

Package ‘DR.SC’

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Type Package

Title Joint Dimension Reduction and Spatial Clustering

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Description Joint dimension reduction and spatial clustering is conducted for Single-cell RNA sequencing and spatial transcriptomics data, and more details can be referred to Wei Liu, Xu Liao, Yi Yang, Huazhen Lin, Joe Yeong, Xiang Zhou, Xingjie Shi and Jin Liu. (2022) <[doi:10.1093/nar/gkac219](https://doi.org/10.1093/nar/gkac219)>. It is not only computationally efficient and scalable to the sample size increment, but also is capable of choosing the smoothness parameter and the number of clusters as well.

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Depends parallel, spatstat.geom, R (>= 4.0.0)

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dlpfc151510

A human dorsolateral prefrontal cortex data

Description

A human dorsolateral prefrontal cortex dataset measured on the Visium platform, which includes 4634 spots and 500 genes, a subset of raw dataset at <https://github.com/LieberInstitute/spatialLIBD>.

Note

nothing

Author(s)

Wei Liu

References

None

Examples

```
data("dlpfc151510")
```

Description

Joint dimension reduction and spatial clustering for scRNA-seq and spatial transcriptomics data

Usage

```
## S3 method for class 'Seurat'  
DR.SC(seu, K, q=15, platform= "Visium",...)
```

Arguments

seu	an object of class "Seurat". The details of this object are given under 'Details'.
q	a positive integer, specify the number of latent features to be extracted, default as 15.
K	a positive integer or integer vector, specify the number of clusters. When K is a vector, it is automatically selected by MBIC criteria. Users can also use BIC and AIC to select K or refine model with MBIC by argument <code>pen.const</code> in the function selectModel .
platform	a string, specify the platform of the provided data, default as "Visium". There are more platforms to be chosen, including ("Visium", "ST", "seqfish", 'merfish', 'slide-seqv2') and "scRNAseq", where the first group means there are spatial coordinates information in the metadata of seu, named "row" and "col" and a Hidden Markov random field is used to model the spatial coordinates, and the last one "scRNAseq" means there is no spatial information in object seu and a multinomial model is used to model the unobserved class labels. The platform helps to calculate the adjacency matrix.
...	Other arguments to pass into DR.SC_fit function.

Details

seu is an object named Seurat, which can easily be created by R package [Seurat](#). If the data is collected by the spatial transcriptomics technologies such as 10X Visium, ST, seqFISH, MERFISH and Slide-seq, then there are spatial coordinates information in the metadata of seu, named "row" and "col". DR-SC model uses a Hidden Markov random field to model the spatial coordinates. If the data is collected by the single cell RNA sequencing technologies which means there is no spatial information in object seu then a multinomial model is used to model the unobserved class labels.

Value

DR.SC returns a revised Seurat object. There are two revisions in the seu. 1. the metadata is added a new column named `spatial.drsc.cluster` that represents the clustering results from DR-SC model, and the `Idents(seu)` is assigned with `spatial.drsc.cluster`. 2. a `DimReduc` object named `dr-sc` is added in the slot `reductions`, which represents the features extracted by DR-SC model.

Note

nothing

Author(s)

Wei Liu

References

Wei Liu, Xu Liao, Yi Yang, Huazhen Lin, Joe Yeong, Xiang Zhou*, Xingjie Shi* and Jin Liu* (2022). Joint dimension reduction and clustering analysis of single-cell RNA-seq and spatial transcriptomics data, *Nucleic Acids Research*

See Also

None

Examples

```
## we generate the spatial transcriptomics data with lattice neighborhood, i.e. ST platform.
seu <- gendata_RNAExp(height=10, width=10,p=50, K=4,platform="ST")
library(Seurat)
seu <- NormalizeData(seu, verbose=FALSE)
# choose 100 highly variable features
# seu <- FindVariableFeatures(seu, nfeatures = 100)
# maxIter = 2 is only used for illustration, and user can use default.
# seu1 <- DR.SC(seu, K=4, platform = 'ST', maxIter=2,verbose=FALSE)

# choose spatially variable features (SVGs)
seu <- FindSVGs(seu, nfeatures = 40, verbose=FALSE)
# use SVGs to fit DR.SC model
# maxIter = 2 is only used for illustration, and user can use default.
seu1 <- DR.SC(seu, K=4,platform = 'ST', maxIter=2, verbose=TRUE)
```

DR.SC_fit

*Joint dimension reduction and spatial clustering***Description**

Joint dimension reduction and spatial clustering for scRNA-seq and spatial transcriptomics data

Usage

```
DR.SC_fit(X, K, Adj_sp=NULL, q=15,
          error.heter= TRUE, beta_grid=seq(0.5, 5, by=0.5),
          maxIter=25, epsLogLik=1e-5, verbose=FALSE, maxIter_ICM=6,
          wpca.int=FALSE, coreNum = 5)
```

Arguments

X	a sparse matrix with class dgCMatrix or matrix, specify the log-normalization gene expression matrix used for DR-SC model.
K	a positive integer allowing scalar or vector, specify the number of clusters in model fitting.
Adj_sp	an optional sparse matrix with class dgCMatrix, specify the adjoint matrix used for DR-SC model. We provide this interface for those users who would like to define the adjoint matrix by their own.
q	a positive integer, specify the number of latent features to be extracted, default as 15.
error.heter	an optional logical value, whether use the heterogenous error for DR-SC model, default as TRUE. If error.heter=FALSE, then the homogenous error is used for probabilistic PCA model in DR-SC.
beta_grid	an optional vector of positive value, the candidate set of the smoothing parameter to be searched by the grid-search optimization approach.
maxIter	an optional positive value, represents the maximum iterations of EM.
epsLogLik	an optional positive vlaue, tolerance vlaue of relative variation rate of the observed pseudo log-loglikelihood value, default as '1e-5'.
verbose	an optional logical value, whether output the information of the ICM-EM algorithm.
maxIter_ICM	an optional positive value, represents the maximum iterations of ICM.
wpca.int	an optional logical value, means whether use the weighted PCA to obtain the initial values of loadings and other paramters, default as FALSE which means the ordinary PCA is used.
coreNum	an optional positive integer, means the number of thread used in parallel computing, default as 5. If the length of K is one, then coreNum will be set as 1 automatically.

Details

Nothing

Value

DR.SC_fit returns a [list](#) with class "drscObject" with the following three components:

Objdrsc	a list including the model fitting results, in which the number of elements is same as the length of K.
out_param	a numeric matrix used for model selection in MBIC.
K_set	a scalar or vector equal to input argument K.

In addition, each element of "Objdrsc" is a list with the following components:

cluster	inferred class labels
hZ	extracted latent features.
beta	estimated smoothing parameter
Mu	mean vectors of mixtures components.
Sigma	covariance matrix of mixtures components.
W	estimated loading matrix
Lam_vec	estimated variance of errors in probabilistic PCA model
loglik	pseudo observed log-likelihood.

Note

nothing

Author(s)

Wei Liu

References

Wei Liu, Xu Liao, Yi Yang, Huazhen Lin, Joe Yeong, Xiang Zhou*, Xingjie Shi* and Jin Liu* (2022). Joint dimension reduction and clustering analysis of single-cell RNA-seq and spatial transcriptomics data, *Nucleic Acids Research*

See Also

None

Examples

```
## we generate the spatial transcriptomics data with lattice neighborhood, i.e. ST platform.
seu <- gendata_RNAExp(height=10, width=10,p=50, K=4)
library(Seurat)
seu <- NormalizeData(seu, verbose=FALSE)
# choose 40 highly variable features using FindVariableFeatures in Seurat
# seu <- FindVariableFeatures(seu, nfeatures = 40)
# or choose 40 spatially variable features using FindSVGs in DR.SC
seu <- FindSVGs(seu, nfeatures = 40, verbose=FALSE)
# users define the adjacency matrix
Adj_sp <- getAdj(seu, platform = 'ST')
var.features <- seu@assays$RNA@var.features
X <- Matrix::t(seu[["RNA"]@data[var.features,])
# maxIter = 2 is only used for illustration, and user can use default.
drscList <- DR.SC_fit(X,Adj_sp=Adj_sp, K=4, maxIter=2, verbose=TRUE)
```

drscPlot

tSNE or UMAP plot visualization

Description

Intuitive way of visualizing how cell types changes across the spatial locations.

Usage

```
drscPlot(seu, dims=1:5, visu.method='tSNE',...)
```

Arguments

seu	an object of class "Seurat" obtained by DR.SC .
dims	a positive integer to specify the number of latent features for visualization.
visu.method	a string including 'tSNE' or "UMAP".
...	Other arguments passing to DimPlot function.

Details

Nothing

Value

return a ggplot2 object.

Note

nothing

Author(s)

Wei Liu

References

None

See Also

None

Examples

```
## we generate the spatial transcriptomics data with lattice neighborhood, i.e. ST platform.
seu <- gendata_RNAExp(height=10, width=10,p=50, K=4)
library(Seurat)
seu <- NormalizeData(seu)
# choose spatially variable features
seu <- FindSVGs(seu)

# use SVGs to fit DR.SC model
# maxIter = 2 is only used for illustration, and user can use default.
seu1 <- DR.SC(seu, K=4,platform = 'ST', maxIter = 2,verbose=FALSE)
drscPlot(seu1)
```

FindSVGs

*Find spatially variable genes***Description**

Identifies features that have spatially variation along spots.

Usage

```
FindSVGs(seu, nfeatures=2000, covariates=NULL, num_core=1, verbose=TRUE)
```

Arguments

seu	an object of class "Seurat".
nfeatures	a positive integer, means how many spatially variable genes to be chosen. If there are less than 2000 features in seu, then all features are identified.
covariates	a covariate matrix named control variable matrix whose number of rows is equal to the number of columns of seu.
num_core	an optional positive integer, specify the cores used for identifying the SVGs in parallel.
verbose	an optional logical value, whether output the related information.

Details

Nothing

Value

return a revised Seurat object by adding three columns named "is.SVGs", "order.SVGs" and "adjusted.pval.SVGs" in the meta.features of default Assay.

Note

nothing

References

Zhu, J., Sun, S., Zhou, X.: Spark-x: non-parametric modeling enables scalable and robust detection of spatialexpression patterns for large spatial transcriptomic studies. *Genome Biology* 22(1), 1–25 (2021)

See Also

[topSVGs](#)

Examples

```
seu <- gdata_RNAExp(height=20, width=20,p=200, K=4)

seu <- FindSVGs(seu, nfeatures=100)
topSVGs(seu)
```

gdata_RNAExp

Generate simulated data

Description

Generate simulated spatial transcriptomics data or scRNAseq data.

Usage

```
gdata_RNAExp(height=30, width=30, platform="ST", p =100, q=10, K=7,
              G=4,sigma2=1, tau=8, seed=1, view=FALSE)
```

Arguments

height, width	Height and width of lattice grids for generating spatial coordinates. $n = \text{height} * \text{width}$ cells for scRNAseq data
platform	set the platform for the simulated data, only support 'ST' and 'scRNAseq'.
p	number of genes to generate.
q	number of true latent features to generate gene expression
K	number of clusters (cell types).
seed	random seed for generate data
G	the number of neighbors. The latter must be one of $G = 4$ or $G = 8$, which respectively correspond to a first order and a second order dependency structure. By default, $G = 4$.
sigma2	Variance of error term in probabilistic PCA model.
tau	a positive factor of mixture mean values.
view	Logical value indicating whether the draw should be printed. Do not display the optional borders.

Details

Nothing

Value

return a "Seurat" object. If `platform="ST"`, then the metadata of this Seurat object will include two columns with names "row" and "col" which are the spatial coordinates; If `platform="scRNAseq"`, then the metadata of this Seurat object will not have them.

Note

nothing

Author(s)

Wei Liu

References

None

See Also

None

Examples

```
## we generate the spatial transcriptomics data with lattice neighborhood, i.e. ST platform.
seu <- gendata_RNAExp(height=20, width=20,p=200, K=4)
seu

## generate scRNAseq data
seu <- gendata_RNAExp(height=20, width=20, platform="scRNAseq", p=100, K=4)
seu
```

getAdj

Calculate the adjacency matrix given the spatial coordinates

Description

Calculate the adjacency matrix for the spatial transcriptomics data measured on 10X Visium or other platforms as a Seurat object.

Usage

```
getAdj(obj, platform = 'Visium')
## getAdj(obj, platform = c('Visium', 'ST', "seqfish",
## 'merfish', 'slide-seqv2', 'seqscope'))
## S3 method for class 'Seurat'
getAdj(obj,platform = 'Visium')
```

Arguments

obj	an object with class "Seurat", there are spatial coordinates information in the metadata of obj, named "row" and "col", where first column is x-axis coordinate, the second column is y-axis coordinate. getAdj_manual and getAdj_auto supports multi-dimensional spatial coordinates with a matrix as input.
platform	a string, specify the platform of the provided data, default as "Visium". There are many platforms to be supported, including ("Visium", "ST", "SeqFISH", 'merFISH', 'slide-seqv2', 'seqscope', "HDST"), which means there are spatial coordinates information in the metadata of seu, named "row" and "col". The platform helps to calculate the adjacency matrix by defining the neighborhoods.

Details

For lattice grids, i.e., two-dimensional coordinates, the interior spot has four neighbors (left, right, up and down), the boundary spot has three neighbors, and the spot in the corner has two neighbors. For hexagon grids, such as spatial coordinate in 10X Visium platform, the interior spot has six neighbors. More flexible definition can be used if there are some additional information. And then use [getAdj_manual](#) function to evaluate the adjacency matrix as a input for [DR.SC_fit](#) to run DR-SC model.

Value

Return a dgCMatrix object recording the information of neighborhoods about each spot.

Note

nothing

Author(s)

Wei Liu

References

None

See Also

[getAdj_auto](#), [getAdj_manual](#).

Examples

```
## S3 method for class "Seurat"
seu <- gendata_RNAExp(height=20, width=20, p=200, K=4)
Adj_sp <- getAdj(seu, platform = 'ST')
```

getAdj_auto

Calculate adjacency matrix by automatically choosing radius

Description

an efficient function to find the radius by bi-section method then find neighborhoods based on the matrix of position, which ensures that each spot has about lower.med~upper.med neighborhoods in the sense of median.

Usage

```
getAdj_auto(pos, lower.med=4, upper.med=6, radius.upper= 100)
```

Arguments

pos	a n-by-2 matrix of position.
lower.med	an integer, the lower bound of median number of neighbors among all spots.
upper.med	an integer, the upper bound of median number of neighbors among all spots.
radius.upper	a real, the upper bound of radius.

Value

A sparse adjacency matrix containing the neighbourhood.

See Also

[getAdj](#), [getAdj_manual](#).

getAdj_manual

Calculate adjacency matrix by user-specified radius

Description

an efficient function to find the neighborhood based on the matrix of position and a pre-defined radius.

Usage

```
getAdj_manual(pos, radius)
```

Arguments

pos	is a n-by-d matrix of position, where n is the number of spots, and d is the dimension of coordinates.
radius	is a threshold of Euclidean distance to decide whether a spot is an neighborhood of another spot. For example, if the Euclidean distance between spot A and B is less than radius, then A is taken as the neighbourhood of B.

Value

A sparse matrix containing the neighbourhood

See Also

[getAdj_auto](#), [getAdj](#).

getneighborhood_fast

getneighborhood_fast

Description

an efficient function to find the neighborhood based on the matrix of position and a pre-defined cutoff

Usage

```
getneighborhood_fast(x, radius)
```

Arguments

x is a n-by-2 matrix of position.
radius is a threshold of Euclidean distance to decide whether a spot is a neighborhood of another spot. For example, if the Euclidean distance between spot A and B is less than cutoff, then A is taken as the neighbourhood of B.

Value

A sparse matrix containing the neighbourhood

mbicPlot *MBIC plot visualization*

Description

Intuitive way of visualizing how modified BIC values changes across different number of clusters

Usage

```
mbicPlot(seu, criteria="MBIC")
```

Arguments

seu an object of class Seurat revised by [DR.SC](#) with argument K=NULL.
criteria a string specifying the information criteria such as AIC, BIC and MBIC to be plotted, default as MBIC.

Details

Nothing

Value

return a ggplot2 object.

Note

nothing

Author(s)

Wei Liu

References

None

See Also

None

Examples

```
## we generate the spatial transcriptomics data with lattice neighborhood, i.e. ST platform.
seu <- gendata_RNAExp(height=20, width=20,p=100, K=4)
library(Seurat)
seu <- NormalizeData(seu)
# choose spatially variable features
seu <- FindSVGs(seu)
## Just for illustrating the usage of mbicPlot
seu[["RNA"]][misc[['icMat']] <- data.frame(K=2:5, MBIC=c(105, 101, 99, 108))
mbicPlot(seu)
```

read10XVisium	<i>Read the spatial transcriptomics data measured on 10X Visium platform</i>
---------------	--

Description

Read the spatial transcriptomics data measured on 10X Visium platform as a Seurat object, where the spatial coordinates are saved in the metadata, named "row" and "col".

Usage

```
read10XVisium(dirname)
```

Arguments

dirname A string, the dictory of Visium datasets

Details

Nothing

Value

return a Seurat object.

Note

nothing

Author(s)

Wei Liu

References

None

See Also

None

Examples

```
## Not run:
## set your file directory, then read it.
data_name <- "D/HCC"
HCC1 <- read10XVisium(data_name)

## End(Not run)
```

readscRNAseq

Read the scRNAseq data measured on scRNA sequencing platform

Description

Read the single cell RNA sequencing data measured on scRNA sequencing platform as a Seurat object.

Usage

```
readscRNAseq(mtx, cells, features, ...)
```

Arguments

mtx	a string, ame or remote URL of the mtx file
cells	a string, Name or remote URL of the cells/barcodes file
features	a string, Name or remote URL of the features/genes file
...	the arguments passing to ReadMtx

Details

Nothing

Value

return a Seurat object including expression matrix.

Note

nothing

Author(s)

Wei Liu

References

None

See Also

None

Examples

```
## Not run:
### set the file directory, then read it.
seu <- readscRNAseq(mtx="GSM3755564_16_Liver_Treg_matrix.mtx.gz",
                    features='GSM3755564_16_Liver_Treg_genes.tsv.gz',
                    cells='GSM3755564_16_Liver_Treg_barcodes.tsv.gz' )
seu

## End(Not run)
```

RunWPCA*Run Weighted Principal Component Analysis*

Description

Run a weighted PCA dimensionality reduction

Usage

```
RunWPCA(object, q=15)
### S3 method for class "Seurat"
## RunWPCA(object, q=15)

### S3 method for class "matrix"
## RunWPCA(object, q=15)

### S3 method for class "dgCMatix"
## RunWPCA(object, q=15)
```

Arguments

object	an object named "Seurat", "matrix" or "dgCMatix". The object of class "Seurat" must include slot "scale.data".
q	an optional positive integer, specify the number of features to be extracted.

Details

Nothing

Value

For Seurat object, return a Seurat object. For object "matrix" and "dgCMatrix", return a object "matrix" with rownames same as the colnames of X, and colnames "WPCA1" to "WPCAq".

Note

nothing

Author(s)

Wei Liu

References

Bai, J. and Liao, Y. (2017). Inferences in panel data with interactive effects using large covariance matrices. *Journal of Econometrics*, 200(1):59–78.

See Also

None

Examples

```
## Not run:
library(Seurat)
seu <- gendata_RNAExp(height=20, width=20,p=100, K=4)
## log-normalization
seu <- NormalizeData(seu)
##
seu <- FindVariableFeatures(seu, nfeatures=80)
## Scale
seu <- ScaleData(seu)
## Run WPCA
seu <- RunWPCA(seu)
seu
## Run tSNE based on wpc
seu <- RunTSNE(seu, reduction='wpc')
seu
## Find SVGs
seu <- FindSVGs(seu, nfeatures=80)
(genes <- topSVGs(seu, ntop=10))
Idents(seu) <- factor(paste0("cluster", seu$true_clusters), levels=paste0("cluster",1:4))
RidgePlot(seu, features = genes[1:2], ncol = 2)
FeaturePlot(seu, features = genes[1:2], reduction = 'tsne' ,ncol=2)

## End(Not run)
```

selectModel	<i>Select the number of clusters</i>
-------------	--------------------------------------

Description

Select the number of clusters by specified criteria.

Usage

```
selectModel(obj, criteria = 'MBIC', pen.const=1)
## S3 method for class 'drscObject'
selectModel(obj, criteria = 'MBIC', pen.const=1)
## S3 method for class 'Seurat'
selectModel(obj, criteria = 'MBIC', pen.const=1)
```

Arguments

	S
	an object with class Seurat by DR.SC or class drscObject by DR.SC_fit .
obj	criteria a string, specify the criteria used for selecting the number of clusters, supporting "MBIC", "BIC" and "AIC".
pen.const	an optional positive value, the adjusted constant used in the MBIC criteria. It usually takes value between 0.1 to 1.

Value

For S3 method of Seurat, it return a revised "Seurat" object with updated `Ident(s)`, `spatial.drsc.cluster` in the metadata and `DimReduc` object named `dr-sc` in the slot reductions. For S3 method of `drscObject`, it returns a list with the following components:

bestK	the selected number of clusters.
cluster	inferred class labels
hZ	extracted latent features.
icMat	a numeric matrix including the criteria value for each number of clusters K.

Note

nothing

Author(s)

Wei Liu

References

None

See Also

[DR.SC](#), [DR.SC_fit](#).

Examples

```
seu <- gendata_RNAExp(height=10, width=10,p=50, K=4)
library(Seurat)
seu <- NormalizeData(seu, verbose=FALSE)
# or choose 40 spatailly variable features using FindSVGs in DR.SC
seu <- FindSVGs(seu, nfeatures = 40, verbose=FALSE)
# users define the adjacency matrix
Adj_sp <- getAdj(seu, platform = 'ST')
var.features <- seu@assays$RNA@var.features
X <- Matrix::t(seu[["RNA"]][@data[var.features,])
# maxIter = 2 is only used for illustration, and user can use default.
drscList <- DR.SC_fit(X,Adj_sp=Adj_sp ,K=4, maxIter=2, verbose=TRUE)
drsc1 <- selectModel(drscList)
str(drsc1)
```

spatialPlotClusters *Spatial coordinates plot visualization*

Description

Intuitive way of visualizing how cell types changes across the spatial locations.

Usage

```
spatialPlotClusters(seu)
```

Arguments

seu an object of class "Seurat" obtained by [DR.SC](#).

Details

Nothing

Value

return a ggplot2 object.

Note

nothing

Author(s)

Wei Liu

References

None

See Also

None

Examples

```
## we generate the spatial transcriptomics data with lattice neighborhood, i.e. ST platform.
seu <- gendata_RNAExp(height=10, width=10,p=50, K=4)
library(Seurat)
seu <- NormalizeData(seu)
# choose spatially variable features using Seurat
seu <- FindSVGs(seu)
# use SVGs to fit DR.SC model
# maxIter = 2 is only used for illustration, and user can use default.
seu1 <- DR.SC(seu, K=4,platform = 'ST', maxIter=2,verbose=FALSE)
spatialPlotClusters(seu1)
```

`sp_means_Rcpp`*Calculate column-wise or row-wise mean*

Description

Calculate column-wise or row-wise mean

Usage`sp_means_Rcpp(sp_data, rowMeans = FALSE)`**Arguments**

<code>sp_data</code>	A sparse matrix
<code>rowMeans</code>	A boolean value, whether to calculate row-wise mean

ValueA $n \times 1$ or $p \times 1$ matrix

sp_sums_Rcpp	<i>Calculate column-wise or row-wise sum</i>
--------------	--

Description

Calculate column-wise or row-wise sum

Usage

```
sp_sums_Rcpp(sp_data, rowSums = FALSE)
```

Arguments

sp_data	A sparse matrix
rowSums	A boolean value, whether to calculate row-wise sum

Value

A $n \times 1$ or $p \times 1$ matrix

topSVGs	<i>Return the top n SVGs</i>
---------	------------------------------

Description

Return top n spatially variable genes given a Seurat object performed by [FindSVGs](#).

Usage

```
topSVGs(seu, ntop=5)
```

Arguments

seu	an object of class "Seurat".
ntop	an optional positive integer, means how many spatially variable genes to access.

Details

Nothing

Value

return a [character](#) vector including the names of SVGs.

Note

nothing

Author(s)

Wei Liu

References

None

See Also

[topSVGs](#)

Examples

```
seu <- gendata_RNAExp(height=20, width=20,p=200, K=4)
seu <- FindSVGs(seu, nfeatures=100, verbose=FALSE)
(genes <- topSVGs(seu, ntop=10))
```

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