

# Package ‘CytoSimplex’

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**Type** Package

**Title** Simplex Visualization of Cell Fate Similarity in Single-Cell Data

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**Description** Create simplex plots to visualize the similarity between single-cells and selected clusters in a 1-/2-/3-simplex space.

Velocity information can be added as an additional layer.

See Liu J, Wang Y et al (2023) <[doi:10.1093/bioinformatics/btaf119](https://doi.org/10.1093/bioinformatics/btaf119)> for more details.

**URL** <https://welch-lab.github.io/CytoSimplex/>,  
<https://github.com/welch-lab/CytoSimplex>

**License** GPL-3

**Encoding** UTF-8

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---

|              |  |
|--------------|--|
| colNormalize | <i>Normalize each column of the input matrix by the column sum</i> |
|--------------|--|

---

### Description

Normalize each column of the input matrix by the column sum

### Usage

```
colNormalize(x, scaleFactor = NULL, log = FALSE, ...)

## Default S3 method:
colNormalize(x, scaleFactor = NULL, log = FALSE, ...)

## S3 method for class 'dgCMatrix'
colNormalize(x, scaleFactor = NULL, log = FALSE, ...)

## S3 method for class 'Seurat'
colNormalize(
  x,
  scaleFactor = NULL,
  log = FALSE,
  assay = NULL,
```

```

    layer = "counts",
    ...
)

## S3 method for class 'SingleCellExperiment'
colNormalize(x, scaleFactor = NULL, log = FALSE, assay.type = "counts", ...)

```

### Arguments

|             |  |
|-------------|--|
| x           | Feature by observation matrix. Alternatively, Seurat object or SingleCellExperiment object with raw counts available are also supported. |
| scaleFactor | Multiplier on normalized data. Default NULL.   |
| log         | Logical. Whether to take log <sub>1p</sub> transformation after scaling. Default FALSE   |
| ...         | Additional arguments passed to methods   |
| assay       | For "Seurat" method, the specific assay to get data from. Default NULL to the default assay.   |
| layer       | For "Seurat" method, which layer of the assay to be used. Default "counts".  |
| assay.type  | For "SingleCellExperiment" method, the assay type to get data from. Default "counts".  |

### Value

Normalized matrix of the same size

A Seurat object with normalized data in the specified slot of the specified assay.

A SingleCellExperiment object with normalized data in the specified assay. "normcounts" if log = FALSE and "logcounts" if log = TRUE.

### Examples

```

rnaNorm <- colNormalize(rnaRaw)

# Seurat example
library(Seurat)
srt <- CreateSeuratObject(rnaRaw)
srt <- colNormalize(srt)

# SingleCellExperiment example
library(SingleCellExperiment)
sce <- SingleCellExperiment(assays = list(counts = rnaRaw))
sce <- colNormalize(sce)

```

---

plotBinary

*Create binary plots*


---

### Description

Create binary plots that show similarity between single cells and two selected terminals in a barycentric coordinate. The two vertices are placed at the left and right of a 2D plot where x-axis measures the similarity. Y-axis is jittered for a clear view. A density (histogram) curve is added for indicating the distribution.

See [plotTernary](#) manual for more details.

### Usage

```
plotBinary(x, ...)

## Default S3 method:
plotBinary(
  x,
  clusterVar,
  vertices,
  features = NULL,
  byCluster = NULL,
  processed = FALSE,
  method = c("euclidean", "cosine", "pearson", "spearman"),
  force = FALSE,
  sigma = 0.08,
  scale = TRUE,
  dotColor = "grey60",
  returnData = FALSE,
  ...
)

## S3 method for class 'Seurat'
plotBinary(
  x,
  layer = "counts",
  assay = NULL,
  clusterVar = NULL,
  processed = FALSE,
  ...
)

## S3 method for class 'SingleCellExperiment'
plotBinary(x, assay.type = "counts", clusterVar = NULL, processed = FALSE, ...)

## S3 method for class 'simMat'
```

```

plotBinary(
  x,
  dotSize = 0.6,
  dotColor = "grey60",
  denslinewidth = 0.8,
  labelColors = c("#3B4992FF", "#EE0000FF"),
  title = NULL,
  ...
)

```

### Arguments

|            |   |
|------------|---|
| x          | Input data. Can be a matrix or dgCMatrix object with cells as columns, a Seurat or SingleCellExperiment object. "simMat" method takes intermediate values.  |
| ...        | Arguments passed to other methods.  |
| clusterVar | A vector/factor assigning the cluster variable to each column of the matrix object. For "Seurat" method, NULL (default) for Idents(x), or a variable name in meta.data slot. For "SingleCellExperiment" method, NULL (default) for colLabels(x), or a variable name in colData slot.  |
| vertices   | Vector of three unique cluster names that will be used for plotting. Or a named list that groups clusters as three terminal vertices. There must not be any overlap between groups.   |
| features   | Valid matrix row subsetting index to select features for similarity calculation. Default NULL uses all available features.  |
| byCluster  | Default NULL to generate one plot with all cells. Set "all" to split cells in plot by cluster and returns a list of subplots for each cluster as well as the plot including all cells. Otherwise, a vector of cluster names to generate a list of subplots for the specified clusters.  |
| processed  | Logical. Whether the input matrix is already processed. TRUE will bypass internal preprocessing and input matrix will be directly used for similarity calculation. Default FALSE and raw count input is recommended. If missing in call, using slot = "counts" in "Seurat" method or using assay.type = "counts" in "SingleCellExperiment" method will force this argument to be FALSE and others for TRUE. |
| method     | Similarity calculation method. Default "euclidean". Choose from "euclidean", "cosine", "pearson", "spearman".   |
| force      | Whether to force calculate the similarity when more than 500 features are detected, which is generally not recommended. Default FALSE.  |
| sigma      | Gaussian kernel parameter that controls the effect of variance. Only effective when using a distance metric (i.e. method is "euclidian" or "cosine"). Larger value tighten the dot spreading on figure. Default 0.08.   |
| scale      | Whether to min-max scale the distance matrix by clusters. Default TRUE.   |
| returnData | Logical. Whether to return similarity data instead of generating plot. Default FALSE.   |

|                   |  |
|-------------------|--|
| layer             | For "Seurat" method, which layer of the assay to be used. Default "counts".                              |
| assay             | For "Seurat" method, the specific assay to get data from. Default NULL to the default assay.             |
| assay.type        | For "SingleCellExperiment" methods. Which assay to use for calculating the similarity. Default "counts". |
| dotSize, dotColor | Dot aesthetics passed to <a href="#">geom_point</a> . Default 0.6 and "grey60".                          |
| densLinewidth     | Density plot line aesthetic. Default 0.8.  |
| labelColors       | Color of the axis lines and vertex labels. Default c("#3B4992FF", "#EE0000FF") (blue and red).           |
| title             | Title text of the plot. Default NULL.  |

### Value

For 'simMat' method, a ggplot object. For other methods, a ggplot object when `splitCluster = FALSE`, or a list of ggplot objects when `splitCluster = TRUE`.

### Examples

```
gene <- selectTopFeatures(rnaRaw, rnaCluster, c("RE", "OS"))
plotBinary(rnaRaw, rnaCluster, c("RE", "OS"), gene)

# Seurat example
library(Seurat)
srt <- CreateSeuratObject(rnaRaw)
Idents(srt) <- rnaCluster
gene <- selectTopFeatures(srt, vertices = c("OS", "RE"))
plotBinary(srt, features = gene, vertices = c("OS", "RE"))

# SingleCellExperiment example
library(SingleCellExperiment)
sce <- SingleCellExperiment(assays = list(counts = rnaRaw))
colLabels(sce) <- rnaCluster
gene <- selectTopFeatures(sce, vertices = c("OS", "RE"))
plotBinary(sce, features = gene, vertices = c("OS", "RE"))
```

---

plotQuaternary

*Create quaternary simplex plots*

---

### Description

Create quaternary plots that show similarity between single cells and selected four terminals in a baricentric coordinate.

See [plotTernary](#) for more details on methodologies.

A dynamic rotating view in a GIF image file can be created with [writeQuaternaryGIF](#). Package `magick` must be installed in advance. Linux users may refer to this [installation guide](#).

**Usage**

```
plotQuaternary(x, ...)

## Default S3 method:
plotQuaternary(
  x,
  clusterVar,
  vertices,
  features = NULL,
  veloGraph = NULL,
  byCluster = NULL,
  processed = FALSE,
  method = c("euclidean", "cosine", "pearson", "spearman"),
  force = FALSE,
  sigma = 0.05,
  scale = TRUE,
  dotColorBy = NULL,
  dotColor = NULL,
  palette = "D",
  direction = 1,
  breaks = NULL,
  legendTitle = NULL,
  returnData = FALSE,
  ...
)
```

**Arguments**

|                      |  |
|----------------------|--|
| x                    | Input data. Can be a matrix or dgCMatrix object with cells as columns, a Seurat or SingleCellExperiment object.  |
| ...                  | Arguments passed on to <a href="#">plotQuaternary.simMat</a>   |
| nGrid                | Number of grids along the x-axis of the tetrahedron. Default 10.   |
| radius               | Arrow length of unit velocity. Lower this when arrows point outside of the tetrahedron. Default 0.2.   |
| dotSize              | Size of each dot. Default 0.6 for static figure, and 4 for interactive view.   |
| labelColors          | Colors of the vertex labels. Default <code>c("#3B4992FF", "#EE0000FF", "#008B45FF", "#631879FF")</code> (blue, red, green and purple).                           |
| arrowLinewidth       | Arrow aesthetics. Default 1.6 for interactive view, 0.6 for static figure.   |
| arrowAngle, arrowLen | Arrow aesthetics passed to <code>TODOOOO grid::arrow</code> . The length of the arrow will be internally converted to unit onject in inches. Default 20 and 0.1. |
| edgeLinewidth        | Controls the linewidth of the edges of the tetrahedron. Default 1.   |
| vertexLabelSize      | Numeric, size of vertex text label relative to default size. Default 1.  |

|                         |  |
|-------------------------|--|
|                         | <p><code>title</code> Title text of the plot. Default NULL.</p> <p><code>titleSize, titleColor</code> Setting on the main title text. Default 1, and "black".</p> <p><code>theta, phi</code> Numeric scalar. The angles defining the viewing direction. <code>theta</code> gives the azimuthal direction and <code>phi</code> the colatitude. Default 20 and 0.</p> <p><code>interactive</code> Logical. Whether to display plotly interactive view. Default TRUE.</p> |
| <code>clusterVar</code> | A vector/factor assigning the cluster variable to each column of the matrix object. For "Seurat" method, NULL (default) for <code>Idents(x)</code> , or a variable name in <code>meta.data</code> slot. For "SingleCellExperiment" method, NULL (default) for <code>colLabels(x)</code> , or a variable name in <code>colData</code> slot.   |
| <code>vertices</code>   | Vector of three unique cluster names that will be used for plotting. Or a named list that groups clusters as three terminal vertices. There must not be any overlap between groups.  |
| <code>features</code>   | Valid matrix row subsetting index to select features for similarity calculation. Default NULL uses all available features.   |
| <code>veloGraph</code>  | Cell x cell <code>dgMatrix</code> object containing velocity information. Shows velocity grid-arrow layer when specified. Default NULL does not show velocity.   |
| <code>byCluster</code>  | Default NULL to generate one plot with all cells. Set "all" to split cells in plot by cluster and returns a list of subplots for each cluster as well as the plot including all cells. Otherwise, a vector of cluster names to generate a list of subplots for the specified clusters.   |
| <code>processed</code>  | Logical. Whether the input matrix is already processed. TRUE will bypass internal preprocessing and input matrix will be directly used for similarity calculation. Default FALSE and raw count input is recommended. If missing in call, using <code>slot = "counts"</code> in "Seurat" method or using <code>assay.type = "counts"</code> in "SingleCellExperiment" method will force this argument to be FALSE and others for TRUE.                                  |
| <code>method</code>     | Similarity calculation method. Default "euclidean". Choose from "euclidean", "cosine", "pearson", "spearman".  |
| <code>force</code>      | Whether to force calculate the similarity when more than 500 features are detected, which is generally not recommended. Default FALSE.   |
| <code>sigma</code>      | Gaussian kernel parameter that controls the effect of variance. Only effective when using a distance metric (i.e. <code>method</code> is "euclidian" or "cosine"). Larger values tighten the dot spreading on figure. Default 0.05.  |
| <code>scale</code>      | Whether to min-max scale the distance matrix by clusters. Default TRUE.  |
| <code>dotColorBy</code> | A vector/factor for coloring dots, can be either categorical (must be character or factor) or continuous. Default NULL.  |
| <code>dotColor</code>   | Character vector of color codes. When <code>dotColorBy</code> is NULL, use one or as many colors as the number of cells. If <code>dotColorBy</code> is categorical, specify as many colors as the number of categories in <code>dotColorBy</code> or <code>ggplot2</code> categorical color palette is used by default. If <code>dotColorBy</code> is continuous, specify together with <code>breaks</code> argument.  |
| <code>palette</code>    | Color palette to use when <code>dotColorBy</code> is given. Default "D" (viridis) for continuous value and <code>ggplot2</code> default for categorical value. See detail for alternatives.  |



|             |   |
|-------------|---|
| direction   | Sets the order of colors in the scale. Default 1 orders as palette default. If -1, the order of colors is reversed.   |
| breaks      | Number of breaks for continuous color scale passed to non-interactive "plot3D::scatter3D" call. Default NULL.   |
| legendTitle | Title on the legend/colorbar. Default NULL uses "cluster" if dotColorBy is missing (default); user-end variable expression if dotColorBy is directly specified from plotQuaternary.default method; variable name if dotColorBy is specified from Seurat or SingleCellExperiment method. |
| returnData  | Logical. Whether to return similarity and aggregated velocity data if applicable instead of generating plot. Default FALSE.   |

### Value

By default, a "plotly" object. When interactive = FALSE, a "quatPlot" object when byCluster is not specified, or a "list" of "quatPlot" objects when byCluster is specified. When returnData = TRUE, a list of similarity matrix and aggregated velocity matrix is returned.

### See Also

Other plotQuaternary: [plotQuaternary.Seurat\(\)](#), [plotQuaternary.SingleCellExperiment\(\)](#)

### Examples

```
gene <- selectTopFeatures(
  x = rnaRaw,
  clusterVar = rnaCluster,
  vertices = c("RE", "OS", "CH", "ORT")
)
plotQuaternary(
  x = rnaRaw,
  clusterVar = rnaCluster,
  vertices = c("RE", "OS", "CH", "ORT"),
  features = gene
)
```

---

plotQuaternary.Seurat *Create quaternary simplex plot with Seurat object*

---

### Description

Create quaternary plots that show similarity between single cells and selected four terminals in a baricentric coordinate.

See [plotTernary](#) for more details on methodologies.

A dynamic rotating view in a GIF image file can be created with [writeQuaternaryGIF](#). Package magick must be installed in advance. Linux users may refer to this [installation guide](#).

**Usage**

```
## S3 method for class 'Seurat'
plotQuaternary(
  x,
  layer = "counts",
  assay = NULL,
  clusterVar = NULL,
  processed = FALSE,
  dotColorBy = NULL,
  legendTitle = NULL,
  ...
)
```

**Arguments**

|                          |   |
|--------------------------|---|
| <code>x</code>           | A Seurat object   |
| <code>layer</code>       | Layer in the specified assay to use. Default "counts".  |
| <code>assay</code>       | The assay to get data from. Default NULL uses the default assay.  |
| <code>clusterVar</code>  | A variable name in meta.data (x[[ <code>[[</code> ]]]). Default NULL uses <code>Idents(x)</code> .  |
| <code>processed</code>   | Logical. Whether the input matrix is already processed. TRUE will bypass internal preprocessing and input matrix will be directly used for similarity calculation. Default FALSE and raw count input is recommended. If missing in call, using <code>slot = "counts"</code> in "Seurat" method or using <code>assay.type = "counts"</code> in "SingleCellExperiment" method will force this argument to be FALSE and others for TRUE. |
| <code>dotColorBy</code>  | A vector/factor for coloring dots, can be either categorical (must be character or factor) or continuous. Default NULL.   |
| <code>legendTitle</code> | Title on the legend/colorbar. Default NULL uses "cluster" if <code>dotColorBy</code> is missing (default); user-end variable expression if <code>dotColorBy</code> is directly specified from <code>plotQuaternary.default</code> method; variable name if <code>dotColorBy</code> is specified from Seurat or SingleCellExperiment method.   |
| <code>...</code>         | Arguments passed on to <code>plotQuaternary.default</code> , <code>plotQuaternary.simMat</code>   |
| <code>vertices</code>    | Vector of three unique cluster names that will be used for plotting.<br>Or a named list that groups clusters as three terminal vertices. There must not be any overlap between groups.  |
| <code>features</code>    | Valid matrix row subsetting index to select features for similarity calculation. Default NULL uses all available features.  |
| <code>byCluster</code>   | Default NULL to generate one plot with all cells. Set "all" to split cells in plot by cluster and returns a list of subplots for each cluster as well as the plot including all cells. Otherwise, a vector of cluster names to generate a list of subplots for the specified clusters.  |
| <code>veloGraph</code>   | Cell x cell <code>dgMatrix</code> object containing velocity information. Shows velocity grid-arrow layer when specified. Default NULL does not show velocity.  |
| <code>method</code>      | Similarity calculation method. Default "euclidean". Choose from "euclidean", "cosine", "pearson", "spearman".   |

**force** Whether to force calculate the similarity when more than 500 features are detected, which is generally not recommended. Default FALSE.

**sigma** Gaussian kernel parameter that controls the effect of variance. Only effective when using a distance metric (i.e. method is "euclidian" or "cosine"). Larger values tighten the dot spreading on figure. Default 0.05.

**scale** Whether to min-max scale the distance matrix by clusters. Default TRUE.

**dotColor** Character vector of color codes. When dotColorBy is NULL, use one or as many colors as the number of cells. If dotColorBy is categorical, specify as many colors as the number of categories in dotColorBy or ggplot2 categorical color palette is used by default. If dotColorBy is continuous, specify together with breaks argument.

**palette** Color palette to use when dotColorBy is given. Default "D" (viridis) for continuous value and ggplot2 default for categorical value. See detail for alternatives.

**direction** Sets the order of colors in the scale. Default 1 orders as palette default. If -1, the order of colors is reversed.

**breaks** Number of breaks for continuous color scale passed to non-interactive "plot3D::scatter3D" call. Default NULL.

**returnData** Logical. Whether to return similarity and aggregated velocity data if applicable instead of generating plot. Default FALSE.

**nGrid** Number of grids along the x-axis of the tetrahedron. Default 10.

**radius** Arrow length of unit velocity. Lower this when arrows point outside of the tetrahedron. Default 0.2.

**dotSize** Size of each dot. Default 0.6 for static figure, and 4 for interactive view.

**labelColors** Colors of the vertex labels. Default c("#3B4992FF", "#EE0000FF", "#008B45FF", "#631879FF") (blue, red, green and purple).

**arrowLinewidth** Arrow aesthetics. Default 1.6 for interactive view, 0.6 for static figure.

**arrowAngle, arrowLen** Arrow aesthetics passed to `TODOOOO grid::arrow`. The length of the arrow will be internally converted to unit object in inches. Default 20 and 0.1.

**edgeLinewidth** Controls the linewidth of the edges of the tetrahedron. Default 1.

**vertexLabelSize** Numeric, size of vertex text label relative to default size. Default 1.

**title** Title text of the plot. Default NULL.

**titleSize, titleColor** Setting on the main title text. Default 1, and "black".

**theta, phi** Numeric scalar. The angles defining the viewing direction. theta gives the azimuthal direction and phi the colatitude. Default 20 and 0.

**interactive** Logical. Whether to display plotly interactive view. Default TRUE.

### Value

By default, a "plotly" object. When `interactive = FALSE`, a "quatPlot" object when `byCluster` is not specified, or a "list" of "quatPlot" objects when `byCluster` is specified. When `returnData =`

TRUE, a list of similarity matrix and aggregated velocity matrix is returned.

### See Also

Other plotQuaternary: [plotQuaternary\(\)](#), [plotQuaternary.SingleCellExperiment\(\)](#)

### Examples

```
# Seurat example
if (requireNamespace("Seurat", quietly = TRUE)) {
  require(Seurat)
  srt <- CreateSeuratObject(rnaRaw)
  Idents(srt) <- rnaCluster
  gene <- selectTopFeatures(srt, vertices = c("OS", "RE", "CH", "ORT"))
  plotQuaternary(srt, features = gene,
                 vertices = c("OS", "RE", "CH", "ORT"))
}
```

---

`plotQuaternary.simMat` *Create quaternary simplex plot with pre-calculated similarity matrix*

---

### Description

Create quaternary simplex plot with pre-calculated similarity matrix

### Usage

```
## S3 method for class 'simMat'
plotQuaternary(
  x,
  veloMat = NULL,
  nGrid = 10,
  radius = 0.2,
  dotSize = NULL,
  colorArg = NULL,
  labelColors = c("#3B4992FF", "#EE0000FF", "#008B45FF", "#631879FF"),
  arrowLinewidth = NULL,
  arrowAngle = 20,
  arrowLen = 0.1,
  vertexLabelSize = NULL,
  edgeLinewidth = 1,
  title = NULL,
  titleSize = 1,
  titleColor = "black",
  theta = 20,
  phi = 0,
  interactive = TRUE,
  ...
)
```

**Arguments**

|                       |  |
|-----------------------|--|
| x                     | simMat object, n cells by 4 vertices, each row summing to 1.   |
| veloMat               | Aggregated velocity matrix. Output of aggrVeloGraph.   |
| nGrid                 | Number of grids along the x-axis of the tetrahedron. Default 10.   |
| radius                | Arrow length of unit velocity. Lower this when arrows point outside of the tetrahedron. Default 0.2.   |
| dotSize               | Size of each dot. Default 0.6 for static figure, and 4 for interactive view.   |
| colorArg              | A "colorArg" object, internally prepared by <code>plotQuaternary.default</code> . Default NULL.  |
| labelColors           | Colors of the vertex labels. Default <code>c("#3B4992FF", "#EE0000FF", "#008B45FF", "#631879FF")</code> (blue, red, green and purple).                           |
| arrowLinewidth        | Arrow aesthetics. Default 1.6 for interactive view, 0.6 for static figure.   |
| arrowAngle, arrowLen  | Arrow aesthetics passed to <code>TODOOOO grid::arrow</code> . The length of the arrow will be internally converted to unit object in inches. Default 20 and 0.1. |
| vertexLabelSize       | Numeric, size of vertex text label relative to default size. Default 1.  |
| edgeLinewidth         | Controls the linewidth of the edges of the tetrahedron. Default 1.   |
| title                 | Title text of the plot. Default NULL.  |
| titleSize, titleColor | Setting on the main title text. Default 1, and "black".  |
| theta, phi            | Numeric scalar. The angles defining the viewing direction. theta gives the azimuthal direction and phi the colatitude. Default 20 and 0.                         |
| interactive           | Logical. Whether to display plotly interactive view. Default TRUE.   |
| ...                   | Not used   |

**Value**

A "quatPlot" object, can be displayed by printing.

---

plotQuaternary.SingleCellExperiment

*Create quaternary simplex plot with SingleCellExperiment object*

---

**Description**

Create quaternary plots that show similarity between single cells and selected four terminals in a baricentric coordinate.

See [plotTernary](#) for more details on methodologies.

A dynamic rotating view in a GIF image file can be created with [writeQuaternaryGIF](#). Package `magick` must be installed in advance. Linux users may refer to this [installation guide](#).

**Usage**

```
## S3 method for class 'SingleCellExperiment'
plotQuaternary(
  x,
  assay.type = "counts",
  clusterVar = NULL,
  processed = FALSE,
  dotColorBy = NULL,
  legendTitle = NULL,
  ...
)
```

**Arguments**

|                          |   |
|--------------------------|---|
| <code>x</code>           | A <code>SingleCellExperiment</code> object  |
| <code>assay.type</code>  | Assay to use for calculating the similarity. Default "counts".  |
| <code>clusterVar</code>  | A variable name in <code>colData(x)</code> . Default NULL uses <code>colLabels(x)</code> .  |
| <code>processed</code>   | Logical. Whether the input matrix is already processed. TRUE will bypass internal preprocessing and input matrix will be directly used for similarity calculation. Default FALSE and raw count input is recommended. If missing in call, using <code>slot = "counts"</code> in "Seurat" method or using <code>assay.type = "counts"</code> in "SingleCellExperiment" method will force this argument to be FALSE and others for TRUE. |
| <code>dotColorBy</code>  | A vector/factor for coloring dots, can be either categorical (must be character or factor) or continuous. Default NULL.   |
| <code>legendTitle</code> | Title on the legend/colorbar. Default NULL uses "cluster" if <code>dotColorBy</code> is missing (default); user-end variable expression if <code>dotColorBy</code> is directly specified from <code>plotQuaternary.default</code> method; variable name if <code>dotColorBy</code> is specified from Seurat or <code>SingleCellExperiment</code> method.  |
| <code>...</code>         | Arguments passed on to <code>plotQuaternary.default</code> , <code>plotQuaternary.simMat</code>   |
|                          | <code>vertices</code> Vector of three unique cluster names that will be used for plotting.<br>Or a named list that groups clusters as three terminal vertices. There must not be any overlap between groups.  |
|                          | <code>features</code> Valid matrix row subsetting index to select features for similarity calculation. Default NULL uses all available features.  |
|                          | <code>byCluster</code> Default NULL to generate one plot with all cells. Set "all" to split cells in plot by cluster and returns a list of subplots for each cluster as well as the plot including all cells. Otherwise, a vector of cluster names to generate a list of subplots for the specified clusters.   |
|                          | <code>velocityGraph</code> Cell x cell <code>dgMatrix</code> object containing velocity information. Shows velocity grid-arrow layer when specified. Default NULL does not show velocity.   |
|                          | <code>method</code> Similarity calculation method. Default "euclidean". Choose from "euclidean", "cosine", "pearson", "spearman".   |
|                          | <code>force</code> Whether to force calculate the similarity when more than 500 features are detected, which is generally not recommended. Default FALSE.   |

**sigma** Gaussian kernel parameter that controls the effect of variance. Only effective when using a distance metric (i.e. `method` is "euclidian" or "cosine"). Larger values tighten the dot spreading on figure. Default 0.05.  
**scale** Whether to min-max scale the distance matrix by clusters. Default TRUE.  
**dotColor** Character vector of color codes. When `dotColorBy` is NULL, use one or as many colors as the number of cells. If `dotColorBy` is categorical, specify as many colors as the number of categories in `dotColorBy` or ggplot2 categorical color palette is used by default. If `dotColorBy` is continuous, specify together with `breaks` argument.  
**palette** Color palette to use when `dotColorBy` is given. Default "D" (viridis) for continuous value and ggplot2 default for categorical value. See detail for alternatives.  
**direction** Sets the order of colors in the scale. Default 1 orders as palette default. If -1, the order of colors is reversed.  
**breaks** Number of breaks for continuous color scale passed to non-interactive "plot3D::scatter3D" call. Default NULL.  
**returnData** Logical. Whether to return similarity and aggregated velocity data if applicable instead of generating plot. Default FALSE.  
**nGrid** Number of grids along the x-axis of the tetrahedron. Default 10.  
**radius** Arrow length of unit velocity. Lower this when arrows point outside of the tetrahedron. Default 0.2.  
**dotSize** Size of each dot. Default 0.6 for static figure, and 4 for interactive view.  
**labelColors** Colors of the vertex labels. Default `c("#3B492FF", "#EE000FF", "#008B45FF", "#631879FF")` (blue, red, green and purple).  
**arrowLinewidth** Arrow aesthetics. Default 1.6 for interactive view, 0.6 for static figure.  
**arrowAngle, arrowLen** Arrow aesthetics passed to `TODOOOO grid::arrow`. The length of the arrow will be internally converted to unit onject in inches. Default 20 and 0.1.  
**edgeLinewidth** Controls the linewidth of the edges of the tetrahedron. Default 1.  
**vertexLabelSize** Numeric, size of vertex text label relative to default size. Default 1.  
**title** Title text of the plot. Default NULL.  
**titleSize, titleColor** Setting on the main title text. Default 1, and "black".  
**theta, phi** Numeric scalar. The angles defining the viewing direction. `theta` gives the azimuthal direction and `phi` the colatitude. Default 20 and 0.  
**interactive** Logical. Whether to display plotly interactive view. Default TRUE.

### Value

By default, a "plotly" object. When `interactive = FALSE`, a "quatPlot" object when `byCluster` is not specified, or a "list" of "quatPlot" objects when `byCluster` is specified. When `returnData = TRUE`, a list of similarity matrix and aggregated velocity matrix is returned.

**See Also**

Other plotQuaternary: [plotQuaternary\(\)](#), [plotQuaternary.Seurat\(\)](#)

**Examples**

```
# SingleCellExperiment example
if (requireNamespace("SingleCellExperiment", quietly = TRUE)) {
  require(SingleCellExperiment)
  sce <- SingleCellExperiment(assays = list(counts = rnaRaw))
  colLabels(sce) <- rnaCluster
  gene <- selectTopFeatures(sce, vertices = c("OS", "RE", "CH", "ORT"))
  plotQuaternary(sce, features = gene,
                 vertices = c("OS", "RE", "CH", "ORT"))
}
```

---

plotTernary

*Create ternary simplex plots*

---

**Description**

Create ternary plots that show similarity between single cells and selected three terminals in a ternary baricentric coordinate.

**Usage**

```
plotTernary(x, ...)

## Default S3 method:
plotTernary(
  x,
  clusterVar,
  vertices,
  features = NULL,
  veloGraph = NULL,
  byCluster = NULL,
  processed = FALSE,
  method = c("euclidean", "cosine", "pearson", "spearman"),
  force = FALSE,
  sigma = 0.08,
  scale = TRUE,
  dotColorBy = NULL,
  dotColor = NULL,
  palette = "D",
  direction = 1,
  breaks = NULL,
  legendTitle = NULL,
```



```

    returnData = FALSE,
    ...
)

```

### Arguments

**x** Input data. Can be a matrix or dgCMatrix object with cells as columns, a Seurat or SingleCellExperiment object. "simMat" method takes intermediate values.

**...** Arguments passed on to `plotTernary.simMat`

**title** Title text of the plot. Default NULL.

**nGrid** Number of grids along the bottom side of the equilateral triangle. Default 10.

**radius** Arrow length of unit velocity. Lower this when arrows point outside of the coordinate. Default 0.1.

**dotSize** Dot aesthetics passed to `geom_point`. Default 0.6 when not interactive, 4 when interactive.

**dotShuffle** Whether to shuffle the order of dots being added to the plot, useful when categorical colors are used and mixing of categories is expected. Default NULL does shuffle when `dotColorBy` given is categorical and does not otherwise.

**labelColors** Colors of the axis lines and vertex labels. Default `c("#3B4992FF", "#EE0000FF", "#008B45FF")` (blue, red and green)

**vertexLabelSize** Size of vertex labels. Default 6 when not interactive, 16 when interactive.

**vertexLabelDrift** Position adjustment of the vertex labels, only applied to non-interactive view. Default 0.03.

**axisBreak** Number of breaks to be labeled along axis. Default 5.

**axisTextShow** Logical, whether to show axis text. Default TRUE.

**axisTextSize** Size of text along each axis break. Default 4 for non-interactive view, 12 for interactive view.

**axisTextDrift** Position adjustment of the axis text, only applied to non-interactive view. Default 0.01.

**gridLineAlpha** Transparency of background grid lines. Default 0.6.

**arrowLinewidth** Line width of the velocity arrows. Default 0.25 for non-interactive view, 2 for interactive view.

**arrowAngle** Controls the angle of the arrowhead, only applied to non-interactive view. Default 20.

**arrowLen** Control length in centimetre from arrow tip to arrow tail, only applied to non-interactive view. Default 0.2.

**titleSize** Size of title text. Default 14 for non-interactive view, 20 for interactive view.

**equilateral** Logical, whether to always display the triangle as equilateral. Default TRUE.

**margin** Margin allowed around of the triangle plotting region when `equilateral = TRUE`

|            |             |   |
|------------|-------------|---|
|            | interactive | Logical. Whether to display plotly interactive view. Default FALSE.   |
| clusterVar |             | A vector/factor assigning the cluster variable to each column of the matrix object. For "Seurat" method, NULL (default) for Idents(x), or a variable name in meta.data slot. For "SingleCellExperiment" method, NULL (default) for colLabels(x), or a variable name in colData slot.  |
| vertices   |             | Vector of three unique cluster names that will be used for plotting. Or a named list that groups clusters as three terminal vertices. There must not be any overlap between groups.   |
| features   |             | Valid matrix row subsetting index to select features for similarity calculation. Default NULL uses all available features.  |
| veloGraph  |             | Cell x cell dgMatrix object containing velocity information. Shows velocity grid-arrow layer when specified. Default NULL does not show velocity.   |
| byCluster  |             | Default NULL to generate one plot with all cells. Set "all" to split cells in plot by cluster and returns a list of subplots for each cluster as well as the plot including all cells. Otherwise, a vector of cluster names to generate a list of subplots for the specified clusters.  |
| processed  |             | Logical. Whether the input matrix is already processed. TRUE will bypass internal preprocessing and input matrix will be directly used for similarity calculation. Default FALSE and raw count input is recommended. If missing in call, using slot = "counts" in "Seurat" method or using assay.type = "counts" in "SingleCellExperiment" method will force this argument to be FALSE and others for TRUE. |
| method     |             | Similarity calculation method. Default "euclidean". Choose from "euclidean", "cosine", "pearson", "spearman".   |
| force      |             | Whether to force calculate the similarity when more than 500 features are detected, which is generally not recommended. Default FALSE.  |
| sigma      |             | Gaussian kernel parameter that controls the effect of variance. Only effective when using a distance metric (i.e. method is "euclidian" or "cosine"). Larger values tighten the dot spreading on figure. Default 0.08.  |
| scale      |             | Whether to min-max scale the distance matrix by clusters. Default TRUE.   |
| dotColorBy |             | A vector/factor for coloring dots, can be either categorical (must be character or factor) or continuous. Default NULL.   |
| dotColor   |             | Character vector of color codes. When dotColorBy is NULL, use one or as many colors as the number of cells. If dotColorBy is categorical, specify as many colors as the number of categories in dotColorBy or ggplot2 categorical color palette is used by default. If dotColorBy is continuous, specify together with breaks argument.   |
| palette    |             | Color palette to use when dotColorBy is given. Default "D" (viridis) for continuous value and ggplot2 default for categorical value. See detail for alternatives.   |
| direction  |             | Sets the order of colors in the scale. Default 1 orders as palette default. If -1, the order of colors is reversed.   |
| breaks     |             | Number of breaks for continuous color scale passed to non-interactive "plot3D::scatter3D" call. Default NULL.   |

|             |   |
|-------------|---|
| legendTitle | Title on the legend/colorbar. Default NULL uses "cluster" if dotColorBy is missing (default); user-end variable expression if dotColorBy is directly specified from plotQuaternary.default method; variable name if dotColorBy is specified from Seurat or SingleCellExperiment method. |
| returnData  | Logical. Whether to return similarity and aggregated velocity data if applicable instead of generating plot. Default FALSE.   |

## Details

**Argument inheritance** - For matrix/dgCMatrix ("default" method), we first calculate the similarity matrix and obtain a "simMat" object. Then the "simMat" method is internally called. For data container objects (e.g. Seurat), we obtain the correct data matrix first and then call the "default" method. The arguments inherits as the flow described above.

**The calculation of similarity matrix** - The similarity is calculated either by converting a distance metric ("euclidean" or "cosine") with Gaussian kernel, or directly computed with correlation metrics ("pearson" or "spearman"). The centroid of each terminal is obtained first, and the specified metric from each cell to each terminal is calculated. The similarity matrix (n cells by v terminals) is lastly normalized to sum to 1 for each cell, so it becomes a baricentric coordinate.

## Value

By default, a "ggplot" object when byCluster is not specified, a list of "ggplot" object when byCluster is specified. When interactive = TRUE, a "plotly" object is returned. When returnData = TRUE, a list of similarity matrix and aggregated velocity matrix is returned.

## See Also

Other plotTernary: [plotTernary.Seurat\(\)](#), [plotTernary.SingleCellExperiment\(\)](#)

## Examples

```
gene <- selectTopFeatures(rnaRaw, rnaCluster, c("OS", "RE", "CH"))
plotTernary(rnaRaw, rnaCluster, c("OS", "RE", "CH"), gene)
```

---

plotTernary.Seurat      *Create ternary simplex plot with Seurat objects*

---

## Description

Create ternary plots that show similarity between single cells and selected three terminals in a ternary baricentric coordinate.

**Usage**

```
## S3 method for class 'Seurat'
plotTernary(
  x,
  layer = "counts",
  assay = NULL,
  clusterVar = NULL,
  processed = FALSE,
  dotColorBy = NULL,
  legendTitle = NULL,
  ...
)
```

**Arguments**

|             |   |
|-------------|---|
| x           | A Seurat object   |
| layer       | Layer in the specified assay to use. Default "counts".  |
| assay       | The assay to get data from. Default NULL uses the default assay.  |
| clusterVar  | A variable name in meta.data (x[[[]]). Default NULL uses Idents(x).   |
| processed   | Logical. Whether the input matrix is already processed. TRUE will bypass internal preprocessing and input matrix will be directly used for similarity calculation. Default FALSE and raw count input is recommended. If missing in call, using slot = "counts" in "Seurat" method or using assay.type = "counts" in "SingleCellExperiment" method will force this argument to be FALSE and others for TRUE. |
| dotColorBy  | A vector/factor for coloring dots, can be either categorical (must be character or factor) or continuous. Default NULL.   |
| legendTitle | Title on the legend/colorbar. Default NULL uses "cluster" if dotColorBy is missing (default); user-end variable expression if dotColorBy is directly specified from plotQuaternary.default method; variable name if dotColorBy is specified from Seurat or SingleCellExperiment method.   |
| ...         | Arguments passed on to <a href="#">plotTernary.default</a> , <a href="#">plotTernary.simMat</a>   |
| vertices    | Vector of three unique cluster names that will be used for plotting.<br>Or a named list that groups clusters as three terminal vertices. There must not be any overlap between groups.  |
| features    | Valid matrix row subsetting index to select features for similarity calculation. Default NULL uses all available features.  |
| byCluster   | Default NULL to generate one plot with all cells. Set "all" to split cells in plot by cluster and returns a list of subplots for each cluster as well as the plot including all cells. Otherwise, a vector of cluster names to generate a list of subplots for the specified clusters.  |
| veloGraph   | Cell x cell dgCMatrx object containing velocity information. Shows velocity grid-arrow layer when specified. Default NULL does not show velocity.   |
| method      | Similarity calculation method. Default "euclidean". Choose from "euclidean", "cosine", "pearson", "spearman".   |

**force** Whether to force calculate the similarity when more than 500 features are detected, which is generally not recommended. Default FALSE.

**sigma** Gaussian kernel parameter that controls the effect of variance. Only effective when using a distance metric (i.e. method is "euclidian" or "cosine"). Larger values tighten the dot spreading on figure. Default 0.08.

**scale** Whether to min-max scale the distance matrix by clusters. Default TRUE.

**dotColor** Character vector of color codes. When dotColorBy is NULL, use one or as many colors as the number of cells. If dotColorBy is categorical, specify as many colors as the number of categories in dotColorBy or ggplot2 categorical color palette is used by default. If dotColorBy is continuous, specify together with breaks argument.

**palette** Color palette to use when dotColorBy is given. Default "D" (viridis) for continuous value and ggplot2 default for categorical value. See detail for alternatives.

**direction** Sets the order of colors in the scale. Default 1 orders as palette default. If -1, the order of colors is reversed.

**breaks** Number of breaks for continuous color scale passed to non-interactive "plot3D::scatter3D" call. Default NULL.

**returnData** Logical. Whether to return similarity and aggregated velocity data if applicable instead of generating plot. Default FALSE.

**title** Title text of the plot. Default NULL.

**nGrid** Number of grids along the bottom side of the equilateral triangle. Default 10.

**radius** Arrow length of unit velocity. Lower this when arrows point outside of the coordinate. Default 0.1.

**dotSize** Dot aesthetics passed to geom\_point. Default 0.6 when not interactive, 4 when interactive.

**dotShuffle** Whether to shuffle the order of dots being added to the plot, useful when categorical colors are used and mixing of categories is expected. Default NULL does shuffle when dotColorBy given is categorical and does not otherwise.

**labelColors** Colors of the axis lines and vertex labels. Default c("#3B4992FF", "#EE0000FF", "#008B45FF") (blue, red and green)

**vertexLabelSize** Size of vertex labels. Default 6 when not interactive, 16 when interactive.

**vertexLabelDrift** Position adjustment of the vertex labels, only applied to non-interactive view. Default 0.03.

**axisBreak** Number of breaks to be labeled along axis. Default 5.

**axisTextShow** Logical, whether to show axis text. Default TRUE.

**axisTextSize** Size of text along each axis break. Default 4 for non-interactive view, 12 for interactive view.

**axisTextDrift** Position adjustment of the axis text, only applied to non-interactive view. Default 0.01.

**gridLineAlpha** Transparency of background grid lines. Default 0.6.

**arrowLinewidth** Line width of the velocity arrows. Default 0.25 for non-interactive view, 2 for interactive view.

**arrowAngle** Controls the angle of the arrowhead, only applied to non-interactive view. Default 20.

**arrowLen** Control length in centimetre from arrow tip to arrow tail, only applied to non-interactive view. Default 0.2.

**titleSize** Size of title text. Default 14 for non-interactive view, 20 for interactive view.

**equilateral** Logical, whether to always display the triangle as equilateral. Default TRUE.

**margin** Margin allowed around of the triangle plotting region when equilateral = TRUE

**interactive** Logical. Whether to display plotly interactive view. Default FALSE.

### Value

By default, a "ggplot" object when `byCluster` is not specified, a list of "ggplot" object when `byCluster` is specified. When `interactive = TRUE`, a "plotly" object is returned. When `returnData = TRUE`, a list of similarity matrix and aggregated velocity matrix is returned.

### See Also

Other plotTernary: [plotTernary\(\)](#), [plotTernary.SingleCellExperiment\(\)](#)

### Examples

```
# Seurat example
library(Seurat)
srt <- CreateSeuratObject(rnaRaw)
Idents(srt) <- rnaCluster
gene <- selectTopFeatures(srt, vertices = c("OS", "RE", "CH"))
plotTernary(srt, features = gene, vertices = c("OS", "RE", "CH"))
```

---

plotTernary.simMat      *Create quaternary simplex plot with pre-calculated similarity matrix*

---

### Description

Create quaternary simplex plot with pre-calculated similarity matrix

### Usage

```
## S3 method for class 'simMat'
plotTernary(
  x,
  title = NULL,
  veloMat = NULL,
```

```

nGrid = 10,
radius = 0.1,
dotSize = NULL,
dotShuffle = NULL,
colorArg = NULL,
labelColors = c("#3B4992FF", "#EE0000FF", "#008B45FF"),
vertexLabelSize = NULL,
vertexLabelDrift = 0.03,
axisBreak = 5,
axisTextShow = TRUE,
axisTextSize = NULL,
axisTextDrift = 0.01,
gridLineAlpha = 0.6,
arrowLinewidth = NULL,
arrowAngle = 20,
arrowLen = 0.2,
titleSize = NULL,
equilateral = TRUE,
margin = 0.1,
interactive = FALSE,
...
)

```

### Arguments

|                  |   |
|------------------|---|
| x                | simMat object, n cells by 4 vertices, each row summing to 1.  |
| title            | Title text of the plot. Default NULL.   |
| veloMat          | Aggregated velocity matrix. Output of <code>aggrVeloGraph</code> .  |
| nGrid            | Number of grids along the bottom side of the equilateral triangle. Default 10.  |
| radius           | Arrow length of unit velocity. Lower this when arrows point outside of the coordinate. Default 0.1.   |
| dotSize          | Dot aesthetics passed to <code>geom_point</code> . Default 0.6 when not interactive, 4 when interactive.  |
| dotShuffle       | Whether to shuffle the order of dots being added to the plot, useful when categorical colors are used and mixing of categories is expected. Default NULL does shuffle when <code>dotColorBy</code> given is categorical and does not otherwise. |
| colorArg         | A "colorArg" object, internally prepared by <code>plotQuaternary.default</code> . Default NULL.   |
| labelColors      | Colors of the axis lines and vertex labels. Default <code>c("#3B4992FF", "#EE0000FF", "#008B45FF")</code> (blue, red and green)   |
| vertexLabelSize  | Size of vertex labels. Default 6 when not interactive, 16 when interactive.   |
| vertexLabelDrift | Position adjustment of the vertex labels, only applied to non-interactive view. Default 0.03.   |
| axisBreak        | Number of breaks to be labeled along axis. Default 5.   |

|                             |   |
|-----------------------------|---|
| <code>axisTextShow</code>   | Logical, whether to show axis text. Default TRUE.   |
| <code>axisTextSize</code>   | Size of text along each axis break. Default 4 for non-interactive view, 12 for interactive view.              |
| <code>axisTextDrift</code>  | Position adjustment of the axis text, only applied to non-interactive view. Default 0.01.                     |
| <code>gridLineAlpha</code>  | Transparency of background grid lines. Default 0.6.   |
| <code>arrowLinewidth</code> | Line width of the velocity arrows. Default 0.25 for non-interactive view, 2 for interactive view.             |
| <code>arrowAngle</code>     | Controls the angle of the arrowhead, only applied to non-interactive view. Default 20.                        |
| <code>arrowLen</code>       | Control length in centimetre from arrow tip to arrow tail, only applied to non-interactive view. Default 0.2. |
| <code>titleSize</code>      | Size of title text. Default 14 for non-interactive view, 20 for interactive view.                             |
| <code>equilateral</code>    | Logical, whether to always display the triangle as equilateral. Default TRUE.                                 |
| <code>margin</code>         | Margin allowed around of the triangle plotting region when <code>equilateral = TRUE</code>                    |
| <code>interactive</code>    | Logical. Whether to display plotly interactive view. Default FALSE.   |
| <code>...</code>            | Not used  |

---

`plotTernary.SingleCellExperiment`

*Create ternary simplex plot with `SingleCellExperiment` objects*

---

## Description

Create ternary plots that show similarity between single cells and selected three terminals in a ternary baricentric coordinate.

## Usage

```
## S3 method for class 'SingleCellExperiment'
plotTernary(
  x,
  assay.type = "counts",
  clusterVar = NULL,
  processed = FALSE,
  dotColorBy = NULL,
  legendTitle = NULL,
  ...
)
```



**Arguments**

|                          |   |
|--------------------------|---|
| <code>x</code>           | A <code>SingleCellExperiment</code> object.   |
| <code>assay.type</code>  | Assay to use for calculating the similarity. Default "counts".  |
| <code>clusterVar</code>  | A variable name in <code>colData(x)</code> . Default NULL uses <code>colLabels(x)</code> .  |
| <code>processed</code>   | Logical. Whether the input matrix is already processed. TRUE will bypass internal preprocessing and input matrix will be directly used for similarity calculation. Default FALSE and raw count input is recommended. If missing in call, using <code>slot = "counts"</code> in "Seurat" method or using <code>assay.type = "counts"</code> in "SingleCellExperiment" method will force this argument to be FALSE and others for TRUE. |
| <code>dotColorBy</code>  | A vector/factor for coloring dots, can be either categorical (must be character or factor) or continuous. Default NULL.   |
| <code>legendTitle</code> | Title on the legend/colorbar. Default NULL uses "cluster" if <code>dotColorBy</code> is missing (default); user-end variable expression if <code>dotColorBy</code> is directly specified from <code>plotQuaternary.default</code> method; variable name if <code>dotColorBy</code> is specified from Seurat or <code>SingleCellExperiment</code> method.  |
| <code>...</code>         | Arguments passed on to <code>plotTernary.default</code> , <code>plotTernary.simMat</code>   |
| <code>vertices</code>    | Vector of three unique cluster names that will be used for plotting.<br>Or a named list that groups clusters as three terminal vertices. There must not be any overlap between groups.  |
| <code>features</code>    | Valid matrix row subsetting index to select features for similarity calculation. Default NULL uses all available features.  |
| <code>byCluster</code>   | Default NULL to generate one plot with all cells. Set "all" to split cells in plot by cluster and returns a list of subplots for each cluster as well as the plot including all cells. Otherwise, a vector of cluster names to generate a list of subplots for the specified clusters.  |
| <code>veloGraph</code>   | Cell x cell <code>dgMatrix</code> object containing velocity information. Shows velocity grid-arrow layer when specified. Default NULL does not show velocity.  |
| <code>method</code>      | Similarity calculation method. Default "euclidean". Choose from "euclidean", "cosine", "pearson", "spearman".   |
| <code>force</code>       | Whether to force calculate the similarity when more then 500 features are detected, which is generally not recommended. Default FALSE.  |
| <code>sigma</code>       | Gaussian kernel parameter that controls the effect of variance. Only effective when using a distance metric (i.e. <code>method</code> is "euclidian" or "cosine"). Larger values tighten the dot spreading on figure. Default 0.08.   |
| <code>scale</code>       | Whether to min-max scale the distance matrix by clusters. Default TRUE.   |
| <code>dotColor</code>    | Character vector of color codes. When <code>dotColorBy</code> is NULL, use one or as many colors as the number of cells. If <code>dotColorBy</code> is categorical, specify as many colors as the number of categories in <code>dotColorBy</code> or <code>ggplot2</code> categorical color palette is used by default. If <code>dotColorBy</code> is continuous, specify together with <code>breaks</code> argument.                 |
| <code>palette</code>     | Color palette to use when <code>dotColorBy</code> is given. Default "D" (viridis) for continuous value and <code>ggplot2</code> default for categorical value. See detail for alternatives.   |

**direction** Sets the order of colors in the scale. Default 1 orders as palette default. If -1, the order of colors is reversed.

**breaks** Number of breaks for continuous color scale passed to non-interactive "plot3D::scatter3D" call. Default NULL.

**returnData** Logical. Whether to return similarity and aggregated velocity data if applicable instead of generating plot. Default FALSE.

**title** Title text of the plot. Default NULL.

**nGrid** Number of grids along the bottom side of the equilateral triangle. Default 10.

**radius** Arrow length of unit velocity. Lower this when arrows point outside of the coordinate. Default 0.1.

**dotSize** Dot aesthetics passed to `geom_point`. Default 0.6 when not interactive, 4 when interactive.

**dotShuffle** Whether to shuffle the order of dots being added to the plot, useful when categorical colors are used and mixing of categories is expected. Default NULL does shuffle when `dotColorBy` given is categorical and does not otherwise.

**labelColors** Colors of the axis lines and vertex labels. Default `c("#3B4992FF", "#EE0000FF", "#008B45FF")` (blue, red and green)

**vertexLabelSize** Size of vertex labels. Default 6 when not interactive, 16 when interactive.

**vertexLabelDrift** Position adjustment of the vertex labels, only applied to non-interactive view. Default 0.03.

**axisBreak** Number of breaks to be labeled along axis. Default 5.

**axisTextShow** Logical, whether to show axis text. Default TRUE.

**axisTextSize** Size of text along each axis break. Default 4 for non-interactive view, 12 for interactive view.

**axisTextDrift** Position adjustment of the axis text, only applied to non-interactive view. Default 0.01.

**gridLineAlpha** Transparency of background grid lines. Default 0.6.

**arrowLinewidth** Line width of the velocity arrows. Default 0.25 for non-interactive view, 2 for interactive view.

**arrowAngle** Controls the angle of the arrowhead, only applied to non-interactive view. Default 20.

**arrowLen** Control length in centimetre from arrow tip to arrow tail, only applied to non-interactive view. Default 0.2.

**titleSize** Size of title text. Default 14 for non-interactive view, 20 for interactive view.

**equilateral** Logical, whether to always display the triangle as equilateral. Default TRUE.

**margin** Margin allowed around of the triangle plotting region when `equilateral = TRUE`

**interactive** Logical. Whether to display plotly interactive view. Default FALSE.

**Value**

By default, a "ggplot" object when byCluster is not specified, a list of "ggplot" object when byCluster is specified. When interactive = TRUE, a "plotly" object is returned. When returnData = TRUE, a list of similarity matrix and aggregated velocity matrix is returned.

**See Also**

Other plotTernary: [plotTernary\(\)](#), [plotTernary.Seurat\(\)](#)

**Examples**

```
# SingleCellExperiment example
library(SingleCellExperiment)
sce <- SingleCellExperiment(assays = list(counts = rnaRaw))
collLabels(sce) <- rnaCluster
gene <- selectTopFeatures(sce, vertices = c("OS", "RE", "CH"))
plotTernary(sce, features = gene, vertices = c("OS", "RE", "CH"))
```

---

print.quatPlot

*Show plist object produced with plot3D package*

---

**Description**

Show plist object produced with plot3D package

**Usage**

```
## S3 method for class 'quatPlot'
print(x, ...)
```

**Arguments**

x                    quatPlot object, returned by [plotQuaternary](#) when interactive = FALSE.  
...                   Graphic parameters passed to [plot](#). mar is pre-specified.

**Value**

No return value. It displays the plot described in a 'plist' object returned by [plotQuaternary](#), internally created by package 'plot3D'.

## Examples

```
gene <- selectTopFeatures(  
  x = rnaRaw,  
  clusterVar = rnaCluster,  
  vertices = c("RE", "OS", "CH", "ORT")  
)  
quat <- plotQuaternary(  
  x = rnaRaw,  
  clusterVar = rnaCluster,  
  vertices = c("RE", "OS", "CH", "ORT"),  
  features = gene,  
  interactive = FALSE  
)  
quat; print(quat)
```

---

|                  |  |
|------------------|--|
| readH5ADObsNames | <i>Extract 'adata.obs_names' from an H5AD file</i> |
|------------------|--|

---

## Description

It frequently happens that velocity analyses stored in H5AD files do not contain the full raw count data suggested for CytoSimplex visualization. Extracting the cell IDs (e.g. barcodes) helps matching the velocity data to raw count data imported from other sources.

## Usage

```
readH5ADObsNames(filename)
```

## Arguments

filename      File path to the H5AD file.

## Value

A character vector of cell IDs.

## See Also

Other H5AD-reader: [readH5ADObsVar\(\)](#), [readH5ADUnsSpMat\(\)](#), [readVelocitytoLoom\(\)](#)

## Examples

```
## Not run:  
h5adFile <- "path/to/analysis.h5ad"  
cellIDs <- readH5ADObsNames(h5adFile)  
  
## End(Not run)
```

---

|                |   |
|----------------|---|
| readH5ADObsVar | <i>Extract a variable from adata.obs stored in an H5AD file</i> |
|----------------|---|

---

## Description

Primarily designed for fetching the annotation used for visualization.

## Usage

```
readH5ADObsVar(filename, obsKey, named = TRUE, categoricalAsFactor = TRUE)
```

## Arguments

|                     |   |
|---------------------|---|
| filename            | File path to the H5AD file.   |
| obsKey              | The variable name to extract, must use only one character string.   |
| named               | Logical, whether to name the vector with cell IDs that came from <code>adata.obs_names</code> . Default TRUE. |
| categoricalAsFactor | Logical, whether to convert categorical variables to factors. Default TRUE.                                   |

## Value

A vector of the extracted variable, or a factor if the variable is encoded to be categorical and `categoricalAsFactor = TRUE`.

## See Also

Other H5AD-reader: [readH5ADObsNames\(\)](#), [readH5ADUnsSpMat\(\)](#), [readVelocityLoom\(\)](#)

## Examples

```
## Not run:  
h5adFile <- "path/to/analysis.h5ad"  
cluster <- readH5ADObsVar(h5adFile, "leiden")  
  
## End(Not run)
```

---

readH5ADUnsSpMat      *Extract a sparse matrix from adata.uns stored in an H5AD file*

---

### Description

Primarily designed for fetching the velocity data presented as a cell-cell transition graph.

### Usage

```
readH5ADUnsSpMat(filename, unsKey)
```

### Arguments

|          |   |
|----------|---|
| filename | File path to the H5AD file.                                       |
| unsKey   | The adata.uns key to extract, must use only one character string. |

### Value

A CSC-matrix of "dgCMatrix" class

### See Also

Other H5AD-reader: [readH5ADObsNames\(\)](#), [readH5ADObsVar\(\)](#), [readVelocitytoLoom\(\)](#)

### Examples

```
## Not run:  
h5adFile <- "path/to/analysis.h5ad"  
velo <- readH5ADUnsSpMat(h5adFile, "velo_s_norm_graph")  
  
## End(Not run)
```

---

readVelocitytoLoom      *Extract the raw counts from a LOOM file*

---

### Description

This function is primarily designed for fetching the raw count data from a LOOM file, output by **Velocity**. We by default use the spliced counts.

**Usage**

```
readVelocityLoom(
  filename,
  matrixPath = "layers/spliced",
  cellID = "CellID",
  featureID = "Gene",
  chunkSize = 1000
)
```

**Arguments**

|            |   |
|------------|---|
| filename   | File path to the LOOM file.   |
| matrixPath | A path in the LOOM file to the matrix to extract, following the inner HDF5 structure. Default "layers/spliced". See Details.              |
| cellID     | The name of the cell ID column in the LOOM column-attributes. The same thing as argument obs_names of scanpy.read_loom. Default "CellID". |
| featureID  | The name of the feature ID column in the LOOM row-attributes. The same thing as argument var_names of scanpy.read_loom. Default "Gene".   |
| chunkSize  | The maximum size of the chunk to load the matrix. Default 1000.   |

**Details**

The velocity output LOOM file is HDF5 based and is roughly organized as follows:

- "matrix": The whole raw counts, which is the sum of spliced, unspliced and ambiguous counts.
- layers: A group like a folder
  - "layers/spliced": The spliced counts.
  - "layers/unspliced": The unspliced counts.
  - "layers/ambiguous": The ambiguous counts.

An AnnData object created with Scanpy by default loads the data with a different structure, so that all the four matrices are accessible in `adata.layers` and set one of them (by default "layers/spliced") to `adata.X`.

**Value**

A sparse matrix of class "dgCMatrix", with cells as columns and genes as rows.

**See Also**

Other H5AD-reader: [readH5ADObsNames\(\)](#), [readH5ADObsVar\(\)](#), [readH5ADUnsSpMat\(\)](#)

**Examples**

```
## Not run:  
loomFile <- "velocity/out/analysis.loom"  
rawCounts <- readVelocityLoom(loomFile)  
  
## End(Not run)
```

---

rnaCluster

*Major cell type annotation of the example mouse bone marrow data*

---

**Description**

Major cell type annotation of the example mouse bone marrow data

**Usage**

```
rnaCluster
```

**Format**

factor object

**Source**

<https://www.nature.com/articles/s41467-023-38034-2>

**References**

Matsushita, Y., Liu, J., Chu, A.K.Y. et al. Bone marrow endosteal stem cells dictate active osteogenesis and aggressive tumorigenesis. *Nat Commun* 14, 2383 (2023).

---

rnaRaw

*Mouse bone marrow scRNAseq example data*

---

**Description**

Mouse bone marrow scRNAseq example data

**Usage**

```
rnaRaw
```

**Format**

[dgCMatrx-class](#) object



**Source**

<https://www.nature.com/articles/s41467-023-38034-2>

**References**

Matsushita, Y., Liu, J., Chu, A.K.Y. et al. Bone marrow endosteal stem cells dictate active osteogenesis and aggressive tumorigenesis. *Nat Commun* 14, 2383 (2023).

---

rnaVelo

*Velocity graph of the example mouse bone marrow data*

---

**Description**

Velocity graph of the example mouse bone marrow data

**Usage**

rnaVelo

**Format**

[dgCMatrx-class](#) object

**Source**

<https://www.nature.com/articles/s41467-023-38034-2>

**References**

Matsushita, Y., Liu, J., Chu, A.K.Y. et al. Bone marrow endosteal stem cells dictate active osteogenesis and aggressive tumorigenesis. *Nat Commun* 14, 2383 (2023).

---

selectTopFeatures

*Pick top differentially presented features for similarity calculation*

---

**Description**

Performs wilcoxon rank-sum test on input matrix. While `clusterVar` and `vertices` together defines the groups of cells to be set as terminals of the simplex, this function will test each of these groups against the rest of the cells. The U-Statistics (`statistic`), p-value (`pval`) and adjusted p-value (`padj`), together with average presence in group (`avgExpr`), log fold-change (`logFC`), AUC (`auc`), percentage in group (`pct_in`) and percentage out of group (`pct_out`) will be calculated. Set `returnStats = TRUE` to return the full statistics table.

Top features are selected by sorting primarily on adjusted p-value, and secondarily on log fold-change, after filtering for up-regulated features.

**Usage**

```
selectTopFeatures(x, clusterVar, vertices, ...)
```

```
## Default S3 method:
selectTopFeatures(
  x,
  clusterVar,
  vertices,
  nTop = 30,
  processed = FALSE,
  lfcThresh = 0.1,
  returnStats = FALSE,
  ...
)
```

```
## S3 method for class 'Seurat'
selectTopFeatures(
  x,
  clusterVar = NULL,
  vertices,
  assay = NULL,
  layer = "counts",
  processed = FALSE,
  ...
)
```

```
## S3 method for class 'SingleCellExperiment'
selectTopFeatures(
  x,
  clusterVar = NULL,
  vertices,
  assay.type = "counts",
  processed = FALSE,
  ...
)
```

**Arguments**

|                         |  |
|-------------------------|--|
| <code>x</code>          | Dense or sparse matrix, observation per column. Preferably a raw count matrix. Alternatively, a Seurat object or a SingleCellExperiment object.  |
| <code>clusterVar</code> | A vector/factor assigning the cluster variable to each column of the matrix object. For "Seurat" method, NULL (default) for <code>Idents(x)</code> , or a variable name in <code>meta.data</code> slot. For "SingleCellExperiment" method, NULL (default) for <code>colLabels(x)</code> , or a variable name in <code>colData</code> slot. |
| <code>vertices</code>   | Vector of cluster names that will be used for plotting. Or a named list that groups clusters as a terminal vertex. There must not be any overlap between groups.   |
| <code>...</code>        | Arguments passed to methods.   |

|             |  |
|-------------|--|
| nTop        | Number of top differentially presented features per terminal. Default 30.  |
| processed   | Logical. Whether the input matrix is already processed. TRUE will bypass internal preprocessing and input matrix will be directly used for rank-sum calculation. Default FALSE and raw count input is recommended. |
| lfcThresh   | Threshold on log fold-change to identify up-regulated features. Default 0.1.   |
| returnStats | Logical. Whether to return the full statistics table rather than returning the selected genes. Default FALSE   |
| assay       | Assay name of the Seurat object to be used. Default NULL.  |
| layer       | For "Seurat" method, which layer of the assay to be used. Default "counts".  |
| assay.type  | Assay name of the SingleCellExperiment object to be used. Default "counts".  |

### Value

When returnStats = FALSE (default), a character vector of at most length(unique(vertices))\*nTop feature names. When returnStats = TRUE, a data.frame of wilcoxon rank sum test statistics.

### Examples

```
selectTopFeatures(rnaRaw, rnaCluster, c("OS", "RE"))

# Seurat example
library(Seurat)
srt <- CreateSeuratObject(rnaRaw)
Idents(srt) <- rnaCluster
gene <- selectTopFeatures(srt, vertices = c("OS", "RE"))

# SingleCellExperiment example
library(SingleCellExperiment)
sce <- SingleCellExperiment(assays = list(counts = rnaRaw))
collabels(sce) <- rnaCluster
gene <- selectTopFeatures(sce, vertices = c("OS", "RE"))
```

---

writeQuaternaryGIF      *Create GIF image for dynamic rotating view of 3D quaternary simplex plot*

---

### Description

Create GIF image for dynamic rotating view of 3D quaternary simplex plot

**Usage**

```
writeQuaternaryGIF(
  x,
  ...,
  cluster = NULL,
  filename = NULL,
  fps = 10,
  degreePerFrame = 10,
  width = 5,
  height = 5,
  res = 100
)
```

**Arguments**

|                    |  |
|--------------------|--|
| x                  | Input object that <code>plotQuaternary</code> accepts.   |
| ...                | All other arguments needed for <code>plotQuaternary</code> . Must be specified with exact argument names instead of a positional manner. |
| cluster            | One cluster that exists in <code>clusterVar</code> if users need to view the plot for a specific group. Default NULL plot all cells.     |
| filename           | Output GIF image file path. Default NULL does not write to file.   |
| fps                | Number of frame per second, must be a factor of 100. Default 10.   |
| degreePerFrame     | Number of degree that the tetrahedron is rotated per frame. Default 10.  |
| width, height, res | <code>grDevices::png</code> parameters to set figure size and resolution. Width and Height are in inches. Default 5, 5, 100.             |

**Value**

A object of class `magick-image` that can be shown in the Viewer panel in RStudio or equivalent display device. If `filename` is specified, the GIF image will be written to the file path.

**Examples**

```
gene <- selectTopFeatures(rnaRaw, rnaCluster, c("RE", "OS", "CH", "ORT"))

writeQuaternaryGIF(rnaRaw, clusterVar = rnaCluster, features = gene,
  vertices = c("RE", "OS", "CH", "ORT"),
  gifPath = tempfile(fileext = ".gif"))
```

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