Package 'SpotSweeper'

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Title Spatially-aware quality control for spatial transcriptomics

Version 1.0.2

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Description Spatially-aware quality control (QC) software for both spot-level
and artifact-level QC in spot-based spatial transcripomics, such as 10x
Visium. These methods calculate local (nearest-neighbors) mean and variance
of standard QC metrics (library size, unique genes, and mitochondrial
percentage) to identify outliers spot and large technical artifacts.

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URL <https://github.com/MicTott/SpotSweeper>

BugReports <https://support.bioconductor.org/tag/SpotSweeper>

biocViews Software, Spatial, Transcriptomics, QualityControl,
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grDevices

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Contents

DLPFC_artifact   *human DLPFC dataset with a technical artifact (hangnail).*

Description

The `DLPFC_artifact` dataset is a `SpatialExperiment` object containing a single-sample subset of the human dorsolateral prefrontal cortex (DLPFC) dataset from Hukki-Myers et al. 2023. This particular sample ('Br2743_ant') is included to demonstrate the identification and removal of technical artifacts within spatial transcriptomics data. The dataset serves as an example for artifact detection using the 'SpotSweeper' workflow.

Usage

```
data(DLPFC_artifact)
```

Format

An SpatialExperiment object.

Source

spatialLIBD

References

Hukki-Myers et al. (2023) bioRxiv (bioRxiv)

Examples

```
data(DLPFC_artifact)
```

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findArtifacts   *Identify and annotate artifacts in spatial transcriptomics data*

Description

This function identifies and annotates potential artifacts in spatial transcriptomics data. Artifacts are detected based on local mito variance, and the results are added to the original SpatialExperiment (sce) object.

Usage

```
findArtifacts(
  spe,
  mito_percent = "expr_chrM_ratio",
  mito_sum = "expr_chrM",
  samples = "sample_id",
  n_rings = 5,
  log = TRUE,
  name = "artifact",
  var_output = TRUE
)
```

Arguments

spe	A SingleCellExperiment object.
mito_percent	The column name representing the mitochondrial percent. Default is 'expr_chrM_ratio'.
mito_sum	The column name representing sum mitochondrial expression. Default is 'expr_chrM'.
samples	The column name representing sample IDs. Default is 'sample_id'.
n_rings	The number of rings for local mito variance calculation. Default is 5.
log	Logical, indicating whether to log1p transform mito_percent. Default is TRUE.
name	Prefix for the local variance column names. Default is 'artifact'.
var_output	Logical, indicating whether to include local variances in the output. Default is TRUE.

Value

Returns the modified SingleCellExperiment object with artifact annotations.

See Also

localVariance

Examples

```
library(SpotSweeper)
library(SpatialExperiment)
library(escheR)

data(DLPFC_artifact)
spe <- DLPFC_artifact

# find artifacts
spe <- findArtifacts(spe,
    mito_percent = "expr_chrM_ratio",
    mito_sum = "expr_chrM",
    n_rings = 2, # 5 recommended, using 1 for time
    name = "artifact"
)
```

---

localOutliers | *localOutliers Function*

Description

This function detects local outliers in spatial transcriptomics data based on standard quality control metrics, such as library size, unique genes, and mitochondrial ratio. Local outliers are defined as spots with low/high quality metrics compared to their surrounding neighbors, based on a modified z-score statistic.

Usage

```
localOutliers(
  spe,
  metric = "detected",
  direction = "lower",
  n_neighbors = 36,
  samples = "sample_id",
  log = TRUE,
  cutoff = 3
)
```

Arguments

spe	SpatialExperiment object
metric	colData QC metric to use for outlier detection
direction	Direction of outlier detection (higher, lower, or both)
n_neighbors	Number of nearest neighbors to use for outlier detection
samples	Column name in colData to use for sample IDs

log	Logical indicating whether to log1p transform the features (default is TRUE)
cutoff	Cutoff for outlier detection (default is 3)

Value

SpatialExperiment object with updated colData containing outputs

Examples

```
library(SpotSweeper)
library(SpatialExperiment)

# load example data
spe <- STexampleData::Visium_humanDLPFC()

# change from gene id to gene names
rownames(spe) <- rowData(spe)$gene_name

# drop out-of-tissue spots
spe <- spe[, spe$in_tissue == 1]
spe <- spe[, !is.na(spe$ground_truth)]

# Identifying the mitochondrial transcripts in our SpatialExperiment.
is.mito <- rownames(spe)[grepl("^MT-", rownames(spe))]

# Calculating QC metrics for each spot using scuttle
spe <- scuttle::addPerCellQCMetrics(spe, subsets = list(Mito = is.mito))
colnames(colData(spe))

# Identifying local outliers using SpotSweeper
spe <- localOutliers(spe,
                     metric = "sum",
                     direction = "lower",
                     log = TRUE
)
```

---

localVariance	*localVariance Function*

Description

This function does calculates the local variance based on kNN.

Usage

```
localVariance(
  spe,
  n_neighbors = 36,
```

```
    metric = c("expr_chrM_ratio"),
    samples = "sample_id",
    log = FALSE,
    name = NULL
)
```

Arguments

spe	SpatialExperiment object with the following columns in colData: sample_id, sum_umi, sum_gene
n_neighbors	Number of nearest neighbors to use for variance calculation
metric	metric to use for variance calculation
samples	Column in colData to use for sample ID
log	Whether to log1p transform the metric
name	Name of the new column to add to colData

Value

SpatialExperiment object with metric variance added to colData

Examples

```
# for more details see extended example in vignettes
library(SpotSweeper)
library(SpatialExperiment)
library(escheR)

# load example data
spe <- STexampleData::Visium_humanDLPFC()

# change from gene id to gene names
rownames(spe) <- rowData(spe)$gene_name

# show column data before SpotSweepR
colnames(colData(spe))

# drop out-of-tissue spots
spe <- spe[, spe$in_tissue == 1]
spe <- spe[, !is.na(spe$ground_truth)]

# Identifying the mitochondrial transcripts in our SpatialExperiment.
is.mito <- rownames(spe)[grepl("^MT-", rownames(spe))]

# Calculating QC metric for each spot using scuttle
spe <- scuttle::addPerCellQCMetrics(spe, subsets = list(Mito = is.mito))
colnames(colData(spe))

spe <- localVariance(spe,
    metric = "subsets_Mito_percent",
    n_neighbors = 36,
```

```
    name = "local_mito_variance_k36"
    )

plotQC(spe, metric="local_mito_variance_k36")
```

---

plotQC   *Plot QC metrics for a Single Sample in a SpatialExperiment object*

Description

This function generates a plot for a specified sample within a SpatialExperiment object, highlighting outliers based on a specified metric. The plot visualizes the metric of interest and indicates outliers with a distinct color.

Usage

```
plotQC(
  spe,
  sample_id = "sample_id",
  sample = unique(spe$sample_id)[1],
  metric = "detected",
  outliers = NULL,
  point_size = 2,
  colors = c("white", "black"),
  stroke = 1
)
```

Arguments

spe	A SpatialExperiment object containing the data to be plotted.
sample_id	A character string specifying the column name in colData(spe) that contains unique sample identifiers. Default is "sample_id".
sample	A character string or numeric value specifying the sample to be plotted. By default, it plots the first unique sample found in spe$sample_id.
metric	A character string specifying the metric to be visualized in the plot. This metric should be a column name in colData(spe).
outliers	A character string specifying the column name in colData(spe) that indicates whether a data point is considered an outlier. Default is NULL.
point_size	A numeric value specifying the size of the points in the plot. Default is 2.
colors	A character vector specifying the colors to be used for the gradient scale. If length is 2, the gradient will be a single color gradient.
stroke	A numeric value specifying the border thickness for outlier points. Default is 1.

Value

The function returns a plot object created by make_escheR and modified with additional layers for visualizing the specified metric and outliers. The plot is not explicitly printed by the function and should be printed by the caller.

Examples

```
library(SpotSweeper)
library(SpatialExperiment)
library(escheR)

# load example data
spe <- STexampleData::Visium_humanDLPFC()

# change from gene id to gene names
rownames(spe) <- rowData(spe)$gene_name

# drop out-of-tissue spots
spe <- spe[, spe$in_tissue == 1]
spe <- spe[, !is.na(spe$ground_truth)]

# Identifying the mitochondrial transcripts in our SpatialExperiment.
is.mito <- rownames(spe)[grepl("^MT-", rownames(spe))]

# Calculating QC metrics for each spot using scuttle
spe <- scuttle::addPerCellQCMetrics(spe, subsets = list(Mito = is.mito))
colnames(colData(spe))

# Identifying local outliers using SpotSweeper
spe <- localOutliers(spe,
                     metric = "sum",
                     direction = "lower",
                     log = TRUE
)

plotQC(spe, metric="sum", outliers="sum_outliers")
```

---

plotQCpdf   *Plot Outlier Metrics to PDF*

Description

This function generates a PDF file containing plots for each sample in the SpatialExperiment object, highlighting outliers based on specified metrics. Each plot visualizes outlier metrics for a single sample, allowing for easy comparison and analysis across samples.

Usage

```
plotQCpdf(
  spe,
  sample_id = "sample_id",
  metric = "detected",
  outliers = "local_outliers",
  colors = c("white", "black"),
  stroke = 1,
  point_size = 2,
  width = 5,
  height = 5,
  fname
)
```

Arguments

spe	A SpatialExperiment object containing the data to be plotted.
sample_id	A character string specifying the column name in colData(spe) that contains unique sample identifiers. Default is 'sample_id'.
metric	A character string specifying the metric to be visualized in the plot. This metric should be a column name in colData(spe).
outliers	A character string specifying the column name in colData(spe) that indicates whether a data point is considered an outlier. Default is local_outliers'.
colors	A character vector specifying the colors to be used for the gradient scale. If length is 2, the gradient will be a single color gradient
stroke	A numeric value specifying the border thickness for outlier points. Default is 1.
point_size	A numeric value specifying the size of the points in the plot. Default is 2.
width	A numeric value indicating the width of the plot. Default is 5.
height	A numeric value indicating the height of the plot. Default is 5.
fname	A character string specifying the path and name of the output PDF file.

Value

ggplot object if specified. Generates a plot otherwise.

Examples

```
library(SpotSweeper)
library(SpatialExperiment)
library(escheR)

# load example data
spe <- STexampleData::Visium_humanDLPFC()

tempFilePath <- file.path(tempdir(), "examplePlot.pdf")

# change from gene id to gene names
```

```
rownames(spe) <- rowData(spe)$gene_name

# drop out-of-tissue spots
spe <- spe[, spe$in_tissue == 1]
spe <- spe[, !is.na(spe$ground_truth)]

# Identifying the mitochondrial transcripts in our SpatialExperiment.
is.mito <- rownames(spe)[grepl("^MT-", rownames(spe))]

# Calculating QC metrics for each spot using scuttle
spe <- scuttle::addPerCellQCMetrics(spe, subsets = list(Mito = is.mito))
colnames(colData(spe))

# Identifying local outliers using SpotSweeper
spe <- localOutliers(spe,
                     metric = "sum",
                     direction = "lower",
                     log = TRUE
)

plotQCpdf(spe,
          metric="sum",
          outliers="sum_outliers",
          fname=tempFilePath)
```

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