Package ‘hoodscanR’

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Title  Spatial cellular neighbourhood scanning in R
Version  1.2.0
Description  hoodscanR is an user-friendly R package providing functions to assist
   cellular neighborhood analysis of any spatial transcriptomics data with single-cell resolution.
   All functions in the package are built based on the SpatialExperiment object,
   allowing integration into various spatial transcriptomics-related packages from Bioconductor.
   The package can result in cell-level neighborhood annotation output, along with functions to
   perform neighborhood colocalization analysis and neighborhood-based cell clustering.

biocViews  Spatial, Transcriptomics, SingleCell, Clustering
License  GPL-3 + file LICENSE
URL  https://github.com/DavisLaboratory/hoodscanR,
     https://davislaboratory.github.io/hoodscanR/

BugReports  https://github.com/DavisLaboratory/hoodscanR/issues

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Method to identify cellular spatial neighbourhood from single cell spatial transcriptomics data.

**Description**

hoodscanR implements a novel method to scan for cell neighbourhood from spatial transcriptomics data at single cell level, such as CosMx and MERFISH etc. hoodscanR takes the cellular position and cell type annotations as inputs, allowing cellular spatial neighbourhood analysis.

**Details**

Key neighborhood analysis functions include `findNearCells, scanHoods, mergeByGroup, calcMetrics, clustByHood`. Key visualisation functions include `plotTissue, plotHoodMat, plotColocal, plotProbDist`.

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calcMetrics

Calculate metrics for probability matrix

Description

Calculate metrics for probability matrix

Usage

calcMetrics(spe, pm = NA, pm_cols = NA, val_names = c("entropy", "perplexity"))

Arguments

spe            A SpatialExperiment object.
pm             Optional. The probability matrix.
pm_cols        The colnames of probability matrix. This is requires for SpatialExperiment input. Assuming that the probability is stored in the colData.
val_names      Character vector with length of 2. Column names used to store calculated entropy and perplexity.

Value

A SpatialExperiment object. Calculated entropy and perplexity are saved as columns in the colData of the SpatialExperiment object. Entropy and perplexity are calculated based on information theory:
P(x) is the probability calculated from the scanHoods function.
Entropy \( H(x) = -P(x)\log_2(P(x)) \)
Perplexity \( P(x) = 2^H(x) \)

By default, the calculated entropy and perplexity will be stored in the colData of the input spe, with column name as entropy and perplexity.

Examples

data("spe_test")
spe <- readHoodData(spe, anno_col = "celltypes")
fnc <- findNearCells(spe, k = 100)
pm <- scanHoods(fnc$distance)
pm2 <- mergeByGroup(pm, fnc$cells)
spe <- mergeHoodSpe(spe, pm2)
spe <- calcMetrics(spe, pm_cols = colnames(pm2))
**Description**

Cluster the probability matrix with K-means

**Usage**

```r
clustByHood(object, ...)  
```

## S4 method for signature 'matrix'

```r
clustByHood(object, k = 2^ncol(object) - 1, iter_max = 1000, nstart = 5)
```

## S4 method for signature 'SpatialExperiment'

```r
clustByHood(
  object,
  pm_cols,
  k = 0,
  iter_max = 1000,
  nstart = 5,
  algo = "Hartigan-Wong",
  val_names = "clusters"
)
```

**Arguments**

- `object`: A probability matrix or a SpatialExperiment.
- `...`: Ignore parameter.
- `k`: The number of clusters. By default is $2^{\text{ncol}(\text{object})}-1$.
- `iter_max`: the maximum number of iterations allowed.
- `nstart`: how many random sets should be chosen.
- `pm_cols`: The colnames of probability matrix. This is requires for SpatialExperiment input. Assuming that the probability is stored in the colData.
- `algo`: Algorithm to be used. Options include Hartigan-Wong, Lloyd, and MacQueen.
- `val_names`: Character. Column names used to store the clusters.

**Value**

A probability matrix or a SpatialExperiment object. For latter, the clustering results are saved in the colData of the SpatialExperiment object.
Examples

m <- matrix(abs(rnorm(1000 * 100)), 1000, 100)

clust <- clustByHood(m, k = 3)

findNearCells

Find the k-th nearest cells for each cell

Description

Find the k-th nearest cells for each cell

Usage

findNearCells(
  dat,
  k = 100,
  targetCell = FALSE,
  reportCellID = FALSE,
  reportDist = TRUE,
  anno_col = 0
)

Arguments

dat A SpatialExperiment object, can be generated using function readHoodData.
k The maximum number of nearest cells to compute.
targetCell Specify the cells to be the target cell for finding nearest cells.
reportCellID Logical. Set to TRUE to report cell id instead of cell types.
reportDist Logical. Set to TRUE to report the distance matrix.
anno_col Character vector. The name of annotation column to use.

Details

The findNearCells function uses the nn2 function from the RANN package, which uses the Approximate Near Neighbor (ANN) C++ library. For more information on the ANN library please see http://www.cs.umd.edu/~mount/ANN/.

Value

A list includes a data.frame and a matrix, describing the cell types and distances of the k-th nearest cells of each cell.
mergeByGroup

Merge probability matrix based on annotations

Usage
mergeByGroup(pm, group_df)

Arguments
- pm: A numeric matrix. Probability matrix generated by the soft_max function.
- group_df: A character matrix. Annotation of the neighboring cells to be used.

Value
A probability matrix, describing the probability of each cell being in each cellular neighborhood.

Examples
data("spe_test")
spe <- readHoodData(spe, anno_col = "celltypes")
fnc <- findNearCells(spe, k = 100)

pm <- scanHoods(fnc$distance)

pm2 <- mergeByGroup(pm, fnc$cells)
mergeHoodSpe

Merge probability matrix into SpatialExperiment object.

Description

Merge probability matrix into SpatialExperiment object.

Usage

mergeHoodSpe(spe, pm, val_names = NULL)

Arguments

- **spe**: A SpatialExperiment object.
- **pm**: Probability matrix. Can be obtained by the function mergeByGroup.
- **val_names**: Character vector with length of the ncol of pm.

Value

A SpatialExperiment object. Cell-level neighborhood information are saved in the colData of the SpatialExperiment object.

Examples

data("spe_test")
spe <- readHoodData(spe, anno_col = "celltypes")
fnc <- findNearCells(spe, k = 100)
pm <- scanHoods(fnc$distance)

pm2 <- mergeByGroup(pm, fnc$cells)
spe <- mergeHoodSpe(spe, pm2)

plotColocal

Plot heatmap for neighbourhood analysis

Description

Plot heatmap for neighbourhood analysis
plotColocal

Usage

plotColocal(object, ...)

## S4 method for signature 'matrix'
plotColocal(object, hm_width = 5, hm_height = 5)

## S4 method for signature 'SpatialExperiment'
plotColocal(
  object,
  pm_cols,
  self_cor = TRUE,
  by_group = NULL,
  hm_width = 5,
  hm_height = 5,
  cluster_row = TRUE,
  cluster_col = TRUE,
  return_matrix = FALSE
)

Arguments

object A probability matrix or SpatialExperiment.
...
ignore parameter.
hm_width Integer. The width of heatmap.
hm_height Integer. The height of heatmap.
pm_cols The colnames of probability matrix. This is requires for SpatialExperiment input. Assuming that the probability is stored in the colData.
self_cor Logical. By default is TRUE, indiciating running a correlation between neighbourhoods to perform a simple co-localization analysis. When this set to FALSE, it will plot the average probability of each neighbourhood by group using the by_group parameter.
by_group Character. This is required when self_cor is set to FALSE.
cluster_row Logical. Cluster rows.
cluster_col Logical. Cluster columns.
return_matrix Logical. Export a numeric matrix.

Value

A ComplexHeatmap plot. When return_matrix is set to TRUE, return a matrix Object.

Examples

data("spe_test")

spe <- readHoodData(spe, anno_col = "celltypes")
fnc <- findNearCells(spe, k = 100)

pm <- scanHoods(fnc$distance)

pm2 <- mergeByGroup(pm, fnc$cells)

spe <- mergeHoodSpe(spe, pm2)

plotColocal(spe, pm_cols = colnames(pm2))

plotColocal(spe, pm_cols = colnames(pm2), self_cor = FALSE, by_group = "cell_annotation")

---

**plotHoodMat**

**Plot probability matrix as a heatmap**

**Description**

Plot probability matrix as a heatmap

**Usage**

`plotHoodMat(object, ...)`

```r
## S4 method for signature 'matrix'
plotHoodMat(
  object,
  targetCells = NA,
  n = 30,
  hm_width = 4,
  hm_height = 15,
  clusterRows = TRUE,
  clusterCols = TRUE,
  title = "Probability of neighborhoods"
)
```

```r
## S4 method for signature 'SpatialExperiment'
plotHoodMat(
  object,
  pm_cols,
  targetCells = NA,
  n = 30,
  hm_width = 4,
  hm_height = 15,
  clusterRows = TRUE,
  clusterCols = TRUE,
  title = "Probability of neighborhoods"
)
```
## Arguments

- **object**
  A probability matrix or SpatialExperiment.

- **targetCells**
  Character. Optional. Can specify one or more cells to be plotted.

- **n**
  Integer. The number of randomly selected cells to be plotted. This parameter will be used when targetCells is not specified.

- **hm_width**
  Integer. The width of heatmap.

- **hm_height**
  Integer. The height of heatmap.

- **clusterRows**
  Logical. Cluster rows or not.

- **clusterCols**
  Logical. Cluster columns or not.

- **title**
  Title of the heatmap.

- **pm_cols**
  The colnames of probability matrix. This is required for SpatialExperiment input. Assuming that the probability is stored in the colData.

## Value

A ComplexHeatmap plot.

## Examples

```r
data("spe_test")

spe <- readHoodData(spe, anno_col = "celltypes")

fnc <- findNearCells(spe, k = 100)

pm <- scanHoods(fnc$distance)

pm2 <- mergeByGroup(pm, fnc$cells)

spe <- mergeHoodSpe(spe, pm2)

plotHoodMat(spe, pm_cols = colnames(pm2))
```

---

### Description

Plot probability distribution
plotProbDist

Usage

plotProbDist(object, ...)  

## S4 method for signature 'matrix'
plotProbDist(object, targetCells = NA, ...)

## S4 method for signature 'SpatialExperiment'
plotProbDist(
object,
pm_cols,
targetCells = NA,
by_cluster = FALSE,
show_clusters = as.character(seq(6)),
plot_all = FALSE,
sample_size = 2,
...  )

Arguments

object  A probability matrix or SpatialExperiment.

...  aesthetic mappings to pass to ggplot2::aes_string().

targetCells  Character. Optional. Can specify one or more cells to be plotted.

pm_cols  The colnames of probability matrix. This is requires for SpatialExperiment input. Assuming that the probability is stored in the colData.

by_cluster  Logical. By default is TRUE, to plot distribution by each cluster.

show_clusters  Character. The cluster to be plotted, by default is 1 to 6.

plot_all  Logical. By default is FALSE, set this to true to plot box plot instead of bar plot to show all cells in each cluster.

sample_size  Integer. By default is 2, sampling two cell from each cluster to be plotted.

Value

A ggplot object.

Examples

data("spe_test")

spe <- readHoodData(spe, anno_col = "celltypes")

fnc <- findNearCells(spe, k = 100)

pm <- scanHoods(fnc$distance)

pm2 <- mergeByGroup(pm, fnc$cells)
spe <- mergeHoodSpe(spe, pm2)
plotProbDist(spe, pm_cols = colnames(pm2))

---

**plotTissue**

*Plot cells based on cell position on tissue.*

**Description**

Plot cells based on cell position on tissue.

**Usage**

```r
plotTissue(
  spe,
  targetcell = FALSE,
  k_near = 100,
  targetsize = 3,
  targetshape = 1,
  targetcolor = "red",
  scaleFactor = 1,
  reverseY = TRUE,
  ...
)
```

**Arguments**

- `spe` SpatialExperiment object.
- `targetcell` Optional. Can input ONE specific cell id to zoom-in on the region of a specific cell.
- `k_near` Optional. If `targetcell` is specified, the `k_near` cells around the `targetcell` will be plotted.
- `targetsizem` Dot size of the `targetcell`.
- `targetshape` Shape of the `targetcell`.
- `targetcolor` Colour of the `targetcell`.
- `scaleFactor` Scale factor to align with the image.
- `reverseY` Reverse y coordinates.
- `...` aesthetic mappings to pass to `ggplot2::aes_string()`.

**Value**

A ggplot object.
readHoodData

Examples

data("spe_test")

plotTissue(spe, color = celltypes)

readHoodData

Read cellular position and annotation data into a list object.

Description

Read cellular position and annotation data into a list object.

Usage

readHoodData(
  spe = NA,
  anno_col = NA,
  cell_pos_dat = NA,
  cell_anno_dat = NA,
  pos_col = NA
)

Arguments

spe          SpatialExperiment object.
anno_col     Character. The column name of the annotation to be used in the
             following neighbourhood analysis.
cell_pos_dat data.frame object contains the cellular positions.
cell_anno_dat data.frame object contains the cell annotations.
pos_col     Character. If the x and y are in the colData instead of in the SpatialCoords
             of spe, can specify this parameter.

Value

A SpatialExperiment object.

Examples

data("spe_test")

spe <- readHoodData(spe, anno_col = "celltypes")
scanHoods  
*Scan cellular neighbourhoods.*

**Description**

Scan cellular neighbourhoods.

**Usage**

```r
scanHoods(
  m,
  mode = c("proximityFocused", "smoothFadeout"),
  tau = NA,
  t_init = NA
)
```

**Arguments**

- `m` Distance matrix. Can be obtained from function `findNearCells`.
- `mode` Character. Either `proximityFocused` or `smoothFadeout`. By default is `proximityFocused`.
- `tau` The hyperparameter `tau`, by default is $\text{median}(m^2)/5$.
- `t_init` An initial `tau`. In the `smoothFadeout` mode, user can provide an initial `tau` for optimization.

**Value**

A probability matrix.

**Examples**

```r
m <- matrix(abs(rnorm(1000 * 100)), 1000, 100)
pm <- scanHoods(m)
```

---

**spe_test**  
*Example test spatial transcriptomics data*

**Description**

hoodscanR-package has 1 datasets:

- spe_test Example test spatial transcriptomics data in SpatialExperiment format. This test data is randomly subsetting from the publicly available CosMx non-small cell lung cancer data. Source data: https://nanostring.com/products/cosmx-spatial-molecular-imager/nsclc-ffpe-dataset/.
Usage
data("spe_test")

Format
A SpatialExperiment object

Value
A SpatialExperiment object

Examples
data(spe_test)
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