# Package 'CSOA'

October 24, 2025

**Description** Cell Set Overlap Analysis (CSOA) is a tool for calculating per-cell gene signature scores in an scRNA-seq dataset. CSOA constructs a set for each gene in the signature, consisting of the cells that highly express the gene. Next, all overlaps of pairs of cell sets are computed, ranked, filtered and scored. The CSOA percell score is calculated by summing up all products of the overlap scores and the min-maxnormalized expression of the two involved genes. CSOA can run on a Seurat object, a SingleCellExperiment object, a matrix and a dgCMatrix. License MIT + file LICENSE **Imports** bayesbio, dplyr, ggplot2, henna, kerntools, methods, qs, reshape2, rlang, Seurat, SeuratObject, SummarizedExperiment, sgof, spatstat.utils, stats, textshape, wesanderson **Encoding UTF-8** RoxygenNote 7.3.3 Suggests BiocStyle, knitr, patchwork, rmarkdown, scRNAseq, scuttle, stringr, testthat (>= 3.0.0) biocViews Software, SingleCell, GeneSetEnrichment, GeneExpression VignetteBuilder knitr URL https://github.com/andrei-stoica26/CSOA BugReports https://github.com/andrei-stoica26/CSOA/issues Config/testthat/edition 3 git\_url https://git.bioconductor.org/packages/CSOA git\_branch devel

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```
attachCellScores.default
```

Attach CSOA scores to object

#### **Description**

This function attaches the data frame of CSOA scores to the input object.

#### Usage

```
## Default S3 method:
attachCellScores(scObj, scoreDF, ...)

## S3 method for class 'Seurat'
attachCellScores(scObj, scoreDF, ...)

## S3 method for class 'SingleCellExperiment'
attachCellScores(scObj, scoreDF, ...)

## S3 method for class 'matrix'
attachCellScores(scObj, scoreDF, ...)

## S3 method for class 'dgCMatrix'
attachCellScores(scObj, scoreDF, ...)

attachCellScores(scObj, ...)
```

## Arguments

scObj A Seurat object, SingleCellExperiment object, or expression matrix.

scoreDF Data frame of CSOA scores.

Additional arguments.

#### **Details**

If the input object is of the Seurat or SingleCellExpression class, it will be returned with added CSOA scores. Otherwise, a list containing the expression matrix and the CSOA scores data frame will be returned.

#### Value

A Seurat object with CSOA scores added to metadata.

A SingleCellExperiment object with CSOA scores added to colData.

A list containing the expression matrix and the CSOA scores data frame.

A list containing the expression matrix and the CSOA scores data frame.

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#### **Examples**

```
library(Seurat)
mat <- matrix(0, 500, 300)
rownames(mat) <- paste0('G', seq(500))
colnames(mat) <- paste0('C', seq(300))
mat[sample(8000)] <- sample(20, 8000, TRUE)
seuratObj <- CreateSeuratObject(mat)
seuratObj <- NormalizeData(seuratObj)
scores <- data.frame(CSOA = runif(300))
seuratObj <- attachCellScores(seuratObj, scores)
head(seuratObj$CSOA)</pre>
```

basicHeatmap

Plot a simple heatmap

#### **Description**

This function plots a simple heatmap, with clustering but no dendograms.

## Usage

```
basicHeatmap(
  mat,
  aesNames = c("x", "y", "Score"),
  title = "Heatmap",
  axisTextSize = 7,
  palType = "fillCont",
  wesPal = "Royal1",
  wesLow = 3,
  wesHigh = 2,
  ...
)
```

#### **Arguments**

A matrix. mat aesNames A character vector of length 3 representing the y, x and fill aes elements. title Plot title. axisTextSize Axis text size. Palette type: color or fill, continuous or discrete. Accepted values are 'colorpalType Cont', 'fillCont', 'colDis' and 'fillDis'. The function shows a warning and does not change the color scheme if a different value is passed here. wesPal A wesanderson palette. wesLow Index of color marking low values. wesHigh Index of color marking high values. Other arguments passed to henna::centerTitle.

bfCorrectDF 5

## Value

A ggplot object.

## **Examples**

```
mat <- matrix(0, 10, 20)
mat[sample(length(mat), 50)] <- runif(50, max = 2.5)
basicHeatmap(mat)</pre>
```

bfCorrectDF

Perform multiple testing correction and filtering with Bonferroni

## **Description**

This function performs the Bonferroni correction for multiple testing in a dataframe column of p-values and filters the data-frame based on p-values.

## Usage

```
bfCorrectDF(
   df,
   nTests = nrow(df),
   pvalThr = 0.05,
   colStr = "pval",
   newColStr = "pvalAdj"
)
```

## **Arguments**

df A dataframe with a column of p-values.

nTests Number of tests. Default to the number of rows in the data frame.

pvalThr p-value threshold.

colStr Name of the column of p-values.

newColStr Name of the column of adjusted p-values that will be created.

#### Value

The data frame with Benjamini-Yekutieli-corrected p-values.

6 breakWeakTies

bhCorrectDF	Perform multiple testing correction Hochberg	and filtering with Benjamini-

## **Description**

This function performs the Benjamini-Hochberg correction for multiple testing in a dataframe column of p-values and filters the data-frame based on p-values.

#### Usage

```
bhCorrectDF(df, ...)
```

#### **Arguments**

df A dataframe with a column of p-values.

... Additional arguments passed to fdrCorrectDF.

#### Value

The data frame with Benjamini-Hochberg-corrected p-values.

breakWeakTies	Remove overlap pairs with low Jaccard scores	

## **Description**

This function iteratively removes all overlap pairs with neighbor Jaccard score below a fixed cutoff until no overlap pairs can be removed. Subsequently, overlap ranks are recalculated.

## Usage

```
breakWeakTies(overlapDF, cutoff = 1/3, doConnComp = FALSE)
```

## Arguments

overlapDF An overlap data frame.

cutoff A cutoff used in the filtering of edges with low Jaccard scores.

doConnComp Whether to calculate the connected components.

#### **Details**

The functions removes overlaps for which the two involved genes record too few shared neighbors—genes whose cell set significantly overlaps with those of both overlap genes.

byCorrectDF 7

#### Value

An overlap data frame in which edges with low Jaccard scores have been removed.

#### **Examples**

```
overlapDF <- data.frame(gene1=paste0('G', c(1, 3, 7, 6, 8, 2, 4, 3, 4, 5)), gene2=paste0('G', c(2, 7, 2, 5, 4, 5, 1, 2, 2, 8)), ratio=runif(10, 2, 10), pval=runif(10, 0, 1e-10)) breakWeakTies(overlapDF, cutoff=0.1)
```

byCorrectDF

Perform multiple testing correction and filtering with Benjamini-Yekutieli

## **Description**

This function performs the Benjamini-Yekutieli correction for multiple testing in a dataframe column of p-values and filters the data-frame based on p-values.

#### Usage

```
byCorrectDF(df, ...)
```

## **Arguments**

df A dataframe with a column of p-values.

... Additional arguments passed to fdrCorrectDF.

#### Value

The data frame with Benjamini-Yekutieli-corrected p-values.

cellDistribution

Show the distribution of cell sets among cells

## **Description**

This function returns a logical matrix that shows the representation of cell sets among all cells.

```
cellDistribution(cellSets, allCells)
```

8 cellSetsOverlaps

## Arguments

cellSets A list of character vectors.

allCells Names of all cells in the dataset.

#### Value

A logical matrix with genes as rows and cells as columns.

#### **Examples**

```
cellSets <- list(c('A', 'H', 'J'),
c('B', 'D', 'E', 'F', 'J'),
c('C', 'I', 'L'))
allCells <- LETTERS[seq(15)]
cellDistribution(cellSets, allCells)</pre>
```

cellSetsOverlaps

Calculates the significance of overlaps of pairs of cells sets

#### **Description**

This function computes the statistical significance of overlaps of pairs of cell sets.

#### Usage

```
cellSetsOverlaps(cellSets, nCells, pairs = NULL, overlapFileName = NULL)
```

#### **Arguments**

cellSets A list of character arrays.

nCells The total number of cells in the Seurat object.

pairs Pairs of cell sets to be assessed. If NULL (as default), all pairs will be assessed.

overlapFileName

The name of the file where the overlap data frame will be saved. This option can be used to save time when performing exploratory analyses such as trying different jaccardCutoff parameters in breakWeakTies. Default is NULL (the overlap data frame will not be saved).

#### Value

A data frame listing statistics for all cell set overlaps: cell set sizes, recorded and expected shared cells, the recorded-over-expected ratio and the hypergeometric p-value.

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## **Examples**

```
cellSets <- list(G1 = c('A', 'H', 'J'),
G2 = c('B', 'D', 'E', 'F', 'J'),
G3 = c('C', 'I', 'L'))
cellSetsOverlaps(cellSets, 40)</pre>
```

computePairScores

Compute aggregate gene pair scores

## Description

This function assesses the relative contribution of each gene pair to the CSOA score

## Usage

```
computePairScores(
  overlapDF,
  pcPairScores,
  pairFileName = "pairs",
  keepOverlapOrder = FALSE
)
```

## Arguments

overlapDF An overlap data frame.

pcPairScores A date frame of pair scores in each cell for each pair in the overlap data frame.

pairFileName The name of the file where the pair data frame will be saved.

keepOverlapOrder

Whether to keep the rank-based order of overlaps in the pair score file, as opposed to changing it to a pair score-based order.

## Value

A data frame with overlap and pair scores and ranks.

10 edgeLists.default

computePCPairScores Compu

Compute per-cell gene pair scores

#### **Description**

This function scores each gene pair corresponding to a top overlap in each cell.

#### Usage

```
computePCPairScores(overlapDF, normExp)
```

#### **Arguments**

overlapDF

An overlap data frame.

normExp

A min-max normalized expression matrix of the genes involved in top overlaps.

#### **Details**

The score is calculated by multiplying the overlap score with the min-max-normalized expression of the two corresponding genes.

#### Value

A data frame with gene pairs as rows and cells as columns.

edgeLists.default

Extract the edge list from overlap data frame or list of overlap data frames

#### **Description**

This function creates a list of data frames with three columns: gene1, gene2 and group. If overlap0bj is an overlap data frame, the groups correspond to the connected components. If it is a list of overlap data frames, the groups must be specified as groupNames.

```
## Default S3 method:
edgeLists(overlapObj, ...)

## S3 method for class 'data.frame'
edgeLists(overlapObj, ...)

## S3 method for class 'list'
edgeLists(overlapObj, groupNames, cutoff = NULL, ...)
edgeLists(overlapObj, ...)
```

expMat 11

## **Arguments**

overlapObj An overlap data frame or list of overlap data frames.

... Additional arguments.

groupNames Names of groups. If provided, must be a vector of the same length as the list of

overlap data frames.

cutoff Number of retained edges from each overlap data frame after refiltering. If NULL

(as default), no refiltering will be performed.

#### Value

A list of data frames.

expMat

Extracts the data expression matrix from object

## **Description**

This function extracts the data expression matrix from object as a non-sparse matrix. Selected genes can be specified as input.

#### Usage

```
expMat(scObj, ...)
## Default S3 method:
expMat(scObj, genes = NULL, ...)
## S3 method for class 'Seurat'
expMat(scObj, ...)
## S3 method for class 'SingleCellExperiment'
expMat(scObj, ...)
## S3 method for class 'dgCMatrix'
expMat(scObj, ...)
## S3 method for class 'matrix'
expMat(scObj, ...)
```

#### **Arguments**

sc0bj A Seurat object, SingleCellExperiment object, or expression matrix.

... Additional arguments.

genes Genes retained in the expression matrix. If NULL, all genes will be retained

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## Value

An expression matrix.

## **Examples**

```
library(Seurat)
mat <- matrix(0, 6, 4)
mat[sample(length(mat), 7)] <- sample(3, 7, TRUE)
seuratObj <- CreateSeuratObject(counts = mat)
seuratObj <- NormalizeData(seuratObj)
expMat(seuratObj)</pre>
```

fdrCorrectDF

Perform multiple testing correction by controlling the false discovery rate

## Description

This function perform multiple testing correction by controlling the false discovery rate.

## Usage

```
fdrCorrectDF(
   df,
   fdrControlFun,
   pvalThr = 0.05,
   colStr = "pval",
   newColStr = "pvalAdj"
)
```

## **Arguments**

df A dataframe with a column of p-values.

pvalThr p-value threshold.

colStr Name of the column of p-values.

newColStr Name of the column of adjusted p-values that will be created.

#### Value

The data frame with p-values corrected using the method of choice (Benjamini-Hochberg or Benjamini-Yekutieli)

featureWes 13

featureWes A

A feature plot with a more distinctive color scheme.

## Description

This function customizes the appearance of Seurat::FeaturePlot for improved distinctiveness and aesthetics.

## Usage

```
featureWes(
   seuratObj,
   feature,
   title = feature,
   idClass = NULL,
   labelSize = 3.5,
   titleSize = 12,
   wesPal = "Royal1",
   wesLow = 3,
   wesHigh = 2,
   ...
)
```

## Arguments

seuratObj	A Seurat object.
feature	Seurat feature.
title	Plot title.
idClass	Column to be used for labelling. If NULL, no column-based labels will be generated.
labelSize	Size of labels. Ignored if idClass is NULL.
titleSize	Title size.
wesPal	A wesanderson palette.
wesLow	Index of color marking low values.
wesHigh	Index of color marking high values.
•••	Additional arguments passed to Seurat::FeaturePlot.

## Value

A ggplot object.

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#### **Examples**

```
library(Seurat)
mat <- matrix(0, 3000, 800)
mat[sample(length(mat), 90000)] <- sample(8, 90000, TRUE)
seuratObj <- CreateSeuratObject(counts = mat)
seuratObj <- FindVariableFeatures(seuratObj, nfeatures=200)
seuratObj <- NormalizeData(seuratObj)
seuratObj <- ScaleData(seuratObj)
seuratObj <- RunPCA(seuratObj, verbose=FALSE)
seuratObj <- RunUMAP(seuratObj, dims=1:20, verbose=FALSE)
featureWes(seuratObj, 'Feature3')</pre>
```

geneRadialPlot

Radial plot for an overlap data frame

#### **Description**

This function draws a radial plot for an overlap data frame to illustrate gene participation in top overlaps.

## Usage

```
geneRadialPlot(
  overlapObj,
  title = "Top overlap genes plot",
  degreeLegendTitle = "Number of top overlaps",
  groupLegendTitle = "Group",
  extraCircles = 2,
  groupNames = NULL,
  cutoff = NULL,
  ...
)
```

## Arguments

```
overlapObj
                  An overlap data frame or list of overlap data frames.
                  Plot title.
title
degreeLegendTitle
                  The title of the degree legend.
groupLegendTitle
                  The title of the group legend. If NULL, no groups will be distinguished.
extraCircles
                  Number of extra circles to be displayed on the plot.
                  Names of groups. If provided, must be a vector of the same length as the list of
groupNames
                   overlap data frames.
cutoff
                  Number of retained edges from each overlap data frame after refiltering. If NULL
                   (as default), no refiltering will be performed.
                   Additional parameters passed to henna::radialPlot.
```

generateOverlaps 15

#### **Details**

The function can separate genes by groups. The groups can be, for instance, different gene sets, or different connected components of the same overlap data frame. A wrapper around henna::radialPlot

#### Value

A ggplot object.

#### **Examples**

```
edgesDF <- data.frame(gene1 = paste0('G', c(1, 2, 3, 4, 7, 8, 10,
11, 11, 10, 10, 10)),
gene2 = paste0('G', c(2, 5, 1, 8, 4, 9, 12,
13, 14, 13, 16, 14)))
edgesDF <- henna::connectedComponents(edgesDF, 'group')
geneRadialPlot(edgesDF, groupLegendTitle='Component', extraCircles=1)</pre>
```

generateOverlaps

Generate overlaps of cell sets for input genes

## Description

This function constructs, for each gene in the expression matrix, a set of cells expressing the gene at or above the input percentile. Subsequently, overlaps of pairs of the constructed cell sets are assessed for statistical significance.

#### Usage

```
generateOverlaps(
  geneSetExp,
  percentile = 90,
  pairs = NULL,
  overlapFileName = NULL)
```

## **Arguments**

geneSetExp

A gene expression non-sparse matrix with the rows restricted to the genes for

which cell sets will be computed.

percentile

A positive number under 100.

pairs

Pairs of cell sets to be assessed. If NULL (as default), all pairs will be assessed.

overlapFileName

The name of the file where the overlap data frame will be saved. This option can be used to save time when performing exploratory analyses such as trying different jaccardCutoff parameters in breakWeakTies. Default is NULL (the overlap data frame will not be saved).

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## **Details**

Wrapper around percentileSets and cellSetsOverlaps.

#### Value

A data frame listing statistics for all cell set overlaps

## **Examples**

```
mat <- matrix(0, 2000, 500)
rownames(mat) <- paste0('G', seq(2000))
colnames(mat) <- paste0('C', seq(500))
mat[sample(length(mat), 270000)] <- sample(50, 270000, TRUE)
mat <- mat[paste0('G', sample(2000, 5)), ]
generateOverlaps(mat)</pre>
```

getPairs

Get all unordered pairs of two elements from a vector

## Description

This function returns all unorderded pairs of two elements from a vector.

## Usage

```
getPairs(v)
```

## **Arguments**

V

A vector.

#### Value

A list of vectors of length 2.

## **Examples**

```
v <- c('ASD', 'VBN', 'HJKL')
getPairs(v)</pre>
```

mtCorrectDF 17

mtCorrectDF	Perform multiple testing correction	
-------------	-------------------------------------	--

## Description

This function performs correction for multiple testing in a dataframe column of p-values and filters the data-frame based on p-values.

## Usage

```
mtCorrectDF(df, mtMethod = c("bf", "bh", "by"), ...)
```

#### **Arguments**

df A dataframe with a column of p-values.

mtMethod Multiple testing correction method. Options are Bonferroni ('bf'), Benjamini-

Hochberg('bh'), and Benjamini-Yekutieli ('by').

... Additional arguments passed to the multiple testing correction method.

#### **Details**

This function calls bfCorrectDF, bhCorrectDF or byCorrectDF, depending on the selected option. See their documentation for additional parameters.

networkPlotDF Prepare overlap data frame for network plot

#### **Description**

This function prepares a ranked and filtered overlap data frame for network plot.

#### Usage

```
networkPlotDF(overlapDF, rankCol = "rank", edgeScale = 2)
```

#### **Arguments**

overlapDF Overlap data frame.

rankCol Name of the rank column.

edgeScale Scaling factor used in generating edge weights.

#### Value

A data frame ready to serve as input to networkPlot.

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overlapCutoffPlot

Plot the selection of overlaps

## Description

This function illustrates the process of selecting the overlap rank cutoff by plotting rank frequencies against ranks and showcasing the convex hull of the rank-frequency points.

## Usage

```
overlapCutoffPlot(
  overlapDF,
  title = "Overlap cutoff plot",
  palette = c("purple", "yellow"),
  hullWidth = 0.8,
  xLab = "Overlap rank",
  yLab = "Frequency",
  legendLabs = c("Accepted overlaps", "Discarded overlaps"),
  pointShape = 24,
  ...
)
```

#### **Arguments**

overlapDF Processed overlap data frame created with processOverlaps.

title Plot title.

palette Color palette. Must have two colors, the first one representing accepted overlaps

and the other representing discarded overlaps.

hullWidth Width of the convex hull.

xLab x axis label. yLab y axis label. legendLabs Legend labels. pointShape Point shape.

... Additional arguments passed to henna::hullPlot.

#### **Details**

A wrapper around henna::hullPlot.

#### Value

A ggplot object.

overlapGenes 19

## **Examples**

```
overlapDF <- data.frame(gene1=paste0('G', c(1, 3, 7, 6, 8, 2, 4, 3, 4, 5)), gene2=paste0('G', c(2, 7, 2, 5, 4, 5, 1, 2, 2, 8)), rank=c(1, 2, 3, 4, 4, 6, 7, 7, 7, 10)) overlapCutoffPlot(overlapDF)
```

overlapGenes

Get all genes from an overlap data frame

## **Description**

This function gets all genes from an overlap data frame.

## Usage

```
overlapGenes(overlapDF, components = NULL)
```

## **Arguments**

overlapDF Overlap data frame.

components A numeric vector representing the connected components of the overlap data

frame graph.

## Value

A character vector of genes.

## **Examples**

```
overlapDF <- data.frame(gene1 = paste0('G', c(1, 2, 3)), gene1 = paste0('G', c(2, 7, 8))) overlapGenes(overlapDF)
```

overlapNetworkPlot

Plot the overlaps as a network

#### **Description**

This function plots the graph of the overlap data frame, with genes as vertices and overlaps as edges.

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#### Usage

```
overlapNetworkPlot(
  overlapDF,
  title = "Top overlaps network plot",
  nodeColor = "orange",
  edgeColor = "green4",
  ...
)
```

## Arguments

overlapDF Overlap data frame.

title Plot title.

nodeColor The color of nodes. If NULL, the default henna::networkPlot color scheme will

be used, which uses different colors for nodes belonging to different connected

components.

edgeColor The color of edges.

... Additional parameters passed to henna::networkPlot.

#### **Details**

A thin wrapper around henna::networkPlot.

#### Value

An overlap network plot.

#### **Examples**

```
overlapDF <- data.frame(gene1 = paste0('G', c(1, 2, 5, 6, 7, 17)), gene2 = paste0('G', c(2, 5, 8, 11, 11, 11)), rank = c(1, 1, 3, 3, 3, 3)) overlapNetworkPlot(overlapDF)
```

overlapPairs

Extract gene pairs from overlap data frame

## Description

This function extracts the gene pairs from an overlap data frame.

```
overlapPairs(overlapDF)
```

percentileSets 21

## **Arguments**

overlapDF Overlap data frame.

#### Value

A list of gene pairs.

## **Examples**

```
overlapDF <- data.frame(gene1 = paste0('G', c(1, 2, 3)), gene1 = paste0('G', c(2, 7, 8))) overlapPairs(overlapDF)
```

percentileSets

Generates cell expressing input genes at an input percentile

## Description

This function constructs, for each gene in the expression matrix, a set of cells expressing the gene at or above the input percentile.

#### Usage

```
percentileSets(geneSetExp, percentile = 90)
```

## **Arguments**

geneSetExp A gene expression non-sparse matrix with the rows restricted to the genes for

which cell sets will be computed.

percentile A positive number under 100.

#### Value

A named list of character vectors of length equaling the number of input genes. Each vector stores the cells expressing the gene at or above the input percentile.

## **Examples**

```
mat <- matrix(0, 1000, 500)
rownames(mat) <- paste0('G', seq(1000))
colnames(mat) <- paste0('C', seq(500))
mat[sample(length(mat), 70000)] <- sample(50, 70000, TRUE)
mat <- mat[paste0('G', sample(1000, 3)), ]
percentileSets(mat)</pre>
```

22 processOverlaps

processOver]	anc
brocessoveri	Labs

Process data frame of overlaps of cell sets

## **Description**

This function filters, ranks and scores previously generated overlaps of cell sets.

## Usage

```
processOverlaps(
  overlapDF,
  mtMethod = c("by", "bh", "bf"),
  jaccardCutoff = NULL,
  osMethod = c("log", "minmax"),
  ...
)
```

#### **Arguments**

overlapDF Overlap data frame.

mtMethod Multiple testing correction method. Options are Bonferroni ('bf'), Benjamini-Hochberg('bh'), and the default Benjamini-Yekutieli ('by').

jaccardCutoff A cutoff used in the filtering of edges with low Jaccard scores. If NULL (as default), no filtering of such edges will be performed.

osMethod Method used to compute overlap scores. Options are "log" and "minmax".

Additional arguments passed to mtCorrectDF.

#### **Details**

Wrapper around byCorrectDF, rankOverlaps, prepareFiltering, filterOverlaps and scoreOverlaps. If jaccardCutoff is not NULL, it also calls breakWeakTies between filterOverlaps and scoreOverlaps.

#### Value

A data frame consisting of filtered, ranked and scored cell sets overlaps

#### **Examples**

```
overlapDF <- data.frame(gene1=paste0('G',
c(1, 3, 7, 6, 8, 2, 4, 3, 4, 5)),
gene2=paste0('G',
c(2, 7, 2, 5, 4, 5, 1, 2, 2, 8)),
ratio=runif(10, 2, 10),
pval=runif(10, 0, 1e-10))
processOverlaps(overlapDF)</pre>
```

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qGrab

Read and delete a .qs file

## **Description**

This functions reads a .qs file, deletes it, and returns its content.

## Usage

```
qGrab(qsFile)
```

## **Arguments**

qsFile

Name of .qs file with path.

#### Value

The content of the .qs file.

## **Examples**

```
library(qs)
qsave(c(1, 2, 3), 'temp.qs')
qGrab('temp.qs')
```

runCSOA

Run the CSOA pipeline

## Description

This function generates cell set overlaps for input gene sets based on percentiles of gene expression, computes the significance of these overlaps, ranks, filters and scores the overlaps, and builds a percell score by summing the products of overlap scores and the min-max-normalized expression of the corresponding pairs of genes.

```
runCSOA(
   scObj,
   geneSets,
   percentile = 90,
   mtMethod = c("by", "bh", "bf"),
   jaccardCutoff = NULL,
   osMethod = c("log", "minmax"),
   overlapFileName = NULL,
```

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```
pairFileTemplate = NULL,
keepOverlapOrder = FALSE,
...
)
```

#### **Arguments**

sc0bj A Seurat object, SingleCellExperiment object, or expression matrix.

geneSets Named list of character vectors of which each must contain at least two genes.

percentile A positive number under 100.

mtMethod Multiple testing correction method. Options are Bonferroni ('bf'), Benjamini-

Hochberg('bh'), and the default Benjamini-Yekutieli ('by').

jaccardCutoff A cutoff used in the filtering of edges with low Jaccard scores. If NULL (as

default), no filtering of such edges will be performed.

osMethod Method used to compute overlap scores. Options are "log" and "minmax".

overlapFileName

The name of the file where the overlap data frame will be saved. This option can be used to save time when performing exploratory analyses such as trying different jaccardCutoff parameters in breakWeakTies. Default is NULL (the overlap data frame will not be saved).

pairFileTemplate

Character object used in the naming of the files where the pair data frames will be saved. Default is NULL (the pair data frames will not be saved).

keepOverlapOrder

Keep the rank-based order of overlaps in the pair score file, as opposed to changing it to a pair score-based order. Ignored if pairFileTemplate is NULL.

... Additional arguments.

#### **Details**

Wrapper around expMat, generateOverlaps, scoreCells and attachCellScores.

#### Value

An object of the same class as scObj with per-gene-set CSOA scores assigned for each cell.

#### **Examples**

```
mat <- matrix(0, 500, 300)
rownames(mat) <- paste0('G', seq(500))
colnames(mat) <- paste0('C', seq(300))
mat[sample(8000)] <- runif(8000, max=13)
genes <- paste0('G', seq(200))
mat[genes, 20:50] <- matrix(runif(200 * 31, min = 14, max = 15),
nrow = 200, ncol = 31)
geneSet1 <- paste0('G', seq(1, 150))
geneSet2 <- paste0('G', seq(50, 200))
df <- runCSOA(mat, list(a = geneSet1, b = geneSet2))</pre>
```

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head(df)

scoreCells

Generate CSOA scores from overlap data frame and list of pairs

## **Description**

This function scores an overlap data frame using its associated list of pairs. The overlap data frame is split based on the overlaps corresponding to each gene set and scored, and the output is rejoined as a data frame.

## Usage

```
scoreCells(
  geneSetExp,
  overlapDF,
  setPairs,
  geneSetNames,
  mtMethod = c("by", "bh", "bf"),
  jaccardCutoff = NULL,
  osMethod = c("log", "minmax"),
  pairFileTemplate = NULL,
  keepOverlapOrder = FALSE,
  ...
)
```

#### **Arguments**

geneSetExp A gene expression non-sparse matrix with the rows restricted to the genes for

which cell sets will be computed.

overlapDF Overlap data frame.

setPairs A list of overlaps corresponding to each input gene set.

geneSetNames Character vector of names of gene sets.

mtMethod Multiple testing correction method. Options are Bonferroni ('bf'), Benjamini-

Hochberg('bh'), and the default Benjamini-Yekutieli ('by').

jaccardCutoff A cutoff used in the filtering of edges with low Jaccard scores. If NULL (as

default), no filtering of such edges will be performed.

osMethod Method used to compute overlap scores. Options are "log" and "minmax".

pairFileTemplate

Character object used in the naming of the files where the pair data frames will be saved. Default is NULL (the pair data frames will not be saved).

keepOverlapOrder

Keep the rank-based order of overlaps in the pair score file, as opposed to chang-

ing it to a pair score-based order. Ignored if pairFileTemplate is NULL.

... Additional arguments passed to mtCorrectDF.

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#### **Details**

This function calls scoreCells to score each gene set data frame split from the full overlap data frame

#### Value

A data frame whose columns correspond to the CSOA scores of the input gene sets.

#### **Examples**

```
mat <- matrix(0, 500, 300)
rownames(mat) <- paste0('G', seq(500))</pre>
colnames(mat) <- paste0('C', seq(300))</pre>
mat[sample(8000)] <- runif(8000, max=13)</pre>
genes <- paste0('G', seq(200))</pre>
mat[genes, 20:50] <- matrix(runif(200 * 31, min=14, max=15),</pre>
nrow=200, ncol=31)
geneSet1 \leftarrow paste0('G', seq(1, 150))
geneSet2 <- paste0('G', seq(50, 200))</pre>
geneSets <- list(geneSet1, geneSet2)</pre>
geneSets <- lapply(geneSets, sort)</pre>
setPairs <- lapply(geneSets, getPairs)</pre>
pairs <- Reduce(union, setPairs)</pre>
genes <- union(geneSet1, geneSet2)</pre>
mat <- mat[genes, ]</pre>
overlapDF <- generateOverlaps(mat, pairs=pairs)</pre>
scoreDF <- scoreCells(mat, overlapDF, setPairs, c('set1', 'set2'))</pre>
head(scoreDF)
```

scoreCellsCore

Generate CSOA scores from overlap data frame for a single gene set

## **Description**

This function computes per-cell CSOA scores from overlap data frame for a single gene set.

```
scoreCellsCore(
  geneSetExp,
  overlapDF,
  colStr = "CSOA",
  mtMethod = c("by", "bh", "bf"),
  jaccardCutoff = NULL,
  osMethod = c("log", "minmax"),
  pairFileName = NULL,
  keepOverlapOrder = FALSE,
  ...
)
```

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## Arguments

geneSetExp	A gene expression non-sparse matrix with the rows restricted to the genes for which cell sets will be computed.
overlapDF	Overlap data frame.
colStr	Name of column where CSOA scores will be stored.
mtMethod	Multiple testing correction method. Options are Bonferroni ('bf'), Benjamini-Hochberg('bh'), and the default Benjamini-Yekutieli ('by').
jaccardCutoff	A cutoff used in the filtering of edges with low Jaccard scores. If NULL (as default), no filtering of such edges will be performed.
osMethod	Method used to compute overlap scores. Options are "log" and "minmax".
pairFileName	The name of the file where the pair data frame will be saved.
keepOverlapOrd	er
	Whether to keep the rank-based order of overlaps in the pair score file, as opposed to changing it to a pair score-based order.

Additional arguments passed to mtCorrectDF. . . .

## Value

A data frame with a column corresponding to the CSOA scores.

scoreModules	Run CSOA separately on the connected components of the overlap graph
--------------	--

## Description

This function runs CSOA on the connected components of the graph having the filtered overlaps as edges.

```
scoreModules(
  scObj,
 networkDF,
  components,
 colStrTemplate = "CSOA_component",
)
```

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## Arguments

A Seurat object, SingleCellExperiment object, or expression matrix.

A data frame with gene1, gene2 and component columns.

A numeric vector representing the connected components of the overlap data frame graph.

ColStrTemplate Character used in the naming of the component gene sets.

Additional parameters passed to runCSOAMultiple.

#### Value

An object of the same class as scObj with CSOA scores corresponding to the genes defining each connected components assigned for each cell.

#### **Examples**

```
mat <- matrix(0, 500, 300)
rownames(mat) <- paste0('G', seq(500))</pre>
colnames(mat) <- paste0('C', seq(300))</pre>
mat[sample(8000)] <- runif(8000, max=13)</pre>
genes1 <- paste0('G', seq(100))</pre>
mat[genes1, 20:50] <- matrix(runif(100 * 31, min = 14, max = 15),
nrow = 100, ncol = 31)
genes2 <- paste0('G', seq(101, 200))</pre>
mat[genes2, 70:100] \leftarrow matrix(runif(100 * 31, min = 14, max = 15),
nrow = 100, ncol = 31)
genes <- union(genes1, genes2)</pre>
mat <- mat[genes, ]</pre>
overlapDF <- generateOverlaps(mat)</pre>
overlapDF <- processOverlaps(overlapDF)</pre>
overlapDF <- henna::connectedComponents(overlapDF)</pre>
df <- scoreModules(mat, overlapDF, unique(overlapDF$component))[[2]]</pre>
head(df)
```

wesBinaryGradient

Adds a gradient color scale using two wesanderson colors

## **Description**

This function a gradient color scale to a ggplot object using a wesanderson palette, an index marking low values, and an index marking high values. The indices are used to select colors from the wesanderson palette of choice.

```
wesBinaryGradient(p, palType, wesPal = "Royal1", wesLow = 3, wesHigh = 2, ...)
```

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## Arguments

p A ggplot object.

palType Palette type: color or fill, continuous or discrete. Accepted values are 'color-

Cont', 'fillCont', 'colDis' and 'fillDis'. The function shows a warning and does

not change the color scheme if a different value is passed here.

wesPal A wesanderson palette.

wesLow Index of color marking low values.
wesHigh Index of color marking high values.
... Arguments passed to other functions.

## Value

A ggplot object with a new color scheme.

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