

# Package ‘scClassify’

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**Type** Package

**Title** scClassify: single-cell Hierarchical Classification

**Version** 1.25.0

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**Description**

scClassify is a multiscale classification framework for single-cell RNA-seq data based on ensemble learning and cell type hierarchies, enabling sample size estimation required for accurate cell type classification and joint classification of cells using multiple references.

**License** GPL-3

**Encoding** UTF-8

**LazyData** false

**Depends** R (>= 4.0)

**Imports** S4Vectors, limma, ggraph, igraph, methods, cluster, minpack.lm, mixtools, BiocParallel, proxy, proxyC, Matrix, ggplot2, hopach, diptest, mgcv, stats, graphics, statmod, Cepo

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---

.scClassifyTrainModel *The scClassifyTrainModel class*

---

### Description

The scClassifyTrainModel class is designed to stored training model for scClassify

### Usage

```
.scClassifyTrainModel(
  name,
  cellTypeTree,
  cellTypeTrain,
  features,
  model,
  modelweights,
  metaData
)
```

### Arguments

name	Name of the training dataset
cellTypeTree	A list indicate a cell type tree
cellTypeTrain	A vector of cell type in training dataset

features	A vector of character indicates the features that are trained for this data
model	A list stored the training model, including the features that are selected and the cell expression matrix that are used for training
modelweights	A vector of numeric indicates the weights of each model
metaData	A DataFrame stored meta data of training model

**Value**

A `scClassifyTrainModel` object

**Author(s)**

Yingxin Lin

---

<code>cellTypeTrain</code>	<i>Accessors of <code>cellTypeTrain</code> for <code>scClassifyTrainModel</code></i>
----------------------------	--

---

**Description**

Methods to access various components of the ‘`scClassifyTrainModel`’ object.

**Usage**

```
cellTypeTrain(x)
```

**Arguments**

x A ‘`scClassifyTrainModel`’ object.

**Value**

`cellTypeTrain` of the `scClassifyTrainModel` slot

**Examples**

```
data(trainClassExample_xin)
cellTypeTrain(trainClassExample_xin)
```

---

`cellTypeTree`*Accessors of cellTypeTree for scClassifyTrainModel*

---

**Description**

Methods to access various components of the 'scClassifyTrainModel' object.

**Usage**

```
cellTypeTree(x)
```

**Arguments**

`x` A 'scClassifyTrainModel' object.

**Value**

cellTypeTree of the scClassifyTrainModel slot

**Examples**

```
data(trainClassExample_xin)
cellTypeTree(trainClassExample_xin)
```

---

`features`*Accessors of features for scClassifyTrainModel*

---

**Description**

Methods to access various components of the 'scClassifyTrainModel' object.

**Usage**

```
features(x)
```

**Arguments**

`x` A 'scClassifyTrainModel' object.

**Value**

features of the scClassifyTrainModel slot

**Examples**

```
data(trainClassExample_xin)
features(trainClassExample_xin)
```

---

getN	<i>Function to get the required N given by the accuracy and the learning curve model</i>
------	--

---

**Description**

Function to get the required N given by the accuracy and the learning curve model

**Usage**

```
getN(res, acc = 0.9)
```

**Arguments**

res	model results returned by learning_curve function
acc	accuracy that are quired

**Value**

sample size that are required

**Examples**

```
set.seed(2019)
n <- seq(20, 10000, 100)
accMat <- do.call(cbind, lapply(1:length(n), function(i){
  tmp_n <- rep(n[i], 50)
  y <- -2/(tmp_n^0.8) + 0.95 + rnorm(length(tmp_n), 0, 0.02)
}))
res <- learningCurve(accMat = accMat, n)
N <- getN(res, acc = 0.9)
```

---

learningCurve	<i>Fit learning curve for accuracy matrix</i>
---------------	---

---

**Description**

Fit learning curve for accuracy matrix

**Usage**

```
learningCurve(
  accMat,
  n,
  auto_initial = TRUE,
  a = NULL,
  b = NULL,
  c = NULL,
  d_list = NULL,
```

```

fitmodel = c("nls", "nls_mix", "gam"),
plot = TRUE,
verbose = TRUE
)

```

### Arguments

accMat	Matrix of accuracy rate where column indicate different sample size
n	Vector indicates the sample size
auto_initial	whether automatical intialise
a	input the parameter a starting point
b	input the parameter a starting point
c	input the parameter a starting point
d_list	range of d
fitmodel	"nls", "nls_mix", "gam"
plot	indicates whether plot or not
verbose	indicates whether verbose or not

### Value

list of results

### Author(s)

Yingxin Lin

### Examples

```

set.seed(2019)
n <- seq(20, 10000, 100)
accMat <- do.call(cbind, lapply(1:length(n), function(i){
  tmp_n <- rep(n[i], 50)
  y <- -2/(tmp_n^0.8) + 0.95 + rnorm(length(tmp_n), 0, 0.02)
}))
res <- learningCurve(accMat = accMat, n)
N <- getN(res, acc = 0.9)

```

---

model

*Accessors of model for scClassifyTrainModel*

---

### Description

Methods to access various components of the 'scClassifyTrainModel' object.

### Usage

```
model(x)
```

**Arguments**

x                    A 'scClassifyTrainModel' object.

**Value**

model of the scClassifyTrainModel slot

**Examples**

```
data(trainClassExample_xin)
model(trainClassExample_xin)
```

---

modelweights	<i>Accessors of modelweights for scClassifyTrainModel</i>
--------------	---

---

**Description**

Methods to access various components of the 'scClassifyTrainModel' object.

**Usage**

```
modelweights(x)
```

**Arguments**

x                    A 'scClassifyTrainModel' object.

**Value**

modelweights of the scClassifyTrainModel slot

**Examples**

```
data(trainClassExample_xin)
modelweights(trainClassExample_xin)
```

---

name	<i>Accessors of name for scClassifyTrainModel</i>
------	---

---

**Description**

Methods to access various components of the 'scClassifyTrainModel' object.

**Usage**

```
name(x)
```

**Arguments**

x                    A 'scClassifyTrainModel' object.

**Value**

name of the scClassifyTrainModel slot

**Examples**

```
data(trainClassExample_xin)
name(trainClassExample_xin)
```

---

plotCellTypeTree            *To plot cell type tree*

---

**Description**

To plot cell type tree

**Usage**

```
plotCellTypeTree(cutree_list, group_level = NULL)
```

**Arguments**

cutree\_list        A list indicates the hierarchical cell type tree  
group\_level        Indicate whether plot or not

**Value**

A ggplot object visualising the HOPACH tree

**Examples**

```
data("trainClassExample_xin")
plotCellTypeTree(cellTypeTree(trainClassExample_xin))
```

---

predict\_scClassify      *Testing scClassify model*

---

### Description

Testing scClassify model

### Usage

```
predict_scClassify(
  exprsMat_test,
  trainRes,
  cellTypes_test = NULL,
  k = 10,
  prob_threshold = 0.7,
  cor_threshold_static = 0.5,
  cor_threshold_high = 0.7,
  features = "limma",
  algorithm = "WKNN",
  similarity = "pearson",
  cutoff_method = c("dynamic", "static"),
  weighted_ensemble = FALSE,
  weights = NULL,
  parallel = FALSE,
  BPPARAM = BiocParallel::SerialParam(),
  verbose = FALSE
)
```

### Arguments

exprsMat_test	A list or a matrix indicates the log-transformed expression matrices of the query datasets
trainRes	A 'scClassifyTrainModel' or a 'list' indicates scClassify trained model
cellTypes_test	A list or a vector indicates cell types of the query datasets (Optional).
k	An integer indicates the number of neighbour
prob_threshold	A numeric indicates the probability threshold for KNN/WKNN/DWKNN.
cor_threshold_static	A numeric indicates the static correlation threshold.
cor_threshold_high	A numeric indicates the highest correlation threshold
features	A vector indicates the gene selection method, set as "limma" by default. This should be one or more of "limma", "DV", "DD", "chisq", "BI".
algorithm	A vector indicates the KNN method that are used, set as "WKNN" by default. This should be one or more of "WKNN", "KNN", "DWKNN".
similarity	A vector indicates the similarity measure that are used, set as "pearson" by default. This should be one or more of "pearson", "spearman", "cosine", "jaccard", "kendall", "binomial", "weighted_rank", "manhattan"
cutoff_method	A vector indicates the method to cutoff the correlation distribution. Set as "dynamic" by default.

weighted_ensemble	A logical input indicates in ensemble learning, whether the results is combined by a weighted score for each base classifier.
weights	A vector indicates the weights for ensemble
parallel	A logical input indicates whether running in parallel or not
BPPARAM	A BiocParallelParam class object from the BiocParallel package is used. Default is SerialParam().
verbose	A logical input indicates whether the intermediate steps will be printed

**Value**

list of results

**Author(s)**

Yingxin Lin

**Examples**

```
data("scClassify_example")
wang_cellTypes <- scClassify_example$wang_cellTypes
exprsMat_wang_subset <- scClassify_example$exprsMat_wang_subset
data("trainClassExample_xin")

pred_res <- predict_scClassify(exprsMat_test = exprsMat_wang_subset,
  trainRes = trainClassExample_xin,
  cellTypes_test = wang_cellTypes,
  algorithm = "WKNN",
  features = c("limma"),
  similarity = c("pearson"),
  prob_threshold = 0.7,
  verbose = TRUE)
```

---

predict\_scClassifyJoint

*Testing scClassify model (joint training)*

---

**Description**

Testing scClassify model (joint training)

**Usage**

```
predict_scClassifyJoint(
  exprsMat_test,
  trainRes,
  cellTypes_test = NULL,
  k = 10,
  prob_threshold = 0.7,
  cor_threshold_static = 0.5,
  cor_threshold_high = 0.7,
```

```

features = "limma",
algorithm = "WKNN",
similarity = "pearson",
cutoff_method = c("dynamic", "static"),
parallel = FALSE,
BPPARAM = BiocParallel::SerialParam(),
verbose = FALSE
)

```

### Arguments

**exprsMat\_test** A list or a matrix indicates the expression matrices of the testing datasets

**trainRes** A 'scClassifyTrainModel' or a 'list' indicates scClassify training model

**cellTypes\_test** A list or a vector indicates cell types of the testing datasets (Optional).

**k** An integer indicates the number of neighbour

**prob\_threshold** A numeric indicates the probability threshold for KNN/WKNN/DWKNN.

**cor\_threshold\_static** A numeric indicates the static correlation threshold.

**cor\_threshold\_high** A numeric indicates the highest correlation threshold

**features** A vector indicates the method to select features, set as "limma" by default. This should be one or more of "limma", "DV", "DD", "chisq", "BI".

**algorithm** A vector indicates the KNN method that are used, set as "WKNN" by default. This should be one or more of "WKNN", "KNN", "DWKNN".

**similarity** A vector indicates the similarity measure that are used, set as "pearson" by default. This should be one or more of "pearson", "spearman", "cosine", "jaccard", "kendall", "binomial", "weighted\_rank", "manhattan"

**cutoff\_method** A vector indicates the method to cutoff the correlation distribution. Set as "dynamic" by default.

**parallel** A logical input indicates whether running in parallel or not

**BPPARAM** A BiocParallelParam class object from the BiocParallel package is used. Default is SerialParam().

**verbose** A logical input indicates whether the intermediate steps will be printed

### Value

list of results

### Author(s)

Yingxin Lin

### Examples

```

data("scClassify_example")
wang_cellTypes <- scClassify_example$wang_cellTypes
exprsMat_wang_subset <- scClassify_example$exprsMat_wang_subset
data("trainClassExample_xin")
data("trainClassExample_wang")

```

```

trainClassExampleJoint <- scClassifyTrainModelList(trainClassExample_wang,
trainClassExample_xin)

pred_res_joint <- predict_scClassifyJoint(exprsMat_test = exprsMat_wang_subset,
trainRes = trainClassExampleJoint,
cellTypes_test = wang_cellTypes,
algorithm = "WKNN",
features = c("limma"),
similarity = c("pearson"),
prob_threshold = 0.7,
verbose = FALSE)

table(pred_res_joint$jointRes$cellTypes, wang_cellTypes)

```

---

runHOPACH

---

*Create HOPACH tree*


---

### Description

A function generating HOPACH tree using the average expression matrix for each cell type.

### Usage

```
runHOPACH(data, plot = TRUE, kmax = 5)
```

### Arguments

data	A matrix of average expression matrix (each row indicates the gene, each column indicates the cell type)
plot	Indicate whether plot or not
kmax	Integer between 1 and 9 specifying the maximum number of children at each node in the tree.

### Value

Return a list where

- `cutree_list`: A list indicates the hierarchical cell type tree
- `plot`: A `ggplot` visualise the cell type tree

### Author(s)

Yingxin Lin

### References

van der Laan, M. J. and Pollard, K. S. (2003) ‘A new algorithm for hybrid hierarchical clustering with visualization and the bootstrap’, *Journal of Statistical Planning and Inference*. doi: 10.1016/S0378-3758(02)00388-9.

**Examples**

```

data("scClassify_example")
wang_cellTypes <- factor(scClassify_example$wang_cellTypes)
exprsMat_wang_subset <- scClassify_example$exprsMat_wang_subset
avgMat_wang <- apply(exprsMat_wang_subset, 1, function(x)
  aggregate(x, list(wang_cellTypes), mean)$x)
rownames(avgMat_wang) <- levels(wang_cellTypes)
res_hopach <- runHOPACH(avgMat_wang)
res_hopach$plot

```

runSampleCal

*Run sample size calculation for pilot data for reference dataset***Description**

Run sample size calculation for pilot data for reference dataset

**Usage**

```

runSampleCal(
  exprsMat,
  cellTypes,
  n_list = c(20, 40, 60, 80, 100, seq(200, 500, 100)),
  num_repeat = 20,
  level = NULL,
  cellType_tree = NULL,
  BPPARAM = BiocParallel::SerialParam(),
  subset_test = FALSE,
  num_test = NULL,
  ...
)

```

**Arguments**

exprsMat	A matrix of expression matrix of pilot dataset (log-transformed, or normalised)
cellTypes	A vector of cell types of pilot dataset
n_list	A vector of integer indicates the sample size to run.
num_repeat	An integer indicates the number of run for each sample size will be repeated.
level	An integer indicates the accuracy rate is calculate based on the n-th level from top of cell type tree. If it is NULL (by default), it will be the bottom of the cell type tree. It can not be larger than the total number of levels of the tree.
cellType_tree	A list indicates the cell type tree (optional), if it is NULL, the accuracy rate is calculate based on the provided cellTypes.
BPPARAM	A BiocParallelParam class object from the BiocParallel package is used. Default is SerialParam().
subset_test	A logical input indicates whether we used a subset of data (fixed number for each sample size) to test instead of all remaining data. By default, it is FALSE.
num_test	An integer indicates the size of the test data.
...	other parameter from scClassify

**Value**

A matrix of accuracy matrix, where columns corresponding to different sample sizes, rows corresponding to the number of repetition.

**Examples**

```
data("scClassify_example")
xin_cellTypes <- scClassify_example$xin_cellTypes
exprsMat_xin_subset <- scClassify_example$exprsMat_xin_subset

exprsMat_xin_subset <- as(exprsMat_xin_subset, "dgCMatrix")
set.seed(2019)
accMat <- runSampleCal(exprsMat_xin_subset,
xin_cellTypes,
n_list = seq(20, 100, 20),
num_repeat = 5, BPPARAM = BiocParallel::SerialParam())
```

---

scClassify

*Train and test scClassify model*


---

**Description**

Train and test scClassify model

**Usage**

```
scClassify(
  exprsMat_train = NULL,
  cellTypes_train = NULL,
  exprsMat_test = NULL,
  cellTypes_test = NULL,
  tree = "HOPACH",
  algorithm = "WKNN",
  selectFeatures = "limma",
  similarity = "pearson",
  cutoff_method = c("dynamic", "static"),
  weighted_ensemble = FALSE,
  weights = NULL,
  weighted_jointClassification = TRUE,
  cellType_tree = NULL,
  k = 10,
  topN = 50,
  hopach_kmax = 5,
  pSig = 0.01,
  prob_threshold = 0.7,
  cor_threshold_static = 0.5,
  cor_threshold_high = 0.7,
  returnList = TRUE,
  parallel = FALSE,
  BPPARAM = BiocParallel::SerialParam(),
  verbose = FALSE
)
```

**Arguments**

exprsMat_train	A matrix of log-transformed expression matrix of reference dataset
cellTypes_train	A vector of cell types of reference dataset
exprsMat_test	A list or a matrix indicates the expression matrices of the query datasets
cellTypes_test	A list or a vector indicates cell types of the query datasets (Optional).
tree	A vector indicates the method to build hierarchical tree, set as "HOPACH" by default. This should be one of "HOPACH" and "HC" (using hclust).
algorithm	A vector indicates the KNN method that are used, set as "WKNN" by default. This should be one or more of "WKNN", "KNN", "DWKNN".
selectFeatures	A vector indicates the gene selection method, set as "limma" by default. This should be one or more of "limma", "DV", "DD", "chisq", "BI" and "Cepo".
similarity	A vector indicates the similarity measure that are used, set as "pearson" by default. This should be one or more of "pearson", "spearman", "cosine", "jaccard", "kendall", "binomial", "weighted_rank", "manhattan"
cutoff_method	A vector indicates the method to cutoff the correlation distribution. Set as "dynamic" by default.
weighted_ensemble	A logical input indicates in ensemble learning, whether the results is combined by a weighted score for each base classifier.
weights	A vector indicates the weights for ensemble
weighted_jointClassification	A logical input indicates in joint classification using multiple training datasets, whether the results is combined by a weighted score for each training model.
cellType_tree	A list indicates the cell type tree provided by user. (By default, it is NULL) (Only for one training data input)
k	An integer indicates the number of neighbour
topN	An integer indicates the top number of features that are selected
hopach_kmax	An integer between 1 and 9 specifying the maximum number of children at each node in the HOPACH tree.
pSig	A numeric indicates the cutoff of pvalue for features
prob_threshold	A numeric indicates the probability threshold for KNN/WKNN/DWKNN.
cor_threshold_static	A numeric indicates the static correlation threshold.
cor_threshold_high	A numeric indicates the highest correlation threshold
returnList	A logical input indicates whether the output will be class of list
parallel	A logical input indicates whether running in parallel or not
BPPARAM	A BiocParallelParam class object from the BiocParallel package is used. Default is SerialParam().
verbose	A logical input indicates whether the intermediate steps will be printed

**Value**

A list of the results, including testRes storing the results of the testing information, and trainRes storing the training model information.

**Author(s)**

Yingxin Lin

**Examples**

```

data("scClassify_example")
xin_cellTypes <- scClassify_example$xin_cellTypes
exprsMat_xin_subset <- scClassify_example$exprsMat_xin_subset
wang_cellTypes <- scClassify_example$wang_cellTypes
exprsMat_wang_subset <- scClassify_example$exprsMat_wang_subset

scClassify_res <- scClassify(exprsMat_train = exprsMat_xin_subset,
  cellTypes_train = xin_cellTypes,
  exprsMat_test = list(wang = exprsMat_wang_subset),
  cellTypes_test = list(wang = wang_cellTypes),
  tree = "HOPACH",
  algorithm = "WKNN",
  selectFeatures = c("limma"),
  similarity = c("pearson"),
  returnList = FALSE,
  verbose = FALSE)

```

---

scClassifyTrainModel-class

*An S4 class to stored training model for scClassify*


---

**Description**

An S4 class to stored training model for scClassify

**Slots**

name Name of the training dataset

cellTypeTrain A vector of cell type in training dataset

cellTypeTree A list indicate a cell type tree

features A vector of character indicates the features that are trained for this data

model A list stored the training model, including the features that are selected and the cell expression matrix that are used for training

modelweights A vector of numeric indicates the weights of each model

metaData A DataFrame stored meta data of training model

---

`scClassifyTrainModelList`*The scClassifyTrainModelList class*

---

**Description**

The scClassifyTrainModelList class

**Usage**

```
scClassifyTrainModelList(...)
```

**Arguments**

...                   scClassifyTrainModel objects

**Value**

A scClassifyTrainModelList object

**Examples**

```
data("trainClassExample_xin")
data("trainClassExample_wang")
trainClassExampleList <- scClassifyTrainModelList(trainClassExample_xin,
trainClassExample_wang
)
```

---

`scClassifyTrainModelList-class`*An S4 class to stored a list of training models from scClassify*

---

**Description**

An S4 class to stored a list of training models from scClassify

---

scClassify\_example     *Example data used in scClassify package*

---

**Description**

A list includes expression matrix and cell type of subsets of wang et al., xin et al.

**Usage**

```
data(scClassify_example, package = 'scClassify')
```

**Format**

An object of class list of length 4.

**Source**

Wang YJ, Schug J, Won K-J, Liu C, Naji A, Avrahami D, Golson ML & Kaestner KH (2016) Single cell transcriptomics of the human endocrine pancreas. Diabetes: db160405

Xin Y, Kim J, Okamoto H, Ni M, Wei Y, Adler C, Murphy AJ, Yancopoulos GD, Lin C & Gromada J (2016) RNA sequencing of single human islet cells reveals type 2 diabetes genes. Cell Metab. 24: 608–615

---

trainClassExample\_wang

*Subset of pretrained model of Wang et al.*

---

**Description**

An object of class scClassifyTrainModel for Wang et al.

**Usage**

```
data(trainClassExample_wang, package = 'scClassify')
```

**Format**

An object of class scClassifyTrainModel of length 1.

**Source**

Wang YJ, Schug J, Won K-J, Liu C, Naji A, Avrahami D, Golson ML & Kaestner KH (2016) Single cell transcriptomics of the human endocrine pancreas. Diabetes: db160405

---

trainClassExample\_xin *Subset of pretrained model of Xin et al.*

---

**Description**

An object of scClassifyTrainModel for Xin et al.

**Usage**

```
data(trainClassExample_xin, package = 'scClassify')
```

**Format**

An object of class scClassifyTrainModel of length 1.

**Source**

Xin Y, Kim J, Okamoto H, Ni M, Wei Y, Adler C, Murphy AJ, Yancopoulos GD, Lin C & Gromada J (2016) RNA sequencing of single human islet cells reveals type 2 diabetes genes. *Cell Metab.* 24: 608–615

---

train\_scClassify *Training scClassify model*

---

**Description**

Training scClassify model

**Usage**

```
train_scClassify(  
  exprsMat_train,  
  cellTypes_train,  
  tree = "HOPACH",  
  selectFeatures = "limma",  
  topN = 50,  
  hopach_kmax = 5,  
  pSig = 0.05,  
  cellType_tree = NULL,  
  weightsCal = FALSE,  
  parallel = FALSE,  
  BPPARAM = BiocParallel::SerialParam(),  
  verbose = TRUE,  
  returnList = TRUE,  
  ...  
)
```

**Arguments**

exprsMat_train	A matrix of log-transformed expression matrix of reference dataset
cellTypes_train	A vector of cell types of reference dataset
tree	A vector indicates the method to build hierarchical tree, set as "HOPACH" by default. This should be one of "HOPACH" and "HC" (using stats::hclust).
selectFeatures	A vector indicates the gene selection method, set as "limma" by default. This should be one or more of "limma", "DV", "DD", "chisq", "BI", "Cepo".
topN	An integer indicates the top number of features that are selected
hopach_kmax	An integer between 1 and 9 specifying the maximum number of children at each node in the HOPACH tree.
pSig	A numeric indicates the cutoff of pvalue for features
cellType_tree	A list indicates the cell type tree provided by user. (By default, it is NULL)
weightsCal	A logical input indicates whether we need to calculate the weights for the model.
parallel	A logical input indicates whether the algorithms will run in parallel
BPPARAM	A BiocParallelParam class object from the BiocParallel package is used. Default is SerialParam().
verbose	A logical input indicates whether the intermediate steps will be printed
returnList	A logical input indicates whether the output will be class of list
...	Other input for predict_scClassify for the case when weights calculation of the pretrained model is performed

**Value**

list of results or an object of scClassifyTrainModel

**Author(s)**

Yingxin Lin

**Examples**

```
data("scClassify_example")
xin_cellTypes <- scClassify_example$xin_cellTypes
exprsMat_xin_subset <- scClassify_example$exprsMat_xin_subset
trainClass <- train_scClassify(exprsMat_train = exprsMat_xin_subset,
cellTypes_train = xin_cellTypes,
selectFeatures = c("limma", "BI"),
returnList = FALSE
)
```

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