

# Package ‘standR’

June 2, 2023

**Title** Spatial transcriptome analyses of Nanostring's DSP data in R

**Version** 1.5.1

**Description** standR is an user-friendly R package providing functions to assist conducting good-practice analysis of Nanostring's GeoMX DSP data. All functions in the package are built based on the SpatialExperiment object, allowing integration into various spatial transcriptomics-related packages from Bioconductor. standR allows data inspection, quality control, normalization, batch correction and evaluation with informative visualizations.

**biocViews** Spatial, Transcriptomics, GeneExpression,  
DifferentialExpression, QualityControl, Normalization,  
ExperimentHubSoftware

**License** MIT + file LICENSE

**URL** <https://github.com/DavisLaboratory/standR>

**BugReports** <https://github.com/DavisLaboratory/standR/issues>

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**Author** Ning Liu [aut, cre] (<<https://orcid.org/0000-0002-9487-9305>>),  
 Dharmesh D Bhuvu [aut] (<<https://orcid.org/0000-0002-6398-9157>>),  
 Ahmed Mohamed [aut]

**Maintainer** Ning Liu <liu.n@wehi.edu.au>

## R topics documented:

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|                |                                                                            |
|----------------|----------------------------------------------------------------------------|
| standR-package | <i>Tools for analyzing NanoString's GeoMX spatial transcriptomics data</i> |
|----------------|----------------------------------------------------------------------------|

---

## Description

standR implements a series of functions to facilitate inspection, analysis and visualization of the NanoString's GeoMX DSP datasets. standR takes the either the csv files from the Nanostring or DGEList object as input, allowing for multiple methods to be analyzed together.

**Details**

standR represents the GeoMX DSP data as SpatialExperiment objects, which can easily be integrated with a wide variety of Bioconductor packages. standR generates various plots, such as QC distribution plots, dimension reduction plots and RLE plots, for quality control of genes and region of interest (ROI) samples. Multiple normalization and batch correction methods are also provided in the package as well, with the ability to compute statistics for assessing the normalization/batch correction results.

**Author(s)**

Ning Liu <liu.n@wehi.edu.au>

---

addPerROIQC

---

*Add QC statistics to the Spatial Experiment object*


---

**Description**

Add QC statistics to the Spatial Experiment object

**Usage**

```
addPerROIQC(
  spe_object,
  sample_fraction = 0.9,
  rm_genes = TRUE,
  min_count = 5,
  design = NULL
)
```

**Arguments**

|                 |                                                                                                                                         |
|-----------------|-----------------------------------------------------------------------------------------------------------------------------------------|
| spe_object      | A SpatialExperiment object                                                                                                              |
| sample_fraction | Double. Genes with low count in more than this threshold of the samples will be removed. Default is 0.9                                 |
| rm_genes        | Logical. Decide whether genes with low count in more than sample_fraction of the samples are removed from the dataset. Default is TRUE. |
| min_count       | Integer. Minimum read count to calculate count threshold. Default is 5.                                                                 |
| design          | Generate using model.matrix, if this is specify, edgeR::filterByExpr will be used to filter genes.                                      |

**Value**

A SpatialExperiment object

**Examples**

```
data("dkd_spe_subset")
spe_filtered <- addPerROIQC(dkd_spe_subset)
spe_filtered
```

---

```
computeClusterEvalStats
```

*Calculate statistics for evaluating batch correction*

---

**Description**

Calculate statistics for evaluating batch correction

**Usage**

```
computeClusterEvalStats(
  spe_object,
  foiColumn,
  precomputed = NULL,
  n_dimension = c(1, 2),
  assay = 2
)
```

**Arguments**

|             |                                                                                                                                      |
|-------------|--------------------------------------------------------------------------------------------------------------------------------------|
| spe_object  | A Spatial Experiment object.                                                                                                         |
| foiColumn   | A column name indicating the factor of interest to be tested, can be biological factor or batch factor.                              |
| precomputed | a dimensional reduction results from stats::prcomp. result in reducedDims(object) to plot. Default is NULL, we will compute for you. |
| n_dimension | The top n dimensions to be plotted                                                                                                   |
| assay       | a numeric or character, specifying the assay to use (for SummarizedExperiment and its derivative classes).                           |

**Value**

A dataframe object containing the clustering evaluating statistics.

**Examples**

```
library(scater)
data("dkd_spe_subset")
computeClusterEvalStats(dkd_spe_subset, "SlideName")
```

---

|                |                                                   |
|----------------|---------------------------------------------------|
| dkd_spe_subset | <i>Description of the standR example datasets</i> |
|----------------|---------------------------------------------------|

---

**Description**

standR-package has 1 datasets:

- dkd\_spe\_subset Example subset of a GeoMX DSP WTA dataset,

**Usage**

```
data("dkd_spe_subset")
```

**Format**

A SpatialExperiment object with 3000 rows and 70 samples:

**Source**

<http://nanosttring-public-share.s3-website-us-west-2.amazonaws.com/GeoScriptHub/KidneyDataset/>

**Examples**

```
data(dkd_spe_subset)
```

---

|         |                                                                               |
|---------|-------------------------------------------------------------------------------|
| drawPCA | <i>Compute and plot the results of a PCA analysis on gene expression data</i> |
|---------|-------------------------------------------------------------------------------|

---

**Description**

Compute and plot the results of a PCA analysis on gene expression data

**Usage**

```
drawPCA(object, dims = c(1, 2), ...)

## S4 method for signature 'ExpressionSet'
drawPCA(object, dims = c(1, 2), precomputed = NULL, textScale = 1, ...)

## S4 method for signature 'SummarizedExperiment'
drawPCA(
  object,
  dims = c(1, 2),
  assay = 1,
```

```

    precomputed = NULL,
    textScale = 1,
    ...
)

## S4 method for signature 'SingleCellExperiment'
drawPCA(
  object,
  dims = c(1, 2),
  assay = 1,
  precomputed = NULL,
  textScale = 1,
  ...
)

## S4 method for signature 'SpatialExperiment'
drawPCA(
  object,
  dims = c(1, 2),
  assay = 1,
  precomputed = NULL,
  textScale = 1,
  ...
)

```

### Arguments

|             |                                                                                                                       |
|-------------|-----------------------------------------------------------------------------------------------------------------------|
| object      | a DGEList, SummarizedExperiment or ExpressionSet object containing gene expression data.                              |
| dims        | a numeric, containing 2 values specifying the dimensions to plot.                                                     |
| ...         | aesthetic mappings to pass to <code>ggplot2::aes_string()</code> .                                                    |
| precomputed | a dimensional reduction results from <code>stats::prcomp</code> . result in <code>reducedDims(object)</code> to plot. |
| textScale   | a numeric, specifying the relative scale factor to apply to text on the plot.                                         |
| assay       | a numeric or character, specifying the assay to use (for SummarizedExperiment and its derivative classes).            |

### Value

a ggplot2 object

### Examples

```

data("dkd_spe_subset")
drawPCA(dkd_spe_subset)

```

findBestK

*Testing multiple K for RUV4 batch correction to find the best K.***Description**

Testing multiple K for RUV4 batch correction to find the best K.

**Usage**

```
findBestK(
  spe,
  maxK = 10,
  factor_of_int,
  factor_batch,
  NCGs,
  point_size = 3,
  line_col = "black",
  point_col = "black",
  text_size = 13
)
```

**Arguments**

|               |                                                                                           |
|---------------|-------------------------------------------------------------------------------------------|
| spe           | A Spatial Experiment object.                                                              |
| maxK          | Integer. The max k to test, will test k from 1 to maxK, by default is 10.                 |
| factor_of_int | Column name(s) to indicate the factors of interest. This is required for the RUV4 method. |
| factor_batch  | Column name to indicate the batch.                                                        |
| NCGs          | Negative control genes. This is required for the RUV4 method.                             |
| point_size    | Numeric. Plotting parameter.                                                              |
| line_col      | Character. Plotting parameter.                                                            |
| point_col     | Character. Plotting parameter.                                                            |
| text_size     | Numeric. Plotting parameter.                                                              |

**Value**

A ggplot object.

**Examples**

```
data("dkd_spe_subset")
spe <- findNCGs(dkd_spe_subset, top_n = 100)
findBestK(spe,
  factor_of_int = c("disease_status"),
  factor_batch = "SlideName", NCGs = S4Vectors::metadata(spe)$NCGs
)
```

---

|          |                                                               |
|----------|---------------------------------------------------------------|
| findNCGs | <i>Get negative control genes from each batch of the data</i> |
|----------|---------------------------------------------------------------|

---

### Description

Get negative control genes from each batch of the data

### Usage

```
findNCGs(spe, n_assay = 2, batch_name = "SlideName", top_n = 200)
```

### Arguments

|            |                                                                           |
|------------|---------------------------------------------------------------------------|
| spe        | A Spatial Experiment object.                                              |
| n_assay    | Integer to indicate the nth count table in the assay(spe) to be used.     |
| batch_name | Column name indicating batches.                                           |
| top_n      | Integer indicate how many genes to be included as negative control genes. |

### Value

A Spatial Experiment object, conatining negative control genes in the metadata.

### Examples

```
data("dkd_spe_subset")

spe <- findNCGs(dkd_spe_subset, top_n = 100)
S4Vectors::metadata(spe)$NCGs
```

---

|                      |                                        |
|----------------------|----------------------------------------|
| geomxBatchCorrection | <i>Batch correction for GeoMX data</i> |
|----------------------|----------------------------------------|

---

### Description

Batch correction for GeoMX data

**Usage**

```
geomxBatchCorrection(
  spe,
  k,
  factors,
  NCGs,
  n_assay = 2,
  batch = NULL,
  batch2 = NULL,
  covariates = NULL,
  design = matrix(1, ncol(spe), 1),
  method = c("RUV4", "Limma", "RUVg"),
  isLog = TRUE
)
```

**Arguments**

|            |                                                                                                                                                                          |
|------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| spe        | A Spatial Experiment object.                                                                                                                                             |
| k          | The number of unwanted factors to use. Can be 0. This is required for the RUV4 method.                                                                                   |
| factors    | Column name(s) to indicate the factors of interest. This is required for the RUV4 method.                                                                                |
| NCGs       | Negative control genes. This is required for the RUV4 method.                                                                                                            |
| n_assay    | Integer to indicate the nth count table in the assay(spe) to be used.                                                                                                    |
| batch      | A vector indicating batches. This is required for the Limma method.                                                                                                      |
| batch2     | A vector indicating the second series of batches. This is specific for the Limma method.                                                                                 |
| covariates | A matrix or vector of numeric covariates to be adjusted for.                                                                                                             |
| design     | A design matrix relating to treatment conditions to be preserved, can be generated using <code>stats::model.matrix</code> function with all biological factors included. |
| method     | Can be either RUV4 or Limma or RUVg, by default is RUV4.                                                                                                                 |
| isLog      | Logical vector, indicating if the count table is log or not.                                                                                                             |

**Value**

A Spatial Experiment object, containing the normalized count and normalization factor. For method RUV4 and RUVg, the W matrices will be saved in the colData of the object.

**Note**

The normalised count is not intended to be used directly for linear modelling. For linear modelling, it is better to include the batch factors/W matrices in the linear model.

## References

Gagnon-Bartsch, J. A., Jacob, L., & Speed, T. P. (2013). Removing unwanted variation from high dimensional data with negative controls. Berkeley: Tech Reports from Dep Stat Univ California, 1-112.

Ritchie, M. E., Phipson, B., Wu, D. I., Hu, Y., Law, C. W., Shi, W., & Smyth, G. K. (2015). limma powers differential expression analyses for RNA-sequencing and microarray studies. *Nucleic acids research*, 43(7), e47-e47.

## Examples

```
data("dkd_spe_subset")
spe <- findNCGs(dkd_spe_subset, top_n = 100)
spe_ruv <- geomxBatchCorrection(spe,
  k = 3,
  factors = c("disease_status", "region"),
  NCGs = S4Vectors::metadata(spe)$NCGs
)
```

---

geomxNorm

*Perform normalization to GeoMX data*

---

## Description

Perform normalization to GeoMX data

## Usage

```
geomxNorm(
  spe_object,
  method = c("TMM", "RPKM", "TPM", "CPM", "upperquartile", "sizefactor"),
  log = TRUE
)
```

## Arguments

|            |                                                                                                                                                                                                               |
|------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| spe_object | A SpatialExperiment object.                                                                                                                                                                                   |
| method     | Normalization method to use. Options: TMM, RPKM, TPM, CPM, upperquartile, sizefactor. RPKM and TPM require gene length information, which should be added into rowData(spe). Note that TMM here is TMM + CPM. |
| log        | Log-transformed or not.                                                                                                                                                                                       |

## Value

A SpatialExperiment object, with the second assay being the normalized count matrix. The normalised count is stored in the assay slot called "logcounts" by default. With method TMM and sizefactor, the norm.factor will be saved in the metadata of the SpatialExperiment object.

**Note**

The normalised count is not intended to be used directly for linear modelling. For linear modelling, it is better to include the normalized factors in the "norm.factors" column of the DGEList object.

**References**

Robinson, M. D., McCarthy, D. J., & Smyth, G. K. (2010). edgeR: a Bioconductor package for differential expression analysis of digital gene expression data. *Bioinformatics*, 26(1), 139-140.

Love, M., Anders, S., & Huber, W. (2014). Differential analysis of count data—the DESeq2 package. *Genome Biol*, 15(550), 10-1186.

**Examples**

```
data("dkd_spe_subset")

spe_tmm <- geomxNorm(dkd_spe_subset, method = "TMM")
spe_upq <- geomxNorm(dkd_spe_subset, method = "upperquartile")
spe_deseqnorm <- geomxNorm(dkd_spe_subset, method = "sizefactor")
```

---

plotClusterEvalStats    *Compare and evaluate different batch corrected data with plotting.*

---

**Description**

Compare and evaluate different batch corrected data with plotting.

**Usage**

```
plotClusterEvalStats(
  spe_list,
  bio_feature_name,
  batch_feature_name,
  data_names,
  colors = NA
)
```

**Arguments**

|                    |                                       |
|--------------------|---------------------------------------|
| spe_list           | A list of Spatial Experiment object.  |
| bio_feature_name   | The common biological variation name. |
| batch_feature_name | The common batch variation name.      |
| data_names         | Data names.                           |
| colors             | Color values of filing the bars.      |

**Value**

A ggplot object.

**Examples**

```
library(scater)
data("dkd_spe_subset")
spe <- dkd_spe_subset
spe2 <- spe
spe3 <- spe
plotClusterEvalStats(list(spe, spe2, spe3),
  bio_feature_name = "region",
  batch_feature_name = "SlideName", c("test1", "test2", "test3")
)
```

---

|        |                                                                                                |
|--------|------------------------------------------------------------------------------------------------|
| plotDR | <i>Compute and plot the results of any dimension reduction methods on gene expression data</i> |
|--------|------------------------------------------------------------------------------------------------|

---

**Description**

Compute and plot the results of any dimension reduction methods on gene expression data

**Usage**

```
plotDR(object, dims = c(1, 2), ...)

## S4 method for signature 'SingleCellExperiment'
plotDR(object, dims, dimred = "PCA", textScale = 1, ...)

## S4 method for signature 'SpatialExperiment'
plotDR(object, dims, dimred = "PCA", textScale = 1, ...)
```

**Arguments**

|           |                                                                                                                 |
|-----------|-----------------------------------------------------------------------------------------------------------------|
| object    | a DGEList, SummarizedExperiment or ExpressionSet object containing gene expression data.                        |
| dims      | a numeric, containing 2 values specifying the dimensions to plot.                                               |
| ...       | aesthetic mappings to pass to <code>ggplot2::aes_string()</code> .                                              |
| dimred    | a string or integer scalar indicating the reduced dimension result in <code>reducedDims(object)</code> to plot. |
| textScale | a numeric, specifying the relative scale factor to apply to text on the plot.                                   |

**Value**

a ggplot2 object

**Examples**

```
library(scater)
data("dkd_spe_subset")
spe <- scater::runPCA(dkd_spe_subset)
plotDR(spe, dimred = "PCA")
```

plotGeneQC

*Plot gene-wise QC plot***Description**

Plot gene-wise QC plot

**Usage**

```
plotGeneQC(
  spe,
  top_n = 9,
  ordannots = c(),
  point_size = 1,
  line_type = "dashed",
  line_col = "darkred",
  line_cex = 1,
  hist_col = "black",
  hist_fill = "skyblue",
  bin_num = 30,
  text_size = 13,
  layout_ncol = 1,
  layout_nrow = 2,
  layout_height = c(1, 1),
  ...
)
```

**Arguments**

|            |                                                                    |
|------------|--------------------------------------------------------------------|
| spe        | A SpatialExperiment object.                                        |
| top_n      | Integer. Indicating the top n genes will be plotted. Default is 9. |
| ordannots  | variables or computations to sort samples by (tidy style).         |
| point_size | Numeric. Point size.                                               |
| line_type  | Character. Line types for ggplot.                                  |
| line_col   | Color for line.                                                    |
| line_cex   | Cex for line.                                                      |
| hist_col   | Color for histogram.                                               |

|               |                                                                |
|---------------|----------------------------------------------------------------|
| hist_fill     | Fill for histogram.                                            |
| bin_num       | Bin numbers for histogram.                                     |
| text_size     | Text size.                                                     |
| layout_ncol   | Integer. Column number for layout. Default is 1.               |
| layout_nrow   | Integer. Row number for layout. Default is 2.                  |
| layout_height | Vector of numerics with length of 2. Default is c(1, .4).      |
| ...           | aesthetic mappings to pass to ggplot2::aes() of the dot plots. |

**Value**

A ggplot object

**Examples**

```
data("dkd_spe_subset")
spe <- addPerROIQC(dkd_spe_subset)
plotGeneQC(spe)
```

---

|         |                                                                               |
|---------|-------------------------------------------------------------------------------|
| plotMDS | <i>Compute and plot the results of a MDS analysis on gene expression data</i> |
|---------|-------------------------------------------------------------------------------|

---

**Description**

Compute and plot the results of a MDS analysis on gene expression data

**Usage**

```
plotMDS(
  object,
  dims = c(1, 2),
  precomputed = NULL,
  textScale = 1,
  assay = 1,
  ...
)

## S4 method for signature 'DGEList'
plotMDS(
  object,
  dims = c(1, 2),
  precomputed = NULL,
  textScale = 1,
  assay = 1,
  ...
)
```

```
## S4 method for signature 'ExpressionSet'
plotMDS(
  object,
  dims = c(1, 2),
  precomputed = NULL,
  textScale = 1,
  assay = 1,
  ...
)

## S4 method for signature 'SummarizedExperiment'
plotMDS(
  object,
  dims = c(1, 2),
  precomputed = NULL,
  textScale = 1,
  assay = 1,
  ...
)

## S4 method for signature 'SingleCellExperiment'
plotMDS(
  object,
  dims = c(1, 2),
  precomputed = NULL,
  textScale = 1,
  assay = 1,
  ...
)

## S4 method for signature 'SpatialExperiment'
plotMDS(
  object,
  dims = c(1, 2),
  precomputed = NULL,
  textScale = 1,
  assay = 1,
  ...
)
```

### Arguments

|             |                                                                                          |
|-------------|------------------------------------------------------------------------------------------|
| object      | a DGEList, SummarizedExperiment or ExpressionSet object containing gene expression data. |
| dims        | a numeric, containing 2 values specifying the dimensions to plot.                        |
| precomputed | a dimensional reduction results from either <code>limma::plotMDS</code> .                |
| textScale   | a numeric, specifying the relative scale factor to apply to text on the plot.            |

|       |                                                                                                            |
|-------|------------------------------------------------------------------------------------------------------------|
| assay | a numeric or character, specifying the assay to use (for SummarizedExperiment and its derivative classes). |
| ...   | aesthetic mappings to pass to ggplot2::aes_string().                                                       |

**Value**

a ggplot2 object

**Examples**

```
data("dkd_spe_subset")
standR::plotMDS(dkd_spe_subset)
```

---

plotPairPCA

---

*Plot pair-wise PCA plots for multiple dimensions*


---

**Description**

Plot pair-wise PCA plots for multiple dimensions

**Usage**

```
plotPairPCA(
  spe_object,
  n_dimension = 3,
  precomputed = NULL,
  assay = 2,
  title = NA,
  title.size = 14,
  rmduplabs = FALSE,
  flipcoord = FALSE,
  ...
)
```

**Arguments**

|             |                                                                                                                                      |
|-------------|--------------------------------------------------------------------------------------------------------------------------------------|
| spe_object  | A SpatialExperiment object.                                                                                                          |
| n_dimension | The top n dimensions to be plotted                                                                                                   |
| precomputed | a dimensional reduction results from stats::prcomp. result in reducedDims(object) to plot. Default is NULL, we will compute for you. |
| assay       | a numeric or character, specifying the assay to use (for SummarizedExperiment and its derivative classes).                           |
| title       | Character vector, title to put at the top.                                                                                           |
| title.size  | Numeric vector, size of the title.                                                                                                   |
| rmduplabs   | Remove duplicated labels from the plot. FALSE by default.                                                                            |
| flipcoord   | Flip the xy coordinates. FALSE by default.                                                                                           |
| ...         | aesthetic mappings to pass to ggplot2::aes().                                                                                        |

**Value**

A ggplot object.

**Examples**

```
data("dkd_spe_subset")
plotPairPCA(dkd_spe_subset)
```

---

plotPCAbiplot

*Plot PCA bi plot*


---

**Description**

Plot PCA bi plot

**Usage**

```
plotPCAbiplot(
  spe_object,
  n_loadings = 10,
  dims = c(1, 2),
  precomputed = NULL,
  assay = 1,
  arrow_x = 0,
  arrow_y = 0,
  ...
)
```

**Arguments**

|             |                                                                                                                                      |
|-------------|--------------------------------------------------------------------------------------------------------------------------------------|
| spe_object  | A SpatialExperiment object.                                                                                                          |
| n_loadings  | Plot the top n gene loadings                                                                                                         |
| dims        | The top n dimensions to be plotted                                                                                                   |
| precomputed | a dimensional reduction results from stats::prcomp. result in reducedDims(object) to plot. Default is NULL, we will compute for you. |
| assay       | a numeric or character, specifying the assay to use (for SummarizedExperiment and its derivative classes).                           |
| arrow_x     | a numeric, indicating the x coordinate of the base of the arrow.                                                                     |
| arrow_y     | a numeric, indicating the y coordinate of the base of the arrow.                                                                     |
| ...         | aesthetic mappings to pass to ggplot2::aes().                                                                                        |

**Value**

A ggplot object.

**Examples**

```
data("dkd_spe_subset")
plotPCAbiplot(dkd_spe_subset)
```

---

|             |                                                                                      |
|-------------|--------------------------------------------------------------------------------------|
| plotRLEExpr | <i>Compute and plot relative log expression (RLE) values of gene expression data</i> |
|-------------|--------------------------------------------------------------------------------------|

---

**Description**

Compute and plot relative log expression (RLE) values of gene expression data

**Usage**

```
plotRLEExpr(object, ordannots = c(), ...)
```

```
## S4 method for signature 'DGEList'
```

```
plotRLEExpr(object, ordannots = c(), ...)
```

```
## S4 method for signature 'ExpressionSet'
```

```
plotRLEExpr(object, ordannots = c(), ...)
```

```
## S4 method for signature 'SummarizedExperiment'
```

```
plotRLEExpr(object, ordannots, assay = 1, ...)
```

**Arguments**

|           |                                                                                                            |
|-----------|------------------------------------------------------------------------------------------------------------|
| object    | a DGEList, SummarizedExperiment or ExpressionSet object containing gene expression data.                   |
| ordannots | variables or computations to sort samples by (tidy style).                                                 |
| ...       | aesthetic mappings to pass to ggplot2::aes_string().                                                       |
| assay     | a numeric or character, specifying the assay to use (for SummarizedExperiment and its derivative classes). |

**Value**

a ggplot2 object, containing the RLE plot.

**Examples**

```
data("dkd_spe_subset")
plotRLEExpr(dkd_spe_subset)
```

---

|           |                                 |
|-----------|---------------------------------|
| plotROIQC | <i>Plot Sample-wise QC plot</i> |
|-----------|---------------------------------|

---

**Description**

Plot Sample-wise QC plot

**Usage**

```
plotROIQC(
  spe_object,
  x_axis = "AOINucleiCount",
  y_axis = "lib_size",
  x_lab = "AOINucleiCount",
  y_lab = "Library size",
  x_threshold = NULL,
  y_threshold = NULL,
  regression_col = "purple",
  hist_col = "black",
  hist_fill = "white",
  bin_num = 50,
  threshold_col = "red",
  threshold_linetype = "dashed",
  layout_ncol = 2,
  layout_nrow = 2,
  layout_height = c(0.8, 2.5),
  layout_width = c(2.5, 0.8),
  ...
)
```

**Arguments**

|                |                                    |
|----------------|------------------------------------|
| spe_object     | A SpatialExperiment object.        |
| x_axis         | Numeric feature to plot as x axis. |
| y_axis         | Numeric feature to plot as y axis. |
| x_lab          | Label name for x axis.             |
| y_lab          | Label name for y axis.             |
| x_threshold    | Threshold to draw.                 |
| y_threshold    | Threshold to draw.                 |
| regression_col | Color for the regression line.     |
| hist_col       | Color for the histograms.          |
| hist_fill      | Fill for the histograms.           |
| bin_num        | Bin numbers for the histograms.    |
| threshold_col  | Threshold line color.              |

|                    |                                                                |
|--------------------|----------------------------------------------------------------|
| threshold_linetype | Threshold line type.                                           |
| layout_ncol        | Column number layout.                                          |
| layout_nrow        | Row number layout.                                             |
| layout_height      | Height layout.                                                 |
| layout_width       | Width layout.                                                  |
| ...                | aesthetic mappings to pass to ggplot2::aes() of the dot plots. |

**Value**

A ggplot object.

**Examples**

```
library(ggplot2)
library(patchwork)
data("dkd_spe_subset")
spe <- addPerROIQC(dkd_spe_subset)

plotROIQC(spe)
```

---

|                |                                                            |
|----------------|------------------------------------------------------------|
| plotSampleInfo | <i>Plot the user-defined meta data using alluvium plot</i> |
|----------------|------------------------------------------------------------|

---

**Description**

Plot the user-defined meta data using alluvium plot

**Usage**

```
plotSampleInfo(spe_object, column2plot, textsize = 3)
```

**Arguments**

|             |                             |
|-------------|-----------------------------|
| spe_object  | A SpatialExperiment object. |
| column2plot | Which columns to plot.      |
| textsize    | text size.                  |

**Value**

A ggplot object

**Examples**

```
library(ggalluvial)

data("dkd_spe_subset")
plotSampleInfo(dkd_spe_subset, column2plot = c("SlideName", "disease_status", "region"))
```

---

plotScreePCA

*Plot the PCA scree plot.*


---

**Description**

Plot the PCA scree plot.

**Usage**

```
plotScreePCA(
  spe_object,
  dims = ncol(spe_object),
  precomputed = NULL,
  assay = 1,
  bar_color = "black",
  bar_fill = "royalblue",
  bar_width = 0.8,
  point_col = "tomato3",
  line_col = "tomato3",
  point_size = 2
)
```

**Arguments**

|             |                                                                                                                                      |
|-------------|--------------------------------------------------------------------------------------------------------------------------------------|
| spe_object  | A SpatialExperiment object.                                                                                                          |
| dims        | The top n dimensions to be plotted                                                                                                   |
| precomputed | a dimensional reduction results from stats::prcomp. result in reducedDims(object) to plot. Default is NULL, we will compute for you. |
| assay       | a numeric or character, specifying the assay to use (for SummarizedExperiment and its derivative classes).                           |
| bar_color   | Color for bar.                                                                                                                       |
| bar_fill    | Fill for bar.                                                                                                                        |
| bar_width   | Bar width.                                                                                                                           |
| point_col   | Color for point.                                                                                                                     |
| line_col    | Color for line.                                                                                                                      |
| point_size  | Point size.                                                                                                                          |

**Value**

A ggplot object.

**Examples**

```
data("dkd_spe_subset")
plotScreePCA(dkd_spe_subset, dims = 10)
```

---

|                     |                                                                                      |
|---------------------|--------------------------------------------------------------------------------------|
| prepareSpatialDecon | <i>Preparing the inputs for SpatialDecon for doing deconvolution on spatial data</i> |
|---------------------|--------------------------------------------------------------------------------------|

---

**Description**

Preparing the inputs for SpatialDecon for doing deconvolution on spatial data

**Usage**

```
prepareSpatialDecon(
  spe,
  assay2use = "logcounts",
  negProbeName = "NegProbe-WTX",
  pool = NA
)
```

**Arguments**

|              |                                                                                                           |
|--------------|-----------------------------------------------------------------------------------------------------------|
| spe          | SpatialExperiment object.                                                                                 |
| assay2use    | The name of the assay to use. By default is logcounts.                                                    |
| negProbeName | The name of the negative probe gene. By default is NegProbe-WTX.                                          |
| pool         | A vector indicates the pools of the genes. This is required when there are more than one Negative Probes. |

**Value**

A list of two dataframes. The first data.frame is the normalised count, the second data.frame is the background for the data.

**Examples**

```
library(ExperimentHub)
eh <- ExperimentHub()

query(eh, "standR")
countFile <- eh[["EH7364"]]
sampleAnnoFile <- eh[["EH7365"]]
```

```
spe <- readGeoMx(countFile, sampleAnnoFile, rmNegProbe = FALSE)

out <- prepareSpatialDecon(spe)
```

---

|           |                                                                                |
|-----------|--------------------------------------------------------------------------------|
| readGeoMx | <i>Import GeoMX DSP data into a sapatial experiment object from file paths</i> |
|-----------|--------------------------------------------------------------------------------|

---

## Description

Import GeoMX DSP data into a sapatial experiment object from file paths

## Usage

```
readGeoMx(
  countFile,
  sampleAnnoFile,
  featureAnnoFile = NA,
  rmNegProbe = TRUE,
  NegProbeName = "NegProbe-WTX",
  colnames.as.rownames = c("TargetName", "SegmentDisplayName", "TargetName"),
  coord.colnames = c("ROICoordinateX", "ROICoordinateY")
)
```

## Arguments

|                      |                                                                                                                                                                         |
|----------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| countFile            | tsv file or a dataframe object. Count matrix, with samples in columns and features/genes in rows. The first column is gene names/ids.                                   |
| sampleAnnoFile       | tsv file or a dataframe object. Sample annotations.                                                                                                                     |
| featureAnnoFile      | tsv file or a dataframe object. Feature/Gene annotations.                                                                                                               |
| rmNegProbe           | Logical. Default is TRUE, indicating there are negative probe genes in the data.                                                                                        |
| NegProbeName         | Character. Name of negative probe genes, default is NegProbe-WTX.                                                                                                       |
| colnames.as.rownames | Vector of characters, length of 3. Column names used to capture gene names, sample names and gene names in countFile, sampleAnnoFile and featureAnnoFile, respectively. |
| coord.colnames       | Vector of characters, length of 2. Column names used to capture ROI coordinates.                                                                                        |

## Value

A SpatialExperiment object.

**Examples**

```
library(ExperimentHub)

eh <- ExperimentHub()
query(eh, "standR")
countFile <- eh[["EH7364"]]
sampleAnnoFile <- eh[["EH7365"]]

spe <- readGeoMx(countFile, sampleAnnoFile, rmNegProbe = FALSE)
```

---

|                  |                                                                                    |
|------------------|------------------------------------------------------------------------------------|
| readGeoMxFromDGE | <i>Import GeoMX DSP data into a spatial experiment object from DGE-List object</i> |
|------------------|------------------------------------------------------------------------------------|

---

**Description**

Import GeoMX DSP data into a spatial experiment object from DGEList object

**Usage**

```
readGeoMxFromDGE(dge_object, spatialCoord = NULL)
```

**Arguments**

|              |                                                                                                   |
|--------------|---------------------------------------------------------------------------------------------------|
| dge_object   | a DGEList object (created using edgeR::DGEList).                                                  |
| spatialCoord | a matrix with coordinates of samples, rowname must be consistent with the colnames of dge_object. |

**Value**

A SpatialExperiment object.

**Examples**

```
# making a simple DGEList object
ng <- 1000
ns <- 10
Counts <- matrix(rnbinom(ng * ns, mu = 5, size = 2), ng, ns)
rownames(Counts) <- seq(ng)
y <- edgeR::DGEList(counts = Counts, group = rep(seq(2), each = 5))

# transfer into spatial experiment object
coords <- matrix(rnorm(2 * ns), 10, 2)
spe <- readGeoMxFromDGE(dge_object = y, spatialCoord = coords)
spe
```

---

|         |                                                                              |
|---------|------------------------------------------------------------------------------|
| spe2dge | <i>Transfer SpatialExperiment object into DGEList object for DE analysis</i> |
|---------|------------------------------------------------------------------------------|

---

**Description**

Transfer SpatialExperiment object into DGEList object for DE analysis

**Usage**

```
spe2dge(spe, norm = NULL)
```

**Arguments**

|      |                                                                                                                               |
|------|-------------------------------------------------------------------------------------------------------------------------------|
| spe  | SpatialExperiment object.                                                                                                     |
| norm | Prior normalization method to be used. Norm factor calculated from the previous normalization step to be used in the DGEList. |

**Value**

A DGEList.

**Examples**

```
data("dkd_spe_subset")

dge <- spe2dge(dkd_spe_subset)

spe_tmm <- geomxNorm(dkd_spe_subset, method = "TMM")
dge <- spe2dge(spe_tmm, norm = "TMM")
```

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