_top\_genes \hspace{0.2in} show \hspace{0.2in} top \hspace{0.2in} genes \hspace{0.2in} per \hspace{0.2in} gene \hspace{0.2in}
```

## Value

data.table with filtered information

224 signPCA

signPCA signPCA

#### **Description**

identify significant prinicipal components (PCs)

## Usage

```
signPCA(
 gobject,
 name = "pca",
 method = c("screeplot", "jackstraw"),
 expression_values = c("normalized", "scaled", "custom"),
 reduction = c("cells", "genes"),
 pca_method = c("irlba", "factominer"),
  rev = FALSE,
  genes_to_use = NULL,
  center = T,
  scale_unit = T,
 ncp = 50,
  scree_ylim = c(0, 10),
  jack_iter = 10,
  jack_threshold = 0.01,
  jack_ylim = c(0, 1),
  verbose = TRUE,
  show_plot = NA,
 return_plot = NA,
 save_plot = NA,
 save_param = list(),
 default_save_name = "signPCA"
)
```

## Arguments

gobject

name of PCA object if available name method to use to identify significant PCs method expression\_values expression values to use reduction cells or genes pca\_method which implementation to use do a reverse PCA rev subset of genes to use for PCA genes\_to\_use center center data before PCA scale features before PCA scale\_unit number of principal components to calculate ncp y-axis limits on scree plot scree\_ylim

giotto object

silhouetteRank 225

```
jack_iter
                  number of interations for jackstraw
jack_threshold p-value threshold to call a PC significant
                  y-axis limits on jackstraw plot
jack_ylim
verbose
                  verbosity
show_plot
                  show plot
return_plot
                  return ggplot object
                  directly save the plot [boolean]
save_plot
                  list of saving parameters from all_plots_save_function()
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

## **Details**

Two different methods can be used to assess the number of relevant or significant prinicipal components (PC's).

- 1. Screeplot works by plotting the explained variance of each individual PC in a barplot allowing you to identify which PC provides a significant contribution (a.k.a. 'elbow method').
- 2. The Jackstraw method uses the permutationPA function. By systematically permuting genes it identifies robust, and thus significant, PCs.

#### Value

ggplot object for scree method and maxtrix of p-values for jackstraw

silhouetteRank silhouetteRank

## **Description**

Previously: calculate\_spatial\_genes\_python. This method computes a silhouette score per gene based on the spatial distribution of two partitions of cells (expressed L1, and non-expressed L0). Here, rather than L2 Euclidean norm, it uses a rank-transformed, exponentially weighted function to represent the local physical distance between two cells. New multi aggregator implementation can be found at silhouetteRankTest

## Usage

```
silhouetteRank(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  metric = "euclidean",
  subset_genes = NULL,
  rbp_p = 0.95,
  examine_top = 0.3,
  python_path = NULL
)
```

226 silhouetteRankTest

#### **Arguments**

```
gobject giotto object
expression_values
expression values to use

metric distance metric to use

subset_genes only run on this subset of genes
rbp_p fractional binarization threshold
examine_top top fraction to evaluate with silhouette
python_path specify specific path to python if required
```

## Value

data.table with spatial scores

silhouetteRankTest silhouetteRankTest

## **Description**

Multi parameter aggregator version of silhouetteRank

#### Usage

```
silhouetteRankTest(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  subset_genes = NULL,
  overwrite_input_bin = TRUE,
  rbp_ps = c(0.95, 0.99),
  examine_tops = c(0.005, 0.01, 0.05, 0.1, 0.3),
  matrix_type = "dissim",
  num_core = 4,
  parallel_path = "/usr/bin",
  output = NULL,
  query_sizes = 10L,
  verbose = FALSE
)
```

```
gobject giotto object
expression_values
expression values to use
subset_genes only run on this subset of genes
overwrite_input_bin
overwrite input bin
rbp_ps fractional binarization thresholds
examine_tops top fractions to evaluate with silhouette
```

```
matrix_type type of matrix
num_core number of cores to use
parallel_path path to GNU parallel function
output output directory
query_sizes size of query
verbose be verbose
```

#### Value

data.table with spatial scores

```
simulate One Gene Pattern Giotto Object \\ simulate One Gene Pattern Giotto Object
```

## Description

Create a simulated spatial pattern for one selected gnee

## Usage

```
simulateOneGenePatternGiottoObject(
  gobject,
  pattern_name = "pattern",
  pattern_cell_ids = NULL,
  gene_name = NULL,
  spatial_prob = 0.95,
  gradient_direction = NULL,
  show_pattern = TRUE,
  pattern_colors = c(`in` = "green", out = "red"),
  ...
)
```

# **Arguments**

```
giotto object
gobject
pattern_name
                  name of spatial pattern
pattern_cell_ids
                  cell ids that make up the spatial pattern
                  selected gene
gene_name
spatial_prob
                  probability for a high expressing gene value to be part of the spatial pattern
gradient_direction
                  direction of gradient
                  show the discrete spatial pattern
show_pattern
pattern_colors 2 color vector for the spatial pattern
                  additional parameters for (re-)normalizing
. . .
```

#### Value

Reprocessed Giotto object for which one gene has a forced spatial pattern

228 spark

#### **Description**

Compute spatially expressed genes with SPARK method

## Usage

```
spark(
  gobject,
  percentage = 0.1,
  min_count = 10,
  expression_values = "raw",
  num_core = 5,
  covariates = NULL,
  return_object = c("data.table", "spark"),
  ...
)
```

#### **Arguments**

```
gobject
                  giotto object
percentage
                  The percentage of cells that are expressed for analysis
min_count
                  minimum number of counts for a gene to be included
expression_values
                  type of values to use (raw by default)
                  number of cores to use
num_core
                  The covariates in experiments, i.e. confounding factors/batch effect. Column
covariates
                  name of giotto cell metadata.
                  type of result to return (data.table or spark object)
return_object
                  Additional parameters to the spark.vc function
```

#### **Details**

This function is a wrapper for the method implemented in the SPARK package:

- 1. CreateSPARKObject create a SPARK object from a Giotto object
- 2. spark.vc Fits the count-based spatial model to estimate the parameters, see spark.vc for additional parameters
- 3. spark.test Testing multiple kernel matrices

#### Value

data.table with SPARK spatial genes results or the SPARK object

spatCellCellcom 229

spatCellCellcom spatCellCellcom

#### **Description**

Spatial Cell-Cell communication scores based on spatial expression of interacting cells

#### Usage

```
spatCellCellcom(
 gobject,
  spatial_network_name = "Delaunay_network",
  cluster_column = "cell_types",
 random_iter = 1000,
  gene_set_1,
  gene_set_2,
  log2FC_addendum = 0.1,
 min_observations = 2,
 detailed = FALSE,
 adjust_method = c("fdr", "bonferroni", "BH", "holm", "hochberg", "hommel", "BY",
    "none"),
  adjust_target = c("genes", "cells"),
 do_parallel = TRUE,
  cores = NA,
  set_seed = TRUE,
 seed_number = 1234,
  verbose = c("a little", "a lot", "none")
```

```
gobject
                  giotto object to use
spatial_network_name
                  spatial network to use for identifying interacting cells
cluster_column cluster column with cell type information
                  number of iterations
random_iter
                  first specific gene set from gene pairs
gene_set_1
gene_set_2
                  second specific gene set from gene pairs
log2FC_addendum
                  addendum to add when calculating log2FC
min_observations
                  minimum number of interactions needed to be considered
detailed
                  provide more detailed information (random variance and z-score)
                  which method to adjust p-values
adjust_method
adjust_target
                  adjust multiple hypotheses at the cell or gene level
                  run calculations in parallel with mclapply
do_parallel
                  number of cores to use if do_parallel = TRUE
cores
```

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set\_seed set a seed for reproducibility

seed\_number seed number

verbose verbose

#### **Details**

Statistical framework to identify if pairs of genes (such as ligand-receptor combinations) are expressed at higher levels than expected based on a reshuffled null distribution of gene expression values in cells that are spatially in proximity to eachother..

- LR\_comb:Pair of ligand and receptor
- lig\_cell\_type: cell type to assess expression level of ligand
- lig\_expr: average expression of ligand in lig\_cell\_type
- · ligand: ligand name
- rec\_cell\_type: cell type to assess expression level of receptor
- rec\_expr: average expression of receptor in rec\_cell\_type
- receptor: receptor name
- LR\_expr: combined average ligand and receptor expression
- lig\_nr: total number of cells from lig\_cell\_type that spatially interact with cells from rec\_cell\_type
- rec\_nr: total number of cells from rec\_cell\_type that spatially interact with cells from lig\_cell\_type
- rand\_expr: average combined ligand and receptor expression from random spatial permutations
- av\_diff: average difference between LR\_expr and rand\_expr over all random spatial permutations
- sd\_diff: (optional) standard deviation of the difference between LR\_expr and rand\_expr over all random spatial permutations
- z\_score: (optinal) z-score
- log2fc: log2 fold-change (LR\_expr/rand\_expr)
- pvalue: p-value
- LR\_cell\_comb: cell type pair combination
- p.adj: adjusted p-value
- PI: significanc score: log2fc \* -log10(p.adj)

## Value

Cell-Cell communication scores for gene pairs based on spatial interaction

spatCellPlot 231

spatCellPlot

spatCellPlot

#### **Description**

Visualize cells according to spatial coordinates

#### Usage

```
spatCellPlot(...)
```

```
Arguments passed on to spatCellPlot2D
gobject giotto object
show_image show a tissue background image
gimage a giotto image
image_name name of a giotto image
sdimx x-axis dimension name (default = 'sdimx')
sdimy y-axis dimension name (default = 'sdimy')
spat_enr_names names of spatial enrichment results to include
cell_annotation_values numeric cell annotation columns
cell_color_gradient vector with 3 colors for numeric data
gradient_midpoint midpoint for color gradient
gradient_limits vector with lower and upper limits
select_cell_groups select subset of cells/clusters based on cell_color param-
select_cells select subset of cells based on cell IDs
point_shape shape of points (border, no_border or voronoi)
point_size size of point (cell)
point_alpha transparancy of spatial points
point_border_col color of border around points
point_border_stroke stroke size of border around points
show_cluster_center plot center of selected clusters
show_center_label plot label of selected clusters
center_point_size size of center points
center_point_border_col border color of center points
center_point_border_stroke border stroke size of center points
label_size size of labels
label_fontface font of labels
show_network show underlying spatial network
spatial_network_name name of spatial network to use
network_color color of spatial network
network_alpha alpha of spatial network
show_grid show spatial grid
spatial_grid_name name of spatial grid to use
```

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```
grid_color color of spatial grid
show_other_cells display not selected cells
other_cell_color color of not selected cells
other_point_size point size of not selected cells
other_cells_alpha alpha of not selected cells
coord_fix_ratio fix ratio between x and y-axis
show_legend show legend
legend_text size of legend text
legend_symbol_size size of legend symbols
background_color color of plot background
vor_border_color border colorr for voronoi plot
vor_max_radius maximum radius for voronoi 'cells'
vor_alpha transparancy of voronoi 'cells'
axis_text size of axis text
axis_title size of axis title
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_plot show plot
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
default_save_name default save name for saving, don't change, change save_name
    in save_param
```

## Details

Description of parameters.

#### Value

ggplot

#### See Also

Other spatial cell annotation visualizations: spatCellPlot2D()

#### **Examples**

```
cell_annotation_values = c('1','2'))
```

spatCellPlot2D

spatCellPlot2D

#### **Description**

Visualize cells according to spatial coordinates

## Usage

```
spatCellPlot2D(
 gobject,
 show_image = F,
  gimage = NULL,
  image_name = "image",
  sdimx = "sdimx",
  sdimy = "sdimy",
  spat_enr_names = NULL,
 cell_annotation_values = NULL,
 cell_color_gradient = c("blue", "white", "red"),
 gradient_midpoint = NULL,
 gradient_limits = NULL,
  select_cell_groups = NULL,
  select_cells = NULL,
 point_shape = c("border", "no_border", "voronoi"),
 point_size = 3,
 point_alpha = 1,
 point_border_col = "black",
 point_border_stroke = 0.1,
  show_cluster_center = F,
  show_center_label = F,
  center_point_size = 4,
  center_point_border_col = "black",
  center_point_border_stroke = 0.1,
  label_size = 4,
  label_fontface = "bold",
  show_network = F,
  spatial_network_name = "Delaunay_network",
 network_color = NULL,
 network_alpha = 1,
 show_grid = F,
  spatial_grid_name = "spatial_grid",
 grid_color = NULL,
  show_other_cells = T,
 other_cell_color = "lightgrey",
 other_point_size = 1,
 other_cells_alpha = 0.1,
  coord_fix_ratio = NULL,
  show_legend = T,
```

```
legend_text = 8,
 legend_symbol_size = 1,
 background_color = "white",
  vor_border_color = "white",
  vor_max_radius = 200,
  vor_alpha = 1,
 axis_text = 8,
  axis_title = 8,
  cow_n_col = 2,
  cow_rel_h = 1,
  cow_rel_w = 1,
  cow_align = "h",
  show_plot = NA,
 return_plot = NA,
  save_plot = NA,
  save_param = list(),
 default_save_name = "spatCellPlot2D"
)
```

```
giotto object
gobject
                  show a tissue background image
show_image
                  a giotto image
gimage
image_name
                  name of a giotto image
sdimx
                  x-axis dimension name (default = 'sdimx')
                  y-axis dimension name (default = 'sdimy')
sdimv
spat_enr_names names of spatial enrichment results to include
cell_annotation_values
                  numeric cell annotation columns
cell_color_gradient
                  vector with 3 colors for numeric data
gradient_midpoint
                  midpoint for color gradient
gradient_limits
                  vector with lower and upper limits
select_cell_groups
                  select subset of cells/clusters based on cell_color parameter
                  select subset of cells based on cell IDs
select_cells
                  shape of points (border, no_border or voronoi)
point_shape
point_size
                  size of point (cell)
point_alpha
                  transparancy of spatial points
point_border_col
                  color of border around points
point_border_stroke
                  stroke size of border around points
show_cluster_center
                  plot center of selected clusters
```

show\_center\_label plot label of selected clusters center\_point\_size size of center points center\_point\_border\_col border color of center points center\_point\_border\_stroke border stroke size of center points label\_size size of labels label\_fontface font of labels show\_network show underlying spatial network spatial\_network\_name name of spatial network to use color of spatial network network\_color network\_alpha alpha of spatial network show\_grid show spatial grid spatial\_grid\_name name of spatial grid to use color of spatial grid grid\_color show\_other\_cells display not selected cells other\_cell\_color color of not selected cells other\_point\_size point size of not selected cells other\_cells\_alpha alpha of not selected cells coord\_fix\_ratio fix ratio between x and y-axis show\_legend show legend legend\_text size of legend text legend\_symbol\_size size of legend symbols

background\_color color of plot background

vor\_border\_color

border colorr for voronoi plot

vor\_max\_radius maximum radius for voronoi 'cells' vor\_alpha transparancy of voronoi 'cells'

axis\_text size of axis text axis\_title size of axis title

cow\_n\_col cowplot param: how many columns
cow\_rel\_h cowplot param: relative height
cow\_rel\_w cowplot param: relative width
cow\_align cowplot param: how to align

#### **Details**

Description of parameters.

#### Value

ggplot

#### See Also

Other spatial cell annotation visualizations: spatCellPlot()

# **Examples**

spatDimCellPlot

spatDimCellPlot

## **Description**

Visualize numerical features of cells according to spatial AND dimension reduction coordinates in 2D

# Usage

```
spatDimCellPlot(...)
```

### **Arguments**

Arguments passed on to spatDimCellPlot2D gobject giotto object show\_image show a tissue background image gimage a giotto image image\_name name of a giotto image plot\_alignment direction to align plot spat\_enr\_names names of spatial enrichment results to include cell\_annotation\_values numeric cell annotation columns dim\_reduction\_to\_use dimension reduction to use dim\_reduction\_name dimension reduction name dim1\_to\_use dimension to use on x-axis dim2\_to\_use dimension to use on y-axis sdimx = spatial dimension to use on x-axissdimy = spatial dimension to use on y-axis cell\_color\_gradient vector with 3 colors for numeric data gradient\_midpoint midpoint for color gradient gradient\_limits vector with lower and upper limits select\_cell\_groups select subset of cells/clusters based on cell\_color paramselect cells select subset of cells based on cell IDs dim\_point\_shape dim reduction points with border or not (border or no\_border) dim\_point\_size size of points in dim. reduction space dim\_point\_alpha transparancy of dim. reduction points dim\_point\_border\_col border color of points in dim. reduction space dim\_point\_border\_stroke border stroke of points in dim. reduction space spat\_point\_shape shape of points (border, no\_border or voronoi) spat\_point\_size size of spatial points spat\_point\_alpha transparancy of spatial points spat\_point\_border\_col border color of spatial points spat\_point\_border\_stroke border stroke of spatial points dim\_show\_cluster\_center show the center of each cluster dim\_show\_center\_label provide a label for each cluster dim\_center\_point\_size size of the center point dim\_center\_point\_border\_col border color of center point dim\_center\_point\_border\_stroke stroke size of center point dim\_label\_size size of the center label dim\_label\_fontface font of the center label spat\_show\_cluster\_center show the center of each cluster spat\_show\_center\_label provide a label for each cluster spat\_center\_point\_size size of the spatial center points spat\_center\_point\_border\_col border color of the spatial center points spat\_center\_point\_border\_stroke stroke size of the spatial center points spat\_label\_size size of the center label spat\_label\_fontface font of the center label

```
show_NN_network show underlying NN network
nn_network_to_use type of NN network to use (kNN vs sNN)
nn_network_name name of NN network to use, if show_NN_network = TRUE
dim_edge_alpha column to use for alpha of the edges
spat_show_network show spatial network
spatial_network_name name of spatial network to use
spat_network_color color of spatial network
spat_network_alpha alpha of spatial network
spat_show_grid show spatial grid
spatial_grid_name name of spatial grid to use
spat_grid_color color of spatial grid
show_other_cells display not selected cells
other_cell_color color of not selected cells
dim_other_point_size size of not selected dim cells
spat_other_point_size size of not selected spat cells
spat_other_cells_alpha alpha of not selected spat cells
coord_fix_ratio ratio for coordinates
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_legend show legend
legend_text size of legend text
legend_symbol_size size of legend symbols
dim_background_color background color of points in dim. reduction space
spat_background_color background color of spatial points
vor_border_color border colorr for voronoi plot
vor_max_radius maximum radius for voronoi 'cells'
vor_alpha transparancy of voronoi 'cells'
axis_text size of axis text
axis_title size of axis title
show_plot show plot
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
{\tt default\_save\_name} \ \ default \ save \ name \ for \ saving, \ don't \ change, \ change \ save\_name
    in save_param
```

## **Details**

Description of parameters.

# Value

ggplot

#### See Also

Other spatial and dimension reduction cell annotation visualizations: spatDimCellPlot2D()

#### **Examples**

spatDimCellPlot2D

spatDimCellPlot2D

## **Description**

Visualize numerical features of cells according to spatial AND dimension reduction coordinates in 2D

## Usage

```
spatDimCellPlot2D(
 gobject,
  show_image = F,
 gimage = NULL,
  image_name = "image",
  plot_alignment = c("vertical", "horizontal"),
  spat_enr_names = NULL,
  cell_annotation_values = NULL,
 dim_reduction_to_use = "umap",
 dim_reduction_name = "umap",
  dim1_to_use = 1,
  dim2_to_use = 2,
  sdimx = "sdimx",
  sdimy = "sdimy"
  cell_color_gradient = c("blue", "white", "red"),
  gradient_midpoint = NULL,
 gradient_limits = NULL,
  select_cell_groups = NULL,
  select_cells = NULL,
 dim_point_shape = c("border", "no_border"),
 dim_point_size = 1,
  dim_point_alpha = 1,
  dim_point_border_col = "black",
 dim_point_border_stroke = 0.1,
  spat_point_shape = c("border", "no_border", "voronoi"),
  spat_point_size = 1,
```

```
spat_point_alpha = 1,
spat_point_border_col = "black",
spat_point_border_stroke = 0.1,
dim_show_cluster_center = F,
dim_show_center_label = T,
dim_center_point_size = 4,
dim_center_point_border_col = "black",
dim_center_point_border_stroke = 0.1,
dim_label_size = 4,
dim_label_fontface = "bold",
spat_show_cluster_center = F,
spat_show_center_label = F,
spat_center_point_size = 4,
spat_center_point_border_col = "black",
spat_center_point_border_stroke = 0.1,
spat_label_size = 4,
spat_label_fontface = "bold",
show_NN_network = F,
nn_network_to_use = "sNN",
nn_network_name = "sNN.pca",
dim_edge_alpha = 0.5,
spat_show_network = F,
spatial_network_name = "Delaunay_network",
spat_network_color = "red",
spat_network_alpha = 0.5,
spat_show_grid = F,
spatial_grid_name = "spatial_grid",
spat_grid_color = "green",
show_other_cells = TRUE,
other_cell_color = "grey";
dim_other_point_size = 0.5,
spat_other_point_size = 0.5,
spat_other_cells_alpha = 0.5,
show_legend = T,
legend_text = 8,
legend_symbol_size = 1,
dim_background_color = "white",
spat_background_color = "white",
vor_border_color = "white",
vor_max_radius = 200,
vor_alpha = 1,
axis_text = 8,
axis_title = 8,
coord_fix_ratio = NULL,
cow_n_col = 2,
cow_rel_h = 1,
cow_rel_w = 1,
cow_align = "h",
show_plot = NA,
return_plot = NA,
save_plot = NA,
save_param = list(),
```

```
default_save_name = "spatDimCellPlot2D"
)
```

#### **Arguments**

giotto object gobject show\_image show a tissue background image gimage a giotto image image\_name name of a giotto image plot\_alignment direction to align plot spat\_enr\_names names of spatial enrichment results to include cell\_annotation\_values numeric cell annotation columns dim\_reduction\_to\_use dimension reduction to use dim\_reduction\_name dimension reduction name dim1\_to\_use dimension to use on x-axis dim2\_to\_use dimension to use on y-axis sdimx = spatial dimension to use on x-axis = spatial dimension to use on y-axis sdimy cell\_color\_gradient vector with 3 colors for numeric data gradient\_midpoint midpoint for color gradient gradient\_limits vector with lower and upper limits select\_cell\_groups select subset of cells/clusters based on cell\_color parameter select cells select subset of cells based on cell IDs dim\_point\_shape dim reduction points with border or not (border or no\_border) dim\_point\_size size of points in dim. reduction space dim\_point\_alpha transparancy of dim. reduction points dim\_point\_border\_col border color of points in dim. reduction space dim\_point\_border\_stroke border stroke of points in dim. reduction space spat\_point\_shape shape of points (border, no\_border or voronoi) spat\_point\_size size of spatial points spat\_point\_alpha transparancy of spatial points spat\_point\_border\_col

border color of spatial points

spat\_point\_border\_stroke border stroke of spatial points dim\_show\_cluster\_center show the center of each cluster dim\_show\_center\_label provide a label for each cluster dim\_center\_point\_size size of the center point dim\_center\_point\_border\_col border color of center point dim\_center\_point\_border\_stroke stroke size of center point dim\_label\_size size of the center label dim\_label\_fontface font of the center label spat\_show\_cluster\_center show the center of each cluster spat\_show\_center\_label provide a label for each cluster spat\_center\_point\_size size of the spatial center points spat\_center\_point\_border\_col border color of the spatial center points spat\_center\_point\_border\_stroke stroke size of the spatial center points spat\_label\_size size of the center label spat\_label\_fontface font of the center label show\_NN\_network show underlying NN network nn\_network\_to\_use type of NN network to use (kNN vs sNN) nn\_network\_name name of NN network to use, if show\_NN\_network = TRUE dim\_edge\_alpha column to use for alpha of the edges spat\_show\_network show spatial network spatial\_network\_name name of spatial network to use spat\_network\_color color of spatial network spat\_network\_alpha alpha of spatial network spat\_show\_grid show spatial grid spatial\_grid\_name name of spatial grid to use

spat\_grid\_color

color of spatial grid

```
show_other_cells
                  display not selected cells
other_cell_color
                  color of not selected cells
dim_other_point_size
                  size of not selected dim cells
spat_other_point_size
                  size of not selected spat cells
spat_other_cells_alpha
                  alpha of not selected spat cells
show_legend
                  show legend
legend_text
                  size of legend text
legend_symbol_size
                  size of legend symbols
dim_background_color
                  background color of points in dim. reduction space
spat_background_color
                  background color of spatial points
vor_border_color
                  border colorr for voronoi plot
vor_max_radius maximum radius for voronoi 'cells'
vor_alpha
                  transparancy of voronoi 'cells'
axis_text
                  size of axis text
axis_title
                  size of axis title
coord_fix_ratio
                  ratio for coordinates
                  cowplot param: how many columns
cow_n_col
                  cowplot param: relative height
cow_rel_h
                  cowplot param: relative width
cow_rel_w
cow_align
                  cowplot param: how to align
show_plot
                  show plot
return_plot
                  return ggplot object
                  directly save the plot [boolean]
save_plot
                  list of saving parameters, see showSaveParameters
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

## **Details**

Description of parameters.

#### Value

ggplot

#### See Also

Other spatial and dimension reduction cell annotation visualizations: spatDimCellPlot()

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#### **Examples**

spatDimGenePlot

spatDimGenePlot

## **Description**

Visualize cells according to spatial AND dimension reduction coordinates in ggplot mode

## Usage

```
spatDimGenePlot(...)
```

```
Arguments passed on to spatDimGenePlot2D
gobject giotto object
show_image show a tissue background image
gimage a giotto image
image_name name of a giotto image
expression_values gene expression values to use
plot_alignment direction to align plot
genes genes to show
dim_reduction_to_use dimension reduction to use
dim_reduction_name dimension reduction name
dim1_to_use dimension to use on x-axis
dim2_to_use dimension to use on y-axis
dim_point_shape dim reduction points with border or not (border or no_border)
dim_point_size dim reduction plot: point size
dim_point_alpha transparancy of dim. reduction points
dim_point_border_col color of border around points
dim_point_border_stroke stroke size of border around points
show_NN_network show underlying NN network
show_spatial_network show underlying spatial netwok
nn_network_to_use type of NN network to use (kNN vs sNN)
```

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```
network_name name of NN network to use, if show NN network = TRUE
dim_network_color color of NN network
dim_edge_alpha dim reduction plot: column to use for alpha of the edges
scale_alpha_with_expression scale expression with ggplot alpha parameter
sdimx spatial x-axis dimension name (default = 'sdimx')
sdimy spatial y-axis dimension name (default = 'sdimy')
spatial_network_name name of spatial network to use
spatial_network_color color of spatial network
show_spatial_grid show spatial grid
grid_color color of spatial grid
spatial_grid_name name of spatial grid to use
spat_point_shape spatial points with border or not (border or no_border)
spat_point_size spatial plot: point size
spat_point_alpha transparancy of spatial points
spat_point_border_col color of border around points
spat_point_border_stroke stroke size of border around points
spat_edge_alpha edge alpha
cell_color_gradient vector with 3 colors for numeric data
gradient_midpoint midpoint for color gradient
gradient_limits vector with lower and upper limits
show_legend show legend
legend_text size of legend text
dim_background_color color of plot background for dimension plot
spat_background_color color of plot background for spatial plot
vor_border_color border colorr for voronoi plot
vor max radius maximum radius for voronoi 'cells'
vor_alpha transparancy of voronoi 'cells'
axis_text size of axis text
axis_title size of axis title
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_plot show plots
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
default_save_name default save name for saving, don't change, change save_name
    in save_param
```

#### **Details**

Description of parameters.

#### Value

ggplot

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#### See Also

```
spatDimGenePlot3D
```

Other spatial and dimension reduction gene expression visualizations: spatDimGenePlot2D(), spatDimGenePlot3D()

### **Examples**

spatDimGenePlot2D

spatDimGenePlot2D

## Description

Visualize cells according to spatial AND dimension reduction coordinates in ggplot mode

## Usage

```
spatDimGenePlot2D(
 gobject,
  show_image = F,
 gimage = NULL,
  image_name = "image",
 expression_values = c("normalized", "scaled", "custom"),
 plot_alignment = c("vertical", "horizontal"),
  genes,
 dim_reduction_to_use = "umap",
 dim_reduction_name = "umap",
 dim1_to_use = 1,
 dim2_to_use = 2,
 dim_point_shape = c("border", "no_border"),
 dim_point_size = 1,
 dim_point_alpha = 1,
 dim_point_border_col = "black",
 dim_point_border_stroke = 0.1,
  show_NN_network = F,
  show_spatial_network = F,
 dim_network_color = "gray",
 nn_network_to_use = "sNN",
 network_name = "sNN.pca",
 dim_edge_alpha = NULL,
  scale_alpha_with_expression = FALSE,
  sdimx = "sdimx",
  sdimy = "sdimy",
```

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```
spatial_network_name = "Delaunay_network",
spatial_network_color = NULL,
show_spatial_grid = F,
grid_color = NULL,
spatial_grid_name = "spatial_grid",
spat_point_shape = c("border", "no_border", "voronoi"),
spat_point_size = 1,
spat_point_alpha = 1,
spat_point_border_col = "black",
spat_point_border_stroke = 0.1,
spat_edge_alpha = NULL,
cell_color_gradient = c("blue", "white", "red"),
gradient_midpoint = NULL,
gradient_limits = NULL,
cow_n_col = 2,
cow_rel_h = 1,
cow_rel_w = 1,
cow_align = "h",
show_legend = T,
legend_text = 8,
dim_background_color = "white",
spat_background_color = "white",
vor_border_color = "white",
vor_max_radius = 200,
vor_alpha = 1,
axis_text = 8,
axis_title = 8,
show_plot = NA,
return_plot = NA,
save_plot = NA,
save_param = list(),
default_save_name = "spatDimGenePlot2D"
```

```
giotto object
gobject
show_image
                 show a tissue background image
gimage
                 a giotto image
image_name
                 name of a giotto image
expression_values
                 gene expression values to use
plot_alignment direction to align plot
                 genes to show
genes
dim_reduction_to_use
                 dimension reduction to use
dim_reduction_name
                 dimension reduction name
dim1_to_use
                 dimension to use on x-axis
dim2_to_use
                 dimension to use on y-axis
```

dim\_point\_shape dim reduction points with border or not (border or no\_border) dim\_point\_size dim reduction plot: point size dim\_point\_alpha transparancy of dim. reduction points dim\_point\_border\_col color of border around points dim\_point\_border\_stroke stroke size of border around points show\_NN\_network show underlying NN network show\_spatial\_network show underlying spatial netwok dim\_network\_color color of NN network nn\_network\_to\_use type of NN network to use (kNN vs sNN) name of NN network to use, if show\_NN\_network = TRUE network\_name dim\_edge\_alpha dim reduction plot: column to use for alpha of the edges scale\_alpha\_with\_expression scale expression with ggplot alpha parameter sdimx spatial x-axis dimension name (default = 'sdimx') sdimy spatial y-axis dimension name (default = 'sdimy') spatial\_network\_name name of spatial network to use spatial\_network\_color color of spatial network show\_spatial\_grid show spatial grid grid\_color color of spatial grid spatial\_grid\_name name of spatial grid to use spat\_point\_shape spatial points with border or not (border or no\_border) spat\_point\_size spatial plot: point size spat\_point\_alpha transparancy of spatial points spat\_point\_border\_col color of border around points spat\_point\_border\_stroke stroke size of border around points spat\_edge\_alpha edge alpha cell\_color\_gradient vector with 3 colors for numeric data

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```
gradient_midpoint
                  midpoint for color gradient
gradient_limits
                  vector with lower and upper limits
cow_n_col
                  cowplot param: how many columns
                  cowplot param: relative height
cow_rel_h
cow_rel_w
                  cowplot param: relative width
cow_align
                  cowplot param: how to align
show_legend
                  show legend
legend_text
                  size of legend text
dim_background_color
                  color of plot background for dimension plot
spat_background_color
                  color of plot background for spatial plot
vor_border_color
                  border colorr for voronoi plot
vor_max_radius maximum radius for voronoi 'cells'
vor_alpha
                  transparancy of voronoi 'cells'
axis_text
                  size of axis text
axis_title
                  size of axis title
                  show plots
show_plot
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
                  list of saving parameters, see showSaveParameters
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

#### **Details**

Description of parameters.

#### Value

ggplot

## See Also

```
spatDimGenePlot3D
```

Other spatial and dimension reduction gene expression visualizations: spatDimGenePlot3D(), spatDimGenePlot()

### **Examples**

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spatDimGenePlot3D

spatDimGenePlot3D

## **Description**

Visualize cells according to spatial AND dimension reduction coordinates in ggplot mode

## Usage

```
spatDimGenePlot3D(
 gobject,
 expression_values = c("normalized", "scaled", "custom"),
 plot_alignment = c("horizontal", "vertical"),
 dim_reduction_to_use = "umap",
 dim_reduction_name = "umap",
 dim1_to_use = 1,
 dim2_to_use = 2,
 dim3_to_use = NULL,
  sdimx = "sdimx",
  sdimy = "sdimy",
  sdimz = "sdimz",
 genes,
 cluster_column = NULL,
  select_cell_groups = NULL,
  select_cells = NULL,
  show_other_cells = T,
 other_cell_color = "lightgrey",
 other_point_size = 1.5,
  show_NN_network = FALSE,
 nn_network_to_use = "sNN",
 nn_network_color = "lightgrey",
  nn_network_alpha = 0.5,
 network_name = "sNN.pca",
 label_size = 16,
  genes_low_color = "blue",
  genes_mid_color = "white",
 genes_high_color = "red",
 dim_point_size = 3,
  show_spatial_network = FALSE,
  spatial_network_name = "Delaunay_network",
  spatial_network_color = "lightgray",
  spatial_network_alpha = 0.5,
  show_spatial_grid = FALSE,
  spatial_grid_name = "spatial_grid",
  spatial_grid_color = NULL,
  spatial_grid_alpha = 0.5,
  spatial_point_size = 3,
  legend_text_size = 12,
  axis_scale = c("cube", "real", "custom"),
  custom_ratio = NULL,
  x_{ticks} = NULL,
```

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```
y_ticks = NULL,
      z_ticks = NULL,
      show_plot = NA,
      return_plot = NA,
      save_plot = NA,
      save_param = list(),
      default_save_name = "spatDimGenePlot3D"
Arguments
    gobject
                     giotto object
    expression_values
                     gene expression values to use
    plot_alignment direction to align plot
    dim_reduction_to_use
                     dimension reduction to use
    dim_reduction_name
                     dimension reduction name
                     dimension to use on x-axis
    dim1_to_use
    dim2_to_use
                     dimension to use on y-axis
    dim3_to_use
                     dimension to use on z-axis
    sdimx
                     spatial dimension to use on x-axis
    sdimy
                     spatial dimension to use on y-axis
    sdimz
                     spatial dimension to use on z-axis
                     genes to show
    genes
    cluster_column cluster column to select groups
    select_cell_groups
                     select subset of cells/clusters based on cell_color parameter
                     select subset of cells based on cell IDs
    select_cells
    show_other_cells
                     display not selected cells
    other_cell_color
                     color of not selected cells
    other_point_size
                     size of not selected cells
    show_NN_network
                     show underlying NN network
    nn_network_to_use
                     type of NN network to use (kNN vs sNN)
    nn_network_color
                     color of NN network
    nn_network_alpha
                     alpha of NN network
                     name of NN network to use, if show_NN_network = TRUE
    network_name
    label_size
                     size of labels
    genes_low_color
                     color for low expression levels
```

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```
genes_mid_color
                  color for medium expression levels
genes_high_color
                  color for high expression levels
dim_point_size dim reduction plot: point size
show_spatial_network
                  show spatial network (boolean)
spatial_network_name
                  name of spatial network to use
spatial_network_color
                  color of spatial network
spatial_network_alpha
                  alpha of spatial network
show_spatial_grid
                  show spatial grid (boolean)
spatial_grid_name
                  name of spatial grid to use
spatial_grid_color
                  color of spatial grid
spatial_grid_alpha
                  alpha of spatial grid
spatial_point_size
                  spatial plot: point size
legend_text_size
                  size of legend
axis_scale
                  the way to scale the axis
custom_ratio
                  customize the scale of the plot
                  set the number of ticks on the x-axis
x_ticks
                  set the number of ticks on the y-axis
y_ticks
z_ticks
                  set the number of ticks on the z-axis
                  show plots
show_plot
return_plot
                  return plotly object
save_plot
                  directly save the plot [boolean]
                  list of saving parameters, see showSaveParameters
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

## **Details**

Description of parameters.

#### Value

plotly

## See Also

Other spatial and dimension reduction gene expression visualizations: spatDimGenePlot2D(), spatDimGenePlot()

spatDimPlot

spatDimPlot

#### **Description**

Visualize cells according to spatial AND dimension reduction coordinates 2D

#### Usage

```
spatDimPlot(...)
```

# Arguments

```
Arguments passed on to spatDimPlot2D
gobject giotto object
show_image show a tissue background image
gimage a giotto image
image_name name of a giotto image
plot_alignment direction to align plot
dim_reduction_to_use dimension reduction to use
dim_reduction_name dimension reduction name
dim1_to_use dimension to use on x-axis
dim2_to_use dimension to use on y-axis
sdimx = spatial dimension to use on x-axis
sdimy = spatial dimension to use on y-axis
spat_enr_names names of spatial enrichment results to include
cell_color color for cells (see details)
color_as_factor convert color column to factor
cell_color_code named vector with colors
cell_color_gradient vector with 3 colors for numeric data
gradient_midpoint midpoint for color gradient
gradient_limits vector with lower and upper limits
select_cell_groups select subset of cells/clusters based on cell_color param-
    eter
select_cells select subset of cells based on cell IDs
dim_point_shape point with border or not (border or no_border)
dim_point_size size of points in dim. reduction space
dim_point_alpha transparancy of point in dim. reduction space
dim_point_border_col border color of points in dim. reduction space
dim_point_border_stroke border stroke of points in dim. reduction space
spat_point_shape shape of points (border, no_border or voronoi)
spat_point_size size of spatial points
spat_point_alpha transparancy of spatial points
spat_point_border_col border color of spatial points
spat_point_border_stroke border stroke of spatial points
dim_show_cluster_center show the center of each cluster
```

```
dim_show_center_label provide a label for each cluster
dim_center_point_size size of the center point
dim_center_point_border_col border color of center point
dim_center_point_border_stroke stroke size of center point
dim_label_size size of the center label
dim_label_fontface font of the center label
spat_show_cluster_center show the center of each cluster
spat_show_center_label provide a label for each cluster
spat_center_point_size size of the center point
spat_center_point_border_col border color of spatial center points
spat_center_point_border_stroke border strike size of spatial center points
spat_label_size size of the center label
spat_label_fontface font of the center label
show_NN_network show underlying NN network
nn_network_to_use type of NN network to use (kNN vs sNN)
network_name name of NN network to use, if show NN network = TRUE
nn_network_alpha column to use for alpha of the edges
show_spatial_network show spatial network
spat_network_name name of spatial network to use
spat_network_color color of spatial network
spat_network_alpha alpha of spatial network
show_spatial_grid show spatial grid
spat_grid_name name of spatial grid to use
spat_grid_color color of spatial grid
show_other_cells display not selected cells
other_cell_color color of not selected cells
dim_other_point_size size of not selected dim cells
spat_other_point_size size of not selected spat cells
spat_other_cells_alpha alpha of not selected spat cells
dim_show_legend show legend of dimension reduction plot
spat_show_legend show legend of spatial plot
legend_text size of legend text
legend_symbol_size size of legend symbols
dim_background_color background color of points in dim. reduction space
spat_background_color background color of spatial points
vor_border_color border colorr for voronoi plot
vor_max_radius maximum radius for voronoi 'cells'
vor_alpha transparancy of voronoi 'cells'
axis_text size of axis text
axis_title size of axis title
show_plot show plot
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
default_save_name default save name for saving, don't change, change save_name
    in save_param
```

#### **Details**

Description of parameters.

#### Value

ggplot

## See Also

```
spatDimPlot2D and spatDimPlot3D for 3D visualization.
```

Other spatial and dimension reduction visualizations: spatDimPlot2D(), spatDimPlot3D()

## **Examples**

spatDimPlot2D

spatDimPlot2D

## **Description**

Visualize cells according to spatial AND dimension reduction coordinates 2D

#### Usage

```
spatDimPlot2D(
 gobject,
  show_image = F,
 gimage = NULL,
  image_name = "image",
 plot_alignment = c("vertical", "horizontal"),
 dim_reduction_to_use = "umap",
 dim_reduction_name = "umap",
 dim1_to_use = 1,
 dim2\_to\_use = 2,
  sdimx = "sdimx",
  sdimy = "sdimy",
  spat_enr_names = NULL,
 cell_color = NULL,
 color_as_factor = T,
 cell_color_code = NULL,
 cell_color_gradient = c("blue", "white", "red"),
  gradient_midpoint = NULL,
 gradient_limits = NULL,
  select_cell_groups = NULL,
  select_cells = NULL,
```

```
dim_point_shape = c("border", "no_border"),
dim_point_size = 1,
dim_point_alpha = 1,
dim_point_border_col = "black",
dim_point_border_stroke = 0.1,
spat_point_shape = c("border", "no_border", "voronoi"),
spat_point_size = 1,
spat_point_alpha = 1,
spat_point_border_col = "black",
spat_point_border_stroke = 0.1,
dim_show_cluster_center = F,
dim_show_center_label = T,
dim_center_point_size = 4,
dim_center_point_border_col = "black",
dim_center_point_border_stroke = 0.1,
dim_label_size = 4,
dim_label_fontface = "bold",
spat_show_cluster_center = F,
spat_show_center_label = F,
spat_center_point_size = 4,
spat_center_point_border_col = "blue",
spat_center_point_border_stroke = 0.1,
spat_label_size = 4,
spat_label_fontface = "bold",
show_NN_network = F,
nn_network_to_use = "sNN",
network_name = "sNN.pca",
nn_network_alpha = 0.05,
show_spatial_network = F,
spat_network_name = "Delaunay_network",
spat_network_color = "blue",
spat_network_alpha = 0.5,
show_spatial_grid = F,
spat_grid_name = "spatial_grid",
spat_grid_color = "blue",
show_other_cells = T,
other_cell_color = "lightgrey",
dim_other_point_size = 1,
spat_other_point_size = 1,
spat_other_cells_alpha = 0.5,
dim\_show\_legend = F,
spat_show_legend = F,
legend_text = 8,
legend_symbol_size = 1,
dim_background_color = "white",
spat_background_color = "white",
vor_border_color = "white",
vor_max_radius = 200,
vor_alpha = 1,
axis_text = 8,
axis_title = 8,
show_plot = NA,
```

```
return_plot = NA,
save_plot = NA,
save_param = list(),
default_save_name = "spatDimPlot2D"
)
```

## **Arguments**

gobject giotto object show a tissue background image show\_image a giotto image gimage image\_name name of a giotto image plot\_alignment direction to align plot dim\_reduction\_to\_use dimension reduction to use dim\_reduction\_name dimension reduction name dim1\_to\_use dimension to use on x-axis dim2\_to\_use dimension to use on y-axis sdimx = spatial dimension to use on x-axis sdimy = spatial dimension to use on y-axis spat\_enr\_names names of spatial enrichment results to include color for cells (see details) cell\_color color\_as\_factor convert color column to factor cell\_color\_code named vector with colors cell\_color\_gradient vector with 3 colors for numeric data gradient\_midpoint midpoint for color gradient gradient\_limits vector with lower and upper limits select\_cell\_groups select subset of cells/clusters based on cell\_color parameter select subset of cells based on cell IDs select\_cells dim\_point\_shape point with border or not (border or no\_border) dim\_point\_size size of points in dim. reduction space dim\_point\_alpha transparancy of point in dim. reduction space dim\_point\_border\_col border color of points in dim. reduction space dim\_point\_border\_stroke border stroke of points in dim. reduction space spat\_point\_shape shape of points (border, no\_border or voronoi)

spat\_point\_size size of spatial points spat\_point\_alpha transparancy of spatial points spat\_point\_border\_col border color of spatial points spat\_point\_border\_stroke border stroke of spatial points dim\_show\_cluster\_center show the center of each cluster dim\_show\_center\_label provide a label for each cluster dim\_center\_point\_size size of the center point dim\_center\_point\_border\_col border color of center point dim\_center\_point\_border\_stroke stroke size of center point dim\_label\_size size of the center label dim\_label\_fontface font of the center label spat\_show\_cluster\_center show the center of each cluster spat\_show\_center\_label provide a label for each cluster spat\_center\_point\_size size of the center point spat\_center\_point\_border\_col border color of spatial center points spat\_center\_point\_border\_stroke border strike size of spatial center points spat\_label\_size size of the center label spat\_label\_fontface font of the center label show\_NN\_network show underlying NN network

> type of NN network to use (kNN vs sNN) name of NN network to use, if show\_NN\_network = TRUE

nn\_network\_alpha

network\_name

nn\_network\_to\_use

column to use for alpha of the edges

show spatial network spat\_network\_name

show\_spatial\_network

name of spatial network to use

spat\_network\_color

color of spatial network

```
spat_network_alpha
                  alpha of spatial network
show_spatial_grid
                  show spatial grid
spat_grid_name name of spatial grid to use
spat_grid_color
                  color of spatial grid
show_other_cells
                  display not selected cells
other_cell_color
                  color of not selected cells
dim_other_point_size
                  size of not selected dim cells
spat_other_point_size
                  size of not selected spat cells
spat_other_cells_alpha
                  alpha of not selected spat cells
dim_show_legend
                  show legend of dimension reduction plot
spat_show_legend
                  show legend of spatial plot
                  size of legend text
legend_text
legend_symbol_size
                  size of legend symbols
dim_background_color
                  background color of points in dim. reduction space
spat_background_color
                  background color of spatial points
vor_border_color
                  border colorr for voronoi plot
vor_max_radius maximum radius for voronoi 'cells'
vor_alpha
                  transparancy of voronoi 'cells'
axis_text
                  size of axis text
axis_title
                  size of axis title
show_plot
                  show plot
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
                  list of saving parameters, see showSaveParameters
save_param
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

## **Details**

Description of parameters.

#### Value

ggplot

#### See Also

```
spatDimPlot3D
```

Other spatial and dimension reduction visualizations: spatDimPlot3D(), spatDimPlot()

#### **Examples**

spatDimPlot3D

spatDimPlot3D

## **Description**

Visualize cells according to spatial AND dimension reduction coordinates in plotly mode

## Usage

```
spatDimPlot3D(
 gobject,
 plot_alignment = c("horizontal", "vertical"),
 dim_reduction_to_use = "umap",
 dim_reduction_name = "umap",
 dim1_to_use = 1,
 dim2_to_use = 2,
 dim3_to_use = 3,
  sdimx = "sdimx",
  sdimy = "sdimy"
  sdimz = "sdimz",
  spat_enr_names = NULL,
  show_NN_network = FALSE,
 nn_network_to_use = "sNN",
 network_name = "sNN.pca",
 nn_network_color = "lightgray",
 nn_network_alpha = 0.5,
  show_cluster_center = F,
  show_center_label = T,
 center_point_size = 4,
 label_size = 16,
  select_cell_groups = NULL,
  select_cells = NULL,
  show_other_cells = T,
 other_cell_color = "lightgrey",
 other_point_size = 1.5,
  cell_color = NULL,
  color_as_factor = T,
  cell_color_code = NULL,
```

```
dim_point_size = 3,
      show_spatial_network = F,
      spatial_network_name = "Delaunay_network",
      spatial_network_color = "lightgray",
      spatial_network_alpha = 0.5,
      show_spatial_grid = F,
      spatial_grid_name = "spatial_grid",
      spatial_grid_color = NULL,
      spatial_grid_alpha = 0.5,
      spatial_point_size = 3,
      axis_scale = c("cube", "real", "custom"),
     custom_ratio = NULL,
      x_ticks = NULL,
     y_ticks = NULL,
     z_ticks = NULL,
      legend_text_size = 12,
      show_plot = NA,
     return_plot = NA,
     save_plot = NA,
     save_param = list(),
     default_save_name = "spatDimPlot3D"
   )
Arguments
   gobject
                    giotto object
   plot_alignment direction to align plot
   dim_reduction_to_use
                    dimension reduction to use
   dim_reduction_name
                    dimension reduction name
   dim1_to_use
                    dimension to use on x-axis
                    dimension to use on y-axis
   dim2_to_use
   dim3_to_use
                    dimension to use on z-axis
                    = spatial dimension to use on x-axis
   sdimx
                    = spatial dimension to use on y-axis
   sdimy
                    = spatial dimension to use on z-axis
   sdimz
   spat_enr_names names of spatial enrichment results to include
    show_NN_network
                    show underlying NN network
   nn_network_to_use
                    type of NN network to use (kNN vs sNN)
   network_name
                    name of NN network to use, if show_NN_network = TRUE
   nn_network_color
                    color of nn network
   nn_network_alpha
                    column to use for alpha of the edges
   show_cluster_center
```

show the center of each cluster

show\_center\_label provide a label for each cluster center\_point\_size size of the center point label\_size size of the center label select\_cell\_groups select subset of cells/clusters based on cell\_color parameter select\_cells select subset of cells based on cell IDs show\_other\_cells display not selected cells other\_cell\_color color of not selected cells other\_point\_size size of not selected cells color for cells (see details) cell\_color color\_as\_factor convert color column to factor cell\_color\_code named vector with colors dim\_point\_size size of points in dim. reduction space show\_spatial\_network show spatial network spatial\_network\_name name of spatial network to use spatial\_network\_color color of spatial network spatial\_network\_alpha alpha of spatial network show\_spatial\_grid show spatial grid spatial\_grid\_name name of spatial grid to use spatial\_grid\_color color of spatial grid spatial\_grid\_alpha alpha of spatial grid spatial\_point\_size size of spatial points the way to scale the axis axis\_scale customize the scale of the plot custom\_ratio x\_ticks set the number of ticks on the x-axis y\_ticks set the number of ticks on the y-axis set the number of ticks on the z-axis z\_ticks

legend\_text\_size

show\_plot

size of legend

show plot

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```
return_plot return ggplot object
```

save\_plot directly save the plot [boolean]

save\_param list of saving parameters, see showSaveParameters

default\_save\_name

default save name for saving, don't change, change save\_name in save\_param

## **Details**

Description of parameters.

#### Value

plotly

#### See Also

Other spatial and dimension reduction visualizations: spatDimPlot2D(), spatDimPlot()

spatGenePlot spatGenePlot

# Description

Visualize cells and gene expression according to spatial coordinates

## Usage

```
spatGenePlot(...)
```

#### **Arguments**

```
Arguments passed on to spatGenePlot2D
. . .
                 gobject giotto object
                 show_image show a tissue background image
                 gimage a giotto image
                 image_name name of a giotto image
                 sdimx x-axis dimension name (default = 'sdimx')
                 sdimy y-axis dimension name (default = 'sdimy')
                 expression_values gene expression values to use
                 genes genes to show
                 cell_color_gradient vector with 3 colors for numeric data
                 gradient_midpoint midpoint for color gradient
                 gradient_limits vector with lower and upper limits
                 show_network show underlying spatial network
                 network_color color of spatial network
                 spatial_network_name name of spatial network to use
                 edge_alpha alpha of edge
                 show_grid show spatial grid
```

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```
grid_color color of spatial grid
spatial_grid_name name of spatial grid to use
midpoint expression midpoint
scale_alpha_with_expression scale expression with ggplot alpha parameter
point_shape shape of points (border, no_border or voronoi)
point_size size of point (cell)
point_alpha transparancy of points
point_border_col color of border around points
point_border_stroke stroke size of border around points
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_legend show legend
legend_text size of legend text
background_color color of plot background
vor_border_color border colorr for voronoi plot
vor_max_radius maximum radius for voronoi 'cells'
vor_alpha transparancy of voronoi 'cells'
axis_text size of axis text
axis_title size of axis title
show_plot show plots
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
default_save_name default save name for saving, don't change, change save_name
    in save_param
```

#### Details

Description of parameters.

#### Value

ggplot

#### See Also

```
spatGenePlot3D and spatGenePlot2D

Other spatial gene expression visualizations: spatGenePlot2D(), spatGenePlot3D()
```

# **Examples**

```
data(mini_giotto_single_cell)

all_genes = slot(mini_giotto_single_cell, 'gene_ID')
selected_genes = all_genes[1:2]
spatGenePlot(mini_giotto_single_cell, genes = selected_genes, point_size = 3)
```

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spatGenePlot2D

spatGenePlot2D

## **Description**

Visualize cells and gene expression according to spatial coordinates

## Usage

```
spatGenePlot2D(
 gobject,
  show_image = F,
 gimage = NULL,
  image_name = "image",
  sdimx = "sdimx",
  sdimy = "sdimy",
  expression_values = c("normalized", "scaled", "custom"),
 genes,
  cell_color_gradient = c("blue", "white", "red"),
 gradient_midpoint = NULL,
 gradient_limits = NULL,
 show_network = F,
 network_color = NULL,
  spatial_network_name = "Delaunay_network",
  edge_alpha = NULL,
 show\_grid = F,
 grid_color = NULL,
  spatial_grid_name = "spatial_grid",
 midpoint = 0,
  scale_alpha_with_expression = FALSE,
 point_shape = c("border", "no_border", "voronoi"),
  point_size = 1,
 point_alpha = 1,
 point_border_col = "black",
 point_border_stroke = 0.1,
  show_legend = T,
 legend_text = 8,
 background_color = "white",
 vor_border_color = "white",
  vor_alpha = 1,
  vor_max_radius = 200,
 axis_text = 8,
 axis_title = 8,
 cow_n_col = 2,
 cow_rel_h = 1,
 cow_rel_w = 1,
  cow_align = "h",
  show_plot = NA,
 return_plot = NA,
  save_plot = NA,
  save_param = list(),
```

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```
default_save_name = "spatGenePlot2D"
)
```

## **Arguments**

gobject giotto object

show\_image show a tissue background image

gimage a giotto image

image\_name name of a giotto image

sdimx x-axis dimension name (default = 'sdimx')
sdimy y-axis dimension name (default = 'sdimy')

expression\_values

gene expression values to use

genes genes to show

cell\_color\_gradient

vector with 3 colors for numeric data

gradient\_midpoint

midpoint for color gradient

gradient\_limits

vector with lower and upper limits

show\_network show underlying spatial network

network\_color color of spatial network

spatial\_network\_name

name of spatial network to use

edge\_alpha alpha of edge show\_grid show spatial grid grid\_color color of spatial grid

spatial\_grid\_name

name of spatial grid to use

midpoint expression midpoint
scale\_alpha\_with\_expression

scale expression with ggplot alpha parameter

point\_shape shape of points (border, no\_border or voronoi)

point\_alpha transparancy of points

point\_border\_col

color of border around points

point\_border\_stroke

stroke size of border around points

show\_legend show legend

legend\_text size of legend text

background\_color

color of plot background

vor\_border\_color

border colorr for voronoi plot

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```
vor_alpha
                  transparancy of voronoi 'cells'
vor_max_radius maximum radius for voronoi 'cells'
axis_text
                  size of axis text
                  size of axis title
axis_title
cow_n_col
                  cowplot param: how many columns
cow_rel_h
                  cowplot param: relative height
cow_rel_w
                  cowplot param: relative width
cow_align
                  cowplot param: how to align
show_plot
                  show plots
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
save_param
                  list of saving parameters, see showSaveParameters
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

## **Details**

Description of parameters.

## Value

ggplot

# See Also

```
spatGenePlot3D
```

Other spatial gene expression visualizations: spatGenePlot3D(), spatGenePlot()

# **Examples**

```
data(mini_giotto_single_cell)
all_genes = slot(mini_giotto_single_cell, 'gene_ID')
selected_genes = all_genes[1:2]
spatGenePlot2D(mini_giotto_single_cell, genes = selected_genes, point_size = 3)
```

spatGenePlot3D spatGenePlot3D

# Description

Visualize cells and gene expression according to spatial coordinates

268 spatGenePlot3D

#### Usage

```
spatGenePlot3D(
 gobject,
 expression_values = c("normalized", "scaled", "custom"),
  genes,
  show_network = FALSE,
 network_color = NULL,
  spatial_network_name = "Delaunay_network",
 edge_alpha = NULL,
 cluster_column = NULL,
  select_cell_groups = NULL,
 select_cells = NULL,
  show_other_cells = T,
 other_cell_color = "lightgrey",
 other_point_size = 1,
 genes_high_color = NULL,
 genes_mid_color = "white",
 genes_low_color = "blue",
  show_grid = FALSE,
  spatial_grid_name = "spatial_grid",
  point_size = 2,
  show_legend = TRUE,
  axis_scale = c("cube", "real", "custom"),
 custom_ratio = NULL,
 x_ticks = NULL,
 y_ticks = NULL,
 z_ticks = NULL,
  show_plot = NA,
 return_plot = NA,
 save_plot = NA,
 save_param = list(),
 default_save_name = "spatGenePlot3D"
)
```

# **Arguments**

```
gobject
                  giotto object
expression_values
                  gene expression values to use
genes
                  genes to show
                  show underlying spatial network
show_network
network_color
                  color of spatial network
spatial_network_name
                  name of spatial network to use
edge_alpha
                  alpha of edges
cluster_column cluster column to select groups
select_cell_groups
                  select subset of cells/clusters based on cell_color parameter
                  select subset of cells based on cell IDs
select_cells
```

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show\_other\_cells

display not selected cells

other\_cell\_color

color of not selected cells

 $other\_point\_size$ 

size of not selected cells

genes\_high\_color

color represents high gene expression

genes\_mid\_color

color represents middle gene expression

genes\_low\_color

color represents low gene expression

show\_grid show spatial grid

spatial\_grid\_name

name of spatial grid to use

point\_size size of point (cell)

show\_legend show legend

axis\_scale the way to scale the axis

custom\_ratio customize the scale of the plot

x\_ticks set the number of ticks on the x-axis
y\_ticks set the number of ticks on the y-axis
z\_ticks set the number of ticks on the z-axis

show\_plot show plots

return\_plot return ggplot object

save\_plot directly save the plot [boolean]

save\_param list of saving parameters, see showSaveParameters

default\_save\_name

default save name for saving, don't change, change save\_name in save\_param

#### **Details**

Description of parameters.

#### Value

ggplot

# See Also

Other spatial gene expression visualizations: spatGenePlot2D(), spatGenePlot()

270 spatialAEH

spatialAEH

spatialAEH

# Description

Compute spatial variable genes with spatialDE method

## Usage

```
spatialAEH(
  gobject = NULL,
  SpatialDE_results = NULL,
  name_pattern = "AEH_patterns",
  expression_values = c("raw", "normalized", "scaled", "custom"),
  pattern_num = 6,
  l = 1.05,
  python_path = NULL,
  return_gobject = TRUE
)
```

## **Arguments**

# Details

This function is a wrapper for the SpatialAEH method implemented in the ...

## Value

An updated giotto object

spatialDE 271

spatialDE	spatialDE
spatialDE	spati

# Description

Compute spatial variable genes with spatialDE method

#### Usage

```
spatialDE(
  gobject = NULL,
  expression_values = c("raw", "normalized", "scaled", "custom"),
  size = c(4, 2, 1),
  color = c("blue", "green", "red"),
  sig_alpha = 0.5,
  unsig_alpha = 0.5,
  python_path = NULL,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "SpatialDE"
)
```

# Arguments

```
gobject
                  Giotto object
expression_values
                  gene expression values to use
size
                  size of plot
color
                  low/medium/high color scheme for plot
sig_alpha
                  alpha value for significance
unsig_alpha
                  alpha value for unsignificance
                  specify specific path to python if required
python_path
show_plot
                  show plot
                  return ggplot object
return_plot
save_plot
                  directly save the plot [boolean]
save_param
                  list of saving parameters, see showSaveParameters
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

## **Details**

This function is a wrapper for the SpatialDE method implemented in the ...

# Value

a list of data.frames with results and plot (optional)

272 spatNetwDistributions

```
spatNetwDistributions\ spatNetwDistributionsDistance
```

#### **Description**

This function return histograms displaying the distance distribution for each spatial k-neighbor

#### Usage

```
spatNetwDistributions(
  gobject,
  spatial_network_name = "spatial_network",
  distribution = c("distance", "k_neighbors"),
  hist_bins = 30,
  test_distance_limit = NULL,
  ncol = 1,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "spatNetwDistributions"
)
```

#### **Arguments**

```
gobject
                  Giotto object
spatial_network_name
                  name of spatial network
                  show the distribution of cell-to-cell distance or number of k neighbors
distribution
                  number of binds to use for the histogram
hist_bins
test_distance_limit
                  effect of different distance threshold on k-neighbors
ncol
                  number of columns to visualize the histograms in
show_plot
                  show plot
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
                  list of saving parameters from all_plots_save_function
save_param
default_save_name
                  default save name for saving, alternatively change save_name in save_param
```

## **Details**

The **distance** option shows the spatial distance distribution for each nearest neighbor rank (1st, 2nd, 3th, ... neigbor). With this option the user can also test the effect of a distance limit on the spatial network. This distance limit can be used to remove neigbor cells that are considered to far away. The **k\_neighbors** option shows the number of k neighbors distribution over all cells.

#### Value

```
ggplot plot
```

```
spat {\tt NetwDistributionsDistance} \\ spat {\tt NetwDistributionsDistance}
```

## **Description**

This function return histograms displaying the distance distribution for each spatial k-neighbor

## Usage

```
spatNetwDistributionsDistance(
  gobject,
  spatial_network_name = "spatial_network",
  hist_bins = 30,
  test_distance_limit = NULL,
  ncol = 1,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "spatNetwDistributionsDistance")
```

# Arguments

```
gobject
                  Giotto object
spatial_network_name
                  name of spatial network
hist_bins
                  number of binds to use for the histogram
test_distance_limit
                  effect of different distance threshold on k-neighbors
ncol
                  number of columns to visualize the histograms in
show_plot
                  show plot
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
                  list of saving parameters from all_plots_save_function
save_param
default_save_name
                  default save name for saving, alternatively change save_name in save_param
```

#### Value

ggplot plot

274 spatPlot

```
spat {\tt NetwDistributions} Kneighbors \\ spat {\tt NetwDistributions} Kneighbors
```

## **Description**

This function returns a histogram displaying the number of k-neighbors distribution for each cell

## Usage

```
spatNetwDistributionsKneighbors(
  gobject,
  spatial_network_name = "spatial_network",
  hist_bins = 30,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "spatNetwDistributionsKneighbors")
```

# Arguments

```
gobject
                  Giotto object
spatial_network_name
                  name of spatial network
hist_bins
                  number of binds to use for the histogram
show_plot
                  show plot
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
                  list of saving parameters from all_plots_save_function
save_param
default_save_name
                  default save name for saving, alternatively change save_name in save_param
```

## Value

ggplot plot

spatPlot spatPlot

## **Description**

Visualize cells according to spatial coordinates

#### Usage

```
spatPlot(...)
```

spatPlot 275

#### Arguments

Arguments passed on to spatPlot2D gobject giotto object show\_image show a tissue background image gimage a giotto image image\_name name of a giotto image group\_by create multiple plots based on cell annotation column group\_by\_subset subset the group\_by factor column sdimx x-axis dimension name (default = 'sdimx') sdimy y-axis dimension name (default = 'sdimy') spat\_enr\_names names of spatial enrichment results to include cell\_color color for cells (see details) color\_as\_factor convert color column to factor cell\_color\_code named vector with colors cell\_color\_gradient vector with 3 colors for numeric data gradient\_midpoint midpoint for color gradient gradient\_limits vector with lower and upper limits select\_cell\_groups select subset of cells/clusters based on cell color parameter select\_cells select subset of cells based on cell IDs point\_shape shape of points (border, no border or voronoi) point\_size size of point (cell) point\_alpha transparancy of point point\_border\_col color of border around points point\_border\_stroke stroke size of border around points show\_cluster\_center plot center of selected clusters show\_center\_label plot label of selected clusters center\_point\_size size of center points center\_point\_border\_col border color of center points center\_point\_border\_stroke border stroke size of center points label\_size size of labels label\_fontface font of labels show\_network show underlying spatial network spatial\_network\_name name of spatial network to use network\_color color of spatial network network\_alpha alpha of spatial network show\_grid show spatial grid spatial\_grid\_name name of spatial grid to use grid\_color color of spatial grid show\_other\_cells display not selected cells other\_cell\_color color of not selected cells other\_point\_size point size of not selected cells other\_cells\_alpha alpha of not selected cells coord\_fix\_ratio fix ratio between x and y-axis title title of plot

276 spatPlot

```
show_legend show legend
legend_text size of legend text
legend_symbol_size size of legend symbols
background_color color of plot background
vor_border_color border colorr for voronoi plot
vor_max_radius maximum radius for voronoi 'cells'
vor_alpha transparancy of voronoi 'cells'
axis_text size of axis text
axis_title size of axis title
cow_n_col cowplot param: how many columns
cow_rel_h cowplot param: relative height
cow_rel_w cowplot param: relative width
cow_align cowplot param: how to align
show_plot show plot
return_plot return ggplot object
save_plot directly save the plot [boolean]
save_param list of saving parameters, see showSaveParameters
default_save_name default save name for saving, don't change, change save_name
    in save_param
```

## **Details**

Description of parameters.

## Value

ggplot

# See Also

```
spatPlot3D
```

Other spatial visualizations: spatPlot2D(), spatPlot3D()

## **Examples**

```
data(mini_giotto_single_cell)
spatPlot(mini_giotto_single_cell)
spatPlot(mini_giotto_single_cell, cell_color = 'cell_types', point_size = 3)
```

spatPlot2D 277

spatPlot2D

spatPlot2D

## **Description**

Visualize cells according to spatial coordinates

## Usage

```
spatPlot2D(
 gobject,
  show_image = F,
  gimage = NULL,
  image_name = "image",
 group_by = NULL,
 group_by_subset = NULL,
  sdimx = "sdimx",
  sdimy = "sdimy",
  spat_enr_names = NULL,
 cell_color = NULL,
 color_as_factor = T,
 cell_color_code = NULL,
 cell_color_gradient = c("blue", "white", "red"),
  gradient_midpoint = NULL,
  gradient_limits = NULL,
 select_cell_groups = NULL,
  select_cells = NULL,
 point_shape = c("border", "no_border", "voronoi"),
 point_size = 3,
 point_alpha = 1,
 point_border_col = "black",
  point_border_stroke = 0.1,
  show_cluster_center = F,
  show_center_label = F,
  center_point_size = 4,
  center_point_border_col = "black",
  center_point_border_stroke = 0.1,
  label_size = 4,
 label_fontface = "bold",
  show_network = F,
  spatial_network_name = "Delaunay_network",
 network_color = NULL,
 network_alpha = 1,
  show_grid = F,
  spatial_grid_name = "spatial_grid",
 grid_color = NULL,
  show_other_cells = T,
 other_cell_color = "lightgrey",
 other_point_size = 1,
 other_cells_alpha = 0.1,
  coord_fix_ratio = NULL,
```

278 spatPlot2D

```
title = NULL,
  show_legend = T,
  legend_text = 8,
  legend_symbol_size = 1,
  background_color = "white",
  vor_border_color = "white",
  vor_max_radius = 200,
  vor_alpha = 1,
  axis_text = 8,
  axis_title = 8,
  cow_n_col = 2,
  cow_rel_h = 1,
  cow_rel_w = 1,
  cow_align = "h",
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
 save_param = list(),
 default_save_name = "spatPlot2D"
)
```

## **Arguments**

```
gobject
                  giotto object
                  show a tissue background image
show_image
                  a giotto image
gimage
                  name of a giotto image
image_name
                  create multiple plots based on cell annotation column
group_by
group_by_subset
                  subset the group_by factor column
                  x-axis dimension name (default = 'sdimx')
sdimx
                  y-axis dimension name (default = 'sdimy')
sdimy
spat_enr_names names of spatial enrichment results to include
cell_color
                  color for cells (see details)
color_as_factor
                  convert color column to factor
cell_color_code
                  named vector with colors
cell_color_gradient
                  vector with 3 colors for numeric data
gradient_midpoint
                  midpoint for color gradient
gradient_limits
                  vector with lower and upper limits
select_cell_groups
                  select subset of cells/clusters based on cell_color parameter
select_cells
                  select subset of cells based on cell IDs
                  shape of points (border, no_border or voronoi)
point_shape
point_size
                  size of point (cell)
```

spatPlot2D 279

point\_border\_col color of border around points point\_border\_stroke stroke size of border around points show\_cluster\_center plot center of selected clusters show\_center\_label plot label of selected clusters center\_point\_size size of center points center\_point\_border\_col border color of center points  ${\tt center\_point\_border\_stroke}$ border stroke size of center points label\_size size of labels label\_fontface font of labels show\_network show underlying spatial network spatial\_network\_name name of spatial network to use color of spatial network network\_color alpha of spatial network network\_alpha show\_grid show spatial grid spatial\_grid\_name name of spatial grid to use grid\_color color of spatial grid show\_other\_cells display not selected cells other\_cell\_color color of not selected cells other\_point\_size point size of not selected cells other\_cells\_alpha alpha of not selected cells coord\_fix\_ratio fix ratio between x and y-axis title title of plot show\_legend show legend legend\_text size of legend text legend\_symbol\_size size of legend symbols background\_color color of plot background vor\_border\_color border colorr for voronoi plot vor\_max\_radius maximum radius for voronoi 'cells'

transparancy of point

point\_alpha

280 spatPlot3D

```
vor_alpha
                  transparancy of voronoi 'cells'
                  size of axis text
axis_text
axis_title
                  size of axis title
cow_n_col
                  cowplot param: how many columns
                  cowplot param: relative height
cow_rel_h
cow_rel_w
                  cowplot param: relative width
                  cowplot param: how to align
cow_align
                  show plot
show_plot
return_plot
                  return ggplot object
                  directly save the plot [boolean]
save_plot
save_param
                  list of saving parameters, see showSaveParameters
default_save_name
                  default save name for saving, don't change, change save_name in save_param
```

#### **Details**

Description of parameters.

#### Value

ggplot

## See Also

```
spatPlot3D
```

Other spatial visualizations: spatPlot3D(), spatPlot()

# **Examples**

```
data(mini_giotto_single_cell)
spatPlot2D(mini_giotto_single_cell)
spatPlot2D(mini_giotto_single_cell, cell_color = 'cell_types', point_size = 3)
```

spatPlot3D spatPlot3D

# Description

Visualize cells according to spatial coordinates

spatPlot3D 281

#### Usage

```
spatPlot3D(
 gobject,
  sdimx = "sdimx",
  sdimy = "sdimy",
  sdimz = "sdimz"
  spat_enr_names = NULL,
 point_size = 3,
 cell_color = NULL,
 cell_color_code = NULL,
  select_cell_groups = NULL,
  select_cells = NULL,
 show_other_cells = T,
 other_cell_color = "lightgrey",
 other_point_size = 0.5,
 other_cell_alpha = 0.5,
 show_network = F,
  spatial_network_name = "Delaunay_network",
 network_color = NULL,
 network_alpha = 1,
  show\_grid = F,
  spatial_grid_name = "spatial_grid",
 grid_color = NULL,
 grid_alpha = 1,
  title = "",
  show_legend = T,
 axis_scale = c("cube", "real", "custom"),
 custom_ratio = NULL,
 x_ticks = NULL,
 y_ticks = NULL,
 z_{ticks} = NULL,
 show_plot = NA,
 return_plot = NA,
 save_plot = NA,
 save_param = list(),
 default_save_name = "spat3D"
)
```

#### **Arguments**

```
gobject
                  giotto object
sdimx
                  x-axis dimension name (default = 'sdimx')
sdimy
                  y-axis dimension name (default = 'sdimy')
                  z-axis dimension name (default = 'sdimy')
sdimz
spat_enr_names names of spatial enrichment results to include
                  size of point (cell)
point_size
cell_color
                  color for cells (see details)
cell_color_code
                  named vector with colors
select_cell_groups
                  select subset of cells/clusters based on cell_color parameter
```

282 spatPlot3D

```
select subset of cells based on cell IDs
select_cells
show_other_cells
                  display not selected cells
other_cell_color
                  color of not selected cells
other_point_size
                  size of not selected cells
other\_cell\_alpha
                  alpha of not selected cells
show_network
                  show underlying spatial network
spatial_network_name
                  name of spatial network to use
network_color
                  color of spatial network
                  opacity of spatial network
network_alpha
show_grid
                  show spatial grid
spatial_grid_name
                  name of spatial grid to use
grid_color
                  color of spatial grid
grid_alpha
                  opacity of spatial grid
title
                  title of plot
                  show legend
show_legend
axis_scale
                  the way to scale the axis
custom_ratio
                  customize the scale of the plot
x_ticks
                  set the number of ticks on the x-axis
y_ticks
                  set the number of ticks on the y-axis
                  set the number of ticks on the z-axis
z_ticks
show_plot
                  show plot
return_plot
                  return ggplot object
save_plot
                  directly save the plot [boolean]
                  list of saving parameters, see showSaveParameters
save_param
```

default save name for saving, don't change, change save\_name in save\_param

## Value

ggplot

default\_save\_name

# See Also

Other spatial visualizations: spatPlot2D(), spatPlot()

```
specific Cell Cell communication Scores\\ specific Cell Cell communication Scores
```

#### **Description**

Specific Cell-Cell communication scores based on spatial expression of interacting cells

## Usage

```
specificCellCellcommunicationScores(
  spatial_network_name = "Delaunay_network",
 cluster_column = "cell_types",
 random_iter = 100,
 cell_type_1 = "astrocyte",
  cell_type_2 = "endothelial",
 gene_set_1,
 gene_set_2,
  log2FC_addendum = 0.1,
 min_observations = 2,
 detailed = FALSE,
 adjust_method = c("fdr", "bonferroni", "BH", "holm", "hochberg", "hommel", "BY",
    "none"),
  adjust_target = c("genes", "cells"),
  set_seed = FALSE,
  seed_number = 1234,
  verbose = T
)
```

#### **Arguments**

```
giotto object to use
gobject
spatial_network_name
                  spatial network to use for identifying interacting cells
cluster_column cluster column with cell type information
random_iter
                  number of iterations
cell_type_1
                  first cell type
cell_type_2
                  second cell type
                  first specific gene set from gene pairs
gene_set_1
gene_set_2
                  second specific gene set from gene pairs
log2FC_addendum
                  addendum to add when calculating log2FC
min_observations
                  minimum number of interactions needed to be considered
detailed
                  provide more detailed information (random variance and z-score)
                  which method to adjust p-values
adjust_method
```

adjust\_target adjust multiple hypotheses at the cell or gene level

set\_seed set a seed for reproducibility

seed\_number seed number

verbose verbose

#### **Details**

Statistical framework to identify if pairs of genes (such as ligand-receptor combinations) are expressed at higher levels than expected based on a reshuffled null distribution of gene expression values in cells that are spatially in proximity to eachother.

- LR\_comb:Pair of ligand and receptor
- lig\_cell\_type: cell type to assess expression level of ligand
- lig expr: average expression of ligand in lig cell type
- · ligand: ligand name
- rec\_cell\_type: cell type to assess expression level of receptor
- rec\_expr: average expression of receptor in rec\_cell\_type
- · receptor: receptor name
- LR\_expr: combined average ligand and receptor expression
- lig\_nr: total number of cells from lig\_cell\_type that spatially interact with cells from rec\_cell\_type
- rec\_nr: total number of cells from rec\_cell\_type that spatially interact with cells from lig\_cell\_type
- rand\_expr: average combined ligand and receptor expression from random spatial permutations
- av\_diff: average difference between LR\_expr and rand\_expr over all random spatial permutations
- sd\_diff: (optional) standard deviation of the difference between LR\_expr and rand\_expr over all random spatial permutations
- z\_score: (optinal) z-score
- log2fc: log2 fold-change (LR\_expr/rand\_expr)
- pvalue: p-value
- LR\_cell\_comb: cell type pair combination
- p.adj: adjusted p-value
- PI: significanc score: log2fc \* -log10(p.adj)

# Value

Cell-Cell communication scores for gene pairs based on spatial interaction

stitchFieldCoordinates 285

```
stitchFieldCoordinates
```

stitchFieldCoordinates

#### **Description**

Helper function to stitch field coordinates together to form one complete picture

## Usage

```
stitchFieldCoordinates(
  location_file,
  offset_file,
  cumulate_offset_x = F,
  cumulate_offset_y = F,
  field_col = "Field of View",
  X_coord_col = "X",
  Y_coord_col = "Y",
  reverse_final_x = F,
  reverse_final_y = T
)
```

## **Arguments**

```
location dataframe with X and Y coordinates
location_file
offset_file
                  dataframe that describes the offset for each field (see details)
cumulate_offset_x
                  (boolean) Do the x-axis offset values need to be cumulated?
cumulate_offset_y
                  (boolean) Do the y-axis offset values need to be cumulated?
                  column that indicates the field within the location_file
field_col
                  column that indicates the x coordinates
X_coord_col
                  column that indicates the x coordinates
Y_coord_col
reverse_final_x
                  (boolean) Do the final x coordinates need to be reversed?
reverse_final_y
                  (boolean) Do the final y coordinates need to be reversed?
```

#### **Details**

Stitching of fields:

- 1. have cell locations: at least 3 columns: field, X, Y
- 2. create offset file: offset file has 3 columns: field, x\_offset, y\_offset
- 3. create new cell location file by stitching original cell locations with stitchFieldCoordinates
- 4. provide new cell location file to createGiottoObject

#### Value

Updated location dataframe with new X ['X\_final'] and Y ['Y\_final'] coordinates

286 subClusterCells

```
stitchTileCoordinates stitchTileCoordinates
```

## **Description**

Helper function to stitch tile coordinates together to form one complete picture

## Usage

```
stitchTileCoordinates(location_file, Xtilespan, Ytilespan)
```

## Arguments

 $\begin{array}{ll} \mbox{location\_file} & \mbox{location dataframe with } X \mbox{ and } Y \mbox{ coordinates} \\ \mbox{Xtilespan} & \mbox{numerical value specifying the width of each tile} \\ \mbox{Ytilespan} & \mbox{numerical value specifying the height of each tile} \\ \end{array}$ 

subClusterCells

subClusterCells

#### **Description**

subcluster cells

## Usage

```
subClusterCells(
 gobject,
 name = "sub_clus",
 cluster_method = c("leiden", "louvain_community", "louvain_multinet"),
 cluster_column = NULL,
  selected_clusters = NULL,
 hvg_param = list(reverse_log_scale = T, difference_in_cov = 1, expression_values =
    "normalized"),
 hvg_min_perc_cells = 5,
 hvg_mean_expr_det = 1,
 use_all_genes_as_hvg = FALSE,
 min_nr_of_hvg = 5,
 pca_param = list(expression_values = "normalized", scale_unit = T),
 nn_param = list(dimensions_to_use = 1:20),
 k_neighbors = 10,
 resolution = 1,
 n_{iterations} = 1000,
 gamma = 1,
 omega = 1,
 python_path = NULL,
 nn_network_to_use = "sNN",
 network_name = "sNN.pca",
 return_gobject = TRUE,
  verbose = T
)
```

subClusterCells 287

#### **Arguments**

gobject giotto object

name name for new clustering result

cluster\_method clustering method to use

cluster\_column cluster column to subcluster

selected\_clusters

only do subclustering on these clusters

hvg\_param parameters for calculateHVG

hvg\_min\_perc\_cells

threshold for detection in min percentage of cells

hvg\_mean\_expr\_det

threshold for mean expression level in cells with detection

use\_all\_genes\_as\_hvg

forces all genes to be HVG and to be used as input for PCA

min\_nr\_of\_hvg minimum number of HVG, or all genes will be used as input for PCA

pca\_param parameters for runPCA

nn\_param parameters for parameters for createNearestNetwork

k\_neighbors number of k for createNearestNetwork

resolution resolution

n\_iterations number of interations to run the Leiden algorithm.

gamma gamma omega omega

python\_path specify specific path to python if required

nn\_network\_to\_use

type of NN network to use (kNN vs sNN)

 ${\tt network\_name} \quad \quad name \ of \ NN \ network \ to \ use$ 

return\_gobject boolean: return giotto object (default = TRUE)

verbose verbose

## Details

This function performs subclustering on selected clusters. The systematic steps are:

- 1. subset Giotto object
- 2. identify highly variable genes
- 3. run PCA
- 4. create nearest neighbouring network
- 5. do clustering

#### Value

giotto object with new subclusters appended to cell metadata

## See Also

 ${\tt doLouvainCluster\_multinet}, {\tt doLouvainCluster\_community} \ and \ @see also \ {\tt doLeidenCluster\_community} \\$ 

288 subsetGiottoLocs

subsetGiotto

subsetGiotto

# Description

subsets Giotto object including previous analyses.

# Usage

```
subsetGiotto(gobject, cell_ids = NULL, gene_ids = NULL, verbose = FALSE)
```

# **Arguments**

```
gobject giotto object
cell_ids cell IDs to keep
gene_ids gene IDs to keep
verbose be verbose
```

## Value

giotto object

# **Examples**

subsetGiottoLocs

subsetGiottoLocs

# Description

subsets Giotto object based on spatial locations

subsetGiottoLocs 289

#### Usage

```
subsetGiottoLocs(
  gobject,
  x_max = NULL,
  x_min = NULL,
  y_max = NULL,
  y_min = NULL,
  z_max = NULL,
  z_min = NULL,
  return_gobject = T,
  verbose = FALSE
)
```

#### **Arguments**

gobject	giotto object
x_max	maximum x-coordinate
x_min	minimum x-coordinate
y_max	maximum y-coordinate
y_min	minimum y-coordinate
z_max	maximum z-coordinate
z_min	minimum z-coordinate
return_gobject	return Giotto object
verbose	be verbose

## **Details**

if return\_gobject = FALSE, then a filtered combined metadata data.table will be returned

## Value

giotto object

## **Examples**

```
data(mini_giotto_single_cell)
# spatial plot
spatPlot(mini_giotto_single_cell)
# subset giotto object based on spatial locations
subset_obj = subsetGiottoLocs(mini_giotto_single_cell,
x_max = 1500, x_min = 1000,
y_max = -500, y_min = -1000)
# spatial plot of subset giotto object
spatPlot(subset_obj)
```

290 t\_giotto

trendSceek trendSceek

#### **Description**

Compute spatial variable genes with trendsceek method

## Usage

```
trendSceek(
  gobject,
  expression_values = c("normalized", "raw"),
  subset_genes = NULL,
  nrand = 100,
  ncores = 8,
  ...
)
```

#### **Arguments**

gobject Giotto object
expression\_values
gene expression values to use
subset\_genes subset of genes to run trendsceek on
nrand An integer specifying the number of random resamplings of the mark distribution as to create the null-distribution.

ncores An integer specifying the number of cores to be used by BiocParallel
... Additional parameters to the trendsceek\_test function

#### **Details**

This function is a wrapper for the trendsceek\_test method implemented in the trendsceek package

# Value

data.frame with trendsceek spatial genes results

## **Description**

t function that works with multiple matrix representations

#### Usage

```
t_giotto(mymatrix)
```

updateGiottoImage 291

## **Arguments**

```
mymatrix matrix object
```

#### Value

transposed matrix

 $update {\tt GiottoImage}$ 

updateGiottoImage

# Description

Updates the boundaries of a giotto image attached to a giotto object

## Usage

```
updateGiottoImage(
  gobject,
  image_name,
  xmax_adj = 0,
  xmin_adj = 0,
  ymax_adj = 0,
  ymin_adj = 0,
  return_gobject = TRUE
)
```

#### **Arguments**

```
gobject giotto object
image_name spatial locations

xmax_adj adjustment of the maximum x-value to align the image

xmin_adj adjustment of the minimum x-value to align the image

ymax_adj adjustment of the maximum y-value to align the image

ymin_adj adjustment of the minimum y-value to align the image

return_gobject return a giotto object
```

#### Value

```
a giotto object or an updated giotto image if return_gobject = F
```

292 viewHMRFresults2D

viewHMRFresults

viewHMRFresults

## Description

View results from doHMRF.

## Usage

```
viewHMRFresults(
  gobject,
  HMRFoutput,
  k = NULL,
  betas_to_view = NULL,
  third_dim = FALSE,
  ...
)
```

# Arguments

```
gobject giotto object
```

HMRF output from doHMRF k number of HMRF domains

betas\_to\_view results from different betas that you want to view

third\_dim 3D data (boolean)

... additional paramters (see details)

#### Value

spatial plots with HMRF domains

#### See Also

```
spatPlot2D and spatPlot3D
```

viewHMRFresults2D

viewHMRFresults2D

## Description

View results from doHMRF.

## Usage

```
viewHMRFresults2D(gobject, HMRFoutput, k = NULL, betas_to_view = NULL, ...)
```

viewHMRFresults3D 293

## **Arguments**

gobject giotto object

HMRF output from doHMRF k number of HMRF domains

betas\_to\_view results from different betas that you want to view

... additional parameters to spatPlot2D()

#### Value

spatial plots with HMRF domains

#### See Also

spatPlot2D

viewHMRFresults3D

viewHMRFresults3D

## Description

View results from doHMRF.

# Usage

```
viewHMRFresults3D(gobject, HMRFoutput, k = NULL, betas_to_view = NULL, ...)
```

## Arguments

gobject giotto object

HMRF output from doHMRF k number of HMRF domains

betas\_to\_view results from different betas that you want to view

... additional parameters to spatPlot3D()

#### Value

spatial plots with HMRF domains

## See Also

spatPlot3D

294 violinPlot

violinPlot

violinPlot

#### **Description**

Creates violinplot for selected clusters

## Usage

```
violinPlot(
  gobject,
  expression_values = c("normalized", "scaled", "custom"),
  genes,
  cluster_column,
  cluster_custom_order = NULL,
  color_violin = c("genes", "cluster"),
  cluster_color_code = NULL,
  strip_position = c("top", "right", "left", "bottom"),
  strip\_text = 7,
  axis_text_x_size = 10,
  axis_text_y_size = 6,
  show_plot = NA,
  return_plot = NA,
  save_plot = NA,
  save_param = list(),
  default_save_name = "violinPlot"
)
```

# Arguments

```
gobject
                  giotto object
expression_values
                  expression values to use
                  genes to plot
genes
cluster_column name of column to use for clusters
cluster_custom_order
                  custom order of clusters
color_violin
                  color violin according to genes or clusters
cluster_color_code
                  color code for clusters
strip_position position of gene labels
strip_text
                  size of strip text
\verb"axis_text_x_size"
                  size of x-axis text
axis_text_y_size
                  size of y-axis text
show_plot
                  show plot
return_plot
                  return ggplot object
```

writeHMRFresults 295

#### Value

ggplot

#### **Examples**

writeHMRFresults

writeHMRFresults

## Description

write results from doHMRF to a data.table.

## Usage

```
writeHMRFresults(
  gobject,
  HMRFoutput,
  k = NULL,
  betas_to_view = NULL,
  print_command = F
)
```

#### **Arguments**

```
gobject giotto object
```

HMRF output From doHMRF

k k to write results for

betas\_to\_view results from different betas that you want to view

print\_command see the python command

296 writeHMRFresults

# Value

data.table with HMRF results for each  $\boldsymbol{b}$  and the selected  $\boldsymbol{k}$ 

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