

# Package ‘CiteFuse’

January 19, 2026

**Type** Package

**Title** CiteFuse: multi-modal analysis of CITE-seq data

**Version** 1.22.0

**Description** CiteFuse package implements a suite of methods and tools for CITE-seq data from pre-processing to integrative analytics, including doublet detection, network-based modality integration, cell type clustering, differential RNA and protein expression analysis, ADT evaluation, ligand-receptor interaction analysis, and interactive web-based visualisation of the analyses.

**License** GPL-3

**Encoding** UTF-8

**Depends** R (>= 4.0)

**Imports** SingleCellExperiment (>= 1.8.0), SummarizedExperiment (>= 1.16.0), Matrix, mixtools, cowplot, ggplot2, gridExtra, grid, dbSCAN, uwot, Rtsne, S4Vectors (>= 0.24.0), igraph, scales, scran (>= 1.14.6), graphics, methods, stats, utils, reshape2, ggridges, randomForest, pheatmap, ggraph, grDevices, rhdf5, rlang, Rcpp, compositions

**LinkingTo** Rcpp

**RoxygenNote** 7.2.3

**Suggests** knitr, rmarkdown, DT, mclust, scatter, ExPosition, BiocStyle, pkgdown

**VignetteBuilder** knitr

**LazyData** false

**biocViews** SingleCell, GeneExpression

**BugReports** <https://github.com/SydneyBioX/CiteFuse/issues>

**git\_url** <https://git.bioconductor.org/packages/CiteFuse>

**git\_branch** RELEASE\_3\_22

**git\_last\_commit** 7efd8aa

**git\_last\_commit\_date** 2025-10-29

**Repository** Bioconductor 3.22

**Date/Publication** 2026-01-19

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## Contents

|                                |           |
|--------------------------------|-----------|
| CiteFuse . . . . .             | 2         |
| CITEseq_example . . . . .      | 4         |
| crossSampleDoublets . . . . .  | 4         |
| DEbubblePlot . . . . .         | 5         |
| DEcomparisonPlot . . . . .     | 6         |
| DEgenes . . . . .              | 7         |
| DEgenesCross . . . . .         | 8         |
| geneADTnetwork . . . . .       | 9         |
| igraphClustering . . . . .     | 11        |
| importanceADT . . . . .        | 12        |
| ligandReceptorTest . . . . .   | 13        |
| lr_pair_subset . . . . .       | 14        |
| normaliseExprs . . . . .       | 15        |
| plotHTO . . . . .              | 15        |
| plotHTOSingle . . . . .        | 16        |
| preprocessing . . . . .        | 17        |
| readFrom10X . . . . .          | 18        |
| reducedDimSNF . . . . .        | 19        |
| sce_control_subset . . . . .   | 19        |
| sce_ctl_subset . . . . .       | 20        |
| selectDEgenes . . . . .        | 20        |
| spectralClustering . . . . .   | 21        |
| visImportance . . . . .        | 22        |
| visLigandReceptor . . . . .    | 23        |
| visualiseDim . . . . .         | 24        |
| visualiseExprs . . . . .       | 25        |
| visualiseExprsList . . . . .   | 26        |
| visualiseKNN . . . . .         | 27        |
| withinSampleDoublets . . . . . | 28        |
| <b>Index</b>                   | <b>29</b> |

CiteFuse

*CiteFuse*

## Description

A function to runSNF for CITE seq data

## Usage

```
CiteFuse(
  sce,
  altExp_name = "ADT",
  W_list = NULL,
  gene_select = TRUE,
  dist_cal_RNA = "correlation",
  dist_cal_ADT = "propr",
  ADT_subset = NULL,
  K_knn = 20,
```

```

    K_knn_Aff = 30,
    sigma = 0.45,
    t = 10,
    metadata_names = NULL,
    verbose = TRUE,
    topN = 2000
)

```

### Arguments

|                |                                                                                                                     |
|----------------|---------------------------------------------------------------------------------------------------------------------|
| sce            | a SingleCellExperiment                                                                                              |
| altExp_name    | expression name of ADT matrix                                                                                       |
| W_list         | affinity list, if it is NULL, the function will calculate it.                                                       |
| gene_select    | whether highly variable genes will be selected for RNA-seq to calculate similarity matrix using 'scran' package     |
| dist_cal_RNA   | similarity metrics used for RNA matrix                                                                              |
| dist_cal_ADT   | similarity metrics used for ADT matrix                                                                              |
| ADT_subset     | A vector indicates the subset that will be used.                                                                    |
| K_knn          | Number of nearest neighbours                                                                                        |
| K_knn_Aff      | Number of nearest neighbors for computing affinity matrix                                                           |
| sigma          | Variance for local model for computing affinity matrix                                                              |
| t              | Number of iterations for the diffusion process.                                                                     |
| metadata_names | A vector indicates the names of metadata returned                                                                   |
| verbose        | whether print out the process                                                                                       |
| topN           | top highly variable genes are used variable gene selection (see 'modelGeneVar' in 'scran' package for more details) |

### Value

A SingleCellExperiment object with fused matrix results stored

### References

B Wang, A Mezlini, F Demir, M Fiume, T Zu, M Brudno, B Haibe-Kains, A Goldenberg (2014) Similarity Network Fusion: a fast and effective method to aggregate multiple data types on a genome wide scale. Nature Methods. Online. Jan 26, 2014

### Examples

```

data("sce_ctcl_subset", package = "CiteFuse")
sce_ctcl_subset <- CiteFuse(sce_ctcl_subset)

```

---

|                 |                                              |
|-----------------|----------------------------------------------|
| CITEseq_example | <i>A subset of ECCITE-seq data (control)</i> |
|-----------------|----------------------------------------------|

---

**Description**

Data from Mimitou et al. ECCITE-seq PBMC control sample data, which is a list of three matrices of RNA, ADT and HTO

**Usage**

```
data(CITEseq_example, package = 'CiteFuse')
```

**Format**

An object of class list of length 3.

**Source**

Gene Expression Omnibus with the accession code GSE126310.

**References**

Mimitou, E. P., Cheng, A., Montalbano, A., et al. (2019). Multiplexed detection of proteins, transcriptomes, clonotypes and CRISPR perturbations in single cells. *Nature Methods*, 16(5), 409–412.

---

|                     |                            |
|---------------------|----------------------------|
| crossSampleDoublets | <i>crossSampleDoublets</i> |
|---------------------|----------------------------|

---

**Description**

A function that perform normalisation for alternative expression

**Usage**

```
crossSampleDoublets(sce, altExp_name = NULL, totalExp_threshold = 10)
```

**Arguments**

|                    |                                                                                                               |
|--------------------|---------------------------------------------------------------------------------------------------------------|
| sce                | A SingleCellExperiment object                                                                                 |
| altExp_name        | Name of alternative expression that will be used to perform normalisation. If it is NULL, it will set to HTO. |
| totalExp_threshold | the threshold indicates for the HTO less than this threshold will be filtered from the analysis               |

**Value**

A SingleCellExperiment Object

**Examples**

```
data(CITEseq_example, package = "CiteFuse")
sce_citeseq <- preprocessing(CITEseq_example)
sce_citeseq <- normaliseExprs(sce = sce_citeseq,
altExp_name = "HTO",
transform = "log")
sce_citeseq <- crossSampleDoublets(sce_citeseq)
```

DEbubblePlot

*DEbubblePlot***Description**

A function to generate circlepack plot to visualise the marker for each cluster

**Usage**

```
DEbubblePlot(de_list)
```

**Arguments**

`de_list`                      A list of results from 'DE genes ()'

**Value**

A ggplot to visualise the DE results via bubble plot

**Examples**

```
library(S4Vectors)
data(sce_control_subset, package = "CiteFuse")
sce_control_subset <- DEgenes(sce_control_subset,
altExp_name = "none",
group = sce_control_subset$SNF_W_louvain,
return_all = TRUE,
exprs_pct = 0.5)

sce_control_subset <- selectDEgenes(sce_control_subset,
altExp_name = "none")

sce_control_subset <- DEgenes(sce_control_subset,
altExp_name = "ADT",
group = sce_control_subset$SNF_W_louvain,
return_all = TRUE,
exprs_pct = 0.5)

sce_control_subset <- selectDEgenes(sce_control_subset,
altExp_name = "ADT")

rna_DEgenes <- metadata(sce_control_subset)[["DE_res_RNA_filter"]]
adt_DEgenes <- metadata(sce_control_subset)[["DE_res_ADT_filter"]]
```

```

rna_DEgenes <- lapply(rna_DEgenes, function(x){
  x$name <- gsub("hg19_", "", x$name)
  x})
DEbubblePlot(list(RNA = rna_DEgenes, ADT = adt_DEgenes))

```

---

DEcomparisonPlot

*DEcomparisonPlot*


---

## Description

A function to visualise the pairwise comparison of pvalue in different data modality.

## Usage

```
DEcomparisonPlot(de_list, feature_list)
```

## Arguments

|                           |                                                                                                   |
|---------------------------|---------------------------------------------------------------------------------------------------|
| <code>de_list</code>      | A list including two lists results from ‘DE genes ()’.                                            |
| <code>feature_list</code> | A list including two lists features indicating the selected subset of features will be visualised |

## Value

A ggplot2 to visualise the comparison plot of DE.

## Examples

```

library(S4Vectors)
data(sce_control_subset)
sce_control_subset <- DEgenes(sce_control_subset,
group = sce_control_subset$SNF_W_louvain,
return_all = TRUE,
exprs_pct = 0.5)

sce_control_subset <- selectDEgenes(sce_control_subset)

sce_control_subset <- DEgenes(sce_control_subset,
altExp_name = "ADT",
group = sce_control_subset$SNF_W_louvain,
return_all = TRUE,
exprs_pct = 0.5)

sce_control_subset <- selectDEgenes(sce_control_subset,
altExp_name = "ADT")

rna_list <- c("hg19_CD4",
"hg19_CD8A",
"hg19_HLA-DRB1",
"hg19_ITGAX",
"hg19_NCAM1",
"hg19_CD27",

```

```

"hg19_CD19")

adt_list <- c("CD4", "CD8", "MHCII (HLA-DR)", "CD11c", "CD56", "CD27", "CD19")

rna_DEgenes_all <- S4Vectors::metadata(sce_control_subset)[["DE_res_RNA"]]
adt_DEgenes_all <- S4Vectors::metadata(sce_control_subset)[["DE_res_ADT"]]

feature_list <- list(RNA = rna_list, ADT = adt_list)
de_list <- list(RNA = rna_DEgenes_all, ADT = adt_DEgenes_all)

DEcomparisonPlot(de_list = de_list,
                  feature_list = feature_list)

```

DEgenes

*DEgenes*

## Description

A function to perform DE analysis on CITE seq data

## Usage

```

DEgenes(
  sce,
  altExp_name = "none",
  exprs_value = "logcounts",
  group = NULL,
  method = "wilcox",
  exprs_pct = 0.1,
  exprs_threshold = 0,
  return_all = FALSE,
  pval_adj = 0.05,
  mean_diff = 0,
  pct_diff = 0.1,
  topN = 10
)

```

## Arguments

|                 |                                                                                                   |
|-----------------|---------------------------------------------------------------------------------------------------|
| sce             | A SingleCellExperiment object                                                                     |
| altExp_name     | A character indicates which expression matrix is used. by default is none (i.e. RNA).             |
| exprs_value     | A character indicates which expression value in assayNames is used.                               |
| group           | A vector indicates the grouping of the data                                                       |
| method          | A character indicates the method used in DE analysis                                              |
| exprs_pct       | A numeric indicates the threshold expression percentage of a gene to be considered in DE analysis |
| exprs_threshold | A numeric indicates the threshold of expression. By default is 0.                                 |

|            |                                                                           |
|------------|---------------------------------------------------------------------------|
| return_all | Whether return full list of DE genes                                      |
| pval_adj   | A numeric indicates the threshold of adjusted p-value.                    |
| mean_diff  | A numeric indicates the threshold of difference of average expression.    |
| pct_diff   | A numeric indicates the threshold of difference of percentage expression. |
| topN       | A numeric indicates the top number of genes will be included in the list. |

### Value

A SingleCellExperiment with DE results stored in meta data DE\_res

### Examples

```
data(sce_control_subset)
sce_control_subset <- DEgenes(sce_control_subset,
group = sce_control_subset$SNF_W_louvain,
return_all = TRUE,
exprs_pct = 0.5)

sce_control_subset <- selectDEgenes(sce_control_subset)
```

---

DEgenesCross

*DEgenesCross*


---

### Description

A function to perform DE analysis on a list of CITE seq data

### Usage

```
DEgenesCross(
  sce_list,
  altExp_name = "none",
  exprs_value = "logcounts",
  method = "wilcox",
  colData_name = NULL,
  group_to_test = NULL,
  exprs_pct = 0.1,
  exprs_threshold = 0,
  return_all = FALSE,
  pval_adj = 0.05,
  mean_diff = 0,
  pct_diff = 0.1,
  topN = 10
)
```



**Arguments**

|                 |                                                                                                           |
|-----------------|-----------------------------------------------------------------------------------------------------------|
| sce_list        | A Slist of ingleCellExperiment object                                                                     |
| altExp_name     | A character indicates which expression matrix is used. by default is none (i.e. RNA).                     |
| exprs_value     | A character indicates which expression value in assayNames is used.                                       |
| method          | A character indicates the method used in DE analysis                                                      |
| colData_name    | A vector of character indicates the colData that stored the group information of each sce of the sce_list |
| group_to_test   | A vector of character indicates which group in each sce is used to compared across the sce list.          |
| exprs_pct       | A numeric indicates the threshold expression percentage of a gene to be considered in DE analysis         |
| exprs_threshold | A numeric indicates the threshold of expression. By default is 0.                                         |
| return_all      | Whether return full list of DE genes                                                                      |
| pval_adj        | A numeric indicates the threshold of adjusted p-value.                                                    |
| mean_diff       | A numeric indicates the threshold of difference of average expression.                                    |
| pct_diff        | A numeric indicates the threshold of difference of percentage expression.                                 |
| topN            | A numeric indicates the top number of genes will be included in the list.                                 |

**Value**

A SingleCellExeperiment with DE results stored in meta data DE\_res

**Examples**

```
data("sce_control_subset", package = "CiteFuse")
data("sce_ctcl_subset", package = "CiteFuse")

de_res <- DEgenesCross(sce_list = list(control = sce_control_subset,
ctcl = sce_ctcl_subset),
colData_name = c("SNF_W_louvain", "SNF_W_louvain"),
group_to_test = c("2", "6"))
```

---

geneADTnetwork

*geneADTnetwork*


---

**Description**

A function to visualise the features distribuion

**Usage**

```
geneADTnetwork(
  sce,
  RNA_exprs_value = "logcounts",
  altExp_name = "ADT",
  altExp_exprs_value = "logcounts",
  RNA_feature_subset = NULL,
  ADT_feature_subset = NULL,
  cell_subset = NULL,
  cor_threshold = 0.5,
  cor_method = c("pearson", "kendall", "spearman"),
  RNA_exprs_pct = 0.1,
  ADT_exprs_pct = 0.1,
  RNA_exprs_threshold = 0,
  ADT_exprs_threshold = 0,
  network_layout = NULL,
  return_igraph = FALSE
)
```

**Arguments**

|                     |                                                                                                                                                                          |
|---------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| sce                 | A singlecellexperiment object                                                                                                                                            |
| RNA_exprs_value     | A character indicates which expression value for RNA in assayNames is used.                                                                                              |
| altExp_name         | A character indicates which expression matrix is used. by default is none (i.e. RNA).                                                                                    |
| altExp_exprs_value  | A character indicates which expression value in assayNames is used.                                                                                                      |
| RNA_feature_subset  | A vector of characters indicates the subset of features of RNA that are used for visualisation                                                                           |
| ADT_feature_subset  | A vector of characters indicates the subset of features of ADT that are used for visualisation                                                                           |
| cell_subset         | A vector of characters indicates the subset of cells that are used for visualisation                                                                                     |
| cor_threshold       | Thresholds of correlation.                                                                                                                                               |
| cor_method          | a character string indicating which correlation coefficient (or covariance) is to be computed. One of "pearson" (default), "kendall", or "spearman": can be abbreviated. |
| RNA_exprs_pct       | A numeric indicates the threshold expression percentage of a gene to be considered in correlation analysis                                                               |
| ADT_exprs_pct       | A numeric indicates the threshold expression percentage of a gene to be considered in correlation analysis                                                               |
| RNA_exprs_threshold | A numeric indicates the threshold of RNA expression. By default is 0.                                                                                                    |
| ADT_exprs_threshold | A numeric indicates the threshold of ADT expression. By default is 0.                                                                                                    |
| network_layout      | layout of the network                                                                                                                                                    |
| return_igraph       | indicates whether return the igraph object                                                                                                                               |

**Value**

A igraph object of gene-ADT network

**Examples**

```
library(SingleCellExperiment)
set.seed(2020)
data(sce_control_subset, package = "CiteFuse")
RNA_feature_subset <- sample(rownames(sce_control_subset), 50)
ADT_feature_subset <- rownames(altExp(sce_control_subset, "ADT"))

geneADTnetwork(sce_control_subset,
               RNA_feature_subset = RNA_feature_subset,
               ADT_feature_subset = ADT_feature_subset,
               cor_method = "pearson",
               network_layout = igraph::layout_with_fr)
```

---

igraphClustering

*igraphClustering*


---

**Description**

A function to perform igraph clustering

**Usage**

```
igraphClustering(
  sce,
  metadata = "SNF_W",
  method = c("louvain", "leiden", "walktrap", "spinglass", "optimal", "leading_eigen",
            "label_prop", "fast_greedy", "edge_betweenness"),
  ...
)
```

**Arguments**

|          |                                                                                                      |
|----------|------------------------------------------------------------------------------------------------------|
| sce      | A singlecellexperiment object                                                                        |
| metadata | indicates the meta data name of affinity matrix to virsualise                                        |
| method   | A character indicates the method for finding communities from igraph. Default is louvain clustering. |
| ...      | Other inputs for the igraph functions                                                                |

**Value**

A vector indicates the membership (clustering) results

**Examples**

```
data(sce_control_subset, package = "CiteFuse")
sce_control_subset <- CiteFuse(sce_control_subset)
SNF_W_louvain <- igraphClustering(sce_control_subset,
method = "louvain")
```

importanceADT

*importanceADT***Description**

A function to calculate the importance score of ADT

**Usage**

```
importanceADT(
  sce,
  altExp_name = "ADT",
  exprs_value = "logcounts",
  method = c("randomForest", "PCA"),
  group = NULL,
  subsample = TRUE,
  times = 10,
  prop = 0.8,
  k_pca = 5,
  remove_first_PC = TRUE,
  ...
)
```

**Arguments**

|                 |                                                                                                    |
|-----------------|----------------------------------------------------------------------------------------------------|
| sce             | A singlecellexperiment object                                                                      |
| altExp_name     | A character indicates which expression matrix is used. by default is none (i.e. RNA).              |
| exprs_value     | A character indicates which expression value in assayNames is used.                                |
| method          | A character indicates the method of ADT importance calculation, either randomForest or PCA         |
| group           | A vector indicates the grouping of the data (for random forest)                                    |
| subsample       | Whether perform subsampling (for random forest)                                                    |
| times           | A numeric indicates the times of subsampling is performed (for random forest)                      |
| prop            | A numeric indicates the proportion of cells are subsampled from the whole data (for random forest) |
| k_pca           | Number of principal component will be used to calculate the loading scores (for PCA)               |
| remove_first_PC | A logical input indicates whether the first component will be removed from calculation (for PCA).  |
| ...             | other arguments to 'randomForest()' or 'prcomp()' function                                         |

**Details**

For random forest, the importance scores are based on features importance. For PCA, it implements the method proposed in Levin et al (based on the loading of features).

**Value**

A SingleCellExperiment object

**References**

Levine, J.H., Simonds, E.F., Bendall, S.C., Davis, K.L., El-ad, D.A., Tadmor, M.D., Litvin, O., Fienberg, H.G., Jager, A., Zunder, E.R. and Finck, R., 2015. Data-driven phenotypic dissection of AML reveals progenitor-like cells that correlate with prognosis. *Cell*, 162(1), pp.184-197.

**Examples**

```
data("sce_control_subset", package = "CiteFuse")
sce_control_subset <- importanceADT(sce_control_subset,
group = sce_control_subset$SNF_W_louvain,
subsample = TRUE)
```

---

|                    |                           |
|--------------------|---------------------------|
| ligandReceptorTest | <i>ligandReceptorTest</i> |
|--------------------|---------------------------|

---

**Description**

A function to perform ligand receptor analysis

**Usage**

```
ligandReceptorTest(
  sce,
  ligandReceptor_list,
  cluster,
  RNA_exprs_value = "minMax",
  use_alt_exp = TRUE,
  altExp_name = "ADT",
  altExp_exprs_value = "zi_minMax",
  num_permute = 1000,
  p_sig = 0.05
)
```

**Arguments**

|                     |                                                                             |
|---------------------|-----------------------------------------------------------------------------|
| sce                 | A singlecellexperiment object                                               |
| ligandReceptor_list | A data.frame indicates the ligand receptor list                             |
| cluster             | A vector indicates the cluster results                                      |
| RNA_exprs_value     | A character indicates which expression value for RNA in assayNames is used. |

|                    |                                                                                                             |
|--------------------|-------------------------------------------------------------------------------------------------------------|
| use_alt_exp        | A logical vector indicates whether receptors expression will use alternative expression matrix to quantify. |
| altExp_name        | A character indicates which expression matrix is used. by default is ADT .                                  |
| altExp_exprs_value | A character indicates which expression value in assayNames is used.                                         |
| num_permute        | Number of permutation.                                                                                      |
| p_sig              | A numeric indicates threshold of the pvalue significance                                                    |

**Value**

A SingleCellExperiment object with ligand receptor results

**Examples**

```
data(lr_pair_subset, package = "CiteFuse")
data(sce_control_subset, package = "CiteFuse")

sce_control_subset <- normaliseExprs(sce = sce_control_subset,
  altExp_name = "ADT",
  transform = "zi_minMax")

sce_control_subset <- normaliseExprs(sce = sce_control_subset,
  altExp_name = "none",
  exprs_value = "logcounts",
  transform = "minMax")

sce_control_subset <- ligandReceptorTest(sce = sce_control_subset,
  ligandReceptor_list = lr_pair_subset,
  cluster = sce_control_subset$SNF_W_louvain,
  RNA_exprs_value = "minMax",
  use_alt_exp = TRUE,
  altExp_name = "ADT",
  altExp_exprs_value = "zi_minMax",
  num_permute = 100)
```

---

|                |                                          |
|----------------|------------------------------------------|
| lr_pair_subset | <i>A subset of Ligand Receptor Pairs</i> |
|----------------|------------------------------------------|

---

**Description**

A subset of Ligand Receptor Pairs

**Usage**

```
data(lr_pair_subset, package = 'CiteFuse')
```

**Format**

An object of class matrix (inherits from array) with 50 rows and 2 columns.

---

|                |                       |
|----------------|-----------------------|
| normaliseExprs | <i>normaliseExprs</i> |
|----------------|-----------------------|

---

**Description**

A function that perform normalisation for alternative expression

**Usage**

```
normaliseExprs(
  sce,
  altExp_name = NULL,
  exprs_value = "counts",
  transform = c("log", "clr", "zi_minMax", "minMax"),
  log_offset = NULL
)
```

**Arguments**

|             |                                                                                                         |
|-------------|---------------------------------------------------------------------------------------------------------|
| sce         | A SingleCellExperiment object                                                                           |
| altExp_name | Name of alternative expression that will be used to perform normalisation                               |
| exprs_value | A character indicates which expression value in assayNames is used.                                     |
| transform   | type of transformation, either log or clr (Centered log ratio transform)                                |
| log_offset  | Numeric scalar specifying the pseudo-count to add when log-transforming expression values. Default is 1 |

**Value**

a SingleCellExperiment object

**Examples**

```
data(CITEseq_example, package = "CiteFuse")
sce_citeseq <- preprocessing(CITEseq_example)
sce_citeseq <- normaliseExprs(sce = sce_citeseq,
  altExp_name = "ADT",
  transform = "log")
```

---

|         |                |
|---------|----------------|
| plotHTO | <i>plotHTO</i> |
|---------|----------------|

---

**Description**

A function to plot HTO expression

**Usage**

```
plotHTO(sce, which_idx = seq_len(2), altExp_name = NULL, ncol = 2)
```

**Arguments**

|             |             |
|-------------|-------------|
| sce         | sce         |
| which_idx   | which_idx   |
| altExp_name | altExp_name |
| ncol        | ncol        |

**Value**

A plot visualising the HTO expression

**Examples**

```
data(CITEseq_example, package = "CiteFuse")
sce_citeseq <- preprocessing(CITEseq_example)
sce_citeseq <- normaliseExprs(sce = sce_citeseq,
  altExp_name = "HTO",
  transform = "log")
plotHTO(sce_citeseq, 1:4)
```

---

|               |                      |
|---------------|----------------------|
| plotHTOSingle | <i>plotHTOSingle</i> |
|---------------|----------------------|

---

**Description**

A function to plot HTO expression

**Usage**

```
plotHTOSingle(sce, which_idx = seq_len(2), altExp_name = NULL)
```

**Arguments**

|             |             |
|-------------|-------------|
| sce         | sce         |
| which_idx   | which_idx   |
| altExp_name | altExp_name |

**Value**

A plot visualising the HTO expression



---

|               |                                                               |
|---------------|---------------------------------------------------------------|
| preprocessing | <i>A function to preprocess the list of expression matrix</i> |
|---------------|---------------------------------------------------------------|

---

## Description

This function will keep the samples that are common across the list of expression matrix, and filter the features that are all zeros across samples, and finally construct a SingleCellExperiment object

## Usage

```
preprocessing(  
  exprsMat = NULL,  
  return_sce = TRUE,  
  assay_matrix = 1,  
  filter_features = TRUE,  
  rowData = NULL,  
  colData = NULL  
)
```

## Arguments

|                 |                                                                                                                             |
|-----------------|-----------------------------------------------------------------------------------------------------------------------------|
| exprsMat        | A list or a matrix indicates the expression matrices of the testing datasets (each matrix must be matrix or dgCMatrx class) |
| return_sce      | A logical input indicates whether a SingleCellExperiment object will be return                                              |
| assay_matrix    | A integer indicates which list will be used as 'assay' input of 'SingleCellExperiment'                                      |
| filter_features | A logical input indicates whether the features with all zeros will be removed                                               |
| rowData         | A DataFrame indicates the rowData to be stored in the sce object                                                            |
| colData         | A DataFrame indicates the colData to be stored in the sce object                                                            |

## Value

either a SingleCellExperiment object or a preprocessed expression matrix

## Examples

```
data(CITEseq_example, package = "CiteFuse")  
sce_citeseq <- preprocessing(CITEseq_example)
```

---

|             |                    |
|-------------|--------------------|
| readFrom10X | <i>readFrom10X</i> |
|-------------|--------------------|

---

## Description

A function to read the data from 10X

## Usage

```
readFrom10X(
  dir,
  type = c("auto", "sparse", "HDF5"),
  feature_named_by = c("gene_id", "gene_symbol"),
  filter_features = TRUE
)
```

## Arguments

|                               |                                                                                                           |
|-------------------------------|-----------------------------------------------------------------------------------------------------------|
| <code>dir</code>              | A character indicates the directory of the 10X files                                                      |
| <code>type</code>             | A character indicates the format of the data, sparse or HDF5                                              |
| <code>feature_named_by</code> | A character indicates whehter the genes will be named by <code>gene_id</code> or <code>gene_symbol</code> |
| <code>filter_features</code>  | A logical input indicates whether the features with all zeros will be removed                             |

## Value

a SingleCellExperiment object

## Examples

```
## Not run:
tmpdir <- tempdir()
tenXdata <- "http://cf.10xgenomics.com/samples/cell-exp/3.1.0/connect_5k_pbmc_NGSC3_ch1/"
file <- "connect_5k_pbmc_NGSC3_ch1_filtered_feature_bc_matrix.tar.gz"
download.file(paste0(tenXdata, file), file.path(tmpdir, file))
untar(file.path(tmpdir, file),
      exdir = tmpdir)
sce_citeseq_10X <- readFrom10X(file.path(tmpdir,
"filtered_feature_bc_matrix/"))
sce_citeseq_10X

## End(Not run)
```

---

|               |                      |
|---------------|----------------------|
| reducedDimSNF | <i>reducedDimSNF</i> |
|---------------|----------------------|

---

**Description**

A function to reduce the dimension of the similarity matrix

**Usage**

```
reducedDimSNF(sce, metadata = "SNF_W", method = "UMAP", dimNames = NULL, ...)
```

**Arguments**

|          |                                                                        |
|----------|------------------------------------------------------------------------|
| sce      | A singlecellexperiment object                                          |
| metadata | indicates the meta data name of affinity matrix to virsualise          |
| method   | the method of visualisation, which can be UMAP, tSNE and diffusion map |
| dimNames | indicates the name of the reduced dimension results.                   |
| ...      | other parameters for tsne(), umap()                                    |

**Value**

A SingleCellExperiment object

**Examples**

```
data(sce_control_subset, package = "CiteFuse")
sce_control_subset <- CiteFuse(sce_control_subset)
sce_control_subset <- reducedDimSNF(sce_control_subset,
method = "tSNE",
dimNames = "tSNE_joint")
```

---

|                    |                                                  |
|--------------------|--------------------------------------------------|
| sce_control_subset | <i>A SingleCellExperiment of ECCITE-seq data</i> |
|--------------------|--------------------------------------------------|

---

**Description**

Data from Mimitou et al. ECCITE-seq PBMC Control sample data

**Usage**

```
data(sce_control_subset, package = 'CiteFuse')
```

**Format**

An object of class SingleCellExperiment with 1508 rows and 128 columns.

**Source**

Gene Expression Omnibus with the accession code GSE126310.

## References

Mimitou, E. P., Cheng, A., Montalbano, A., et al. (2019). Multiplexed detection of proteins, transcriptomes, clonotypes and CRISPR perturbations in single cells. *Nature Methods*, 16(5), 409–412.

---

|                 |                                                  |
|-----------------|--------------------------------------------------|
| sce_ctcl_subset | <i>A SingleCellExperiment of ECCITE-seq data</i> |
|-----------------|--------------------------------------------------|

---

## Description

Data from Mimitou et al. ECCITE-seq PBMC CTCL sample data

## Usage

```
data(sce_ctcl_subset, package = 'CiteFuse')
```

## Format

An object of class `SingleCellExperiment` with 1450 rows and 173 columns.

## Source

Gene Expression Omnibus with the accession code GSE126310.

## References

Mimitou, E. P., Cheng, A., Montalbano, A., et al. (2019). Multiplexed detection of proteins, transcriptomes, clonotypes and CRISPR perturbations in single cells. *Nature Methods*, 16(5), 409–412.

---

|               |                      |
|---------------|----------------------|
| selectDEgenes | <i>selectDEgenes</i> |
|---------------|----------------------|

---

## Description

A function to select DE genes

## Usage

```
selectDEgenes(
  sce = NULL,
  de_res = NULL,
  altExp_name = "none",
  pval_adj = 0.05,
  mean_diff = 0,
  pct_diff = 0.1,
  topN = 10
)
```

**Arguments**

|             |                                                                                       |
|-------------|---------------------------------------------------------------------------------------|
| sce         | A SingleCellExperiment object with DE results stored in meta data DE_res list.        |
| de_res      | DE_res returned by DEgenesCross().                                                    |
| altExp_name | A character indicates which expression matrix is used. by default is none (i.e. RNA). |
| pval_adj    | A numeric indicates the threshold of adjusted p-value.                                |
| mean_diff   | A numeric indicates the threshold of difference of average expression.                |
| pct_diff    | A numeric indicates the threshold of difference of percentage expression.             |
| topN        | A numeric indicates the top number of genes will be included in the list.             |

**Value**

A SingleCellExperiment With filtered DE results in DE\_res\_filter list of metadata

**Examples**

```
data(sce_control_subset)
sce_control_subset <- DEgenes(sce_control_subset,
group = sce_control_subset$SNF_W_louvain,
return_all = TRUE,
exprs_pct = 0.5)

sce_control_subset <- selectDEgenes(sce_control_subset)
```

---

spectralClustering      *spectralClustering*

---

**Description**

A function to perform spectral clustering

**Usage**

```
spectralClustering(affinity, K = 20, delta = 1e-05)
```

**Arguments**

|          |                    |
|----------|--------------------|
| affinity | An affinity matrix |
| K        | number of clusters |
| delta    | delta              |

**Value**

A list indicates the spectral clustering results

**Examples**

```
data(sce_control_subset, package = "CiteFuse")
sce_control_subset <- CiteFuse(sce_control_subset)
SNF_W <- S4Vectors::metadata(sce_control_subset)[["SNF_W"]]
SNF_W_clust <- spectralClustering(SNF_W, K = 5)
```

---

visImportance

*visImportance*


---

**Description**

A function to visualise the features distribtuion

**Usage**

```
visImportance(
  sce,
  plot = c("boxplot", "heatmap"),
  altExp_name = "ADT",
  exprs_value = "logcounts"
)
```

**Arguments**

|             |                                                                                       |
|-------------|---------------------------------------------------------------------------------------|
| sce         | A singlecellexperiment object                                                         |
| plot        | A string indicates the type of the plot (either boxplot or heatmap)                   |
| altExp_name | A character indicates which expression matrix is used. by default is none (i.e. RNA). |
| exprs_value | A character indicates which expression value in assayNames is used.                   |

**Value**

A plot (either ggplot or pheatmap) to visualise the ADT importance results

**Examples**

```
data("sce_control_subset", package = "CiteFuse")
sce_control_subset <- importanceADT(sce_control_subset,
  group = sce_control_subset$SNF_W_louvain,
  subsample = TRUE)
visImportance(sce_control_subset, plot = "boxplot")
```

---

|                   |                          |
|-------------------|--------------------------|
| visLigandReceptor | <i>visLigandReceptor</i> |
|-------------------|--------------------------|

---

## Description

A function to visualise ligand receptor analysis

## Usage

```
visLigandReceptor(
  sce,
  type = c("pval_heatmap", "pval_dotplot", "group_network", "group_heatmap",
    "lr_network"),
  receptor_type = NULL
)
```

## Arguments

|               |                                                                                                                                                                                          |
|---------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| sce           | A singlecellexperiment object                                                                                                                                                            |
| type          | A character indicates the type of the plot for ligand receptor results visualisation, option includes "pval_heatmap", "pval_dotplot", "group_network", "group_heatmap", and "lr_network" |
| receptor_type | A character indicates which receptor expression's ligand receptor results are used to generate the figures.                                                                              |

## Value

A plot visualise the ligand receptor results

## Examples

```
data(lr_pair_subset, package = "CiteFuse")
data(sce_control_subset, package = "CiteFuse")

sce_control_subset <- normaliseExprs(sce = sce_control_subset,
  altExp_name = "ADT",
  transform = "zi_minMax")

sce_control_subset <- normaliseExprs(sce = sce_control_subset,
  altExp_name = "none",
  exprs_value = "logcounts",
  transform = "minMax")

sce_control_subset <- ligandReceptorTest(sce = sce_control_subset,
  ligandReceptor_list = lr_pair_subset,
  cluster = sce_control_subset$SNF_W_louvain,
  RNA_exprs_value = "minMax",
  use_alt_exp = TRUE,
  altExp_name = "ADT",
  altExp_exprs_value = "zi_minMax",
  num_permute = 100)

visLigandReceptor(sce_control_subset,
```

```
type = "pval_heatmap",
receptor_type = "ADT")
```

---

visualiseDim

*visualiseDim*


---

## Description

A function to visualise the reduced dimension

## Usage

```
visualiseDim(
  sce,
  dimNames = NULL,
  colour_by = NULL,
  shape_by = NULL,
  data_from = c("colData", "assay", "altExp"),
  assay_name = NULL,
  altExp_name = NULL,
  altExp_assay_name = NULL,
  dim = seq_len(2)
)
```

## Arguments

|                   |                                                                                                              |
|-------------------|--------------------------------------------------------------------------------------------------------------|
| sce               | A singlecellexperiment object                                                                                |
| dimNames          | indicates the name of the reduced dimension results.                                                         |
| colour_by         | A character indicates how the cells coloured by. The information either stored in colData, assay, or altExp. |
| shape_by          | A character indicates how the cells shaped by. The information either stored in colData, assay, or altExp.   |
| data_from         | A character indicates where the colour by data stored                                                        |
| assay_name        | A character indicates the assay name of the expression                                                       |
| altExp_name       | A character indicates the name of alternative expression                                                     |
| altExp_assay_name | A character indicates the assay name of alternative expression                                               |
| dim               | a vector of numeric with length of 2 indicates which component is being plot                                 |

## Value

A ggplot of the reduced dimension visualisation



**Examples**

```
data(sce_control_subset, package = "CiteFuse")
sce_control_subset <- CiteFuse(sce_control_subset)
sce_control_subset <- reducedDimSNF(sce_control_subset,
method = "tSNE",
dimNames = "tSNE_joint")
visualiseDim(sce_control_subset, dimNames = "tSNE_joint",
colour_by = "SNF_W_clust")
```

---

visualiseExprs

*visualiseExprs*


---

**Description**

A function to visualise the features distribution

**Usage**

```
visualiseExprs(
  sce,
  plot = c("boxplot", "violin", "jitter", "density", "pairwise"),
  altExp_name = c("none"),
  exprs_value = "logcounts",
  group_by = NULL,
  facet_by = NULL,
  feature_subset = NULL,
  cell_subset = NULL,
  n = NULL,
  threshold = NULL
)
```

**Arguments**

|                |                                                                                                                |
|----------------|----------------------------------------------------------------------------------------------------------------|
| sce            | A singlecellexperiment object                                                                                  |
| plot           | Type of plot, includes boxplot, violin, jitter, density, and pairwise. By default is boxplot                   |
| altExp_name    | A character indicates which expression matrix is used. by default is none (i.e. RNA).                          |
| exprs_value    | A character indicates which expression value in assayNames is used.                                            |
| group_by       | A character indicates how is the expression will be group in the plots (stored in colData).                    |
| facet_by       | A character indicates how is the expression will be lay out panels in a grid in the plots (stored in colData). |
| feature_subset | A vector of characters indicates the subset of features that are used for visualisation                        |
| cell_subset    | A vector of characters indicates the subset of cells that are used for visualisation                           |
| n              | A numeric indicates the top expressed features to show.                                                        |
| threshold      | Thresholds of high expresion for features (only is used for pairwise plot).                                    |

**Value**

A ggplot to visualise the features distribution

**Examples**

```
data(sce_control_subset)
visualiseExprs(sce_control_subset,
  plot = "boxplot",
  group_by = "SNF_W_louvain",
  feature_subset = c("hg19_CD8A"))

visualiseExprs(sce_control_subset,
  plot = "density",
  altExp_name = "ADT",
  group_by = "SNF_W_louvain",
  feature_subset = c("CD8", "CD4"))
```

---

|                    |                           |
|--------------------|---------------------------|
| visualiseExprsList | <i>visualiseExprsList</i> |
|--------------------|---------------------------|

---

**Description**

A function to visualise the features distribution for a list of SingleCellExperiment

**Usage**

```
visualiseExprsList(
  sce_list,
  plot = c("boxplot", "violin", "jitter", "density"),
  altExp_name = "none",
  exprs_value = "logcounts",
  group_by = NULL,
  feature_subset = NULL,
  cell_subset = NULL,
  n = NULL
)
```

**Arguments**

|                |                                                                                              |
|----------------|----------------------------------------------------------------------------------------------|
| sce_list       | A list of SingleCellExperiment object                                                        |
| plot           | Type of plot, includes boxplot, violin, jitter, density, and pairwise. By default is boxplot |
| altExp_name    | A character indicates which expression matrix is used. by default is none (i.e. RNA).        |
| exprs_value    | A character indicates which expression value in assayNames is used.                          |
| group_by       | A character indicates how the expression will be grouped in the plots (stored in colData).   |
| feature_subset | A vector of characters indicates the subset of features that are used for visualisation      |
| cell_subset    | A vector of characters indicates the subset of cells that are used for visualisation         |
| n              | A numeric indicates the top expressed features to show.                                      |

**Value**

A ggplot to visualise the features distribution

**Examples**

```
data(sce_control_subset, package = "CiteFuse")
data(sce_ctl_subset, package = "CiteFuse")
visualiseExprsList(sce_list = list(control = sce_control_subset,
  ctl = sce_ctl_subset),
  plot = "boxplot",
  altExp_name = "none",
  exprs_value = "logcounts",
  feature_subset = c("hg19_CD8A"),
  group_by = c("SNF_W_louvain", "SNF_W_louvain"))
```

---

|              |                     |
|--------------|---------------------|
| visualiseKNN | <i>visualiseKNN</i> |
|--------------|---------------------|

---

**Description**

A function to perform louvain clustering

**Usage**

```
visualiseKNN(sce, colour_by = NULL, metadata = "SNF_W")
```

**Arguments**

|           |                                                              |
|-----------|--------------------------------------------------------------|
| sce       | A singlecellexperiment object                                |
| colour_by | the name of coldata that is used to colour the node          |
| metadata  | indicates the meta data name of affinity matrix to visualise |

**Value**

A igraph plot

**Examples**

```
data(sce_control_subset, package = "CiteFuse")
sce_control_subset <- CiteFuse(sce_control_subset)
SNF_W_louvain <- igraphClustering(sce_control_subset,
  method = "louvain")
visualiseKNN(sce_control_subset, colour_by = "SNF_W_louvain")
```

---

|                      |                             |
|----------------------|-----------------------------|
| withinSampleDoublets | <i>withinSampleDoublets</i> |
|----------------------|-----------------------------|

---

**Description**

doublet identification within batch

**Usage**

```
withinSampleDoublets(sce, altExp_name = NULL, eps = 200, minPts = 50)
```

**Arguments**

|             |                               |
|-------------|-------------------------------|
| sce         | a SingleCellExperiment        |
| altExp_name | expression name of HTO matrix |
| eps         | eps of DBSCAN                 |
| minPts      | minPts of DBSCAN              |

**Value**

A SingleCellExperiment object

**Examples**

```
data(CITEseq_example, package = "CiteFuse")
sce_citeseq <- preprocessing(CITEseq_example)
sce_citeseq <- normaliseExprs(sce = sce_citeseq,
altExp_name = "HTO",
transform = "log")
sce_citeseq <- crossSampleDoublets(sce_citeseq)
sce_citeseq <- withinSampleDoublets(sce_citeseq,
minPts = 10)
```

# Index

## \* datasets

- CITEseq\_example, [4](#)
- lr\_pair\_subset, [14](#)
- sce\_control\_subset, [19](#)
- sce\_ctcl\_subset, [20](#)

CiteFuse, [2](#)  
CITEseq\_example, [4](#)  
crossSampleDoublets, [4](#)

DEbubblePlot, [5](#)  
DEcomparisonPlot, [6](#)  
DEgenes, [7](#)  
DEgenesCross, [8](#)

geneADTnetwork, [9](#)

igraphClustering, [11](#)  
importanceADT, [12](#)

ligandReceptorTest, [13](#)  
lr\_pair\_subset, [14](#)

normaliseExprs, [15](#)

plotHTO, [15](#)  
plotHTOSingle, [16](#)  
preprocessing, [17](#)

readFrom10X, [18](#)  
reducedDimSNF, [19](#)

sce\_control\_subset, [19](#)  
sce\_ctcl\_subset, [20](#)  
selectDEgenes, [20](#)  
spectralClustering, [21](#)

visImportance, [22](#)  
visLigandReceptor, [23](#)  
visualiseDim, [24](#)  
visualiseExprs, [25](#)  
visualiseExprsList, [26](#)  
visualiseKNN, [27](#)

withinSampleDoublets, [28](#)