Package 'SlimR'

October 8, 2025

Version 1.0.8

Title Machine Learning-Assisted, Marker-Based Tool for Single-Cell and
Spatial Transcriptomics Annotation

Description Annotates single-cell and spatial-transcriptomic (ST) data using marker datasets. Supports unified markers list ('Markers_list') creation from built-in databases (e.g., 'Cell-marker2', 'PanglaoDB', 'scIBD', 'TCellSI'), Seurat objects, or user-supplied Excel files. SlimR can predict calculate parameters by machine learning algorithms (e.g., 'Random Forest', 'Gradient Boosting', 'Support Vector Machine', 'Ensemble Learning'), and based on Markers_list, calculate gene expression of different cell types and predict annotation information and calculate corresponding AUC and annotate it, then verify it. At the same time, it can calculate gene expression corresponding to the cell type to generate a reference map for manual annotation (e.g., 'Heat Map', 'Feature Plots', 'Combined Plots'). For more details see Kabacoff (2020, ISBN:9787115420572).

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URL https://github.com/Zhaoqing-wang/SlimR

BugReports https://github.com/Zhaoqing-wang/SlimR/issues

Depends R (>= 3.5)

Imports cowplot, dplyr, ggplot2, patchwork, pheatmap, readxl, scales,
Seurat, tidyr, tools, tibble

Suggests crayon, caret, gbm, lattice

Encoding UTF-8

LazyData true

RoxygenNote 7.3.3

Date 2025-10-08

NeedsCompilation no

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Repository CRAN

Date/Publication 2025-10-08 12:00:13 UTC

Contents

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calculate_cluster_variability
                    Calculate Cluster Variability

Description

Measures the degree of separation between different cell clusters based on expression patterns.

Usage

```
calculate_cluster_variability(data.features, features)
```

Arguments

data.features	Data frame containing expression data and cluster labels
features	Feature names to include in analysis

Value

Numeric value representing cluster separation strength

See Also

Other Section_1_Functions_Use_in_Package: calculate_expression(), calculate_expression_skewness(),
calculate_probability(), estimate_batch_effect(), extract_dataset_features(), generate_training_data(),
postprocess_parameters(), predict_optimal_parameters(), train_parameter_model()

---

calculate_expression   *Counts average expression of gene set (Use in package)*

Description

Counts average expression of gene set (Use in package)

Usage

```
calculate_expression(
  object,
  features,
  assay = NULL,
  cluster_col = NULL,
  colour_low = "white",
  colour_high = "navy"
)
```

Arguments

object	Enter a Seurat object.
features	Enter one or a set of markers.
assay	Enter the assay used by the Seurat object, such as "RNA". Default parameters use "assay = NULL".
cluster_col	Enter the meta.data column in the Seurat object to be annotated, such as "seurat_cluster". Default parameters use "cluster_col = NULL".
colour_low	Color for lowest expression level. (default = "white")
colour_high	Color for highest expression level. (default = "black")

Value

Average expression genes and relatied informations in the input "Seurat" object given "cluster_col" and given "features".

See Also

Other Section_1_Functions_Use_in_Package: calculate_cluster_variability(), calculate_expression_skewness(calculate_probability(), estimate_batch_effect(), extract_dataset_features(), generate_training_data(), postprocess_parameters(), predict_optimal_parameters(), train_parameter_model()

---

calculate_expression_skewness

Calculate Expression Distribution Skewness

Description

Computes the average skewness of gene expression distributions across all features.

Usage

```
calculate_expression_skewness(expression_matrix)
```

Arguments

expression_matrix

Matrix of expression values

Value

Mean absolute skewness across all genes

See Also

Other Section_1_Functions_Use_in_Package: calculate_cluster_variability(), calculate_expression(), calculate_probability(), estimate_batch_effect(), extract_dataset_features(), generate_training_data(), postprocess_parameters(), predict_optimal_parameters(), train_parameter_model()

---

calculate_probability *Calculate gene set expression and infer probabilities with control datasets (Use in package)*

Description

Calculate gene set expression and infer probabilities with control datasets (Use in package)

Usage

```
calculate_probability(
  object,
  features,
  assay = NULL,
  cluster_col = NULL,
  min_expression = 0.1,
  specificity_weight = 3
)
```

Arguments

object	Enter a Seurat object.
features	Enter one or a set of markers.
assay	Enter the assay used by the Seurat object, such as "RNA". Default parameters use "assay = NULL".
cluster_col	Enter the meta.data column in the Seurat object to be annotated, such as "seurat_cluster". Default parameters use "cluster_col = NULL".
min_expression	The min_expression parameter defines a threshold value to determine whether a cell's expression of a feature is considered "expressed" or not. It is used to filter out low-expression cells that may contribute noise to the analysis. Default parameters use "min_expression = 0.1".
specificity_weight	
	The specificity_weight parameter controls how much the expression variability (standard deviation) of a feature within a cluster contributes to its "specificity score." It amplifies or suppresses the impact of variability in the final score calculation.Default parameters use "specificity_weight = 3".

Value

Average expression of genes in the input "Seurat" object given "cluster_col" and given "features".

See Also

Other Section_1_Functions_Use_in_Package: calculate_cluster_variability(), calculate_expression(), calculate_expression_skewness(), estimate_batch_effect(), extract_dataset_features(), generate_training_data(), postprocess_parameters(), predict_optimal_parameters(), train_parameter_model()

Cellmarker2   *Cellmarker2 dataset*

Description

A dataset containing marker genes for different cell types from Cellmarker2

Usage

```
Cellmarker2
```

Format

A data frame with 8 columns:

Details

This dataset is used to filter and create a standardized marker list. The dataset can be filtered based on species, tissue class, tissue type, cancer type, and cell type to generate a list of marker genes for specific cell types.

Source

<http://117.50.127.228/CellMarker/>

See Also

Other Section_0_Database: Cellmarker2_raw, Cellmarker2_table, Markers_list_TCellSI, Markers_list_scIBD, PanglaoDB, PanglaoDB_raw, PanglaoDB_table

---

Cellmarker2_raw   *Cellmarker2 raw dataset*

Description

A dataset containing marker genes for different cell types from Cellmarker2

Usage

```
Cellmarker2_raw
```

Format

A data frame with 20 columns contined in the Cellmarker2 database:

Details

This dataset is used to filter and create a standardized marker list. The dataset can be filtered based on species, tissue class, tissue type, cancer type, and cell type to generate a list of marker genes for specific cell types.

Source

http://117.50.127.228/CellMarker/

See Also

Other Section_0_Database: Cellmarker2, Cellmarker2_table, Markers_list_TCellSI, Markers_list_scIBD, PanglaoDB, PanglaoDB_raw, PanglaoDB_table

---

Cellmarker2_table   *Cellmarker2 table*

Description

A dataset containing marker genes for different cell types from Cellmarker2

Usage

Cellmarker2_table

Format

A list contain different types like species, tissue_class, tissue_type, cancer_type, cell_type

Details

This list is used to choose filters for creation of standardized marker list.

Source

http://117.50.127.228/CellMarker/

See Also

Other Section_0_Database: Cellmarker2, Cellmarker2_raw, Markers_list_TCellSI, Markers_list_scIBD, PanglaoDB, PanglaoDB_raw, PanglaoDB_table

---

Celltype_Annotation   *Annotate Seurat Object with SlimR Cell Type Predictions*

Description

This function assigns SlimR predicted cell types to a Seurat object based on cluster annotations, and stores the results in the meta.data slot.

Usage

```
Celltype_Annotation(
  seurat_obj,
  cluster_col,
  SlimR_anno_result,
  plot_UMAP = TRUE,
  annotation_col = "Cell_type_SlimR"
)
```

Arguments

seurat_obj	A Seurat object containing cluster information in meta.data.
cluster_col	Character string indicating the column name in meta.data that contains cluster IDs.
SlimR_anno_result	
	List generated by function Celltype_Calculate() which containing a data.frame in $Prediction_results with: 1.cluster_col (Cluster identifiers (should match cluster_col in meta.data)) 2.Predicted_cell_type (Predicted cell types for each cluster).
plot_UMAP	logical(1); if TRUE, plot the UMAP with cell type annotations.
annotation_col	The location to write in 'meta.data' that contains the predicted cell type. (default = "Cell_type_SlimR")

Value

A Seurat object with updated meta.data containing the predicted cell types.

Note

If plot_UMAP = TRUE, this function will print a UMAP plot as a side effect.

See Also

Other Section_3_Automated_Annotation_Workflow: Celltype_Calculate(), Celltype_Verification(), Parameter_Calculate()

Examples

```
## Not run:
sce <- Celltype_Annotation(seurat_obj = sce,
    cluster_col = "seurat_clusters",
    SlimR_anno_result = SlimR_anno_result,
    plot_UMAP = TRUE,
    annotation_col = "Cell_type_SlimR"
    )

## End(Not run)
```

Celltype_annotation_Cellmarker2

Uses "marker_list" from Cellmarker2 for cell annotation

Description

Uses "marker_list" from Cellmarker2 for cell annotation

Usage

```
Celltype_annotation_Cellmarker2(
  seurat_obj,
  gene_list,
  species,
  cluster_col = "seurat_clusters",
  assay = "RNA",
  save_path = NULL,
  min_counts = 1,
  colour_low = "white",
  colour_high = "navy",
  colour_low_mertic = "white",
  colour_high_mertic = "navy"
)
```

Arguments

seurat_obj	Enter the Seurat object with annotation columns such as "seurat_cluster" in meta.data to be annotated.
gene_list	Enter the standard "Marker_list" generated by the Cellmarker2 database for the SlimR package, generated by the "Markers_filter_Cellmarker2 ()" function.
species	This parameter selects the species "Human" or "Mouse" for standard gene format correction of markers entered by "Marker_list".
cluster_col	Enter annotation columns such as "seurat_cluster" in meta.data of the Seurat object to be annotated. Default parameters use "cluster_col = 'seurat_clusters'".

assay	Enter the assay used by the Seurat object, such as "RNA". Default parameters use "assay = "RNA"".
save_path	The output path of the cell annotation picture. Example parameters use "save_path = './SlimR/Celltype_annotation_Cellmarker2/'".
min_counts	The minimum number of counts of genes in "Marker_list" entered. This number represents the number of the same gene in the same species and the same location in the Cellmarker2 database used for annotation of this cell type. Default parameters use "min_counts = 1".
colour_low	Color for lowest expression level. (default = "white")
colour_high	Color for highest expression level. (default = "navy")
colour_low_mertic	
	Color for lowest mertic level. (default = "white")
colour_high_mertic	
	Color for highest mertic level. (default = "navy")

Value

The cell annotation picture is saved in "save_path".

See Also

Other Section_5_Other_Functions_Provided_by_SlimR: `Celltype_annotation_Excel()`, `Celltype_annotation_Pangla`
`Celltype_annotation_Seurat()`

Examples

```
## Not run:
Celltype_annotation_Cellmarker2(seurat_obj = sce,
    gene_list = Markers_list_Cellmarker2,
    species = "Human",
    cluster_col = "seurat_clusters",
    assay = "RNA",
    save_path = file.path(tempdir(),"SlimR_Celltype_annotation_Cellmarker2")
    colour_low = "white",
    colour_high = "navy",
    colour_low_mertic = "white",
    colour_high_mertic = "navy",
    )

## End(Not run)
```

Celltype_Annotation_Combined

Uses "marker_list" to generate combined plot for cell annotation

---

Description

Uses "marker_list" to generate combined plot for cell annotation

Usage

```
Celltype_Annotation_Combined(
  seurat_obj,
  gene_list,
  species,
  cluster_col = "seurat_clusters",
  assay = "RNA",
  save_path = NULL,
  colour_low = "white",
  colour_high = "navy"
)
```

Arguments

seurat_obj	Enter the Seurat object with annotation columns such as "seurat_cluster" in meta.data to be annotated.
gene_list	A list of cells and corresponding gene controls, the name of the list is cell type, and the first column of the list corresponds to markers. Lists can be generated using functions such as "Markers_filter_Cellmarker2 ()", "Markers_filter_PanglaoDB ()", "read_excel_markers ()", "read_seurat_markers ()", etc.
species	This parameter selects the species "Human" or "Mouse" for standard gene format correction of markers entered by "Marker_list".
cluster_col	Enter annotation columns such as "seurat_cluster" in meta.data of the Seurat object to be annotated. Default parameters use "cluster_col = 'seurat_clusters'".
assay	Enter the assay used by the Seurat object, such as "RNA". Default parameters use "assay = 'RNA'".
save_path	The output path of the cell annotation picture. Example parameters use "save_path = './SlimR/Celltype_annotation_Bar/'".
colour_low	Color for lowest expression level. (default = "white")
colour_high	Color for highest expression level. (default = "navy")

Value

The cell annotation picture is saved in "save_path".

See Also

Other Section_4_Semi_Automated_Annotation_Workflow: `Celltype_Annotation_Features()`,
`Celltype_Annotation_Heatmap()`

Examples

```
## Not run:
Celltype_Annotation_Combined(seurat_obj = sce,
    gene_list = Markers_list,
    species = "Human",
    cluster_col = "seurat_clusters",
    assay = "RNA",
    save_path = file.path(tempdir(),"SlimR_Celltype_Annotation_Combined"),
    colour_low = "white",
    colour_high = "navy"
    )

## End(Not run)
```

Celltype_annotation_Excel

Uses "marker_list" from Excel input for cell annotation

Description

Uses "marker_list" from Excel input for cell annotation

Usage

```
Celltype_annotation_Excel(
  seurat_obj,
  gene_list,
  species,
  cluster_col = "seurat_clusters",
  assay = "RNA",
  save_path = NULL,
  metric_names = NULL,
  colour_low = "white",
  colour_high = "navy",
  colour_low_mertic = "white",
  colour_high_mertic = "navy"
)
```

Arguments

seurat_obj	Enter the Seurat object with annotation columns such as "seurat_cluster" in meta.data to be annotated.
gene_list	Enter the standard "Marker_list" generated by the Excel files database for the SlimR package, generated by the "read_excel_markers()" function.
species	This parameter selects the species "Human" or "Mouse" for standard gene format correction of markers entered by "Marker_list".
cluster_col	Enter annotation columns such as "seurat_cluster" in meta.data of the Seurat object to be annotated. Default parameters use "cluster_col = "seurat_clusters"".
assay	Enter the assay used by the Seurat object, such as "RNA". Default parameters use "assay = 'RNA'".
save_path	The output path of the cell annotation picture. Example parameters use "save_path = './SlimR/Celltype_annotation_Excel/'".
metric_names	Change the row name for the input mertics, not recommended unless necessary. (NULL is used as default parameter)
colour_low	Color for lowest expression level. (default = "white")
colour_high	Color for highest expression level. (default = "navy")
colour_low_mertic	
	Color for lowest mertic level. (default = "white")
colour_high_mertic	
	Color for highest mertic level. (default = "navy")

Value

The cell annotation picture is saved in "save_path".

See Also

Other Section_5_Other_Functions_Provided_by_SlimR: `Celltype_annotation_Cellmarker2()`, `Celltype_annotation_PanglaoDB()`, `Celltype_annotation_Seurat()`

Examples

```
## Not run:
Celltype_annotation_Excel(seurat_obj = sce,
    gene_list = Markers_list_Excel,
    species = "Human",
    cluster_col = "seurat_clusters",
    assay = "RNA",
    save_path = file.path(tempdir(),"SlimR_Celltype_annotation_Excel")
    colour_low = "white",
    colour_high = "navy",
    colour_low_mertic = "white",
    colour_high_mertic = "navy",
    )

## End(Not run)
```

Celltype_Annotation_Features

Annotate cell types using features plot with different marker databases

Description

This function dynamically selects the appropriate annotation method based on the gene_list_type
parameter. It supports marker databases from Cellmarker2, PanglaoDB, Seurat (via FindAllMarkers),
or Excel files.

Usage

```
Celltype_Annotation_Features(
  seurat_obj,
  gene_list,
  gene_list_type = "Default",
  species = NULL,
  cluster_col = "seurat_clusters",
  assay = "RNA",
  save_path = NULL,
  min_counts = 1,
  metric_names = NULL,
  colour_low = "white",
  colour_high = "navy",
  colour_low_mertic = "white",
  colour_high_mertic = "navy",
  ...
)
```

Arguments

seurat_obj	A valid Seurat object with cluster annotations in meta.data.
gene_list	A list of data frames containing marker genes and metrics. Format depends on gene_list_type: - **Cellmarker2**: Generated by Markers_filter_Cellmarker2(). - **PanglaoDB**: Generated by Markers_filter_PanglaoDB(). - **Seurat**: Generated by read_seurat_markers(). - **Excel**: Generated by read_excel_markers().
gene_list_type	Type of marker database to use. Be one of: "Cellmarker2", "PanglaoDB", "Seurat", or "Excel".
species	Species of the dataset: "Human" or "Mouse" for gene name standardization.
cluster_col	Column name in meta.data defining clusters (default: "seurat_clusters").
assay	Assay layer in the Seurat object (default: "RNA").
save_path	Directory to save output PNGs. Must be explicitly specified.
min_counts	Minimum number of counts for Cellmarker2 annotations (default: 1).
metric_names	Optional. Change the row name for the input mertics, not recommended unless necessary. (NULL is used as default parameter; used in "Seurat"/"Excel").

colour_low   Color for lowest expression level. (default = "white")

colour_high   Color for highest expression level. (default = "navy")

colour_low_mertic
                    Color for lowest mertic level. (default = "white")

colour_high_mertic
                    Color for highest mertic level. (default = "navy")

...   Additional parameters passed to the specific annotation function.

Value

Saves cell type annotation PNGs in save_path. Returns invisibly.

See Also

Other Section_4_Semi_Automated_Annotation_Workflow: `Celltype_Annotation_Combined()`,
`Celltype_Annotation_Heatmap()`

Examples

```
## Not run:
# Example for Cellmarker2
Celltype_Annotation_Features(seurat_obj = sce,
    gene_list = Markers_list_Cellmarker2,
    species = "Human",
    cluster_col = "seurat_clusters",
    assay = "RNA",
    save_path = file.path(tempdir(),"SlimR_Celltype_annotation_Cellmarker2"),
    colour_low = "white",
    colour_high = "navy",
    colour_low_mertic = "white",
    colour_high_mertic = "navy",
    )

# Example for PanglaoDB
Celltype_Annotation_Features(seurat_obj = sce,
    gene_list = Markers_list_panglaoDB,
    species = "Human",
    cluster_col = "seurat_clusters",
    assay = "RNA",
    save_path = file.path(tempdir(),"SlimR_Celltype_annotation_PanglaoDB")
    colour_low = "white",
    colour_high = "navy",
    colour_low_mertic = "white",
    colour_high_mertic = "navy",
    )

# Example for Seurat marker list
Celltype_Annotation_Features(seurat_obj = sce,
    gene_list = Markers_list_Seurat,
    species = "Human",
    cluster_col = "seurat_clusters",
```

```
    assay = "RNA",
    save_path = file.path(tempdir(),"SlimR_Celltype_annotation_Seurat")
    colour_low = "white",
    colour_high = "navy",
    colour_low_mertic = "white",
    colour_high_mertic = "navy",
    )

# Example for Excel marker list
Celltype_Annotation_Features(seurat_obj = sce,
    gene_list = Markers_list_Excel,
    species = "Human",
    cluster_col = "seurat_clusters",
    assay = "RNA",
    save_path = file.path(tempdir(),"SlimR_Celltype_annotation_Excel")
    colour_low = "white",
    colour_high = "navy",
    colour_low_mertic = "white",
    colour_high_mertic = "navy",
    )

## End(Not run)
```

Celltype_Annotation_Heatmap

Uses "marker_list" to generate heatmap for cell annotation

Description

Uses "marker_list" to generate heatmap for cell annotation

Usage

```
Celltype_Annotation_Heatmap(
  seurat_obj,
  gene_list,
  species,
  cluster_col = "seurat_clusters",
  assay = "RNA",
  min_expression = 0.1,
  specificity_weight = 3,
  colour_low = "navy",
  colour_high = "firebrick3"
)
```

Arguments

seurat_obj	Enter the Seurat object with annotation columns such as "seurat_cluster" in meta.data to be annotated.
gene_list	A list of cells and corresponding gene controls, the name of the list is cell type, and the first column of the list corresponds to markers. Lists can be generated using functions such as "Markers_filter_Cellmarker2 ()", "Markers_filter_PanglaoDB ()", "read_excel_markers ()", "read_seurat_markers ()", etc.
species	This parameter selects the species "Human" or "Mouse" for standard gene format correction of markers entered by "Marker_list".
cluster_col	Enter annotation columns such as "seurat_cluster" in meta.data of the Seurat object to be annotated. Default parameters use "cluster_col = 'seurat_clusters'".
assay	Enter the assay used by the Seurat object, such as "RNA". Default parameters use "assay = 'RNA'".
min_expression	The min_expression parameter defines a threshold value to determine whether a cell's expression of a feature is considered "expressed" or not. It is used to filter out low-expression cells that may contribute noise to the analysis. Default parameters use "min_expression = 0.1".
specificity_weight	
	The specificity_weight parameter controls how much the expression variability (standard deviation) of a feature within a cluster contributes to its "specificity score." It amplifies or suppresses the impact of variability in the final score calculation.Default parameters use "specificity_weight = 3".
colour_low	Color for lowest probability level in Heatmap visualization of probability matrix. (default = "navy")
colour_high	Color for highest probability level Heatmap visualization of probability matrix. (default = "firebrick3")

Value

The heatmap of the comparison between "cluster_col" in the Seurat object and the given gene set "gene_list" needs to be annotated.

See Also

Other Section_4_Semi_Automated_Annotation_Workflow: `Celltype_Annotation_Combined`(), `Celltype_Annotation_Features`()

Examples

```
## Not run:
Celltype_Annotation_Heatmap(seurat_obj = sce,
    gene_list = Markers_list,
    species = "Human",
    cluster_col = "seurat_clusters",
    assay = "RNA",
    min_expression = 0.1,
    specificity_weight = 3,
```

```
      colour_low = "navy",
      colour_high = "firebrick3"
      )

   ## End(Not run)
```

Celltype_annotation_PanglaoDB

Uses "marker_list" from PanglaoDB for cell annotation

Description

Uses "marker_list" from PanglaoDB for cell annotation

Usage

```
Celltype_annotation_PanglaoDB(
  seurat_obj,
  gene_list,
  species,
  cluster_col = "seurat_clusters",
  assay = "RNA",
  save_path = NULL,
  metric_names = NULL,
  colour_low = "white",
  colour_high = "navy",
  colour_low_mertic = "white",
  colour_high_mertic = "navy"
)
```

Arguments

seurat_obj	Enter the Seurat object with annotation columns such as "seurat_cluster" in meta.data to be annotated.
gene_list	Enter the standard "Marker_list" generated by the PanglaoDB database for the SlimR package, generated by the "Markers_filter_PanglaoDB ()" function.
species	This parameter selects the species "Human" or "Mouse" for standard gene format correction of markers entered by "Marker_list".
cluster_col	Enter annotation columns such as "seurat_cluster" in meta.data of the Seurat object to be annotated. Default parameters use "cluster_col = 'seurat_clusters'".
assay	Enter the assay used by the Seurat object, such as "RNA". Default parameters use "assay = 'RNA'".
save_path	The output path of the cell annotation picture. Example parameters use "save_path = './SlimR/Celltype_annotation_PanglaoDB/'".

metric_names   Warning: Do not enter information. This parameter is used to check if "Marker_list"
                    conforms to the PanglaoDB database output.

colour_low   Color for lowest expression level. (default = "white")

colour_high   Color for highest expression level. (default = "navy")

colour_low_mertic
                    Color for lowest mertic level. (default = "white")

colour_high_mertic
                    Color for highest mertic level. (default = "navy")

Value

The cell annotation picture is saved in "save_path".

See Also

Other Section_5_Other_Functions_Provided_by_SlimR: `Celltype_annotation_Cellmarker2()`,
`Celltype_annotation_Excel()`, `Celltype_annotation_Seurat()`

Examples

```
## Not run:
Celltype_annotation_PanglaoDB(seurat_obj = sce,
    gene_list = Markers_list_panglaoDB,
    species = "Human",
    cluster_col = "seurat_clusters",
    assay = "RNA",
    save_path = file.path(tempdir(),"SlimR_Celltype_annotation_PanglaoDB")
    colour_low = "white",
    colour_high = "navy",
    colour_low_mertic = "white",
    colour_high_mertic = "navy",
    )

## End(Not run)
```

Celltype_annotation_Seurat
                    Uses "marker_list" from Seurat object for cell annotation

Description

Uses "marker_list" from Seurat object for cell annotation

Usage

```
Celltype_annotation_Seurat(
  seurat_obj,
  gene_list,
  species,
  cluster_col = "seurat_clusters",
  assay = "RNA",
  save_path = NULL,
  metric_names = NULL,
  colour_low = "white",
  colour_high = "navy",
  colour_low_mertic = "white",
  colour_high_mertic = "navy"
)
```

Arguments

seurat_obj	Enter the Seurat object with annotation columns such as "seurat_cluster" in meta.data to be annotated.
gene_list	Enter the standard "Marker_list" generated by the Seurat object database for the SlimR package, generated by the "read_seurat_markers()" function.
species	This parameter selects the species "Human" or "Mouse" for standard gene format correction of markers entered by "Marker_list".
cluster_col	Enter annotation columns such as "seurat_cluster" in meta.data of the Seurat object to be annotated. Default parameters use "cluster_col = 'seurat_clusters'".
assay	Enter the assay used by the Seurat object, such as "RNA". Default parameters use "assay = 'RNA'".
save_path	The output path of the cell annotation picture. Example parameters use "save_path = './SlimR/Celltype_annotation_Seurat/'".
metric_names	Change the row name for the input mertics, not recommended unless necessary. (NULL is used as default parameter)
colour_low	Color for lowest expression level. (default = "white")
colour_high	Color for highest expression level. (default = "navy")
colour_low_mertic	
	Color for lowest mertic level. (default = "white")
colour_high_mertic	
	Color for highest mertic level. (default = "navy")

Value

The cell annotation picture is saved in "save_path".

See Also

Other Section_5_Other_Functions_Provided_by_SlimR: `Celltype_annotation_Cellmarker2()`, `Celltype_annotation_Excel()`, `Celltype_annotation_PanglaoDB()`

Examples

```
## Not run:
Celltype_annotation_Seurat(seurat_obj = sce,
    gene_list = Markers_list_Seurat,
    species = "Human",
    cluster_col = "seurat_clusters",
    assay = "RNA",
    save_path = file.path(tempdir(),"SlimR_Celltype_annotation_Seurat")
    colour_low = "white",
    colour_high = "navy",
    colour_low_mertic = "white",
    colour_high_mertic = "navy",
    )

## End(Not run)
```

Celltype_Calculate	*Uses "marker_list" to calculate probability, prediction results, AUC and generate heatmap for cell annotation*

Description

Uses "marker_list" to calculate probability, prediction results, AUC and generate heatmap for cell annotation

Usage

```
Celltype_Calculate(
  seurat_obj,
  gene_list,
  species,
  cluster_col = "seurat_clusters",
  assay = "RNA",
  min_expression = 0.1,
  specificity_weight = 3,
  threshold = 0.8,
  compute_AUC = TRUE,
  plot_AUC = TRUE,
  AUC_correction = TRUE,
  colour_low = "navy",
  colour_high = "firebrick3"
)
```

Arguments

seurat_obj   Enter the Seurat object with annotation columns such as "seurat_cluster" in meta.data to be annotated.

gene_list	A list of cells and corresponding gene controls, the name of the list is cell type, and the first column of the list corresponds to markers. Lists can be generated using functions such as "Markers_filter_Cellmarker2 ()", "Markers_filter_PanglaoDB ()", "read_excel_markers ()", "read_seurat_markers ()", etc.
species	This parameter selects the species "Human" or "Mouse" for standard gene format correction of markers entered by "Marker_list".
cluster_col	Enter annotation columns such as "seurat_cluster" in meta.data of the Seurat object to be annotated. Default parameters use "cluster_col = 'seurat_clusters'".
assay	Enter the assay used by the Seurat object, such as "RNA". Default parameters use "assay = 'RNA'".
min_expression	The min_expression parameter defines a threshold value to determine whether a cell's expression of a feature is considered "expressed" or not. It is used to filter out low-expression cells that may contribute noise to the analysis. Default parameters use "min_expression = 0.1".
specificity_weight	
	The specificity_weight parameter controls how much the expression variability (standard deviation) of a feature within a cluster contributes to its "specificity score." It amplifies or suppresses the impact of variability in the final score calculation. Default parameters use "specificity_weight = 3".
threshold	This parameter refers to the normalized similarity between the "alternative cell type" and the "predicted cell type" in the returned results. (the default parameter is 0.8)
compute_AUC	Logical indicating whether to calculate AUC values for predicted cell types. AUC measures how well the marker genes distinguish the cluster from others. When TRUE, adds an AUC column to the prediction results. (default: TRUE)
plot_AUC	The logic indicates whether to draw an AUC curve for the predicted cell type. When TRUE, add an AUC_plot to result. (default: TRUE)
AUC_correction	Logical value controlling AUC-based correction. (default = TRUE) When set to TRUE: 1.Computes AUC values for candidate cell types. (probability > threshold) 2.Selects the cell type with the highest AUC as the final predicted type. 3.Records the selected type's AUC value in the "AUC" column.
colour_low	Color for lowest probability level in Heatmap visualization of probability matrix. (default = "navy")
colour_high	Color for highest probability level Heatmap visualization of probability matrix. (default = "firebrick3")

Value

A list containing:

- Expression_list: List of expression matrices for each cell type
- Proportion_list: List of proportion of expression for each cell type
- Expression_scores_matrix: Matrix of expression scores
- Probability_matrix: Matrix of normalized probabilities
- Prediction_results: Data frame with cluster annotations including:

- – cluster_col: Cluster identifier
- – Predicted_cell_type: Primary predicted cell type
- – AUC: Area Under the Curve value (when compute_AUC = TRUE)
- – Alternative_cell_types: Semi-colon separated alternative cell types
- Heatmap_plot: Heatmap visualization of probability matrix
- AUC_plot: AUC visualization of Predicted cell type

See Also

Other Section_3_Automated_Annotation_Workflow: `Celltype_Annotation()`, `Celltype_Verification()`, `Parameter_Calculate()`

Examples

```
## Not run:
SlimR_anno_result <- Celltype_Calculate(seurat_obj = sce,
    gene_list = Markers_list,
    species = "Human",
    cluster_col = "seurat_clusters",
    assay = "RNA",
    min_expression = 0.1,
    specificity_weight = 3,
    threshold = 0.8,
    compute_AUC = TRUE,
    plot_AUC = TRUE,
    AUC_correction = TRUE,
    colour_low = "navy",
    colour_high = "firebrick3"
    )

## End(Not run)
```

Celltype_Verification   *Perform cell type verification and generate the validation dotplot*

Description

This function performs verification of predicted cell types by selecting high log2FC and high expression proportion genes and generates and generate the validation dotplot.

Usage

```
Celltype_Verification(
  seurat_obj,
  SlimR_anno_result,
  assay = "RNA",
  gene_number = 5,
```

```
    colour_low = "white",
    colour_high = "navy",
    annotation_col = "Cell_type_SlimR"
)
```

Arguments

seurat_obj   A Seurat object containing single-cell data.

SlimR_anno_result

                  A list containing SlimR annotation results with: Expression_list - List of ex-
                  pression matrices for each cell type. Prediction_results - Data frame with cluster
                  annotations.

assay   Enter the assay used by the Seurat object, such as "RNA". Default parameters
                  use "assay = 'RNA'".

gene_number   Integer specifying number of top genes to select per cell type.

colour_low   Color for lowest expression level. (default = "white")

colour_high   Color for highest expression level. (default = "navy")

annotation_col Character string specifying the column in meta.data to use for grouping.

Value

A ggplot object showing expression of top variable genes.

See Also

Other Section_3_Automated_Annotation_Workflow: `Celltype_Annotation()`, `Celltype_Calculate()`,
`Parameter_Calculate()`

Examples

```
## Not run:
Celltype_Verification(seurat_obj = sce,
    SlimR_anno_result = SlimR_anno_result,
    assay = "RNA",
    gene_number = 5,
    colour_low = "white",
    colour_high = "navy",
    annotation_col = "Cell_type_SlimR"
    )

## End(Not run)
```

---

estimate_batch_effect *Estimate Batch Effect Strength*

Description

Roughly estimates the potential impact of batch effects using available metadata.

Usage

```
estimate_batch_effect(seurat_obj, assay)
```

Arguments

seurat_obj	Seurat object
assay	Assay name

Value

Batch effect score (0 indicates no detectable batch effect)

See Also

Other Section_1_Functions_Use_in_Package: calculate_cluster_variability(), calculate_expression(), calculate_expression_skewness(), calculate_probability(), extract_dataset_features(), generate_training_data(), postprocess_parameters(), predict_optimal_parameters(), train_parameter_model()

---

extract_dataset_features

Extract Dataset Characteristics for Machine Learning

Description

Computes various statistical features from single-cell data that are used as input for the parameter prediction model.

Usage

```
extract_dataset_features(
  seurat_obj,
  features,
  assay = NULL,
  cluster_col = NULL
)
```

Arguments

`seurat_obj`	Seurat object
`features`	Features to analyze
`assay`	Assay name
`cluster_col`	Cluster column name

Value

List of dataset characteristics including expression statistics, variability measures, and cluster properties

See Also

Other Section_1_Functions_Use_in_Package: `calculate_cluster_variability()`, `calculate_expression()`, `calculate_expression_skewness()`, `calculate_probability()`, `estimate_batch_effect()`, `generate_training_data()`, `postprocess_parameters()`, `predict_optimal_parameters()`, `train_parameter_model()`

---

generate_training_data

Generate Training Data for Machine Learning Model

Description

Creates synthetic training data based on empirical rules about optimal parameter relationships with dataset characteristics.

Usage

```
generate_training_data(dataset_features, n_samples = 1000)
```

Arguments

`dataset_features`	
	List of actual dataset characteristics
`n_samples`	Number of synthetic samples to generate

Value

Data frame with synthetic features and optimal parameter targets

See Also

Other Section_1_Functions_Use_in_Package: `calculate_cluster_variability()`, `calculate_expression()`, `calculate_expression_skewness()`, `calculate_probability()`, `estimate_batch_effect()`, `extract_dataset_features()`, `postprocess_parameters()`, `predict_optimal_parameters()`, `train_parameter_model()`

Markers_filter_Cellmarker2

Create Marker_list from the Cellmarkers2 database

Description

Create Marker_list from the Cellmarkers2 database

Usage

```
Markers_filter_Cellmarker2(
  df,
  species = NULL,
  tissue_class = NULL,
  tissue_type = NULL,
  cancer_type = NULL,
  cell_type = NULL
)
```

Arguments

df	Standardized Cellmarkers2 database. It is read as data(Cellmarkers2) in the SlimR library.
species	Species information in Cellmarkers2 database. The default input is "Human" or "Mouse".The input can be retrieved by "Cellmarkers2_table". For more information,please refer to http://117.50.127.228/CellMarker/ on Cellmarkers2's official website.
tissue_class	Tissue_class information in Cellmarkers2 database. The input can be retrieved by "Cellmarkers2_table". For more information, please refer to http://117.50.127.228/CellMarker/ on Cellmarkers2's official website.
tissue_type	Tissue_type information in Cellmarkers2 database. The input can be retrieved by "Cellmarkers2_table". For more information, please refer to http://117.50.127.228/CellMarker/ on Cellmarkers2's official website.
cancer_type	Cancer_type information in Cellmarkers2 database. The input can be retrieved by "Cellmarkers2_table". For more information, please refer to http://117.50.127.228/CellMarker/ on Cellmarkers2's official website.
cell_type	Cell_type information in Cellmarkers2 database. The input can be retrieved by "Cellmarkers2_table". For more information, please refer to http://117.50.127.228/CellMarker/ on Cellmarkers2's official website.

Value

The standardized "Marker_list" in the SlimR package

See Also

Other Section_2_Standardized_Markers_List_Input: Markers_filter_PanglaoDB(), Read_excel_markers(),
Read_seurat_markers()

Examples

```
Cellmarker2 <- SlimR::Cellmarker2
Markers_list_Cellmarker2 <- Markers_filter_Cellmarker2(
    Cellmarker2,
    species = "Human",
    tissue_class = "Intestine",
    tissue_type = NULL,
    cancer_type = NULL,
    cell_type = NULL
    )
```

Markers_filter_PanglaoDB

Create Marker_list from the PanglaoDB database

Description

Create Marker_list from the PanglaoDB database

Usage

```
Markers_filter_PanglaoDB(df, species_input, organ_input)
```

Arguments

df	Standardized PanglaoDB database. It is read as data(PanglaoDB) in the SlimR library.
species_input	Species information in PanglaoDB database. The default input is "Human" or "Mouse".The input can be retrieved by "PanglaoDB_table". For more information,please refer to https://panglaodb.se/ on PanglaoDB's official website.
organ_input	Organ type information in the PanglaoDB database. The input can be retrieved by "PanglaoDB_table".For more information, please refer to https://panglaodb.se/ on PanglaoDB's official website.

Value

The standardized "Marker_list" in the SlimR package

See Also

Other Section_2_Standardized_Markers_List_Input: Markers_filter_Cellmarker2(), Read_excel_markers(),
Read_seurat_markers()

Examples

```
PanglaoDB <- SlimR::PanglaoDB
Markers_list_panglaoDB <- Markers_filter_PanglaoDB(
    PanglaoDB,
    species_input = 'Human',
    organ_input = 'GI tract'
    )
```

Markers_list_scIBD   *List of cell type markers in the scIBD dataset*

Description

A dataset containing marker genes for different human intestine cell types from scIBD

Usage

```
Markers_list_scIBD
```

Format

A list with one hundred and one tables.

Details

This list is a table of 101 types of human intestine cell types markers obtained from scIBD. The article doi source is "https://doi.org/10.1038/s43588-023-00464-9", and the reference literature is: Nie et al. (2023) doi:10.1038/s43588-023-00464-9. Note: The 'Markers_list_scIBD' was generated using section 2.5.2 and the parameters 'sort_by = "logFC"' and 'gene_filter = 20' were set.

Source

doi:10.1038/s43588023004649

See Also

Other Section_0_Database: Cellmarker2, Cellmarker2_raw, Cellmarker2_table, Markers_list_TCellSI, PanglaoDB, PanglaoDB_raw, PanglaoDB_table

---

Markers_list_TCellSI   *List of cell type markers in the TCellSI dataset*

Description

A dataset containing marker genes for different T cell types from TCellSI

Usage

```
Markers_list_TCellSI
```

Format

A list with ten tables.

Details

This list is a table of 10 types of T cell markers obtained from TCellSI. The data source is "https://github.com/GuoBioinfoLab/
and the reference literature is: Yang et al. (2024) [doi:10.1002/imt2.231](doi:10.1002/imt2.231).

Source

https://github.com/GuoBioinfoLab/TCellSI/

See Also

Other Section_0_Database: Cellmarker2, Cellmarker2_raw, Cellmarker2_table, Markers_list_scIBD,
PanglaoDB, PanglaoDB_raw, PanglaoDB_table

PanglaoDB   *PanglaoDB dataset*

Description

A dataset containing marker genes for different cell types from PanglaoDB

Usage

```
PanglaoDB
```

Format

A data frame with 9 columns:

Details

This dataset is used to filter and create a standardized marker list.'

Source

https://panglaodb.se/

See Also

Other Section_0_Database: Cellmarker2, Cellmarker2_raw, Cellmarker2_table, Markers_list_TCellSI, Markers_list_scIBD, PanglaoDB_raw, PanglaoDB_table

---

PanglaoDB_raw *PanglaoDB raw dataset*

Description

A dataset containing marker genes for different cell types from PanglaoDB

Usage

```
PanglaoDB_raw
```

Format

A data frame with 14 columns contined in the PanglaoDB database:

Details

This dataset is used to filter and create a standardized marker list.'

Source

https://panglaodb.se/

See Also

Other Section_0_Database: Cellmarker2, Cellmarker2_raw, Cellmarker2_table, Markers_list_TCellSI, Markers_list_scIBD, PanglaoDB, PanglaoDB_table

---

PanglaoDB_table   *PanglaoDB table*

Description

A dataset containing marker genes for different cell types from PanglaoDB

Usage

```
PanglaoDB_table
```

Format

A list contain different types like species, organ, cell type.

Details

This list is used to choose filters for creation of standardized marker list.

Source

<https://panglaodb.se/>

See Also

Other Section_0_Database: [Cellmarker2](#), [Cellmarker2_raw](#), [Cellmarker2_table](#), [Markers_list_TCellSI](#), [Markers_list_scIBD](#), [PanglaoDB](#), [PanglaoDB_raw](#)

Parameter_Calculate   *Adaptive Parameter Tuning for Single-Cell Data Annotation in SlimR*

Description

This function uses machine learning to automatically determine optimal min_expression and specificity_weight parameters for single-cell data analysis based on dataset characteristics.

Usage

```
Parameter_Calculate(
  seurat_obj,
  features,
  assay = NULL,
  cluster_col = NULL,
  method = "ensemble",
  n_models = 3,
  return_model = FALSE,
  verbose = TRUE
)
```

Arguments

`seurat_obj`	A Seurat object containing single-cell data
`features`	Character vector of feature names (genes) to analyze
`assay`	Name of assay to use (default: default assay)
`cluster_col`	Column name in metadata containing cluster information
`method`	Machine learning method: "rf" (random forest), "gbm" (gradient boosting), "svm" (support vector machine), or "ensemble" (default)
`n_models`	Number of models for ensemble learning (default: 3)
`return_model`	Whether to return trained model (default: FALSE)
`verbose`	Whether to print progress messages (default: TRUE)

Value

A list containing:

- min_expression: Recommended expression threshold
- specificity_weight: Recommended specificity weight
- performance: Model performance metric (R-squared)
- dataset_features: Extracted dataset characteristics
- model: Trained model (if return_model = TRUE)

See Also

Other Section_3_Automated_Annotation_Workflow: `Celltype_Annotation()`, `Celltype_Calculate()`, `Celltype_Verification()`

Examples

```
## Not run:
# Basic usage
SlimR_params <- Parameter_Calculate(
  seurat_obj = sce,
  features = c("CD3E", "CD4", "CD8A"),
  assay = "RNA",
  cluster_col = "seurat_clusters",
  method = "ensemble",
  n_models = 3,
  return_model = FALSE,
  verbose = TRUE
  )

# Use with custom method
SlimR_params <- Parameter_Calculate(
  seurat_obj = sce,
  features = unique(Markers_list_Cellmarker2$`B cell`$marker),
  assay = "RNA",
  cluster_col = "seurat_clusters",
```

```
  method = "rf",
  return_model = FALSE,
  verbose = TRUE
  )

## End(Not run)
```

---

postprocess_parameters

Post-process Predicted Parameters

Description

Applies constraints and dataset-specific adjustments to ensure predicted parameters are within reasonable ranges.

Usage

```
postprocess_parameters(predicted_params, dataset_features)
```

Arguments

predicted_params
                List of raw predicted parameters

dataset_features
                Characteristics of current dataset

Value

List of finalized parameters after post-processing

See Also

Other Section_1_Functions_Use_in_Package: calculate_cluster_variability(), calculate_expression(), calculate_expression_skewness(), calculate_probability(), estimate_batch_effect(), extract_dataset_features(), generate_training_data(), predict_optimal_parameters(), train_parameter_model()

predict_optimal_parameters

Predict Optimal Parameters Using Trained Model

Description

Applies the trained machine learning model to predict optimal parameters for the current dataset.

Usage

```
predict_optimal_parameters(model, dataset_features)
```

Arguments

model   Trained machine learning model (now a list with two models)

dataset_features

Extracted characteristics of current dataset

Value

List containing predicted min_expression and specificity_weight

See Also

Other Section_1_Functions_Use_in_Package: `calculate_cluster_variability()`, `calculate_expression()`, `calculate_expression_skewness()`, `calculate_probability()`, `estimate_batch_effect()`, `extract_dataset_features()`, `generate_training_data()`, `postprocess_parameters()`, `train_parameter_model()`

Read_excel_markers   *Create "Marker_list" from Excel files ".xlsx"*

Description

Create "Marker_list" from Excel files ".xlsx"

Usage

```
Read_excel_markers(path)
```

Arguments

path   The path information of Marker files stored in ".xlsx" format. The Sheet name in the file is filled with cell type. The first line of each Sheet is the table head, the first column is filled with markers information, and the following column is filled with mertic information.

Value

The standardized "Marker_list" in the SlimR package.

See Also

Other Section_2_Standardized_Markers_List_Input: `Markers_filter_Cellmarker2()`, `Markers_filter_PanglaoDB()`, `Read_seurat_markers()`

Examples

```
## Not run:
Markers_list_Excel <- Read_excel_markers(
    "D:/Laboratory/Marker_load.xlsx"
    )

## End(Not run)
```

Read_seurat_markers   *Create "Marker_list" from Seurat object*

Description

Create "Marker_list" from Seurat object

Usage

```
Read_seurat_markers(
  df,
  sources = c("Seurat", "presto"),
  sort_by = "FSS",
  gene_filter = 20
)
```

Arguments

df	Dataframe generated by "FindAllMarkers" function, recommend to use parameter "group.by = "Cell_type"" and "only.pos = TRUE".
sources	Type of markers sources to use. Be one of: "Seurat" or "presto".
sort_by	Marker sorting parameter, for Seurat sources, select "avg_log2FC" or "p_val_adj" or "FSS" (Feature Significance Score, FSS, product value of log2FC and Expression ratio). Default parameters use "sort_by = 'FSS'".for presto sources, select "logFC" or "padj" or "FSS". Default parameters use "sort_by = 'FSS'".
gene_filter	The number of markers left for each cell type based on the "sort_by" parameter's level of difference. Default parameters use "gene_fliter = 20"

Value

The standardized "Marker_list" in the SlimR package.

See Also

Other Section_2_Standardized_Markers_List_Input: `Markers_filter_Cellmarker2()`, `Markers_filter_PanglaoDB()`, `Read_excel_markers()`

Examples

```
## Not run:
# Example for Seurat sources markers
seurat_markers <- Seurat::FindAllMarkers(
    object = sce,
    group.by = "Cell_type",
    only.pos = TRUE)

Markers_list_Seurat <- Read_seurat_markers(seurat_markers,
    sources = "Seurat",
    sort_by = "avg_log2FC",
    gene_filter = 20
    )

# Example for presto sources markers
seurat_markers <- dplyr::filter(
    presto::wilcoxauc(
      X = sce,
      group_by = "Cell_type",
      seurat_assay = "RNA"
      ),
    padj < 0.05, logFC > 0.5
    )

Markers_list_Seurat <- Read_seurat_markers(seurat_markers,
    sources = "presto",
    sort_by = "logFC",
    gene_filter = 20
    )

## End(Not run)
```

---

train_parameter_model *Train Parameter Prediction Model*

Description

Trains machine learning models to predict optimal parameters based on dataset characteristics.

Usage

```
train_parameter_model(
  training_data,
  method = "ensemble",
  n_models = 3,
  verbose = TRUE
)
```

Arguments

training_data	Data frame with features and target parameters
method	Machine learning method to use
n_models	Number of models for ensemble learning
verbose	Whether to print training progress

Value

List containing trained model and performance metrics

See Also

Other Section_1_Functions_Use_in_Package: calculate_cluster_variability(), calculate_expression(), calculate_expression_skewness(), calculate_probability(), estimate_batch_effect(), extract_dataset_features(), generate_training_data(), postprocess_parameters(), predict_optimal_paramet

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