

# Package ‘tricycle’

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**Type** Package

**Title** tricycle: Transferable Representation and Inference of cell cycle

**Version** 1.19.0

**Description** The package contains functions to infer and visualize cell cycle process using Single Cell RNASeq data. It exploits the idea of transfer learning, projecting new data to the previous learned biologically interpretable space. We provide a pre-learned cell cycle space, which could be used to infer cell cycle time of human and mouse single cell samples. In addition, we also offer functions to visualize cell cycle time on different embeddings and functions to build new reference.

**Depends** R (>= 4.0), SingleCellExperiment

**Imports** methods, circular, ggplot2, ggnewscale, AnnotationDbi, scater, GenomicRanges, IRanges, S4Vectors, scattermore, dplyr, RColorBrewer, grDevices, stats, SummarizedExperiment, utils

**Suggests** testthat (>= 3.0.0), BiocStyle, knitr, rmarkdown, CircStats, cowplot, htmltools, Seurat, org.Hs.eg.db, org.Mm.eg.db

**License** GPL-3

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circle\_scale\_legend *Get the cyclic legend*

---

## Description

This function is a helper function to create the cyclic ggplot color legend.

## Usage

```
circle_scale_legend(hue.colors = c("#2E22EA", "#9E3DFB", "#F86BE2", "#FCCE7B", "#C4E416", "#4BBA0F", "#447D87", "#2C24E9"),
  hue.n = 500, alpha = 0.6, y.inner = 1.5, y.outer = 3, y.text = 3.8, ymax = 4.5, text.size = 3, addStageLabel = FALSE,
  G1.pos = 0, S.pos = 2.2, G2M.pos = 3.9)
```

## Arguments

hue.colors	The string vector gives the cyclic colors. The first color should look very similar to the last one. Default: c("#2E22EA", "#9E3DFB", "#F86BE2", "#FCCE7B", "#C4E416", "#4BBA0F", "#447D87", "#2C24E9")
hue.n	The number of breaks of color scheme. Default: 500
alpha	The alpha value (transparency). Default: 0.6
y.inner	The radius of inner circle of the donut. Default: 1.5
y.outer	The radius of outer circle of the donut. Default: 3

<code>y.text</code>	The radius of text position. Default: 3.8
<code>y.max</code>	The value control the border of the legend. Default: 4.5
<code>text.size</code>	The size of the text Default: 3
<code>addStageLabel</code>	Whether to add approximate discrete stage labels. Default: FALSE
<code>G1.pos</code>	Approximate radius value of G1 label position. Default: 0
<code>S.pos</code>	Approximate radius value of S label position. Default: 2.2
<code>G2M.pos</code>	Approximate radius value of G2M label position. Default: 3.9

### Details

The function will make a donut shape to serve as the cyclic color legend. The arguments should match the argument used in [plot\\_emb\\_circle\\_scale](#).

### Value

A ggplot object

### Author(s)

Shijie C. Zheng

### Examples

```
circle_scale_legend()
```

---

<code>diagnose_totalumi</code>	<i>Diagnostic function for UMI based datasets</i>
--------------------------------	---

---

### Description

The function will fit loess line for total UMIs numbers over cell cycle position to diagnose non-fitting data, of which cells are not cycling.

### Arguments

<code>theta.v</code>	The cell cycle position - a numeric vector with range between 0 to 2pi.
<code>totalumis</code>	The total UMIs number for each cell (without log2 transformation) - a numeric vector with the same length as <code>theta.v</code> .
<code>span</code>	The parameter $\alpha$ which controls the degree of smoothing. See <a href="#">loess</a> . Default: 0.3
<code>length.out</code>	The number of data points on the fitted lines to be output in the prediction <code>data.frame</code> . Default: 200
<code>plot</code>	If TRUE, a ggplot scatter plot will be included in the output list. The figure will plot $\log_2(\text{totalumis}) \sim \text{theta.v}$ with points and the fitted <a href="#">loess</a> line. Default: TRUE

<code>fig.title</code>	The title of the figure. Default: NULL
<code>point.size</code>	The size of the point in scatter plot used by <code>geom_scattermore</code> . Default: 2.1
<code>point.alpha</code>	The alpha value (transparency) of the point in scatter plot used by <code>geom_scattermore</code> . Default: 0.6
<code>line.size</code>	The size of the fitted line, used by <code>geom_path</code> . Default: 0.8
<code>line.alpha</code>	The alpha value (transparency) of the fitted line, used by <code>geom_path</code> . Default: 0.8
<code>x_lab</code>	Title of x-axis. Default: " $\theta$ "
<code>y_lab</code>	Title of y-axis. Default: "log2(totalumis)"
<code>...</code>	Other arguments input to <code>loess</code> .

### Details

This function fit a loess line between cell cycle position and log2 transformed total UMI number, as described in `fit_periodic_loess`. If almost all cells are not cycling in a dataset, the estimated cell cycle positions might be incorrect due to the shifted embedding center. Using the fact that the cell should have highest total UMI number at the end of S phase and almost half of that highest total UMI number at M phase, we could detect those datasets which should be analysed and interpreted carefully when using tricycle package. For such problematic datasets, the default embedding center (0, 0) could lead to wrong inference. Thus, We don't recommend using cell cycle position values if you get warnings from the `diagnose_totalumi` function.

### Value

A diagnostic message and a list with the following elements:

- `fitted` - The fitted values on the loess line. A vector of the length of `y`.
- `residual` - The residual values from the fitted loess line, i.e. `y - y.fit`. A vector of the length of `y`.
- `pred.df` - The prediction data frame by uniformly sampling `theta` from  $0 - 2\pi$ . Names of variables: `x` and `y`. The number of rows equals to `length.out`.
- `loess.o` - The fitted loess object.
- `rsquared` - The coefficient of determination  $R^2$ . Calculated as  $1 - \text{residual sum of squares} / \text{total sum of squares}$ .
- `fig` - When `plot` is TRUE, a ggplot scatter plot object will be returned with other items.

### Author(s)

Shijie C. Zheng

### See Also

[fit\\_periodic\\_loess](#).

**Examples**

```
data(neurosphere_example, package = "tricycle")
neurosphere_example <- estimate_cycle_position(neurosphere_example)
diagnose.l <- diagnose_totalumi(neurosphere_example$tricyclePosition,
  neurosphere_example$TotalUMIs, plot = TRUE)
```

---

```
estimate_cycle_position
```

*Assign cell cycle position*

---

**Description**

Assign cell cycle position by the angle formed by PC1 and PC2 in the cell cycle space. If the cell cycle projection does not exist, the function will project the data.

**Usage**

```
estimate_cycle_position(
  x,
  exprs_values = "logcounts",
  dimred = "tricycleEmbedding",
  center.pc1 = 0,
  center.pc2 = 0,
  altexp = NULL,
  ...
)
```

**Arguments**

x	A numeric matrix of <b>log-expression</b> values where rows are features and columns are cells. Alternatively, a <a href="#">SummarizedExperiment</a> or <a href="#">SingleCellExperiment</a> containing such a matrix.
exprs_values	Integer scalar or string indicating which assay of x contains the <b>log-expression</b> values, which will be used for projection. If the projection already exists, you can ignore this value. Default: 'logcounts'
dimred	The name of reducedDims in <a href="#">SingleCellExperiment</a> ( <code>reducedDims</code> ). If the dimred already exists, it will be used to assign cell cycle position. If dimred does not exist, the projection will be calculated first by <a href="#">project_cycle_space</a> and stored with name dimred in x. Default: 'tricycleEmbedding'
center.pc1	The center of PC1 when defining the angle. Default: 0
center.pc2	The center of PC2 when defining the angle. Default: 0
altexp	String or integer scalar specifying an alternative experiment containing the <b>log-expression</b> data, which will be used for projection. If the projection is already calculated and stored in the <a href="#">SingleCellExperiment</a> as a dimred, leave this value to default NULL.

... Arguments to be used by [project\\_cycle\\_space](#). If  $x$  is a [SingleCellExperiment](#), and the projection is already in the `reducedDim` with name `dimred`. The `dimred` will be directly used to assign cell cycle position without new projection.

### Details

The function will use to assign the cell cycle position by the angle formed by the PC1 and PC2 of cell cycle projections. If the input is a numeric matrix or a [SummarizedExperiment](#), the projection will be calculated with the input `**log-expression**` values. For [SingleCellExperiment](#), the projection will also be calculated if the designated `dimred` does not exist. Otherwise, the `dimred` will be directly used to assign cell cycle position. Therefore, this function is a wrapper function if the input is a [SingleCellExperiment](#). Refer to [project\\_cycle\\_space](#) to all arguments during the projection.

The estimated cell cycle position is bound between 0 and  $2\pi$ . Note that we strive to get high resolution of cell cycle state, and we think the continuous position is more appropriate when describing the cell cycle. However, to help users understand the position variable, we also note that users can approximately relate  $0.5\pi$  to be the start of S stage,  $\pi$  to be the start of G2M stage,  $1.5\pi$  to be the middle of M stage, and  $1.75\pi-0.25\pi$  to be G1/G0 stage.

### Value

If the input is a numeric matrix, the cell cycle position - a numeric vector bound between  $0 \sim 2\pi$  with the same length as the number of input columns will be returned.

If the input is [SummarizedExperiment](#), the original [SummarizedExperiment](#) with cell cycle position stored in `colData` with name `'tricyclePosition'` will be returned.

If the input is [SingleCellExperiment](#), the original [SingleCellExperiment](#) with cell cycle position stored in `colData` with name `'tricyclePosition'` will be returned and the projection will be stored in `reducedDims(..., dimred)` if it does not exist before.

### Author(s)

Shijie C. Zheng

### References

Zheng SC, et al. *Universal prediction of cell cycle position using transfer learning*. *Genome Biology* (2022) 23: 41 doi:[10.1186/s13059-021-02581-y](https://doi.org/10.1186/s13059-021-02581-y).

### See Also

[project\\_cycle\\_space](#), for projecting new data with a pre-learned reference

### Examples

```
data(neurosphere_example, package = "tricycle")
neurosphere_example <- estimate_cycle_position(neurosphere_example)
reducedDimNames(neurosphere_example)
plot(reducedDim(neurosphere_example, "tricycleEmbedding"))
plot(neurosphere_example$tricyclePosition,
      reducedDim(neurosphere_example, "tricycleEmbedding")[, 1])
```

```
plot(neurosphere_example$tricyclePosition,
     reducedDim(neurosphere_example, "tricycleEmbedding")[, 2])
```

---

```
estimate_Schwabe_stage
```

*Assign cell cycle stages using Schwabe method*

---

## Description

The function is a re-implementation of cell cycle stage assignment method proposed in Schwabe et al.(2020), with a little modification. The core assignment method is not designed by the authors of this package!

## Usage

```
estimate_Schwabe_stage(
  x,
  exprs_values = "logcounts",
  batch.v = NULL,
  altexp = NULL,
  cycleGene.l = NULL,
  gname = NULL,
  gname.type = c("ENSEMBL", "SYMBOL"),
  species = c("mouse", "human"),
  AnnotationDb = NULL,
  corThres = 0.2,
  tolerance = 0.3
)
```

## Arguments

x	A numeric matrix of <b>log-expression</b> values where rows are features and columns are cells. Alternatively, a <a href="#">SummarizedExperiment</a> or <a href="#">SingleCellExperiment</a> containing such a matrix.
exprs_values	Integer scalar or string indicating which assay of x contains the <b>log-expression</b> values, which will be used for projection. If the projection already exists, you can ignore this value. Default: 'logcounts'
batch.v	A string specifies which column in colData of <a href="#">SummarizedExperiment</a> or <a href="#">SingleCellExperiment</a> to use as the batch variable. Or it can be a vector, of which the number of elements equals to the number of columns of x. The 5 stage cell cycle assignments are preformed for each batch separately. No NA is permitted. Default: NULL
altexp	String or integer scalar specifying an alternative experiment containing the <b>log-expression</b> data, which will be used for projection. If the projection is already calculated and stored in the <a href="#">SingleCellExperiment</a> as a dimred, leave this value to default NULL.

cycleGene.l	A list contains the marker genes for each stage. The stage names should be included as names of the elements. If user feed custom list, they should make sure that the same gene id type for x and cycleGene.l. If not custom list is given, <a href="#">RevelioGeneList</a> will be used. Default: NULL
gname	Alternative rownames of x. If provided, this will be used to map genes within x with genes in ref.m. If not provided, the rownames of x will be used instead. Default: NULL
gname.type	The type of gene names as in gname or rownames of x. It can be either 'ENSEMBL' or 'SYMBOL'. If the user uses custom ref.m, this value will have no effect. Default: 'ENSEMBL'
species	The type of species in x. It can be either 'mouse' or 'human'. If the user uses custom cycleGene.l, this value will have no effect. Default: 'mouse'
AnnotationDb	An AnnotationDb objects. It is used to map ENSEMBL IDs to gene SYMBOLs. If no AnnotationDb object being given, the function will use <a href="#">org.Hs.eg.db</a> or <a href="#">org.Mm.eg.db</a> for human and mouse respectively.
corThres	For each batch and each stage, correlations between expression of each gene and the mean of all genes belonging to that stage will be calculated to filter the final gene list used for inference. The genes with a correlation between corThres will not be used for calculating z-scores. Default: 0.2
tolerance	For each cell, the function will compare the largest two z-scores. If the difference between those two z-scores is less than tolerance, the cell will be treated unassignable with NA value returned for that cell. Default: 0.3

## Details

The function is a re-implementation of cell cycle stage assignment method proposed in Schwabe et al.(2020), with a little modification. We include this function only for the purpose of convenience. The core assignment method is not designed by the authors of this package! Briefly, the function assigns cells to discretized cell cycle stages by comparing the z-scores calculated for each stage markers. Without cycleGene.l input, [RevelioGeneList](#) will be used. If you use this function, you should cite Schwabe et al.(2020).

## Value

If the input is a numeric matrix, the discretized cell cycle stages - a factor vector corresponding to each cell will be returned.

If the input is [SummarizedExperiment](#), the original [SummarizedExperiment](#) with the discretized cell cycle stages stored in colData with name 'CCStage' will be returned.

If the input is [SingleCellExperiment](#), the original [SingleCellExperiment](#) with the discretized cell cycle stages stored in colData with name 'CCStage' will be returned.

## Author(s)

Shijie C. Zheng

## References

Schwabe D, et al. *The transcriptome dynamics of single cells during the cell cycle*. Molecular Systems Biology (2020) 16: e9946 doi:[10.15252/msb.20209946](https://doi.org/10.15252/msb.20209946).

Zheng SC, et al. *Universal prediction of cell cycle position using transfer learning*. Genome Biology (2022) 23: 41 doi:[10.1186/s13059-021-02581-y](https://doi.org/10.1186/s13059-021-02581-y).

## Examples

```
data(neurosphere_example, package = "tricycle")
neurosphere_example <- estimate_Schwabe_stage(neurosphere_example,
  gname.type = "ENSEMBL", species = "mouse")
neurosphere_example2 <- estimate_Schwabe_stage(neurosphere_example, batch.v = "sample")
neurosphere_example3 <- estimate_Schwabe_stage(neurosphere_example,
  batch.v = neurosphere_example$sample)
neurosphere_example <- project_cycle_space(neurosphere_example)
plot(reducedDim(neurosphere_example, "tricycleEmbedding"),
  col = neurosphere_example$CCStage)
```

---

fit\_periodic\_loess      *Fit periodic loess line with circular predictor*

---

## Description

The function will fit a loess line using cell cycle position and other variables, such as expression levels of a gene or log-transformed totalUMIs numbers. The circular nature of cell cycle position is taken into account by making 3 copies inside the function. For convenience, the function will also return a scatter plot with fitted line if needed.

## Arguments

theta.v	The cell cycle position - a numeric vector with range between 0 to 2pi.
y	The response variable - a numeric vector with the same length as theta.v.
span	The parameter $\alpha$ which controls the degree of smoothing. See <a href="#">loess</a> . Default: 0.3
length.out	The number of data points on the fitted lines to be output in the prediction data.frame. Default: 200
plot	If TRUE, a ggplot scatter plot will be included in the output list. The figure will plot $y \sim \text{theta.v}$ with points and the fitted <a href="#">loess</a> line. Default: FALSE
fig.title	The title of the figure. Default: NULL
point.size	The size of the point in scatter plot used by <a href="#">geom_scattermore</a> . Default: 2.1
point.alpha	The alpha value (transparency) of the point in scatter plot used by <a href="#">geom_scattermore</a> . Default: 0.6
line.size	The size of the fitted line, used by <a href="#">geom_path</a> . Default: 0.8
line.alpha	The alpha value (transparency) of the fitted line, used by <a href="#">geom_path</a> . Default: 0.8

<code>color.vars</code>	Optional. A vector of categorical variable of the same length of <code>theta.v</code> , and it will be used to color points in figure. Default: NULL
<code>color.name</code>	The name of the color variables. Used as the name for legend. Default: NULL
<code>x_lab</code>	Title of x-axis. Default: " $\theta$ "
<code>y_lab</code>	Title of y-axis. Default: "y"
<code>hue.colors</code>	The string vector gives custom colors. If not given, the default <a href="#">scale_color_discrete</a> will be used. Default: NULL
<code>...</code>	Other arguments input to <a href="#">loess</a> .

### Details

This function fit a normal loess line, but take the circularity of cell cycle position into account by making `theta.v` 3 periods (`c(theta.v - 2 * pi, theta.v, theta.v + 2 * pi)`) and repeating `y` 3 times. Only the fitted values corresponding to original `theta.v` will be returned. For convenience, the function will also return a scatter plot with fitted line if needed. Or user can use `pred.df` to visualize the loess line themselves.

### Value

A list with the following elements:

- `fitted` - The fitted values on the loess line. A vector of the length of `y`.
- `residual` - The residual values from the fitted loess line, i.e. `y - y.fit`. A vector of the length of `y`.
- `pred.df` - The prediction data frame by uniformly sampling `theta` from  $0 - 2\pi$ . Names of variables: `x` and `y`. The number of rows equals to `length.out`.
- `loess.o` - The fitted loess object.
- `rsquared` - The coefficient of determination  $R^2$ . Calculated as  $1 - \text{residual sum of squares} / \text{total sum of squares}$ .
- `fig` - When `plot` is TRUE, a ggplot scatter plot object will be returned with other items.

### Author(s)

Shijie C. Zheng

### See Also

[estimate\\_cycle\\_position](#), for inferring cell cycle position.

### Examples

```
data(neurosphere_example, package = "tricycle")
neurosphere_example <- estimate_cycle_position(neurosphere_example)
top2a.idx <- which(rowData(neurosphere_example)$Gene == "Top2a")
fit.l <- fit_periodic_loess(neurosphere_example$tricyclePosition,
  assay(neurosphere_example, "logcounts")[top2a.idx, ], plot = TRUE)
fit.l$fig
```

---

neuroRef	<i>Pre-learned reference projection matrix from the Neurosphere dataset</i>
----------	---

---

**Description**

Default reference projection matrix learned from the Neurosphere dataset.

**Usage**

```
data(neuroRef)
```

**Format**

An object of class “data.frame”, with 5 variables. Normally, user won’t call this data directly as it will be automatically used in [project\\_cycle\\_space](#) if no custom reference projection matrix is provided. Each row is gene, and rotation scores for PC1 and PC2, mouse ENSEMBL IDs, and mouse gene SYMBOLs are included. The ‘SYMBOL’ values are just the upper-case ‘symbol’ values.

**References**

Zheng SC, et al. *Universal prediction of cell cycle position using transfer learning*. *Genome Biology* (2022) 23: 41 doi:[10.1186/s13059-021-02581-y](https://doi.org/10.1186/s13059-021-02581-y).

**Examples**

```
data(neuroRef)
```

---

neurosphere_example	<i>Example SingleCellExperiment dataset</i>
---------------------	---

---

**Description**

This is a subset of mouse Neurosphere data. 200 cells from sample AX1 and AX2 were randomly sampled from the full data. All genes in the GO cell cycle gene list and [RevelioGeneList](#) as well as other random 573 genes were included.

**Usage**

```
data(neurosphere_example)
```

**Format**

A [SingleCellExperiment](#) object of 1500 genes and 400 cells.

**Examples**

```
data(neurosphere_example)
```

---

plot\_ccposition\_den *Plot cell cycle position kernel density stratified by a factor*

---

## Description

The function will compute and plot cell cycle position kernel density.

## Arguments

theta.v	The cell cycle position - a numeric vector with range between 0 to 2pi.
color_var.v	A factor variable to stratify theta.v, such as samples or 'CCStage'. The length of it should equal to the length of theta.v.
color_name	The name of the color_var.v to be used in the legend.
palette.v	A string vector to give the color names. It should has the length of the number of levels of color_var.v. If not given, the 'Set1' palette will be used. (See <a href="#">display.brewer.all</a> ) If the number of levels of color_var.v is greater than 8, only the top 8 levels of most cell will be shown. You can show them all by feeding enough colors in palette.v. Default: NULL
fig.title	The title of the figure. Default: NULL
type	It can be either of 'linear' or 'circular'. 'linear' corresponds to Cartesian coordinate system and 'circular' for polar coordinate system. Default: 'linear'
bw	The smoothing bandwidth to be used. It is equal to the concentration parameter of the von Mises distribution. See <a href="#">density.circular</a> . Default: 30
weighted	Whether the density should be weighted on the percentage of each level of color_var.v. Default: FALSE
line.size	The size of the line used by <a href="#">geom_path</a> . Default: 0.5
line.alpha	The alpha value of the line used by <a href="#">geom_path</a> . Default: 0.5
addRug	Whether to add rug on the bottom of the linear density plot or an inner circle on the circular plot to show the continuous scale of theta. Default: TRUE
RugPalette.v	The palette used for the rug plot. If not given, it will used the same default palette as in <a href="#">plot_emb_circle_scale</a> .
...	Other arguments accepted by <a href="#">geom_path</a> .

## Details

The function first estimates kernel density using the von Mises distribution. Then, it plots out the density in the polar coordinate system or Cartesian coordinate system. Different colors represents different levels of color\_var.v and the dashed black line is the marginal distribution of all cells.

## Value

A ggplot object

**Author(s)**

Shijie C. Zheng

**See Also**

[estimate\\_Schwabe\\_stage](#), for inferring 5 stages of cell cycle

**Examples**

```
data(neurosphere_example, package = "tricycle")
neurosphere_example <- estimate_cycle_position(neurosphere_example)
plot_ccposition_den(neurosphere_example$tricyclePosition, neurosphere_example$sample, "sample")

neurosphere_example <- estimate_Schwabe_stage(neurosphere_example,
  gname.type = "ENSEMBL", species = "mouse")
plot_ccposition_den(neurosphere_example$tricyclePosition, neurosphere_example$CCStage, "CCStage")
```

---

plot\_emb\_circle\_scale *Plot embedding with cyclic cell cycle position*

---

**Description**

Generate scat plot of embedding with cyclic cell cycle position or other cyclic variables

**Usage**

```
plot_emb_circle_scale(
  sce.o,
  color_by = "tricyclePosition",
  facet_by = NULL,
  dimred = 1,
  dim = seq_len(2),
  fig.title = NULL,
  point.size = 2.1,
  point.alpha = 0.6,
  x_lab = NULL,
  y_lab = NULL,
  hue.colors = c("#2E22EA", "#9E3DFB", "#F86BE2", "#FCCE7B", "#C4E416", "#4BBA0F",
    "#447D87", "#2C24E9"),
  hue.n = 500,
  plot.legend = FALSE
)
```

**Arguments**

sce.o	A <a href="#">SingleCellExperiment</a> contains the embedding to be plotted against.
color_by	The name of variable in colData(sce.o) to be used to show colors. Default: "tricyclePosition"
facet_by	The name of variable in colData(sce.o) to be used to facet scatter plots. If used, the function will return a list of ggplot objects. If NULL, no faceted panels will be returned. Default: NULL
dimred	The name or index of reducedDims in <a href="#">SingleCellExperiment</a> ( <a href="#">reducedDims</a> ). Default: 1
dim	The indices of dimred to be plotted. At the moment, it has to be two integers. Default: 1:2
fig.title	The title of the figure. Default: NULL
point.size	The size of the point in scatter plot used by <a href="#">geom_scattermore</a> . Default: 2.1
point.alpha	The alpha value (transparency) of the point in scatter plot used by <a href="#">geom_scattermore</a> . Default: 0.6
x_lab	Title of x-axis. If not given, the colname of dimred will be used. Default: NULL
y_lab	Title of y-axis. If not given, the colname of dimred will be used. Default: NULL
hue.colors	The string vector gives the cyclic colors. The first color should look very similar to the last one. Default: c("#2E22EA", "#9E3DFB", "#F86BE2", "#FCCE7B", "#C4E416", "#4BBA0F", "#447D87", "#2C24E9")
hue.n	The number of breaks of color scheme. Default: 500
plot.legend	Whether the legend should be plotted with the scatter plot. We recommend not to use this legend but use the cyclic legend produced by <a href="#">circle_scale_legend</a> instead. Default: FALSE

**Details**

This function help users plot embedding scatter plot colored by cyclic variables, such as cell cycle position, which is bound between 0 - 2pi. It will take a [SingleCellExperiment](#) object as input, and plot out its dimred such as PCA, UMAP, and etc with a cyclic color scheme.

**Value**

A ggplot object or a list of ggplot objects. If facet\_by is not assigned, a single ggplot plot of the scatter plot will be return, Otherwise, apart from the first scatter plot showing all cells together, other faceted scatter plots will also be given in the list.

**Author(s)**

Shijie C. Zheng

**Examples**

```
data(neurosphere_example, package = "tricycle")
neurosphere_example <- estimate_cycle_position(neurosphere_example)
plot_emb_circle_scale(neurosphere_example, point.size = 3.1, point.alpha = 0.8)
```

---

project\_cycle\_space     *Project data into the cell cycle pattern space*

---

### Description

Project mouse and human single cell RNAseq data into a cell cycle embedding by a pre-learned reference projection matrix.

### Usage

```
project_cycle_space(
  x,
  exprs_values = "logcounts",
  altexp = NULL,
  name = "tricycleEmbedding",
  ref.m = NULL,
  gname = NULL,
  gname.type = c("ENSEMBL", "SYMBOL"),
  species = c("mouse", "human"),
  AnnotationDb = NULL
)
```

### Arguments

x	A numeric matrix of <b>log-expression</b> values where rows are features and columns are cells. Alternatively, a <a href="#">SummarizedExperiment</a> or <a href="#">SingleCellExperiment</a> containing such a matrix.
exprs_values	Integer scalar or string indicating which assay of x contains the <b>log-expression</b> values. Default: 'logcounts'
altexp	String or integer scalar specifying an alternative experiment containing the input data.
name	String specifying the name to be used to store the result in the <a href="#">reducedDims</a> of the output. Default: 'tricycleEmbedding'
ref.m	A custom reference projection matrix to project the new data, where rows are features and columns are dimensions. Users need to use the same type of gname(or rownames of x) as for the ref.m. If no custom ref.m is given, the internal reference neuroRef will be used.
gname	Alternative rownames of x. If provided, this will be used to map genes within x with genes in ref.m. If not provided, the rownames of x will be used instead. Default: NULL
gname.type	The type of gene names as in gname or rownames of x. It can be either 'ENSEMBL' or 'SYMBOL'. If the user uses custom ref.m, this value will have no effect. Default: 'ENSEMBL'
species	The type of species in x. It can be either 'mouse' or 'human'. If the user uses custom ref.m, this value will have no effect. Default: 'mouse'

**AnnotationDb** An AnnotationDb objects. If the user uses the internal reference to project human data, and provide rownames in the format of Ensembl IDs, this object will be used to map Ensembl IDs to gene SYMBOLs. If no AnnotationDb object being given, the function will use [org.Hs.eg.db](#).

### Details

The function will use pre-learned cell cycle pattern to project new data to show the cell cycle progression. If the user uses internal Neurosphere reference, the expression values must be **\*\*log-transformed\*\***. Besides, we would assume the input data has been already preprocessed, library size normalized at least. The projection process is to take sum of weighted mean-centered expression of chosen genes, so the mean expression of a given gene could be affected without library size normalization.

### Value

If the input is a numeric matrix or a [SummarizedExperiment](#), a projection matrix with rows cells and column dimensions will be returned. The actual rotation matrix used to project the data is included in the attributes with name 'rotation'.

For [SingleCellExperiment](#), an updated [SingleCellExperiment](#) is returned containing projection matrix in `reducedDims(..., name)`.

### Author(s)

Shijie C. Zheng

### References

Zheng SC, et al. *Universal prediction of cell cycle position using transfer learning*. *Genome Biology* (2022) 23: 41 doi:[10.1186/s13059-021-02581-y](https://doi.org/10.1186/s13059-021-02581-y).

### See Also

[estimate\\_cycle\\_position](#), for inferring cell cycle position.

### Examples

```
data(neurosphere_example, package = "tricycle")
neurosphere_example <- project_cycle_space(neurosphere_example)
reducedDimNames(neurosphere_example)
head(reducedDim(neurosphere_example, "tricycleEmbedding"))
plot(reducedDim(neurosphere_example, "tricycleEmbedding"))
names(attributes(reducedDim(neurosphere_example, "tricycleEmbedding")))
```

---

RevelioGeneList      *5 stage cell cycle gene marker list from Revelio*

---

### Description

This 5 stage cell cycle gene marker list is directly from Revelio package. Within the list, 5 vectors corresponds to highly expressed genes at the cell cycle stage. The genes are given as the human gene SYMBOLS. This gene list is originally from the Whitfield et al.(2020).

### Usage

```
data(RevelioGeneList)
```

### Format

An list of 5 string vector. The names of the elements are the names of cell cycle stages with the order of: G1S, S, G2, G2M, MG1.

### References

Whitfield ML, et al. *Identification of Genes Periodically Expressed in the Human Cell Cycle and Their Expression in Tumors*. Molecular Biology of the Cell (2002) 13: 1977-2000 doi:[10.1091/mbc.02-02-0030](https://doi.org/10.1091/mbc.02-02-0030).

### Examples

```
data(RevelioGeneList)
```

---

run\_pca\_cc\_genes      *Run PCA on Gene Ontology cell cycle genes*

---

### Description

Run PCA on Gene Ontology cell cycle genes and get a new [SingleCellExperiment](#). User could use this function to learn new reference projection matrix.

### Arguments

sce.o	A <a href="#">SingleCellExperiment</a> contains library size normalized <b>**log-expression**</b> matrix.
gname	Alternative rownames of sce.o. If provided, this will be used to map genes within Gene Ontology cell cycle gene list. If not provided, the rownames of sce.o will be used instead. Default: NULL
exprs_values	Integer scalar or string indicating which assay of sce.o contains the <b>**log-expression**</b> values, which will be used to run PCA. Default: 'logcounts'

gname.type	The type of gene names as in gname or rownames of sce.o. It can be either 'ENSEMBL' or 'SYMBOL'. Default: 'ENSEMBL'
species	The type of species in sce.o. It can be either 'mouse' or 'human'. If the user uses custom cycleGene.l, this value will have no effect. Default: 'mouse'
AnnotationDb	An AnnotationDb objects. It is used to map ENSEMBL IDs to gene SYMBOLs. If no AnnotationDb object being given, the function will use <a href="#">org.Hs.eg.db</a> or <a href="#">org.Mm.eg.db</a> for human and mouse respectively.
ntop	The number of genes with highest variance to use when calculating PCA, as in <a href="#">calculatePCA</a> . Default: 500
ncomponents	The number of component components to obtain, as in <a href="#">calculatePCA</a> . Default: 20
name	String specifying the name to be used to store the result in the <a href="#">reducedDims</a> of the output. Default: 'PCA'

### Details

The function require an output of a [SingleCellExperiment](#) object which contains the library size normalized **log-expression** matrix. The full dataset will be subsetted to genes in the Gene Ontology cell cycle gene list (GO:0007049). The corresponding AnnotationDb object will be [org.Mm.eg.db](#) and [org.Hs.eg.db](#) for mouse and human respectively. If runSeuratBy is set, the data will be integrated to remove batch effect between samples/batches by Seurat.

User can use this function to make new reference projection matrix by getting the 'rotation' attribute in PCA results. Such as `attr(reducedDim(sce.o, 'PCA'), 'rotation')[, 1:2]`. See examples for more details.

### Value

A subset [SingleCellExperiment](#) object with only GO cell cycle genes will be return. The PCA resulting will be save in reducedDims with chosen name `reducedDims(..., name)`. If Seurat integration is performed, another reducedDims with name 'matched.'+name will also be included in the [SingleCellExperiment](#).

### Author(s)

Shijie C. Zheng

### Examples

```
data(neurosphere_example, package = "tricycle")
### Use internal NeuroRef to project and infer tricyclePosition
neurosphere_example <- estimate_cycle_position(neurosphere_example)

### Build new reference
gocc_sce.o <- run_pca_cc_genes(neurosphere_example)
new.ref <- attr(reducedDim(gocc_sce.o, "PCA"), "rotation")[, seq_len(2)]

### Use new reference to project and infer tricyclePosition
new_sce <- estimate_cycle_position(neurosphere_example, ref.m = new.ref,
```

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```
dimred = "tricycleEmbedding2")  
plot(neurosphere_example$tricyclePosition, new_sce$tricyclePosition)
```

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