Package 'SpaTalk'

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```
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Description

createSpaTalk

create SpaTalk object using spatial transcriptomics data.

SpaTalk object

dec_cci 3

Usage

```
createSpaTalk(
   st_data,
   st_meta,
   species,
   if_st_is_sc,
   spot_max_cell,
   celltype = NULL
)
```

Arguments

st_data	A data frame or matrix or dgCMatrix containing counts of spatial transcriptomics, each column representing a spot or a cell, each row representing a gene.
st_meta	A data frame containing coordinate of spatial transcriptomics with three columns, namely 'spot', 'x', 'y' for spot-based spatial transcriptomics data or 'cell', 'x', 'y' for single-cell spatial transcriptomics data.
species	A character meaning species of the spatial transcriptomics data. 'Human' or 'Mouse'
if_st_is_sc	A logical meaning if it is single-cell spatial transcriptomics data. TRUE is FALSE.
spot_max_cell	A integer meaning max cell number for each plot to predict. If if_st_sc is FALSE, please determine the spot_max_cell. For 10X (55um), we recommend 30. For Slide-seq, we recommend 1.
celltype	A character containing the cell type of ST data. To skip the deconvolution step and directly infer cell-cell communication, please define the cell type. Default is NULL.

Value

SpaTalk object

dec_cci	Decomposing	cell-cell	communications	for	spatial	transciptomics
	data					

Description

Identify the cell-cell communications for single-cell or spot-based spatial transciptomics data with proximal ligand-receptor-target interactions.

```
dec_cci(
  object,
  celltype_sender,
  celltype_receiver,
  n_neighbor = 10,
  min_pairs = 5,
  min_pairs_ratio = 0,
  per_num = 1000,
```

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```
pvalue = 0.05,
  co_exp_ratio = 0.1,
  if_doParallel = T,
  use_n_cores = NULL
)
```

Arguments

object SpaTalk object after find_lr_path. celltype_sender Name of celltype_sender. celltype_receiver Name of celltype_receiver. Number of neighbor cells to select as the proximal cell-cell pair. Default is 10. n_neighbor Min proximal cell-cell pairs between for sending and receiving cell types. Demin_pairs fault is 5. min_pairs_ratio Min proximal cell-cell pairs ratio between for sending and receiving cell types. Default is 0. per_num Number of repeat times for permutation test. Default is 1000. Include the significantly proximal LR pairs with this cutoff of p value from pvalue permutation test. Default is 0.05. Min cell ratio in receiving cells with co-expressed source and target genes for co_exp_ratio predicting the downstream pathway activity. if_doParallel Use doParallel. Default is TRUE.

Value

use_n_cores

SpaTalk object containing the inferred LR pairs and pathways.

dec_cci_all	Decomposing cell-cell communications for spatial transciptomics
	data

Number of CPU cores to use. Default is all cores - 2.

Description

Identify the all cell-cell communications for single-cell or spot-based spatial transciptomics data with proximal ligand-receptor-target interactions.

```
dec_cci_all(
  object,
  n_neighbor = 10,
  min_pairs = 5,
  min_pairs_ratio = 0,
  per_num = 1000,
  pvalue = 0.05,
```

dec_celltype 5

```
co_exp_ratio = 0.1,
  if_doParallel = T,
  use_n_cores = NULL
)
```

Arguments

object SpaTalk object after find_lr_path. Number of neighbor cells to select as the proximal cell-cell pair. Default is 10. n_neighbor Min proximal cell-cell pairs between for sending and receiving cell types. Demin_pairs fault is 5. min_pairs_ratio Min proximal cell-cell pairs ratio between for sending and receiving cell types. Default is 0. Number of repeat times for permutation test. Default is 1000. per_num pvalue Include the significantly proximal LR pairs with this cutoff of p value from permutation test. Default is 0.05. Min cell ratio in receiving cells with co-expressed source and target genes for co_exp_ratio predicting the downstream pathway activity.

use_n_cores Number of CPU cores to use. Default is all cores - 2.

Value

SpaTalk object containing the inferred LR pairs and pathways.

dec_celltype Decomposing cell type for spatial transcriptomics data

Description

Identify the cellular composition for single-cell or spot-based spatial transcriptomics data with non-negative regression.

```
dec_celltype(
 object,
  sc_data,
  sc_celltype,
 min_percent = 0.5,
 min_nFeatures = 10,
 if_use_normalize_data = T,
 if_use_hvg = F,
  if_doParallel = T,
 use_n_cores = NULL,
  iter_num = 1000,
 method = 1,
  env = "base"
  anaconda_path = "~/anaconda3",
  dec_result = NULL
)
```

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Arguments

object SpaTalk object generated from createSpaTalk. sc_data A A data frame or matrix or dgCMatrix containing counts of single-cell RNAseq data as the reference, each column representing a cell, each row representing a gene. sc_celltype A character containing the cell type of the reference single-cell RNA-seq data. Min percent to predict new cell type for single-cell st_data or predict new cell min_percent for spot-based st_data. Default is 0.5. Min number of expressed features/genes for each spot/cell in st_data. Default min_nFeatures is 10. if_use_normalize_data Whether to use normalized st_data and sc_data with Seurat normalization. Default is TRUE. set it FALSE when the st_data and sc_data are already normalized matrix with other methods. if_use_hvg Whether to use highly variable genes for non-negative regression. Default is FALSE. if_doParallel Use doParallel. Default is TRUE. Number of CPU cores to use. Default is all cores - 2. use_n_cores Number of iteration to generate the single-cell data for spot-based data. Default iter_num is 1000. method 1 means using the SpaTalk deconvolution method, 2 means using RCTD, 3 means using Seurat, 4 means using SPOTlight, 5 means using deconvSeq, 6 means using stereoscope, 7 means using cell2location When method set to 6, namely use stereoscope python package to deconvolute, env please define the python environment of installed stereoscope. Default is the 'base' environment. Anaconda is recommended. anaconda_path When use python package, please define the path to anaconda, default is ~/anaconda3 dec_result A matrix of deconvolution result from other upcoming methods, row represents spots or cells, column represents cell types of scRNA-seq reference. See demo_dec_result

Value

SpaTalk object containing the decomposing results.

no data of dec_result

Description

Demo data of dec_result

Usage

demo_dec_result()

demo_geneinfo 7

Details

dec_result used in dec_celltype must be a matrix object, each row representing a spot, each column representing a cell type.

Value

A matrix.

Examples

```
dec_result_demo <- demo_dec_result()</pre>
```

demo_geneinfo

Demo data of geneinfo

Description

Demo data of geneinfo

Usage

```
demo_geneinfo()
```

Details

geneinfo used in dec_celltype must be a data. frame object with three columns, namely 'symbol', 'synonyms', 'species'.

Examples

```
geneinfo_demo <- demo_geneinfo()</pre>
```

demo_lrpairs

Demo data of lrpairs

Description

Demo data of Irpairs

Usage

```
demo_lrpairs()
```

Details

lrpairs used in dec_cci must be a data.frame object with three columns, namely 'ligand',
'receptor', 'species'.

Value

A data.frame.

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Examples

```
lrpairs_demo <- demo_lrpairs()</pre>
```

demo_pathways

Demo data of pathways

Description

Demo data of pathways

Usage

```
demo_pathways()
```

Details

```
pathways used in dec_cci must be a data.frame object with seven columns, namely 'src', 'dest', 'pathway', 'source', 'type', 'src_tf', 'dest_tf', 'species'.
```

Value

A data.frame.

Examples

```
pathways_demo <- demo_pathways()</pre>
```

demo_sc_data

Demo data of sc_data

Description

Demo data of sc_data.

Usage

```
demo_sc_data()
```

Details

sc_data used in dec_celltype must be a matrix object, each column representing a cell, each row representing a gene.

Value

A matrix.

Examples

```
sc_data_demo <- demo_sc_data()</pre>
```

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demo_st_data

Demo data of st_data

Description

Demo data of st_data.

Usage

```
demo_st_data()
```

Details

st_data used in dec_celltype must be a matrix object, each column representing a spot, each row representing a gene.

Value

A matrix.

Examples

```
st_data_demo <- demo_st_data()</pre>
```

demo_st_meta

Demo data of st_meta

Description

Demo data of st_meta

Usage

```
demo_st_meta()
```

Details

st_meta used in dec_celltype must be a data.frame object with three columns, namely 'spot', 'x', 'y' for spot-based spatial transcriptomics data.

Value

A data.frame.

Examples

```
st_meta_demo <- demo_st_meta()</pre>
```

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demo_st_sc_data

Demo data of single-cell st_data

Description

Demo data of single-cell st_data.

Usage

```
demo_st_sc_data()
```

Details

st_data used in dec_celltype must be a matrix object, each column representing a cell, each row representing a gene.

Value

A matrix.

Examples

```
st_data_demo <- demo_st_sc_data()</pre>
```

demo_st_sc_meta

Demo data of st_sc_meta

Description

Demo data of st_sc_meta

Usage

```
demo_st_sc_meta()
```

Details

st_sc_meta used in dec_celltype must be a data.frame object with three columns, namely 'cell', 'x', 'y' for single-cell spatial transcriptomics data.

Value

A data.frame.

Examples

```
st_sc_meta_demo <- demo_st_sc_meta()</pre>
```

find_lr_path

find_lr_path Find lrpairs and pathways

Description

Find 1rpairs and pathways with receptors having downstream targets and transcriptional factors.

Usage

```
find_lr_path(
  object,
  lrpairs,
  pathways,
  max_hop = NULL,
  if_doParallel = T,
  use_n_cores = NULL
)
```

Arguments

object	SpaTalk object generated from dec_celltype.
lrpairs	A data.frame of the system data containing ligand-receptor pairs of 'Human' and 'Mouse' from CellTalkDB.
pathways	A data.frame of the system data containing gene-gene interactions and pathways from KEGG and Reactome as well as the information of transcriptional factors.
max_hop	Max hop from the receptor to the downstream target transcriptional factor to find for receiving cells. Default is 3 for human and 4 for mouse.
if_doParallel	Use doParallel. Default is TRUE.
use_n_cores	Number of CPU cores to use. Default is all cores - 2.

Value

SpaTalk object containing the filtered lrpairs and pathways.

|--|--|

Description

Gene symbols of 'Human' and 'Mouse' updated on June 30, 2021 for revising count matrix.

Usage

geneinfo

Format

An object of class data. frame with 250934 rows and 3 columns.

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Source

https://www.ncbi.nlm.nih.gov/gene

Description

Generate pseudo spot st_data with single-cell st_data

Usage

```
generate_spot(st_data, st_meta, x_min, x_res, x_max, y_min, y_res, y_max)
```

Arguments

st_data	A data frame or matrix or dgCMatrix containing counts of spatial transcriptomics, each column representing a cell, each row representing a gene.
st_meta	A data.frame containing coordinate of spatial transcriptomics with three columns, 'cell', 'x', 'y', and celltype.
x_min	Min value of x axis.
x_res	Resolution of x coordinate.
x_max	Max value of x axis.
y_min	Min value of y axis.
y_res	Resolution of y coordinate.
y_max	Max value of y axis.

Value

A list of spot st_data and st_meta

get_lr_path Get LR and downstream pathways

Description

Get LR and downstream pathways and get p value of receptor-related pathways with LR-target genes by the Fisher-exact test.

```
get_lr_path(
  object,
  celltype_sender,
  celltype_receiver,
  ligand,
  receptor,
  min_gene_num = 5
)
```

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Arguments

object SpaTalk object generated from dec_cci.

celltype_sender

Name of celltype_sender.

celltype_receiver

Name of celltype_receiver.

ligand Name of ligand from celltype_sender.

receptor Name of receptor from celltype_receiver.

min_gene_num Min genes number for each pathway.

Value

A list containing two data.frame. One is LR and downstream pathways, another is the p value of receptor-related pathways with LR-target genes.

lrpairs

lrpairs

Description

Ligand-receptor pairs of 'Human' and 'Mouse' containing 3398 human and 2033 mouse pairs.

Usage

lrpairs

Format

An object of class data.frame with 5427 rows and 3 columns.

Source

http://tcm.zju.edu.cn/celltalkdb/

pathways

pathways

Description

KEGG pathways and Reactomes of 'Human' and 'Mouse' for intra-cellular genes and transcription factors.

Usage

pathways

Format

An object of class data.frame with 669197 rows and 8 columns.

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Source

```
https://www.genome.jp/kegg/pathway.html
https://reactome.org/
http://bioinfo.life.hust.edu.cn/AnimalTFDB/#!/
```

plot_ccdist

Plot cell-cell distribution

Description

Point plot with spatial distribution of celltype_sender and celltype_receiver

Usage

```
plot_ccdist(
  object,
  celltype_sender,
  celltype_receiver,
  color = NULL,
  size = 1,
  if_plot_others = T,
  if_plot_density = T,
  if_plot_edge = T,
  if_show_arrow = T,
  arrow_length = 0.05,
  plot_cells = NULL
)
```

Arguments

```
object
                  SpaTalk object generated from dec_celltype.
celltype_sender
                  Name of celltype_sender.
celltype_receiver
                  Name of celltype_receiver.
                  Color for celltype_sender, celltype_receiver, and others. Three values.
color
size
                  Point size. Default is 1.
if_plot_others Whether to plot others. Default is TRUE.
if_plot_density
                  Whether to plot marginal density plots. Default is TRUE.
if_plot_edge
                  Whether to plot edge between neighbors. Default is TRUE.
                  Whether to show the arrow of the plotted edge. Default is TRUE.
if_show_arrow
arrow_length
                  Arrow length.
plot_cells
                  Which cells to plot. Default is all cells. Input a character vector of cell names to
                  plot.
```

plot_cci_lrpairs 15

Description

Heatmap with LR pairs of celltype_sender and celltype_receiver

Usage

```
plot_cci_lrpairs(
  object,
  celltype_sender,
  celltype_receiver,
  top_lrpairs = 20,
  color = NULL,
  border_color = "black",
  type = "sig",
  fontsize_number = 5,
  number_color = "black",
  color_low = NULL,
  color_high = NULL
)
```

Arguments

color Color for the cells in heatmap.

border_color color of cell borders on heatmap, use NA if no border should be drawn.

type Set 'sig' to plot significant LR pairs or set 'number' to plot the number of spatial

LR interactions.

fontsize_number

fontsize of the numbers displayed in cells.

number_color color of the text.

color_low For 'number' type, define the color for the lowest value.

color_high For 'number' type, define the color for the highest value.

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plot_lrpair Plot LR pair

Description

Point plot with LR pair from celltype_sender to celltype_receiver

Usage

```
plot_lrpair(
  object,
  celltype_sender,
  celltype_receiver,
  ligand,
  receptor,
  color = NULL,
  size = 1,
  if_plot_density = T,
  if_plot_edge = T,
  if_show_arrow = T,
  arrow_length = 0.05,
  plot_cells = NULL
)
```

Arguments

object SpaTalk object generated from dec_celltype. celltype_sender Name of celltype_sender. celltype_receiver Name of celltype_receiver. ligand Name of ligand from celltype_sender. receptor Name of receptor from celltype_receiver. color Color for ligand, receptor, and others. Three values. Point size. Default is 1. size if_plot_density Whether to plot marginal density plots. Default is TRUE. if_plot_edge Whether to plot edge between neighbors. Default is TRUE. Whether to show the arrow of the plotted edge. Default is TRUE. if_show_arrow arrow_length Arrow length. plot_cells Which cells to plot. Default is all cells. Input a character vector of cell names to plot.

plot_lrpair_vln 17

plot_lrpair_vln

Plot spatial distance of LR pair with vlnplot

Description

Violin plot spatial distance of LR pair between expressed senders and receivers and between expressed cell-cell pairs.

Usage

```
plot_lrpair_vln(
  object,
  celltype_sender,
  celltype_receiver,
  ligand,
  receptor,
  vln_color = NULL,
  if_plot_boxplot = T,
  box_width = 0.2
```

Arguments

Box width. Default is 0.2.

plot_lr_path

box_width

Plot LR and downstream pathways

Description

Plot network with LR and downstream pathways

plot_path2gene

Usage

```
plot_lr_path(
  object,
  celltype_sender,
  celltype_receiver,
  ligand,
  receptor,
  color = NULL,
  size = 5,
  arrow_length = 0.1
)
```

Arguments

```
object
                  SpaTalk object generated from dec_cci.
celltype_sender
                  Name of celltype_sender.
celltype_receiver
                  Name of celltype_receiver.
ligand
                  Name of ligand from celltype_sender.
receptor
                  Name of receptor from celltype_receiver.
                  Color for points Two values.
color
                  Size of points.
size
arrow_length
                  Arrow length.
```

plot_path2gene

River plot of significantly activated pathways and related downstream genes of receptors.

Description

River plot of significantly activated pathways and related downstream genes of receptors.

```
plot_path2gene(
  object,
  celltype_sender,
  celltype_receiver,
  ligand,
  receptor,
  min_gene_num = 5,
  pvalue = 0.5,
  color = NULL,
  color_flow = "blue"
)
```

plot_st_celltype 19

Arguments

object SpaTalk object generated from dec_cci.

celltype_sender

Name of celltype_sender.

celltype_receiver

Name of celltype_receiver.

ligand Name of ligand from celltype_sender.

receptor Name of receptor from celltype_receiver.

min_gene_num Min genes number for each pathway.

pvalue P value of the Fisher-exact test.

color Color of pathways and genes. Two values.

color_flow Color of the flow.

plot_st_celltype

Plot spatial distribution of a single cell type

Description

Ponit plot with spatial distribution of a single predicted cell type for transcriptomics data

Usage

```
plot_st_celltype(
  object,
  celltype,
  size = 1,
  color_celltype = "blue",
  color_others = "grey"
)
```

Arguments

object SpaTalk object generated from dec_celltype.

celltype Name of cell type in the sc_celltype.

size Point size. Default is 1.

color_celltype Color for the celltype of interest.

color_others Color for the others.

Description

Plot spatial distribution of all predicted cell types for transcriptomics data

Usage

```
plot_st_celltype_all(object, size = 1, color = NULL)
```

Arguments

object SpaTalk object generated from dec_celltype.

size Point size. Default is 1.

color Color for all predicted cell types.

```
plot_st_celltype_density
```

Plot spatial density of a single cell type

Description

Plot spatial density of a single predicted cell type for transcriptomics data

Usage

```
plot_st_celltype_density(
  object,
  celltype,
  type,
  if_plot_point = T,
  point_color = NULL,
  point_size = 1,
  color_low = "grey",
  color_mid = NULL,
  color_high = "blue",
  color_midpoint = NULL,
  size = 1
)
```

Arguments

object SpaTalk object generated from dec_celltype.

celltype Name of cell type in the sc_celltype.

type Select 'contour' or 'raster'.

if_plot_point Whether to plot points when type is 'contour'.

point_color Point color.

plot_st_celltype_percent

```
point_size Point size. Default is 1.

color_low Color for the lowest value.

color_mid Color for the middle value for using scale_color_gradient2. Default is NULL.

color_high Color for the highest value.

color_midpoint Value for the middle scale. Default is NULL.

size Line size when type is 'contour'. Default is 1.
```

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```
plot_st_celltype_percent
```

Plot spatial distribution of a single cell type percent

Description

Plot spatial distribution of a single predicted cell type percent for transcriptomics data

Usage

```
plot_st_celltype_percent(
  object,
  celltype,
  size = 1,
  color_low = NULL,
  color_mid = NULL,
  color_high = NULL,
  color_midpoint = NULL
)
```

Arguments

object SpaTalk object generated from dec_celltype.

celltype Name of cell type in the sc_celltype.

size Point size. Default is 1.

color_low Color for the lowest value.

color_mid Color for the middle value for using scale_color_gradient2. Default is NULL.

color_high Color for the highest value.

color_midpoint Value for the middle scale. Default is NULL.

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plot_st_cor_heatmap

Plot heatpmap of correlation between marker genes and cell types

Description

Plot heatpmap of correlation between the expression of marker genes and the predicted score of cell types among all spatial cells or spots.

Usage

```
plot_st_cor_heatmap(
  object,
  marker_genes,
  celltypes,
  color_low = NULL,
  color_mid = NULL,
  color_high = NULL,
  scale = "none",
  if_show_top = T,
  top_direction = "row",
  border_color = NA
)
```

Arguments

object	SpaTalk object generated from dec_celltype.
marker_genes	A character containing the known marker genes to plot, provide at least two marker genes of interest.
celltypes	A character containing name of cell type in the sc_celltype. Default is to plot all cell types.
color_low	Color for the lowest value.
color_mid	$Color for the \ middle \ value \ for \ using \ scale_color_gradient 2. \ Default \ is \ NULL.$
color_high	Color for the highest value.
scale	Character indicating if the values should be centered and scaled in either the row direction or the column direction, or none. Corresponding values are 'row', 'column' and 'none'.
if_show_top	Whether to plot a symbol to the highest value across rows or columns. Default is TRUE.
top_direction	Direction to identify the highest value, select 'row' or 'column'.
border_color	Color of the cell border. Default is 'NA'.

plot_st_gene 23

plot_st_gene Plot spatial distribution of gene

Description

Point plot with spatial distribution of a gene for transcriptomics data

Usage

```
plot_st_gene(
  object,
  gene,
  size = 1,
  color_low = "grey",
  color_mid = NULL,
  color_high = "blue",
  color_midpoint = NULL,
  if_use_newmeta = T,
  celltype = NULL,
  if_plot_others = T
)
```

Arguments

object SpaTalk object generated fr gene Symbol of gene, e.g., 'AKT	•
gene Symbol of gene, e.g., 'AKT	71'.
size Point size. Default is 1.	
color_low Color for the lowest value.	
color_mid Color for the middle value i	For using scale_color_gradient2. Default is NULL.
color_high Color for the highest value.	
color_midpoint Value for the middle scale.	Default is NULL.
if_use_newmeta Whether to use newmeta o p for spot-based data. Defaul	plot the spatial distribution of gene after dec_celltype t is TRUE.
celltype gene in which celltype to put when using this parameter.	olot. Default is NULL. Set Nif_use_newmeta TRUE
if_plot_others Whether to plot other cells	when to use defined celltype.

Details

Please set if_use_newmeta as FALSE to plot the spatial distribution of gene before dec_celltype for spot-based data.

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plot_st_pie	Plot spatial transcriptomics data	

Description

Plot scatterpie for spatial transcriptomics data

Usage

```
plot_st_pie(object, pie_scale = 1, xy_ratio = 1, color = NULL)
```

Arguments

object SpaTalk object generated from dec_celltype.

pie_scale Scale of each pie to plot. Default is 1.

xy_ratio Ratio of y and x coordinates. Default is 1.

color Filled of colors for pie plot, length of color must be equal to the number of unique cell types in sc_celltype.

Description

Plot scatterpie for spot-based ST data

Usage

```
plot_st_pie_generate(st_meta, pie_scale = 1, xy_ratio = 1, color = NULL)
```

Arguments

st_meta	st_meta generated from generate_spot
pie_scale	Scale of each pie to plot. Default is 1.
xy_ratio	Ratio of y and x coordinates. Default is 1.
color	Filled of colors for pie plot, length of color must be equal to the number of unique cell types in sc_celltype.

rev_gene 25

rev_gene	Pre-processing step: revising gene symbols

Description

Revise genes according to NCBI Gene symbols updated in June 30, 2021 for count matrix, user-custom lrpairs data.frame, and user-custom pathways data.frame.

Usage

```
rev_gene(data = NULL, data_type = NULL, species = NULL, geneinfo = NULL)
```

Arguments

data	A data frame or matrix or dgCMatrix containing count data each column representing a spot or a cell, each row representing a gene; Or a data frame containing ligand-receptor pairs; Or a data frame containing gene-gene interactions and pathways from KEGG and Reactome as well as the information of transcriptional factors.
data_type	A character to define the type of data, select 'count' for the data matrix, 'lrpairs' for the data.frame containing lrpairs, 'pathways' for the data.frame containing pathways.
species	Species of the data. 'Human' or 'Mouse'.
geneinfo	A data frame of the system data containing gene symbols of 'Human' and 'Mouse' updated on June 30, 2021 for revising count matrix.

Value

A new matrix or data.frame.

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Description

Set the expected cell in SpaTalk object

Usage

```
set_expected_cell(object, value)
```

Arguments

object SpaTalk object

value Th number of expected cell for each spot, must be equal to the spot number.

Value

SpaTalk object

26 SpaTalk

SpaTalk

Definition of 'SpaTalk' class

Description

An S4 class containing the data, meta, and results of inferred cell type compositions, LR pairs, and pathways.

Slots

data A list containing the raw and normalized data.

meta A list containing the raw and new meta data.

para A list containing the parameters.

coef A matrix containing the results of deconvolution.

cellpair A list containing the cell-cell pairs based on the spatial distance.

dist A matrix containing the Euclidean distance among cells.

1rpair A data frame containing the inferred LR pairs.

tf A data frame containing the TFs of receptors.

lr_path A list containing the lrpairs and pathways.

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