

# Package ‘PRECAST’

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**Type** Package

**Title** Embedding and Clustering with Alignment for Spatial Datasets

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**Description** An efficient data integration method is provided for multiple spatial transcriptomics data with non-cluster-relevant effects such as the complex batch effects. It unifies spatial factor analysis simultaneously with spatial clustering and embedding alignment, requiring only partially shared cell/domain clusters across datasets. More details can be referred to Wei Liu, et al. (2023) <[doi:10.1038/s41467-023-35947-w](https://doi.org/10.1038/s41467-023-35947-w)>.

**License** GPL-3

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**Imports** GiRaF, MASS, Matrix, mclust, methods, purrr, utils, Seurat, cowplot, patchwork, scater, pbapply, ggthemes, dplyr, ggplot2, stats, DR.SC, scales, ggpubr, graphics, colorspace, Rcpp (>= 1.0.5)

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---

AddAdjList

*Add adjacency matrix list for a PRECASTObj object*


---

**Description**

Add adjacency matrix list for a PRECASTObj object to prepare for PRECAST model fitting.

**Usage**

```
AddAdjList(PRECASTObj, type="fixed_distance", platform="Visium", ...)
```

**Arguments**

PRECASTObj	a PRECASTObj object created by <a href="#">CreatePRECASTObject</a> .
type	an optional string, specify which type of neighbors' definition. Here we provide two definition: one is "fixed_distance", the other is "fixed_number".
platform	a string, specify the platform of the provided data, default as "Visium". There are more platforms to be chosen, including "Visium", "ST" and "Other_SRT" ("Other_SRT" represents the other SRT platforms except for 'Visium' and 'ST'), which means there are spatial coordinates information in the metadata of PRECASTObj. The platform helps to calculate the adjacency matrix by defining the neighborhoods when type="fixed_distance" is chosen.
...	other arguments to be passed to <a href="#">getAdj</a> , <a href="#">getAdj_auto</a> and <a href="#">getAdj_fixedNumber</a> function.

**Details**

When the type = "fixed\_distance", then the spots within the Euclidean distance cutoffs from one spot are regarded as the neighbors of this spot. When the type = "fixed\_number", the K-nearest spots are regarded as the neighbors of each spot.

**Value**

Return a revised PRECASTObj object by adding the adjacency matrix list.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

[AddParSetting](#).

---

AddParSetting

*Add model settings for a PRECASTObj object*

---

**Description**

The main interface function provides several PRECAST submodels, so a model setting is required to specified in advance for a PRECASTObj object.

**Usage**

```
AddParSetting(PRECASTObj, ...)
```

**Arguments**

PRECASTObj      a PRECASTObj object created by [CreatePRECASTObject](#).  
 ...              other arguments to be passed to [model\\_set](#) function.

**Details**

Nothing

**Value**

Return a revised PRECASTObj object.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

**Examples**

```
data(PRECASTObj)
PRECASTObj <-AddParSetting(PRECASTObj)
PRECASTObj@parameterList
```

---

AddTSNE

*Add tSNE embeddings for a Seurat object*

---

**Description**

Run t-SNE dimensionality reduction on selected features.

**Usage**

```
AddTSNE(seuInt, n_comp=3, reduction='PRECAST', assay='PRE_CAST', seed=1)
```

**Arguments**

seuInt            a Seurat object.  
 n\_comp            an optional positive integer, specify the number of features to be extracted.  
 reduction        an optional string, means which dimensional reduction (e.g. PRECAST, PCA) to use for the tSNE. Default is PRECAST.  
 assay            Name of assay that that t-SNE is being run on.  
 seed             an optional integer, the random seed to evaluate tSNE.

**Details**

Nothing

**Value**

Return a revised Seurat object by adding tSNE reduction object.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

---

AddUMAP

*Add UMAP embeddings for a Seurat object*

---

**Description**

Run UMAP dimensionality reduction on selected features.

**Usage**

```
AddUMAP(seuInt, n_comp=3, reduction='PRECAST', assay='PRE_CAST', seed=1)
```

**Arguments**

seuInt	a Seurat object.
n_comp	an optional positive integer, specify the number of features to be extracted.
reduction	an optional string, means which dimensional reduction (e.g. PRECAST, PCA) to use for the UMAP. Default is PRECAST.
assay	Name of assay that that t-SNE is being run on.
seed	an optional integer, the random seed to evaluate UMAP.

**Details**

Nothing

**Value**

Return a revised Seurat object by adding UMAP reduction object.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

---

Add\_embed

*Add embeddings for a Seurat object*

---

**Description**

Add embeddings for a Seurat object.

**Usage**

```
Add_embed(embed, seu, embed_name='tSNE' , assay = "RNA")
```

**Arguments**

embed	an embedding matrix.
seu	a Seurat object.
embed_name	an optional string, the name of embeddings.
assay	Name of assay that that embed is being put

**Details**

Nothing

**Value**

Return a revised Seurat object by adding a embedding matrix to the Reduc slot in Seurat object.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

---

`boxPlot`*Boxplot for a matrix*

---

**Description**

Boxplot for a matrix.

**Usage**

```
boxPlot(mat, ylabel='ARI', cols=NULL, ...)
```

**Arguments**

<code>mat</code>	a matrix with columns.
<code>ylabel</code>	an optional string, the name of ylabel.
<code>cols</code>	colors used in the plot
<code>...</code>	Other parameters passed to <code>geom_boxplot</code> .

**Details**

Nothing

**Value**

Return a ggplot2 object.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

**Examples**

```
mat <- matrix(runif(100*3, 0.6, 1), 100, 3)
colnames(mat) <- paste0("Method", 1:3)
boxPlot(mat)
```

---

chooseColors	<i>Choose color schema from a palette</i>
--------------	---

---

**Description**

Choose color schema from a palette

**Usage**

```
chooseColors(  
  palettes_name = c("Nature 10", "Light 13", "Classic 20", "Blink 23", "Hue n"),  
  n_colors = 7,  
  alpha = 1,  
  plot_colors = FALSE  
)
```

**Arguments**

palettes_name	a string, the palette name, one of "Nature 10", "Light 13", "Classic 20", "Blink 23" and "Hue n", default as 'Nature 10'.
n_colors	a positive integer, the number of colors.
alpha	a positive real, the transparency of the color.
plot_colors	a logical value, whether plot the selected colors.

**Examples**

```
chooseColors()
```

---

coordinate_rotate	<i>Coordinates rotation for visualization</i>
-------------------	---

---

**Description**

Coordinates rotation for visualization.

**Usage**

```
coordinate_rotate(pos, theta=0)
```

**Arguments**

pos	a matrix, the n-by-d coordinates, where n is the number of coordinates, d is the dimension of coordinates.
theta	a real number, the angle for counter-clock-wise rotation.



**Details**

Nothing

**Value**

Return a rotated coordinate matrix.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

**Examples**

```
x <- 1:100
pos <- cbind(x, sin(pi/4*x))
oldpar <- par(mfrow = c(1,2))
plot(pos)
plot(coordinate_rotate(pos, 40))
par(oldpar)
```

---

CreatePRECASTObject    *Create the PRECAST object with preprocessing step.*

---

**Description**

Create the PRECAST object with preprocessing step.

**Usage**

```
CreatePRECASTObject(seuList, project = "PRECAST", gene.number=2000,
  selectGenesMethod='SPARK-X', numCores_sparkx=1,
  customGenelist=NULL, premin.spots = 20,
  premin.features=20, postmin.spots=15, postmin.features=15,
  rawData.preserve=FALSE, verbose=TRUE)
```

**Arguments**

<code>seuList</code>	a list consisting of Seurat objects, where each object is a SRT data batch. The default assay of each Seurat object will be used for data preprocessing and followed model fitting. The specified format about <code>seuList</code> argument can be referred to the details and example.
<code>project</code>	An optional string, name of the project, default as "PRECAST".
<code>gene.number</code>	an optional integer, the number of top spatially variable genes (SVGs) or highly variable genes (HVGs) to be chosen.
<code>selectGenesMethod</code>	an optional integer, the method to select genes for each sample. It supports 'SPARK-X' and 'HVGs' to select genes now. Users can provide self-selected genes using <code>customGenelist</code> argument.
<code>numCores_sparkx</code>	an optional integer, specify the number of CPU cores in SPARK package to use when selecting spatial genes.
<code>customGenelist</code>	an optional string vector, the list of user specified genes to be used for PRECAST model fitting. If this argument is given, SVGs/HVGs will not be selected.
<code>premin.spots</code>	An optional integer, the features (genes) are retained in raw data filtering step with at least <code>premin.spots</code> number of spots, default is 20.
<code>premin.features</code>	An optional integer, the locations are retained in raw data filtering step with at least <code>premin.features</code> number of nonzero-count features (genes), default is 20.
<code>postmin.spots</code>	An optional integer, the features (genes) are retained in filtering step after common genes selected among all data batches with at least <code>postmin.spots</code> number of spots, default is 15.
<code>postmin.features</code>	An optional integer, the locations are retained in filtering step after common genes selected among all data batches with at least <code>postmin.features</code> number of nonzero-count features (genes), default is 15.
<code>rawData.preserve</code>	An optional logical value, whether preserve the raw <code>seuList</code> data.
<code>verbose</code>	whether display the message in the creating process.

**Details**

`seuList` is a [list](#) with Seurat object as component, and each Seurat object includes the raw expression count matrix, spatial coordinates and meta data for each data batch, where the spatial coordinates information must be saved in the metadata of Seurat, named "row" and "col" for each data batch.

**Value**

Returns PRECAST object prepared for PRECAST model fitting. See [PRECASTObj-class](#) for more details.

**Examples**

```

data(PRECASTObj)
library(Seurat)
seuList <- PRECASTObj@seulist
## Check the input of seuList for create PRECAST object.
## Check the default assay for each data batch
lapply(seuList, DefaultAssay)
## Check the spatial coordinates in the meta data named "row" and "col".
head(seuList[[1]]@meta.data)
## Then create PRECAST object using this seuList.
## For convenience, we show the user-specified genes' list for creating PRECAST object.
## Users can use SVGs from SPARK-X or HVGs.
PRECASTObj2 <- CreatePRECASTObject(seuList,
  customGeneList= row.names(seuList[[1]]), verbose=FALSE)

```

---

dimPlot

*Low-dimensional embeddings' plot*


---

**Description**

Low-dimensional embeddings' plot colored by a specified meta data in the Seurat object.

**Usage**

```

dimPlot(seuInt, item=NULL, reduction=NULL, point_size=1, text_size=16,
  cols=NULL, font_family='', border_col="gray10",
  fill_col="white", ...)

```

**Arguments**

seuInt	an object named "Seurat".
item	the item used for coloring the plot in the meta data of seuInt object.
reduction	the reduction used for plot in the seuInt object. If reduction is null, the last added one is used for plotting.
point_size	the size of point in the scatter plot.
text_size	the text size in the plot.
cols	colors used in the plot
font_family	the font family used for the plot.
border_col	the border color in the plot.
fill_col	the color used in backgroup.
...	other arguments passed to <a href="#">plot_scatter</a>

**Details**

Nothing

**Value**

Return a ggplot2 object.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

**Examples**

```
data(PRECASTObj)
PRECASTObj <- SelectModel(PRECASTObj)
seuInt <- IntegrateSpaData(PRECASTObj, species='unknown')
dimPlot(seuInt, reduction = 'PRECAST')
## or use the Seurat::DimPlot(seuInt, reduction = 'PRECAST')
```

---

doHeatmap

*Heatmap for spots-by-feature matrix*

---

**Description**

Plot heatmap for a Seurat object with expression data.

**Usage**

```
doHeatmap(seu, features=NULL, cell_label='Cell type', grp_label = FALSE,
          pt_size=4, grp_color=NULL, ...)
```

**Arguments**

seu	an object named "Seurat". The object of class "Seurat" must include slot "scale.data".
features	an optional string vector, the features to be plotted.
cell_label	an optional string, the name of legend.
grp_label	an optional logical value, whether display the group names.
pt_size	the point size used in the plot
grp_color	the colors to use for the group color bar.
...	Other paramters passed to <a href="#">DoHeatmap</a> .

**Details**

Nothing

**Value**

Return a ggplot2 object.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

[featurePlot](#)

**Examples**

```
library(Seurat)
data(PRECASTObj)
PRECASTObj <- SelectModel(PRECASTObj)
seuInt <- IntegrateSpaData(PRECASTObj, species='unknown')
seuInt <- ScaleData(seuInt)
doHeatmap(seuInt, features=row.names(seuInt)[1:5])
```

---

drawFigs

*Draw a figure using a group of ggplot objects*

---

**Description**

Draw a figure using a group of ggplot objects

**Usage**

```
drawFigs(
  pList,
  layout.dim = NULL,
  common.legend = FALSE,
  legend.position = "right",
  ...
)
```

**Arguments**

<code>pList</code>	a list with component ggplot objects.
<code>layout.dim</code>	a integer vector with length 2, the layout of subplots in rows and columns.
<code>common.legend</code>	a logical value, whether use common legend for all subplots.
<code>legend.position</code>	a string, the position of legend.
<code>...</code>	other arguments that pass to <code>ggarrange</code> .

**Value**

return a new ggplot object.

---

featurePlot	<i>Spatial expression heatmap</i>
-------------	-----------------------------------

---

**Description**

Plot spatial heatmap for a feature of Seurat object with spatial transcriptomics data.

**Usage**

```
featurePlot(seu, feature=NULL, cols=NULL, pt_size=1, title_size =16, quant=0.5,
  assay='RNA' , reduction="position")
```

**Arguments**

<code>seu</code>	an object named "Seurat". The object of class "Seurat" must include slot "scale.data".
<code>feature</code>	an optional string, specify the name of feature to be plotted. If it is null, the first feature will be plotted.
<code>cols</code>	colors used in the plot
<code>pt_size</code>	the size of point in the spatial heatmap plot.
<code>title_size</code>	the title size used for the plot.
<code>quant</code>	the quantile value to generate the gradient color map.
<code>assay</code>	the assay selected for plot.
<code>reduction</code>	the Reduc object for plot.

**Details**

Nothing

**Value**

Return a ggplot2 object.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

**Examples**

```
library(Seurat)
data(PRECASTObj)
PRECASTObj <- SelectModel(PRECASTObj)
seuInt <- IntegrateSpaData(PRECASTObj, species='unknown')
seuInt <- ScaleData(seuInt)
featurePlot(seuInt, assay='PRE_CAST')
```

---

firstup

*Set the first letter of a string vector to capital*

---

**Description**

Set the first letter of a string vector to capital.

**Usage**

```
firstup(x)
```

**Arguments**

x                    a string vector.

**Details**

Nothing

**Value**

Return a string vector with first letter capital.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

**Examples**

```
x <- c("good", "Morning")
firstup(x)
```

---

getAdj\_fixedNumber      *Calculate adjacency matrix by user-specified number of neighbors*

---

**Description**

an efficient function to find the neighborhood based on the matrix of position and a user-specified number of neighbors of each spot.

**Usage**

```
getAdj_fixedNumber(pos, number=6)
```

**Arguments**

pos	is a n-by-d matrix of position, where n is the number of spots, and d is the dimension of coordinates.
number	is the number of neighbors of each spot. Euclidean distance to decide whether a spot is a neighborhood of another spot.

**Value**

A sparse matrix containing the neighbourhood.

**See Also**

[getAdj\\_auto](#), [getAdj](#).



---

getAdj_reg	<i>Calculate adjacency matrix for regular spatial coordinates.</i>
------------	--

---

**Description**

Calculate adjacency matrix for regular spatial coordinates from ST or Visium platform.

**Usage**

```
getAdj_reg(pos, platform= "Visium")
```

**Arguments**

pos	is a n-by-d matrix of position, where n is the number of spots, and d is the dimension of coordinates.
platform	a string, specify the platform of the provided data, default as "Visium", and only support "ST" and "Visium" platform.

**Value**

A sparse matrix containing the neighbourhood.

**See Also**

[getAdj\\_auto](#), [getAdj](#), [getAdj\\_fixedNumber](#).

---

Human_HK_genes	<i>Human housekeeping genes database</i>
----------------	--

---

**Description**

Human housekeeping genes database.

**Details**

This data is a [data.frame](#) and include the Human housekeeping genes information in the columns named "Gene" and "Ensembl".

ICM.EM

*ICM-EM algorithm implementation***Description**

ICM-EM algorithm for fitting PRECAST model

**Usage**

```
ICM.EM(XList, q, K, AdjList=NULL, Adjlist_car=NULL, posList = NULL,
       platform = "ST", beta_grid=seq(0.2,4, by=0.2),maxIter_ICM=6,
       maxIter=20, epsLogLik=1e-5, verbose=TRUE,mix_prop_heter=TRUE,
       Sigma_equal=FALSE, Sigma_diag=TRUE,error_heter=TRUE, Sp2=TRUE,
       wpca_int=FALSE, int.model='EEE', seed=1,coreNum = 1, coreNum_int=coreNum)
```

**Arguments**

XList	an M-length list consisting of multiple matrices with class <code>dgMatrix</code> or <code>matrix</code> that specify the log-normalization gene expression matrix for each data sample used for PRECAST model.
q	a positive integer, specify the number of latent features to be extracted, default as 15.
K	a positive integer allowing scalar or vector, specify the number of clusters in model fitting.
AdjList	an M-length list of sparse matrices with class <code>dgMatrix</code> , specify the adjacency matrix used for Potts model in PRECAST. We provide this interface for those users who would like to define the adjacency matrix by their own.
Adjlist_car	an M-length list of sparse matrices with class <code>dgMatrix</code> , specify the adjacency matrix used for CAR model in PRECAST, default as <code>AdjList</code> in the Potts model. We provide this interface for those users who would like to use the different adjacency matrix in CAR model.
posList	an M-length list composed by spatial coordinate matrix for each data sample.
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"). If <code>AdjList</code> is not given, the <code>platform</code> helps to calculate the adjacency matrix by defining the neighbors.
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_ICM	an optional positive value, represents the maximum iterations of ICM.
maxIter	an optional positive value, represents the maximum iterations of EM.
epsLogLik	an optional positive value, tolerance value 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.

<code>mix_prop_heter</code>	an optional logical value, specify whether betas are distinct, default as TRUE.
<code>Sigma_equal</code>	an optional logical value, specify whether Sigmas are equal, default as FALSE.
<code>Sigma_diag</code>	an optional logical value, specify whether Sigmas are diagonal matrices, default as TRUE.
<code>error_heter</code>	an optional logical value, whether use the heterogeneous error for DR-SC model, default as TRUE. If <code>error_heter=FALSE</code> , then the homogeneous error is used for probabilistic PCA model in PRECAST.
<code>Sp2</code>	an optional logical value, whether add the ICAR model component in the model, default as TRUE. We provide this interface for those users who don't want to include the ICAR model.
<code>wpca_int</code>	an optional logical value, means whether use the weighted PCA to obtain the initial values of loadings and other parameters, default as FALSE which means the ordinary PCA is used.
<code>int.model</code>	an optional string, specify which Gaussian mixture model is used in evaluating the initial values for PRECAST, default as "EEE"; and see <code>Mclust</code> for more models' names.
<code>seed</code>	an optional integer, the random seed in fitting PRECAST model.
<code>coreNum</code>	an optional positive integer, means the number of threads used in parallel computing.
<code>coreNum_int</code>	an optional positive integer, means the number of cores used in parallel computation for initial values when <code>K</code> is a vector, default as same as <code>coreNum</code> .

### Details

Nothing

### Value

ICM.EM returns a [list](#) with class "SeqKiDRSC\_Object" with the number of components equal to the length of `K`, where each component includes the model fitting results for one number of cluster and is a list consisting of following components:

<code>cluster</code>	an M-length list that includes the inferred class labels for each data sample.
<code>hZ</code>	an M-length list that includes the batch corrected low-dimensional embeddings for each data sample.
<code>hV</code>	an M-length list that includes the estimate of the ICAR component for each sample.
<code>Rf</code>	an M-length list that includes the posterior probability of domain clusters for each sample.
<code>beta</code>	an M-length vector that includes the estimated smoothing parameters for each sample.
<code>Mu</code>	mean vectors of mixture components.
<code>Sigma</code>	covariance matrix of mixture components.
<code>W</code>	estimated loading matrix
<code>Lam</code>	estimated variance of errors in probabilistic PCA model
<code>loglik</code>	pseudo observed log-likelihood.

**Note**

nothing

**Author(s)**

Wei Liu

**References**

Wei Liu, Liao, X., Luo, Z. et al, Jin Liu\* (2023). Probabilistic embedding, clustering, and alignment for integrating spatial transcriptomics data with PRECAST. *Nature Communications*, 14, 296

**See Also**

None

**Examples**

```
## we generate the spatial transcriptomics data with lattice neighborhood, i.e. ST platform.
library(Matrix)
q <- 10; K <- 4
data(PRECASTObj)
posList <- lapply(PRECASTObj@seulist, function(x) cbind(x$row, x$col))
AdjList <- lapply(posList, getAdj_reg, platform='ST')
XList <- lapply(PRECASTObj@seulist, function(x) t(x[['RNA']]@data))
XList <- lapply(XList, scale, scale=FALSE)
## For illustration, maxIter is set to 4
resList <- ICM.EM(XList,AdjList = AdjList, maxIter=4,
                 q=q, K=K, verbose=TRUE)
```

---

ICM.EM\_structure

*ICM-EM algorithm implementation with organized paramters*

---

**Description**

Efficient data integration as well as spatial clustering for multiple spatial transcriptomics data

**Usage**

```
ICM.EM_structure(XList, K, AdjList, q=15,parameterList=NULL)
```

**Arguments**

XList	an M-length list consisting of multiple matrices with class <code>dgMatrix</code> or <code>matrix</code> that specify the log-normalization gene expression matrix for each data sample used for PRECAST model.
K	a positive integer allowing scalar or vector, specify the number of clusters in model fitting.
AdjList	an M-length list of sparse matrices with class <code>dgMatrix</code> , specify the adjacency matrix used for Potts model and Intrinsic CAR model in PRECAST model. We provide this interface for those users who would like to define the adjacency matrix by their own.
q	a positive integer, specify the number of latent features to be extracted, default as 15.
parameterList	Other arguments in PRECAST model, it can be set by <a href="#">model_set</a> .

**Details**

Nothing

**Value**

ICM.EM\_structure returns a [list](#) with class "SeqK\_PRECAST\_Object" with the number of components equal to the length of K, where each component includes the model fitting results for one number of cluster and is a list consisting of following components:

cluster	an M-length list that includes the inferred class labels for each data sample.
hZ	an M-length list that includes the batch corrected low-dimensional embeddings for each data sample.
hV	an M-length list that includes the estimate the ICAR component for each sample.
Rf	an M-length list that includes the posterior probability of domain clusters for each sample.
beta	an M-length vector that includes the estimated smoothing parameters for each sample.
Mu	mean vectors of mixtures components.
Sigma	covariance matrix of mixtures components.
W	estimated loading matrix
Lam	estimated variance of errors in probabilistic PCA model
loglik	pseudo observed log-likelihood.

**Note**

nothing

**Author(s)**

Wei Liu

**References**

Wei Liu, Liao, X., Luo, Z. et al, Jin Liu\* (2023). Probabilistic embedding, clustering, and alignment for integrating spatial transcriptomics data with PRECAST. *Nature Communications*, 14, 296

**See Also**

None

**Examples**

```
## we generate the spatial transcriptomics data with lattice neighborhood, i.e. ST platform.
library(Matrix)
q <- 10; K <- 4
data(PRECASTObj)
posList <- lapply(PRECASTObj@seulist, function(x) cbind(x$row, x$col))
AdjList <- lapply(posList, getAdj_reg, platform='ST')
XList <- lapply(PRECASTObj@seulist, function(x) t(x[['RNA']]@data))
XList <- lapply(XList, scale, scale=FALSE)
parList <- model_set(maxIter=4)
resList <- ICM.EM_structure(XList, AdjList = AdjList,
                           q=q, K=K, parameterList=parList)
```

---

IntegrateSpaData

*Integrate multiple SRT data*

---

**Description**

Integrate multiple SRT data based on the PRECASTObj by PRECAST model fitting.

**Usage**

```
IntegrateSpaData(PRECASTObj, species="Human",
                 custom_housekeep=NULL, covariates_use=NULL,
                 seuList=NULL, subsample_rate=1, sample_seed=1)
```

**Arguments**

PRECASTObj	a PRECASTObj object after finishing the PRECAST model fitting and model selection.
species	an optional string, one of 'Human', 'Mouse' and 'Unknown', specify the species of the SRT data to help choose the housekeeping genes. 'Unknown' means only using the PRECAST results reconstruct the alligned gene expression.
custom_housekeep	user-specified housekeeping genes.

covariates_use	a string vector, the colnames in 'PRECASTObj@seulist[[1]]@meta.data', representing other biological covariates to be considered when removing batch effects. This is achieved by adding additional covariates for biological conditions in the regression, such as case or control. Default as 'NULL', denoting no other covariates to be considered.
seuList	an optional Seurat list object, 'seuList' plays a crucial role in the integration process. If 'seuList' is set to 'NULL' and 'PRECASTObj@seuList' is not NULL, then 'seuList' will adopt the values of 'PRECASTObj@seuList'. Subsequently, the genes within 'seuList' will be utilized for integration. Conversely, if 'seuList' is not NULL, the integration will directly employ the genes specified within 'seuList'. In the event that both 'seuList' and 'PRECASTObj@seuList' are set to NULL, integration will proceed using the genes outlined in 'PRECASTObj@seulist', i.e., the variable genes. To preserve the 'seuList' not NULL in 'PRECASTObj@seuList', user can set 'rawData.preserve=TRUE' when running 'CreatePRECASTObject'. This parameter empowers users to integrate the entire set of genes in 'seuList' when implementing the integration, as opposed to exclusively considering the variable genes within 'PRECASTObj@seuList'.
subsample_rate	an optional real number ranging from zero to one, this parameter specifies the subsampling rate during integration to enhance computational efficiency, default as 1 (without subsampling).
sample_seed	an optional integer, with a default value of 1, serves to designate the random seed when 'subsample_rate' is set to a value less than one, ensuring reproducibility in the sampling process.

**Details**

Nothing

**Value**

Return a Seurat object by integrating all SRT data batches into a SRT data, where the column "batch" in the meta.data represents the batch ID, and the column "cluster" represents the clusters obtained by PRECAST.

**Note**

nothing

**Author(s)**

Wei Liu

**References**

Wei Liu, Liao, X., Luo, Z. et al, Jin Liu\* (2023). Probabilistic embedding, clustering, and alignment for integrating spatial transcriptomics data with PRECAST. *Nature Communications*, 14, 296

Gagnon-Bartsch, J. A., Jacob, L., & Speed, T. P. (2013). Removing unwanted variation from high dimensional data with negative controls. Berkeley: Tech Reports from Dep Stat Univ California, 1-112.

**See Also**

None

**Examples**

```
data(PRECASTObj)
PRECASTObj <- SelectModel(PRECASTObj)
seuInt <- IntegrateSpaData(PRECASTObj, species='unknown')
```

---

model\_set

*PRECAST model setting*


---

**Description**

Set the PRECAST model structure and parameters in the algorithm.

**Usage**

```
model_set(Sigma_equal=FALSE, Sigma_diag=TRUE, mix_prop_heter=TRUE,
          error_heter=TRUE, Sp2=TRUE, wpca_int=FALSE, int.model='EEE',
          coreNum = 1, coreNum_int=coreNum,
          beta_grid=seq(0.2,4, by=0.2),
          maxIter_ICM=6, maxIter=20, epsLogLik=1e-5, verbose=TRUE, seed=1)
```

**Arguments**

Sigma_equal	an optional logical value, specify whether Sigmaks are equal, default as FALSE.
Sigma_diag	an optional logical value, specify whether Sigmaks are diagonal matrices, default as TRUE.
mix_prop_heter	an optional logical value, specify whether betas are distinct, default as TRUE.
error_heter	an optional logical value, whether use the heterogeneous error i.e. $\lambda_{darj} \neq \lambda_{dark}$ for each sample $r$ , default as TRUE. If error_heter=FALSE, then the homogeneous error is used for probabilistic PCA model.
Sp2	an optional logical value, whether add the ICAR model component in the model, default as TRUE. We provide this interface for those users who don't want to include the ICAR model.
wpca_int	an optional logical value, means whether use the weighted PCA to obtain the initial values of loadings and other parameters, default as FALSE which means the ordinary PCA is used.
int.model	an optional string, specify which Gaussian mixture model is used in evaluating the initial values for PRECAST, default as "EEE"; and see Mclust for more models' names.
coreNum	an optional positive integer, means the number of threads used in parallel computing.



coreNum_int	an optional positive integer, means the number of cores used in parallel computation for initial values when K is a vector, default as same as coreNum.
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_ICM	an optional positive value, represents the maximum iterations of ICM.
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.
seed	an optional integer, the random seed in fitting PRECAST model.

**Details**

Nothing

**Value**

Return a [list](#) including all paramters' setting.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

**Examples**

```
model_set()
```

---

Mouse\_HK\_genes

*Mouse housekeeping genes database*

---

**Description**

Mouse housekeeping genes database.

**Details**

This data is a [data.frame](#) and include the mouse housekeeping genes information in the columns named "Gene" and "Ensembl".

---

plot_RGB	<i>Spatial RGB heatmap</i>
----------	----------------------------

---

**Description**

Plot spatial RGB heatmap.

**Usage**

```
plot_RGB(position, embed_3d, pointsize=2, textsize=15)
```

**Arguments**

position	a coordinates matrix with two columns: x-coordinate and y-coordinate.
embed_3d	a embedding matrix with three columns: x, y and z embeddings.
pointsize	the size of point in the scatter plot.
textsize	the text size in the plot.

**Details**

Nothing

**Value**

Return a ggplot2 object.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

---

`plot_scatter`*Scatter plot for two-dimensional embeddings*

---

**Description**

Scatter plot for two-dimensional embeddings

**Usage**

```
plot_scatter(embed_use, meta_data, label_name,
             xy_names=c('tSNE1', 'tSNE2'), no_guides = FALSE,
             cols = NULL,
             point_size = 0.5, point_alpha=1,
             base_size = 12, do_points = TRUE, do_density = FALSE, border_col='gray',
             legend_pos='right', legend_dir='vertical', nrow.legend=NULL)
```

**Arguments**

<code>embed_use</code>	an object named "Seurat", "maxtrix" or "dgCMatrix". The object of class "Seurat" must include slot "scale.data".
<code>meta_data</code>	an optional positive integer, specify the number of features to be extracted.
<code>label_name</code>	the size of point in the scatter plot.
<code>xy_names</code>	the text size in the plot.
<code>no_guides</code>	whether display the legend.
<code>cols</code>	colors used in the plot.
<code>point_size</code>	the point size of scatter plot.
<code>point_alpha</code>	the transparency of the plot.
<code>base_size</code>	the base text size.
<code>do_points</code>	Plot point plot.
<code>do_density</code>	Plot density plot
<code>border_col</code>	the border color in the plot.
<code>legend_pos</code>	the position of legend.
<code>legend_dir</code>	the direction of legend.
<code>nrow.legend</code>	the number of rows of legend.

**Details**

Nothing

**Value**

Return a ggplot2 object.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

**Examples**

```
embed_use <- cbind(1:100, sin((1:100)*pi/2))
meta_data <- data.frame(cluster=factor(rep(1:2, each=50)))
plot_scatter(embed_use, meta_data, label_name='cluster')
```

---

PRECAST

*Fit a PRECAST model*

---

**Description**

Fit a PRECAST model.

**Usage**

```
PRECAST(PRECASTObj, K=NULL, q= 15)
```

**Arguments**

PRECASTObj	an object named "PRECASTObj". The object PRECASTObj is created by <a href="#">CreatePRECASTObject</a> .
K	An optional integer or integer vector, specify the candidates of number of clusters. if K=NULL, it will be set to 4~12.
q	An optional integer, specify the number of low-dimensional embeddings to extract in PRECAST.

**Details**

The model fitting results are saved in the slot of resList.

**Value**

Return a revised PRECASTObj object.

**Note**

nothing

**Author(s)**

Wei Liu

**References**

Wei Liu, Liao, X., Luo, Z. et al, Jin Liu\* (2023). Probabilistic embedding, clustering, and alignment for integrating spatial transcriptomics data with PRECAST. *Nature Communications*, 14, 296

**See Also**

None

---

PRECASTObj	<i>A simple PRECASTObj for example</i>
------------	--

---

**Description**

A simple PRECASTObj for example.

**Details**

This PRECASTObj include the basic slots in PRECAST object; see [PRECASTObj-class](#) for more details.

---

PRECASTObj-class	<i>Each PRECASTObj object has a number of slots which store information.</i>
------------------	--

---

**Description**

Each PRECASTObj object has a number of slots which store information. Key slots to access are listed below.

**Slots**

`seuList` A list with Seurat object as component, representing the raw expression count matrix, spatial coordinates and meta data for each data batch, where the spatial coordinates information is saved in the metadata of Seurat, named "row" and "col" for each data batch.

`seulist` A Seurat list after the preprocessing step in preparation for PRECAST model.

`AdjList` The adjacency matrix list for a PRECASTObj object.

`parameterList` The model parameter settings for a PRECASTObj object

`resList` The results after fitting PRECAST models.

`project` Name of the project.

---

selectIntFeatures	<i>Select common genes for multiple data batches</i>
-------------------	--

---

**Description**

selectIntFeatures prioritizes genes based on the number of times they were selected as HVGs/SVGs in all data batches, and chose the top genes as the input for the analysis. We broke ties by examining the ranks of the tied genes in each original dataset and taking those with the highest median rank.

**Usage**

```
selectIntFeatures(seulist, spaFeatureList, IntFeatures=2000)
```

**Arguments**

seulist	a list consisting of Seurat objects, where each object is a SRT data batch.
spaFeatureList	an list consisting of SVGs vectors, where each vector is the top HVGs/SVGs for each SRT data batch.
IntFeatures	the number of common HVGs/SVGs genes to be chosen.

**Details**

Nothing

**Value**

Return a string vector, the selected gene list for integration in PRECAST.

**Note**

nothing

**Author(s)**

Wei Liu

**References**

Wei Liu, Liao, X., Luo, Z. et al, Jin Liu\* (2023). Probabilistic embedding, clustering, and alignment for integrating spatial transcriptomics data with PRECAST. Nature Communications, 14, 296

**See Also**

None

---

SelectModel	<i>Select best PRECAST model from candidated models</i>
-------------	---

---

**Description**

Select best PRECAST model from candidated models with different number of clusters.

**Usage**

```
## S3 method for class 'SeqK_PRECAST_Object'
SelectModel(obj, criteria = 'MBIC', pen_const=1, return_para_est=FALSE)
## S3 method for class 'PRECASTObj'
SelectModel(obj, criteria = 'MBIC', pen_const=1, return_para_est=FALSE)
```

**Arguments**

obj	a SeqK_PRECAST_Object or PRECASTObj object after PRECAST model fitting.
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.
return_para_est	an optional logical value, whether return the other paramters' estimators in PRECAST.

**Details**

Nothing

**Value**

Return a revised PRECASTObj object.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

**Examples**

```
data(PRECASTObj)
PRECASTObj <- SelectModel(PRECASTObj)
```

SpaPlot

*Spatial heatmap***Description**

Plot spatial heatmap for a Seurat object with spatial transcriptomics data.

**Usage**

```
SpaPlot(seuInt, batch=NULL, item=NULL, point_size=2, text_size=12,
        cols=NULL, font_family='', border_col="gray10",
        fill_col='white', ncol=2, combine = TRUE,
        title_name="Sample", ...)
```

**Arguments**

<code>seuInt</code>	an object named "Seurat".
<code>batch</code>	an optional positive integer or integer vector, specify the batches to be extracted. Users can check the batches' names by <code>unique(seuInt\$batch)</code> .
<code>item</code>	an optional string, which column is plotted in the meta data of <code>seuInt</code> . Users can check the meta data by <code>head(seuInt@meta.data)</code> . If <code>item</code> takes value from ("RGB_UMAP", "RGB_tSNE"), this function will plot the RGB plot.
<code>point_size</code>	the size of point in the scatter plot.
<code>text_size</code>	the text size in the plot.
<code>cols</code>	colors used in the plot
<code>font_family</code>	the font family used for the plot, default as Times New Roman.
<code>border_col</code>	the border color in the plot.
<code>fill_col</code>	the color used in background.
<code>ncol</code>	the number of columns in the layout of plots.
<code>combine</code>	an optional logical value, whether plot all on a figure. If TRUE, all figures are plotted; otherwise, return a list with each plot as component.
<code>title_name</code>	an optional string, title name in the plot.
<code>...</code>	other arguments passed to <a href="#">plot_scatter</a>
.	

**Details**

Nothing



**Value**

Return a ggplot2 object or list of ggplots objects.

**Note**

nothing

**Author(s)**

Wei Liu

**See Also**

None

**Examples**

```
data(PRECASTObj)
PRECASTObj <- SelectModel(PRECASTObj)
seuInt <- IntegrateSpaData(PRECASTObj, species='unknown')
SpaPlot(seuInt)
```

---

volinPlot

*Volin/boxplot plot*

---

**Description**

Plot volin/boxplot.

**Usage**

```
volinPlot(mat, ylabel='ARI', cols=NULL)
```

**Arguments**

mat	a matrix with columns.
ylabel	an optional string, the name of ylabel.
cols	colors used in the plot

**Details**

Nothing

**Value**

Return a ggplot2 object.

**Note**

nothing

**See Also**

None

**Examples**

```
mat <- matrix(runif(100*3, 0.6, 1), 100, 3)
colnames(mat) <- paste0("Method", 1:3)
volinPlot(mat)
```

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