# Package 'Voyager'

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Type Package

Title From geospatial to spatial omics

Version 1.6.0

Description SpatialFeatureExperiment (SFE) is a new S4 class for working with spatial single-cell genomics data. The voyager package implements basic exploratory spatial data analysis (ESDA) methods for SFE. Univariate methods include univariate global spatial ESDA methods such as Moran's I, permutation testing for Moran's I, and correlograms. Bivariate methods include Lee's L and cross variogram. Multivariate methods include MULTISPATI PCA and multivariate local Geary's C recently developed by Anselin. The Voyager package also implements plotting functions to plot SFE data and ESDA results.

**Depends** R (>= 4.2.0), SpatialFeatureExperiment (>= 1.5.2)

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calculateBivariate

Bivariate spatial statistics

# **Description**

These functions perform bivariate spatial analysis. In this version, the bivariate global method supported are lee, lee.mc, and lee.test from spdep, and cross variograms from gstat (use cross\_variogram and cross\_variogram\_map for type argument, see variogram\_internal). Global Lee statistic is computed by my own implementation that is much faster than that in spdep. Bivariate local methods supported are lee (use locallee for type argument) and localmoran\_bv a bivariate version of Local Moran in spdep.

```
## S4 method for signature 'ANY'
calculateBivariate(
  х,
  y = NULL,
  type,
  listw = NULL,
  coords_df = NULL,
  BPPARAM = SerialParam(),
  zero.policy = NULL,
  returnDF = TRUE,
  p.adjust.method = "BH",
  name = NULL,
)
## S4 method for signature 'SpatialFeatureExperiment'
calculateBivariate(
  Х,
  type,
  feature1,
```

```
feature2 = NULL,
  colGraphName = 1L,
  colGeometryName = 1L,
  sample_id = "all",
  exprs_values = "logcounts",
 BPPARAM = SerialParam(),
  zero.policy = NULL,
  returnDF = TRUE,
  p.adjust.method = "BH",
  swap_rownames = NULL,
  name = NULL,
)
runBivariate(
  Χ,
  type,
  feature1,
  feature2 = NULL,
  colGraphName = 1L,
  colGeometryName = 1L,
  sample_id = "all",
  exprs_values = "logcounts",
 BPPARAM = SerialParam(),
  swap_rownames = NULL,
  zero.policy = NULL,
  p.adjust.method = "BH",
  name = NULL,
  overwrite = FALSE,
)
```

## **Arguments**

Х

A numeric matrix whose rows are features/genes, or a numeric vector (then y must be specified), or a SpatialFeatureExperiment (SFE) object with such a matrix in an assay.

У

A numeric matrix whose rows are features/genes, or a numeric vector. Bivariate statics will be computed for all pairwise combinations of row names of x and row names of y, except in cross variogram where combinations within x and y are also computed.

type

An SFEMethod object, or a string matching the name of an SFEMethod object. The methods mentioned above correspond to SFEMethod objects already implemented in the Voyager package. Use listSFEMethods to see which methods are available. You can implement new SFEMethod objects to apply Voyager functions to other spatial analysis methods. This is in part inspired by the caret, parsnip, and BiocSingular packages.

listw

Weighted neighborhood graph as a spdep listw object. Not used when the

method specified in type does not use a spatial neighborhood graph, such as the

variogram.

coords\_df A sf data frame specifying location of each cell. Not used when the method

specified in type uses a spatial neighborhood graph. Must be specified other-

wise.

BPPARAM A BiocParallelParam object specifying whether and how computing the met-

ric for numerous genes shall be parallelized.

zero.policy default attr(listw, "zero.policy") as set when listw was created, if at-

tribute not set, use global option value; if TRUE assign zero to the lagged value

of zones without neighbours, if FALSE assign NA

returnDF Logical, when the results are not added to a SFE object, whether the results

should be formatted as a DataFrame.

p.adjust.method

Method to correct for multiple testing, passed to p. adjustSP. Methods allowed

are in p.adjust.methods.

name Name to use to store the results, defaults to the name in the SFEMethod object

passed to argument type. Can be set to distinguish between results from the

same method but with different parameters.

... Other arguments passed to S4 method (for convenience wrappers like calculateMoransI)

or method used to compute metrics as specified by the argument type (as in more general functions like calculateUnivariate). See documentation of functions with the same name as specified in type in the spdep package for

the method specific arguments. For variograms, see .variogram.

feature1 ID or symbol of the first genes in SFE object, for the argument x.

feature2 ID or symbol of the second genes in SFE object, for the argument x. Mandatory

if length of feature1 is 1.

colGraphName Name of the listw graph in the SFE object that corresponds to entities repre-

sented by columns of the gene count matrix. Use colGraphNames to look up names of the available graphs for cells/spots. Note that for multiple sample\_ids,

it is assumed that all of them have a graph of this same name.

colGeometryName

Name of a colGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use colGeometryNames to look up names of the sf data frames associated with cells/spots. In the SFE method of calculateUnivariate, this is to specify location of cells for methods that don't take a spatial neighborhood graph such as the variogram. If the geometry

is not of type POINT, then spatialCoords(x) is used instead.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

exprs\_values Integer scalar or string indicating which assay of x contains the expression val-

ues.

swap\_rownames Column name of rowData(object) to be used to identify features instead of

 ${\tt rownames} ({\tt object}) \ when \ labeling \ plot \ elements. \ If \ not \ found \ in \ {\tt rowData}, then$ 

rownames of the gene count matrix will be used.

overwrite Logical, whether to overwrite existing results with the same name. Defaults to

FALSE.

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#### Value

The calculateBivariate function returns a correlation matrix for global Lee, and the results for the each pair of genes for other methods. Global results are not stored in the SFE object. Some methods return one result for each pair of genes, while some return pairwise results for more than 2 genes jointly. Local results are stored in the localResults field in the SFE object, with name the concatenation the two gene names separated by two underscores (\_\_).

## **Examples**

```
library(SFEData)
library(scater)
library(scran)
library(SpatialFeatureExperiment)
library(SpatialExperiment)
sfe <- McKellarMuscleData()</pre>
sfe <- sfe[,sfe$in_tissue]</pre>
sfe <- logNormCounts(sfe)</pre>
gs <- modelGeneVar(sfe)</pre>
hvgs <- getTopHVGs(gs, fdr.threshold = 0.01)</pre>
g <- colGraph(sfe, "visium") <- findVisiumGraph(sfe)</pre>
# Matrix method
mat <- logcounts(sfe)[hvgs[1:5],]</pre>
df <- df2sf(spatialCoords(sfe), spatialCoordsNames(sfe))</pre>
out <- calculateBivariate(mat, type = "lee", listw = g)</pre>
out <- calculateBivariate(mat, type = "cross_variogram", coords_df = df)</pre>
# SFE method
out <- calculateBivariate(sfe, type = "lee",</pre>
feature1 = c("Myh1", "Myh2", "Csrp3"), swap_rownames = "symbol")
out2 <- calculateBivariate(sfe, type = "lee.test", feature1 = "Myh1",
feature2 = "Myh2", swap_rownames = "symbol")
sfe <- runBivariate(sfe, type = "locallee", feature1 = "Myh1",</pre>
feature2 = "Myh2", swap_rownames = "symbol")
```

calculateMultivariate Multivariate spatial data analysis

## **Description**

These functions perform multivariate spatial data analysis, usually spatially informed dimension reduction.

```
## S4 method for signature 'ANY,SFEMethod'
calculateMultivariate(
   x,
   type,
```

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```
listw = NULL,
  transposed = FALSE,
  zero.policy = TRUE,
  p.adjust.method = "BH",
)
## S4 method for signature 'ANY, character'
calculateMultivariate(x, type, listw = NULL, transposed = FALSE, ...)
## S4 method for signature 'SpatialFeatureExperiment, ANY'
calculateMultivariate(
  х,
  type,
  colGraphName = 1L,
  subset_row = NULL,
  exprs_values = "logcounts",
  sample_action = c("joint", "separate"),
  BPPARAM = SerialParam(),
)
runMultivariate(
  х,
  type,
  colGraphName = 1L,
  subset_row = NULL,
  exprs_values = "logcounts",
  sample_action = c("joint", "separate"),
  BPPARAM = SerialParam(),
  name = NULL,
  dest = c("reducedDim", "colData"),
)
```

#### **Arguments**

Х

A numeric matrix whose rows are features/genes, or a SpatialFeatureExperiment (SFE) object with such a matrix in an assay.

type

An SFEMethod object, or a string matching the name of an SFEMethod object. The methods mentioned above correspond to SFEMethod objects already implemented in the Voyager package. Use listSFEMethods to see which methods are available. You can implement new SFEMethod objects to apply Voyager functions to other spatial analysis methods. This is in part inspired by the caret, parsnip, and BiocSingular packages.

listw

Weighted neighborhood graph as a spdep listw object. Not used when the method specified in type does not use a spatial neighborhood graph, such as the variogram.

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transposed Logical, whether the matrix has genes in columns and cells in rows.

zero.policy default attr(listw, "zero.policy") as set when listw was created, if at-

tribute not set, use global option value; if TRUE assign zero to the lagged value

of zones without neighbours, if FALSE assign NA

p.adjust.method

 $Method\ to\ correct\ for\ multiple\ testing,\ passed\ to\ \verb"p.adjustSP".\ Methods\ allowed$ 

are in p.adjust.methods.

... Extra arguments passed to the specific multivariate method. For example, see

multispati\_rsp for arguments for MULTISPATI PCA. See localC for argu-

ments for "localC\_multi" and "localC\_perm\_multi".

colGraphName Name of the listw graph in the SFE object that corresponds to entities repre-

sented by columns of the gene count matrix. Use colGraphNames to look up names of the available graphs for cells/spots. Note that for multiple sample\_ids,

it is assumed that all of them have a graph of this same name.

subset\_row Vector specifying the subset of features to use for dimensionality reduction. This

can be a character vector of row names, an integer vector of row indices or a

logical vector.

ues.

sample\_action Character, either "joint" or "separate". Spatial methods depend on the spatial co-

ordinates and/or spatial neighborhood graph, which is why SpatialExperiment uses sample\_id to keep coordinates from different samples separate. Some spatial methods can be sensibly run jointly for multiple samples. In this case, "joint" will run the method jointly for all samples, and "separate" will run the method

separately for each sample and concatenate the results.

BPPARAM A BiocParallelParam object specifying whether and how computing the met-

ric for numerous genes shall be parallelized. This is to parallelize computation across multiple samples when there are a large number of samples. Be cautious if using an optimized BLAS for matrix operations that supports multithreading.

name Name to use to store the results, defaults to the name in the SFEMethod object

passed to argument type. Can be set to distinguish between results from the

same method but with different parameters.

dest Character, either "reducedDim" or "colData". If the output of the multivariate

method is a matrix or array, as in spatially informed dimension reduction, then the only option is "reducedDim", so the results will be stored in reducedDim of the SFE object. If the output is a vector, as in the multivariate version of localC, then it will be sotred in colData. Data frame output, such as from

localC\_perm, can be stored in either reducedDim or colData.

### **Details**

For the argument type, this package supports "multispati" for MULTISPATI PCA, "localC\_multi" for a multivariate generalization of Geary's C, "localC\_perm\_multi" for the multivariate Geary's C with permutation testing, and "gwpca" for geographically weighted PCA.

#### Value

In calculateMultivariate, a matrix for cell embeddings whose attributes include loadings and eigenvalues if relevant, ready to be added to the SFE object with reducedDim setter. For run\*, a SpatialFeatureExperiment object with the results added. See Details for where the results are stored.

## References

Dray, S., Said, S. and Debias, F. (2008) Spatial ordination of vegetation data using a generalization of Wartenberg's multivariate spatial correlation. Journal of vegetation science, 19, 45-56.

Anselin, L. (2019), A Local Indicator of Multivariate Spatial Association: Extending Geary's c. Geogr Anal, 51: 133-150. doi:10.1111/gean.12164

## **Examples**

```
# example code
library(SFEData)
library(scater)
library(scran)
sfe <- McKellarMuscleData()
sfe <- logNormCounts(sfe)
gvs <- modelGeneVar(sfe)
hvgs <- getTopHVGs(gvs, fdr.threshold = 0.05)
colGraph(sfe, "visium") <- findVisiumGraph(sfe)
sfe <- runMultivariate(sfe, "multispati", subset_row = hvgs)</pre>
```

calculateUnivariate Univariate spatial stiatistics

## Description

These functions compute univariate spatial statistics, both global and local, on matrices, data frames, and SFE objects. For SFE objects, the statistics can be computed for numeric columns of colData, colGeometries, and annotGeometries, and the results are stored within the SFE object. calculateMoransI and runMoransI are convenience wrappers for calculateUnivariate and runUnivariate respectively.

```
## S4 method for signature 'ANY,SFEMethod'
calculateUnivariate(
    x,
    type,
    listw = NULL,
    coords_df = NULL,
    BPPARAM = SerialParam(),
    zero.policy = NULL,
```

```
returnDF = TRUE,
  p.adjust.method = "BH",
  name = NULL,
)
## S4 method for signature 'ANY, character'
calculateUnivariate(
  х,
  type,
  listw = NULL,
  coords_df = NULL,
  BPPARAM = SerialParam(),
  zero.policy = NULL,
  returnDF = TRUE,
  p.adjust.method = "BH",
  name = NULL,
)
## S4 method for signature 'SpatialFeatureExperiment,ANY'
calculateUnivariate(
  х,
  type,
  features = NULL,
  colGraphName = 1L,
  colGeometryName = 1L,
  sample_id = "all",
  exprs_values = "logcounts",
  BPPARAM = SerialParam(),
  zero.policy = NULL,
  returnDF = TRUE,
  include_self = FALSE,
  p.adjust.method = "BH",
  swap_rownames = NULL,
  name = NULL,
)
## S4 method for signature 'ANY'
calculateMoransI(
  х,
 BPPARAM = SerialParam(),
 zero.policy = NULL,
  name = "moran"
)
```

```
## S4 method for signature 'SpatialFeatureExperiment'
calculateMoransI(
  х,
  features = NULL,
  colGraphName = 1L,
  colGeometryName = 1L,
  sample_id = "all",
  exprs_values = "logcounts",
  BPPARAM = SerialParam(),
  zero.policy = NULL,
  returnDF = TRUE,
  include_self = FALSE,
  p.adjust.method = "BH",
  swap_rownames = NULL,
  name = NULL,
)
colDataUnivariate(
  х,
  type,
  features,
  colGraphName = 1L,
  sample_id = "all",
 BPPARAM = SerialParam(),
  zero.policy = NULL,
  include_self = FALSE,
  p.adjust.method = "BH",
  name = NULL,
)
colDataMoransI(
  Х,
  features,
  colGraphName = 1L,
  sample_id = "all",
 BPPARAM = SerialParam(),
  zero.policy = NULL,
  include_self = FALSE,
  p.adjust.method = "BH",
  name = NULL,
)
colGeometryUnivariate(
  Х,
  type,
```

```
features,
  colGeometryName = 1L,
  colGraphName = 1L,
  sample_id = "all",
 BPPARAM = SerialParam(),
  zero.policy = NULL,
  include_self = FALSE,
  p.adjust.method = "BH",
  name = NULL,
)
colGeometryMoransI(
  features,
  colGeometryName = 1L,
  colGraphName = 1L,
  sample_id = "all",
 BPPARAM = SerialParam(),
  zero.policy = NULL,
  include_self = FALSE,
 p.adjust.method = "BH",
 name = NULL,
)
annotGeometryUnivariate(
 х,
  type,
  features,
  annotGeometryName = 1L,
  annotGraphName = 1L,
  sample_id = "all",
 BPPARAM = SerialParam(),
  zero.policy = NULL,
  include_self = FALSE,
  p.adjust.method = "BH",
 name = NULL,
)
annotGeometryMoransI(
 х,
  features,
  annotGeometryName = 1L,
  annotGraphName = 1L,
  sample_id = "all",
 BPPARAM = SerialParam(),
```

```
zero.policy = NULL,
  include_self = FALSE,
 p.adjust.method = "BH",
  name = NULL,
)
runUnivariate(
  Х,
  type,
  features = NULL,
  colGraphName = 1L,
  colGeometryName = 1L,
  sample_id = "all",
  exprs_values = "logcounts",
 BPPARAM = SerialParam(),
  swap_rownames = NULL,
  zero.policy = NULL,
  include_self = FALSE,
  p.adjust.method = "BH",
  name = NULL,
 overwrite = FALSE,
)
runMoransI(
  features = NULL,
  colGraphName = 1L,
  colGeometryName = 1L,
  sample_id = "all",
  exprs_values = "logcounts",
 BPPARAM = SerialParam(),
  swap_rownames = NULL,
  zero.policy = NULL,
  include_self = FALSE,
  p.adjust.method = "BH",
 name = NULL,
)
reducedDimUnivariate(
  Х,
  type,
  dimred = 1L,
  components = 1L,
  colGraphName = 1L,
  sample_id = "all",
```

```
BPPARAM = SerialParam(),
  zero.policy = NULL,
  include_self = FALSE,
  p.adjust.method = "BH",
  name = NULL,
)
reducedDimMoransI(
  dimred = 1L,
  components = 1L,
  colGraphName = 1L,
  sample_id = "all",
 BPPARAM = SerialParam(),
  zero.policy = NULL,
  include_self = FALSE,
  p.adjust.method = "BH",
  name = NULL,
)
```

#### **Arguments**

x A numeric matrix whose rows are features/genes, or a SpatialFeatureExperiment

(SFE) object with such a matrix in an assay.

type An SFEMethod object, or a string matching the name of an SFEMethod object.

The methods mentioned above correspond to SFEMethod objects already implemented in the Voyager package. Use listSFEMethods to see which methods are available. You can implement new SFEMethod objects to apply Voyager functions to other spatial analysis methods. This is in part inspired by the caret,

parsnip, and BiocSingular packages.

listw Weighted neighborhood graph as a spdep listw object. Not used when the

method specified in type does not use a spatial neighborhood graph, such as the

variogram.

coords\_df A sf data frame specifying location of each cell. Not used when the method

specified in type uses a spatial neighborhood graph. Must be specified other-

wise.

BPPARAM A BiocParallelParam object specifying whether and how computing the met-

ric for numerous genes shall be parallelized.

zero.policy default attr(listw, "zero.policy") as set when listw was created, if at-

tribute not set, use global option value; if TRUE assign zero to the lagged value

of zones without neighbours, if FALSE assign NA

returnDF Logical, when the results are not added to a SFE object, whether the results

should be formatted as a DataFrame.

p.adjust.method

Method to correct for multiple testing, passed to p.adjustSP. Methods allowed are in p.adjust.methods.

name Name to use to store the results, defaults to the name in the SFEMethod object

passed to argument type. Can be set to distinguish between results from the

same method but with different parameters.

... Other arguments passed to S4 method (for convenience wrappers like calculateMoransI)

or method used to compute metrics as specified by the argument type (as in more general functions like calculateUnivariate). See documentation of functions with the same name as specified in type in the spdep package for

(colData\*) or any colGeometry (colGeometry\*) or annotGeometry (annotGeometry\*)

the method specific arguments. For variograms, see .variogram.

features Genes (calculate\* SFE method and run\*) or numeric columns of colData(x)

for which the univariate metric is to be computed. Default to NULL. When NULL, then the metric is computed for all genes with the values in the assay specified in the argument exprs\_values. This can be parallelized with the argument BPPARAM. For genes, if the row names of the SFE object are Ensembl IDs, then the gene symbol can be used and converted to IDs behind the scene with a column in rowData can be specified in swap\_rownames. However, if one symbol matches multiple IDs, a warning will be given and the first match will be used. Internally, the results are always stored by the Ensembl ID rather than symbol.

colGraphName Name of the listw graph in the SFE object that corresponds to entities repre-

sented by columns of the gene count matrix. Use colGraphNames to look up names of the available graphs for cells/spots. Note that for multiple sample\_ids,

it is assumed that all of them have a graph of this same name.

colGeometryName

Name of a colGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use colGeometryNames to look up names of the sf data frames associated with cells/spots. In the SFE method of calculateUnivariate, this is to specify location of cells for methods that don't take a spatial neighborhood graph such as the variogram. If the geometry

is not of type POINT, then spatialCoords(x) is used instead.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

exprs\_values Integer scalar or string indicating which assay of x contains the expression val-

include\_self Logical, whether the spatial neighborhood graph should include edges from each

location to itself. This is for Getis-Ord Gi\* as in localG and localG\_perm, not to be used for any other method.

swap\_rownames Column name of rowData(object) to be used to identify features instead of

rownames(object) when labeling plot elements. If not found in rowData, then

rownames of the gene count matrix will be used.

annotGeometryName

Name of a annotGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use annotGeometryNames to look up

names of the sf data frames associated with annotations.

annotGraphName Name of the listw graph in the SFE object that corresponds to the annotGeometry of interest. Use annotGraphNames to look up names of available annotation

graphs.

overwrite Logical, whether to overwrite existing results with the same name. Defaults to

FALSE.

dimred Name of a dimension reduction, can be seen in reducedDimNames.

components Numeric vector of which components in the dimension reduction to compute

spatial statistics on.

#### **Details**

Most univariate methods in the package spdep are supported here. These methods are global, meaning returning one result for all spatial locations in the dataset: moran, geary, moran.mc, geary.mc, moran.test, geary.test, globalG.test, sp.correlogram. The variogram and variogram map from the gstat package are also supported.

The following methods are local, meaning each location has its own results: moran.plot, localmoran, localmoran\_perm, localC\_perm, localG, localG\_perm, LOSH, LOSH.mc, LOSH.cs. The GWmodel::gwss method will be supported soon, but is not supported yet.

Global results for genes are stored in rowData. For colGeometry and annotGeometry, the results are added to an attribute of the data frame called featureData, which is a DataFrame analogous to rowData for the gene count matrix, and can be accessed with the geometryFeatureData function. New column names in featureData would follow the same rules as in rowData. For colData, the results can be accessed with the colFeatureData function.

Local results are stored in the field localResults field of the SFE object, which can be accessed with localResults or localResult. If the results have p-values, then -log10 p and adjusted -log10 p are added. Note that in the multiple testing correction, p.adjustSP is used.

When the results are stored in the SFE object, parameters used to compute the results as well as to construct the spatial neighborhood graph are also added. For localResults, the parameters are added to the metadata field params of the localResults sorted by name, which defaults to the name in the SFEMethod object as specified in the type argument. For global methods, parameters for results for genes are in the metadata of rowData(x), organized by name (metadata(rowData(x))\*params[[name]]). For colData, the global method parameters are stored in metadata of colData in the field params (metadata(colData(x))\*params[[name]]). For geometries, the global method parameters are in an attribute named "params" of the corresponding sf data frame (attr(df, "params")[[name]]).

### Value

In calculateUnivariate, if returnDF = TRUE, then a DataFrame, otherwise a list each element of which is the results for each feature. For run\*, a SpatialFeatureExperiment object with the results added. See Details for where the results are stored.

#### References

Cliff, A. D., Ord, J. K. 1981 Spatial processes, Pion, p. 17.

Anselin, L. (1995), Local Indicators of Spatial Association-LISA. Geographical Analysis, 27: 93-115. doi:10.1111/j.1538-4632.1995.tb00338.x

Ord, J. K., & Getis, A. 2012. Local spatial heteroscedasticity (LOSH), The Annals of Regional Science, 48 (2), 529-539.

Ord, J. K. and Getis, A. 1995 Local spatial autocorrelation statistics: distributional issues and an application. Geographical Analysis, 27, 286-306

## **Examples**

```
library(SpatialFeatureExperiment)
library(SingleCellExperiment)
library(SFEData)
sfe <- McKellarMuscleData("small")</pre>
colGraph(sfe, "visium") <- findVisiumGraph(sfe)</pre>
features_use <- rownames(sfe)[1:5]</pre>
# Moran's I
moran_results <- calculateMoransI(sfe,</pre>
    features = features_use,
    colGraphName = "visium",
    exprs_values = "counts"
)
# This does not advocate for computing Moran's I on raw counts.
# Just an example for function usage.
sfe <- runMoransI(sfe,</pre>
    features = features_use, colGraphName = "visium",
    exprs_values = "counts"
# Look at the results
head(rowData(sfe))
# Local Moran's I
sfe <- runUnivariate(sfe,</pre>
    type = "localmoran", features = features_use,
    colGraphName = "visium", exprs_values = "counts"
head(localResult(sfe, "localmoran", features_use[1]))
# For colData
sfe <- colDataUnivariate(sfe,</pre>
    type = "localmoran", features = "nCounts",
    colGraphName = "visium"
)
head(localResult(sfe, "localmoran", "nCounts"))
# For annotGeometries
annotGraph(sfe, "myofiber_tri2nb") <-</pre>
    findSpatialNeighbors(sfe,
        type = "myofiber_simplified", MARGIN = 3L,
        method = "tri2nb", dist_type = "idw",
        zero.policy = TRUE
sfe <- annotGeometryUnivariate(sfe,</pre>
    type = "localG", features = "area",
    annotGraphName = "myofiber_tri2nb",
    annotGeometryName = "myofiber_simplified",
    zero.policy = TRUE
)
```

18 clusterCorrelograms

clusterCorrelograms

Find clusters of correlogram patterns

## **Description**

Cluster the correlograms to find patterns in length scales of spatial autocorrelation. All the correlograms clustered must be computed with the same method and have the same number of lags. Correlograms are clustered jointly across samples.

#### Usage

```
clusterCorrelograms(
    sfe,
    features,
    BLUSPARAM,
    sample_id = "all",
    method = "I",
    colGeometryName = NULL,
    annotGeometryName = NULL,
    reducedDimName = NULL,
    swap_rownames = NULL,
    name = "sp.correlogram")
```

# Arguments

sfe	A Spatia	ılFeatureExperim	ent object wi	th correlograms	computed for fea-
-----	----------	------------------	---------------	-----------------	-------------------

tures of interest.

BLUSPARAM A BlusterParam object specifying the algorithm to use.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

method "corr" for correlation, "I" for Moran's I, "C" for Geary's C

colGeometryName

Name of colGeometry from which to look for features.

annotGeometryName

Name of annotGeometry from which to look for features.

reducedDimName Name of a dimension reduction, can be seen in reducedDimNames. colGeometryName

and annotGeometryName have precedence over reducedDimName.

swap\_rownames Column name of rowData(object) to be used to identify features instead of

rownames(object) when labeling plot elements. If not found in rowData, then

rownames of the gene count matrix will be used.

name Name under which the correlogram results are stored, which is by default "sp.correlogram".

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#### Value

A data frame with 3 columns: feature for the features, cluster a factor for cluster membership of the features within each sample, and sample\_id for the sample.

#### **Examples**

```
library(SpatialFeatureExperiment)
library(SFEData)
library(bluster)
sfe <- McKellarMuscleData("small")
colGraph(sfe, "visium") <- findVisiumGraph(sfe)
inds <- c(1, 3, 4, 5)
sfe <- runUnivariate(sfe,
    type = "sp.correlogram",
    features = rownames(sfe)[inds],
    exprs_values = "counts", order = 5
)
clust <- clusterCorrelograms(sfe,
    features = rownames(sfe)[inds],
    BLUSPARAM = KmeansParam(2)
)</pre>
```

clusterMoranPlot

Find clusters on the Moran plot

# **Description**

The Moran plot plots the value at each location on the x axis, and the average of the neighbors of each locations on the y axis. Sometimes clusters can be seen on the Moran plot, indicating different types of neighborhoods.

### Usage

```
clusterMoranPlot(
   sfe,
   features,
   BLUSPARAM,
   sample_id = "all",
   colGeometryName = NULL,
   annotGeometryName = NULL,
   swap_rownames = NULL
)
```

# **Arguments**

sfe

A SpatialFeatureExperiment object with Moran plot computed for the feature of interest. If the Moran plot for that feature has not been computed for that feature in this sample\_id, it will be calculated and stored in rowData. See calculateUnivariate.

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features Features whose Moran plot are to be cluster. Features whose Moran plots have

not been computed will be skipped, with a warning.

BLUSPARAM A BlusterParam object specifying the algorithm to use.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

colGeometryName

Name of colGeometry from which to look for features.

annotGeometryName

Name of annotGeometry from which to look for features.

swap\_rownames Column name of rowData(object) to be used to identify features instead of

rownames(object) when labeling plot elements. If not found in rowData, then

rownames of the gene count matrix will be used.

#### Value

A data frame each column of which is a factor for cluster membership of each feature. The column names are the features.

# **Examples**

```
library(SpatialFeatureExperiment)
library(SingleCellExperiment)
library(SFEData)
library(bluster)
sfe <- McKellarMuscleData("small")
colGraph(sfe, "visium") <- findVisiumGraph(sfe)
# Compute moran plot
sfe <- runUnivariate(sfe,
    type = "moran.plot", features = rownames(sfe)[1],
    exprs_values = "counts"
)
clusts <- clusterMoranPlot(sfe, rownames(sfe)[1],
    BLUSPARAM = KmeansParam(2)
)</pre>
```

clusterVariograms

Cluster variograms of multiple features

# Description

This function clusters variograms of features across samples to find patterns in decays in spatial autocorrelation. The fitted variograms are clustered as different samples can have different distance bins.

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## Usage

```
clusterVariograms(
   sfe,
   features,
   BLUSPARAM,
   n = 20,
   sample_id = "all",
   colGeometryName = NULL,
   annotGeometryName = NULL,
   reducedDimName = NULL,
   swap_rownames = NULL,
   name = "variogram"
)
```

## **Arguments**

sfe A SpatialFeatureExperiment object with correlograms computed for fea-

tures of interest.

features Features whose correlograms to cluster.

BLUSPARAM A BlusterParam object specifying the algorithm to use.

n Number of points on the fitted variogram line.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

colGeometryName

Name of colGeometry from which to look for features.

annotGeometryName

Name of annotGeometry from which to look for features.

reducedDimName Name of a dimension reduction, can be seen in reducedDimNames. colGeometryName

and annotGeometryName have precedence over reducedDimName.

swap\_rownames Column name of rowData(object) to be used to identify features instead of

rownames (object) when labeling plot elements. If not found in rowData, then

rownames of the gene count matrix will be used.

name Name under which the correlogram results are stored, which is by default "sp.correlogram".

### Value

A data frame with 3 columns: feature for the features, cluster a factor for cluster membership of the features within each sample, and sample\_id for the sample.

## **Examples**

```
library(SFEData)
library(scater)
library(bluster)
library(Matrix)
sfe <- McKellarMuscleData()</pre>
```

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```
sfe <- logNormCounts(sfe)
# Just the highly expressed genes
gs <- order(Matrix::rowSums(counts(sfe)), decreasing = TRUE)[1:10]
genes <- rownames(sfe)[gs]

sfe <- runUnivariate(sfe, "variogram", features = genes)
clusts <- clusterVariograms(sfe, genes, BLUSPARAM = HclustParam(),
swap_rownames = "symbol")

# Plot the clustering
plotVariogram(sfe, genes, color_by = clusts, group = "feature",
use_lty = FALSE, swap_rownames = "symbol", show_np = FALSE)</pre>
```

ditto\_colors

Colorblind friendly palette from dittoSeq

### **Description**

Just to get the palette without having to install all those dependencies of dittoSeq.

#### **Usage**

ditto\_colors

#### **Format**

A character vector of hex colors of the palette. There are 40 colors.

# Source

The dittoSeq package.

ElbowPlot

Plot the elbow plot or scree plot for PCA

# Description

Apparently, there is no apparent way to plot the PC elbow plot other than extracting the variance explained attribute of the dimred slot, because even the OSCA book makes the elbow plot this way, which I find kind of cumbersome compared to Seurat. So I'm writing this function to make the elbow plot with SCE less cumbersome.

getDivergeRange 23

## Usage

```
ElbowPlot(
    sce,
    ndims = 20,
    nfnega = 0,
    reduction = "PCA",
    sample_id = "all",
    facet = FALSE,
    ncol = NULL
)
```

## **Arguments**

sce	$A  {\tt SingleCellExperiment}  object,  or  anything  that  inherits  from  {\tt SingleCellExperiment}.$
ndims	Number of components with positive eigenvalues, such as PCs in non-spatial PCA.
nfnega	Number of nega eigenvalues and their eigenvectors to compute. These indicate negative spatial autocorrelation.
reduction	Name of the dimension reduction to use. It must have an attribute called either "percentVar" or "eig" for eigenvalues. Defaults to "PCA".
sample_id	Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute metric for all samples; the metric is computed separately for each sample.
facet	Logical, whether to facet by samples when multiple samples are present. This is relevant when spatial PCA is run separately for each sample, which gives different results from running jointly for all samples.
ncol	Number of columns of facets if facetting.

## Value

A ggplot object. The y axis is eigenvalues or percentage of variance explained if relevant.

# **Examples**

```
library(SFEData)
library(scater)
sfe <- McKellarMuscleData("small")
sfe <- runPCA(sfe, ncomponents = 10, exprs_values = "counts")
ElbowPlot(sfe, ndims = 10)</pre>
```

 ${\tt getDivergeRange}$ 

Get beginning and end of palette to center a divergent palette

# Description

This function is no longer used internally as it's unnecessary for scico divergent palettes. But it can be useful when using divergent palettes outside scico where one must specify beginning and end but not midpoint, to override the default palette.

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#### Usage

```
getDivergeRange(values, diverge_center = 0)
```

## **Arguments**

```
values Numeric vector to be colored. diverge_center Value to center on, defaults to 0.
```

## Value

A numeric vector of length 2, the first element is for beginning, and the second for end. The values are between 0 and 1.

# Examples

```
v <- rnorm(10)
getDivergeRange(v, diverge_center = 0)</pre>
```

listSFEMethods

List all spatial methods in Voyager package

## **Description**

This package ships with many spatial statistics methods as SFEMethod objects. The user can adapt the uniform user interface of this package to other spatial methods by creating new SFEMethod objects. This function lists the names of all methods within Voyager, to use for the type argument in calculateUnivariate, calculateBivariate, and calculateMultivariate.

# Usage

```
listSFEMethods(variate = c("uni", "bi", "multi"), scope = c("global", "local"))
```

#### **Arguments**

```
variate Uni-, bi-, or multi-variate.
scope whether it's local or global.
```

#### Value

A data frame with a column for the name and another for a brief description.

# **Examples**

```
listSFEMethods("uni", "local")
```

listw2sparse 25

listw2sparse

Convert listw into sparse adjacency matrix

# Description

Edge weights are used in the adjacency matrix. Because most elements of the matrix are 0, using sparse matrix greatly reduces memory use.

#### Usage

```
listw2sparse(listw)
```

## **Arguments**

listw

A listw object for spatial neighborhood graph.

#### Value

A sparse dgCMatrix, whose row represents each cell or spot and whose columns represent the neighbors. The matrix does not have to be symmetric. If region.id is present in the listwobject, then it will be the row and column names of the output matrix.

#### **Examples**

```
library(SFEData)
sfe <- McKellarMuscleData("small")
g <- findVisiumGraph(sfe)
mat <- listw2sparse(g)</pre>
```

moranBounds

Compute the bounds of Moran's I given spatial neighborhood graph

## Description

Values Moran's I can take depends on the spatial neighborhood graph. The bounds of Moran's I given the graph, C, are given by the minimum and maximum eigenvalues of the double centered – i.e. subtracting column means and row means – adjacency matrix  $(I - \mathbb{K}\mathbb{K}^T/n)C(I - \mathbb{K}\mathbb{K}^T/n)$ , where  $\mathbb{K}$  is a vector of all 1's. This implementation follows the implementation in adespatial and uses the RSpectra package to more quickly find only the minimum and maximum eigenvalues without performing unnecessary work to find the full spectrum as done in base R's eigen.

```
moranBounds(listw)
```

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#### **Arguments**

listw

A listw object for spatial neighborhood graph.

#### Value

A numeric vector of minimum and maximum Moran's I given the spatial neighborhood graph.

#### Note

After double centering, the adjacency matrix is no longer sparse, so this function can take up a lot of memory for larger datasets.

#### References

de Jong, P., Sprenger, C., & van Veen, F. (1984). On extreme values of Moran's I and Geary's C. Geographical Analysis, 16(1), 17-24.

## **Examples**

```
# example code
library(SFEData)
sfe <- McKellarMuscleData("small")
g <- findVisiumGraph(sfe)
moranBounds(g)</pre>
```

moranPlot

Use ggplot to plot the moran.plot results

#### **Description**

This function uses ggplot2 to plot the Moran plot. The plot would be more aesthetically pleasing than the base R version implemented in spdep. In addition, contours are plotted to show point density on the plot, and the points can be colored by a variable, such as clusters. The contours may also be filled and only influential points plotted. When filled, the viridis E option is used.

```
moranPlot(
    sfe,
    feature,
    graphName = 1L,
    sample_id = "all",
    contour_color = "cyan",
    color_by = NULL,
    colGeometryName = NULL,
    annotGeometryName = NULL,
    plot_singletons = TRUE,
    binned = FALSE,
```

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```
filled = FALSE,
  divergent = FALSE,
  diverge_center = NULL,
  swap_rownames = NULL,
  bins = 100,
  binwidth = NULL,
  hex = FALSE,
  plot_influential = TRUE,
  bins_contour = NULL,
  name = "moran.plot",
   ...
)
```

## **Arguments**

sfe A SpatialFeatureExperiment object.

feature Name of one variable to show on the plot. It will be converted to sentence case

on the x axis and lower case in the y axis appended after "Spatially lagged". One feature at a time since the colors in color\_by may be specific to this feature (e.g.

from clusterMoranPlot).

graphName Name of the colGraph or annotGraph, the spatial neighborhood graph used to

compute the Moran plot. This is to determine which points are singletons to plot

differently on this plot.

sample\_id One sample\_id for the sample whose graph to plot.

contour\_color Color of the point density contours, which can be changed so the contours stand

out from the points.

color\_by Variable to color the points by. It can be the name of a column in colData, a gene,

or the name of a column in the colGeometry specified in colGeometryName. Or it can be a vector of the same length as the number of cells/spots in the sample\_id

of interest.

colGeometryName

Name of a colGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use colGeometryNames to look up names of

the sf data frames associated with cells/spots.

annotGeometryName

Name of a annotGeometry of the SFE object, to annotate the gene expression

plot.

plot\_singletons

Logical, whether to plot items that don't have spatial neighbors.

binned Logical, whether to plot 2D histograms. This argument has precedence to filled.

filled Logical, whether to plot filled contours for the non-influential points and only

plot influential points as points.

divergent Logical, whether a divergent palette should be used.

diverge\_center If divergent = TRUE, the center from which the palette should diverge. If NULL,

then not centering.

28 multispati\_rsp

swap\_rownames Column name of rowData(object) to be used to identify features instead of

 $\verb|rownames(object)| when labeling plot elements. If not found in \verb|rowData|, then \\$ 

rownames of the gene count matrix will be used.

bins If binning the colGeometry in space due to large number of cells or spots, the

number of bins, passed to <code>geom\_bin2d</code> or <code>geom\_hex</code>. If NULL (default), then the colGeometry is plotted without binning. If binning, a point geometry is recommended. If the geometry is not point, then the centroids will be used.

binwidth Width of bins, passed to geom\_bin2d or geom\_hex.

hex Logical, whether to use geom\_hex. Note that geom\_hex is broken in ggplot2

version 3.4.0. Please update ggplot2 if you are getting horizontal stripes when

hex = TRUE.

plot\_influential

Logical, whether to plot influential points with different palette if binned =

TRUE.

bins\_contour Number of bins in the point density contour. Use a smaller number to make

sparser contours.

name Name under which the Moran plot results are stored. By default "moran.plot".

... Other arguments to pass to geom\_density2d.

#### Value

A ggplot object.

## **Examples**

multispati\_rsp 29

## **Description**

This implementation uses the RSpectra package to efficiently compute a small subset of eigenvalues and eigenvectors, as a small subset is typically used. Hence it's much faster and memory efficient than the original implementation in adespatial. However, this implementation here does not support row and column weighting other than the standard ones for PCA., so the adespatial implementation is more general.

#### Usage

```
multispati_rsp(x, listw, nfposi = 30L, nfnega = 30L, scale = TRUE)
```

## **Arguments**

X	A matrix whose columns are features and rows are cells.
listw	A listw object, a spatial neighborhood graph for the cells in x. The length must be equal to the number of row of x.
nfposi	Number of positive eigenvalues and their eigenvectors to compute.
nfnega	Number of nega eigenvalues and their eigenvectors to compute. These indicate negative spatial autocorrelation.
scale	Logical, whether to scale the data.

#### Value

A matrix for the cell embeddings in each spatial PC, with attribute loading for the eigenvectors or gene loadings, and attribute eig for the eigenvalues.

#### Note

Eigen decomposition will fail if any feature has variance zero leading to NaN in the scaled matrix.

## References

Dray, S., Said, S. and Debias, F. (2008) Spatial ordination of vegetation data using a generalization of Wartenberg's multivariate spatial correlation. Journal of vegetation science, 19, 45-56.

#### **Examples**

```
library(SFEData)
library(scater)
sfe <- McKellarMuscleData("small")
sfe <- sfe[,sfe$in_tissue]
sfe <- logNormCounts(sfe)
inds <- order(rowSums(logcounts(sfe)), decreasing = TRUE)[1:50]
mat <- logcounts(sfe)[inds,]
g <- findVisiumGraph(sfe)
out <- multispati_rsp(t(mat), listw = g, nfposi = 10, nfnega = 10)</pre>
```

30 plotCellBin2D

multi\_listw2sparse

Convert multiple listw graphs into a single sparse adjacency matrix

# Description

Each sample in the SFE object has a separate spatial neighborhood graph. Spatial analyses performed jointly on multiple samples require a combined spatial neighborhood graph from the different samples, where the different samples would be disconnected components of the graph. This combined adjacency matrix can be used in MULTISPATI PCA.

## Usage

```
multi_listw2sparse(listws)
```

## **Arguments**

listws

A list of listw objects.

#### Value

A sparse dgCMatrix of the combined spatial neighborhood graph, with the original spatial neighborhood graphs of the samples on the diagonal. When the input is an SFE object, the rows and columns will match the column names of the SFE object.

#### **Examples**

```
# example code
```

plotCellBin2D

Plot cell density as 2D histogram

# **Description**

This function plots cell density in histological space as 2D histograms, especially helpful for larger smFISH-based datasets.

```
plotCellBin2D(
   sfe,
   sample_id = "all",
   bins = 200,
   binwidth = NULL,
   hex = FALSE,
   ncol = NULL,
   bbox = NULL
)
```

plotColDataFreqpoly 31

#### **Arguments**

sfe A SpatialFeatureExperiment object.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

bins Number of bins. Can be a vector of length 2 to specify for x and y axes sepa-

rately.

binwidth Width of bins, passed to geom\_bin2d or geom\_hex.

hex Logical, whether to use hexagonal bins.

ncol Number of columns if plotting multiple features. Defaults to NULL, which means

using the same logic as facet\_wrap, which is used by patchwork's wrap\_plots

by default.

A bounding box to specify a smaller region to plot, useful when the dataset is

large. Can be a named numeric vector with names "xmin", "xmax", "ymin", and "ymax", in any order. If plotting multiple samples, it should be a matrix with sample IDs as column names and "xmin", "ymin", "xmax", and "ymax" as row names. If multiple samples are plotted but bbox is a vector rather than a matrix, then the same bounding box will be used for all samples. You may see points at the edge of the geometries if the intersection between the bounding box and a geometry happens to be a point there. If NULL, then the entire tissue is plotted.

#### Value

A ggplot object.

#### **Examples**

```
library(SFEData)
sfe <- HeNSCLCData()
plotCellBin2D(sfe)</pre>
```

plotColDataFreqpoly

Plot frequency polygons for colData and rowData columns

#### **Description**

This function is recommended instead of plotColDataHistogram when coloring by multiple categories and log transforming the y axis, which causes problems in stacked histograms.

```
plotColDataFreqpoly(
   sce,
   feature,
   color_by = NULL,
   subset = NULL,
```

```
bins = 100,
  binwidth = NULL,
  linewidth = 1.2,
  scales = "free",
  ncol = 1,
  position = "identity"
)
plotRowDataFreqpoly(
  sce,
  feature,
  color_by = NULL,
  subset = NULL,
  bins = 100,
  binwidth = NULL,
  linewidth = 1.2,
  scales = "free",
  ncol = 1,
  position = "identity"
)
```

#### **Arguments**

sce A SingleCellExperiment object.

feature Names of columns in colData or rowData to plot. When multiple features are

specified, they will be plotted in separate facets.

color\_by Name of a categorical column in colData or rowData to color the polygons.

subset Name of a logical column to only plot a subset of the data.

bins Number of bins. Overridden by binwidth. Defaults to 30.

binwidth The width of the bins. Can be specified as a numeric value or as a function that

calculates width from unscaled x. Here, "unscaled x" refers to the original x values in the data, before application of any scale transformation. When specifying a function along with a grouping structure, the function will be called once per group. The default is to use the number of bins in bins, covering the range of the data. You should always override this value, exploring multiple widths to

find the best to illustrate the stories in your data.

The bin width of a date variable is the number of days in each time; the bin

width of a time variable is the number of seconds.

linewidth Line width of the polygons, defaults to a thicker 1.2.

scales Should scales be fixed ("fixed", the default), free ("free"), or free in one

dimension ("free\_x", "free\_y")?

ncol Number of columns in the facetting.

position A position adjustment to use on the data for this layer. This can be used in

various ways, including to prevent overplotting and improving the display. The

position argument accepts the following:

- The result of calling a position function, such as position\_jitter(). This method allows for passing extra arguments to the position.
- A string naming the position adjustment. To give the position as a string, strip the function name of the position\_ prefix. For example, to use position\_jitter(), give the position as "jitter".
- For more information and other ways to specify the position, see the layer position documentation.

#### See Also

plotColDataHistogram

# **Examples**

plotColDataHistogram Plot histograms for colData and rowData columns

# Description

Plot histograms for colData and rowData columns

```
plotColDataHistogram(
    sce,
    feature,
    fill_by = NULL,
    facet_by = NULL,
    subset = NULL,
    bins = 100,
    binwidth = NULL,
    scales = "free",
    ncol = 1,
    position = "stack",
    ...
)

plotRowDataHistogram(
    sce,
    feature,
```

```
fill_by = NULL,
facet_by = NULL,
subset = NULL,
bins = 100,
binwidth = NULL,
scales = "free",
ncol = 1,
position = "stack",
...
)
```

#### **Arguments**

position

sce A SingleCellExperiment object.

feature Names of columns in colData or rowData to plot. When multiple features are

specified, they will be plotted in separate facets.

fill\_by Name of a categorical column in colData or rowData to fill the histogram.

facet\_by

Column in colData or rowData to facet with. When multiple features are plotted, the features will be in different facets. In this case, setting facet\_by will call facet\_grid so the features are in rows and categories in facet\_by will be

columns

subset Name of a logical column to only plot a subset of the data.

bins Numeric vector giving number of bins in both vertical and horizontal directions.

Set to 100 by default.

binwidth The width of the bins. Can be specified as a numeric value or as a function that

calculates width from unscaled x. Here, "unscaled x" refers to the original x values in the data, before application of any scale transformation. When specifying a function along with a grouping structure, the function will be called once per group. The default is to use the number of bins in bins, covering the range of the data. You should always override this value, exploring multiple widths to

find the best to illustrate the stories in your data.

The bin width of a date variable is the number of days in each time; the bin

width of a time variable is the number of seconds.

Should scales be fixed ("fixed", the default), free ("free"), or free in one

dimension ("free\_x", "free\_y")?

ncol Number of columns in the facetting.

A position adjustment to use on the data for this layer. This can be used in various ways, including to prevent overplotting and improving the display. The position argument accepts the following:

- The result of calling a position function, such as position\_jitter(). This method allows for passing extra arguments to the position.
- A string naming the position adjustment. To give the position as a string, strip the function name of the position\_ prefix. For example, to use position\_jitter(), give the position as "jitter".
- For more information and other ways to specify the position, see the layer position documentation.

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. . .

Other arguments passed on to layer()'s params argument. These arguments broadly fall into one of 4 categories below. Notably, further arguments to the position argument, or aesthetics that are required can *not* be passed through . . . . Unknown arguments that are not part of the 4 categories below are ignored.

- Static aesthetics that are not mapped to a scale, but are at a fixed value and apply to the layer as a whole. For example, colour = "red" or linewidth = 3. The geom's documentation has an **Aesthetics** section that lists the available options. The 'required' aesthetics cannot be passed on to the params. Please note that while passing unmapped aesthetics as vectors is technically possible, the order and required length is not guaranteed to be parallel to the input data.
- When constructing a layer using a stat\_\*() function, the ... argument can be used to pass on parameters to the geom part of the layer. An example of this is stat\_density(geom = "area", outline.type = "both"). The geom's documentation lists which parameters it can accept.
- Inversely, when constructing a layer using a geom\_\*() function, the ... argument can be used to pass on parameters to the stat part of the layer.
   An example of this is geom\_area(stat = "density", adjust = 0.5). The stat's documentation lists which parameters it can accept.
- The key\_glyph argument of layer() may also be passed on through .... This can be one of the functions described as key glyphs, to change the display of the layer in the legend.

#### Value

A ggplot object

## See Also

plotColDataFreqpoly

# **Examples**

plotColGraph

Plot spatial graphs

# Description

A ggplot version of spdep::plot.nb, reducing boilerplate for SFE objects.

36 plotColGraph

#### Usage

```
plotColGraph(
  sfe,
  colGraphName = 1L,
  colGeometryName = 1L,
  sample_id = "all",
 weights = FALSE,
  segment_size = 0.5,
  geometry_size = 0.5,
  ncol = NULL,
  bbox = NULL
)
plotAnnotGraph(
  sfe,
  annotGraphName = 1L,
  annotGeometryName = 1L,
  sample_id = "all",
  weights = FALSE,
  segment_size = 0.5,
  geometry_size = 0.5,
 ncol = NULL,
  bbox = NULL
)
```

# **Arguments**

sfe A SpatialFeatureExperiment object.

colGraphName Name of graph associated with columns of the gene count matrix to be plotted.

colGeometryName

Name of a colGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use colGeometryNames to look up names of

the sf data frames associated with cells/spots.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

weights Whether to plot weights. If TRUE, then transparency (alpha) of the segments will

represent edge weights.

segment\_size Thickness of the segments that represent graph edges.

geometry\_size Point size (for POINT geometries) or line thickness (for LINESTRING and

POLYGON) to plot the geometry in the background.

ncol Number of columns if plotting multiple features. Defaults to NULL, which means

using the same logic as facet\_wrap, which is used by patchwork's  $wrap\_plots$ 

by default.

bbox A bounding box to specify a smaller region to plot, useful when the dataset is

large. Can be a named numeric vector with names "xmin", "xmax", "ymin", and "ymax", in any order. If plotting multiple samples, it should be a matrix with

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sample IDs as column names and "xmin", "ymin", "xmax", and "ymax" as row names. If multiple samples are plotted but bbox is a vector rather than a matrix, then the same bounding box will be used for all samples. You may see points at the edge of the geometries if the intersection between the bounding box and a geometry happens to be a point there. If NULL, then the entire tissue is plotted.

annotGraphName Name of the annotation graph to plot. annotGeometryName

Name of the annotGeometry, which is associated with the graph specified with annotGraphName, for spatial coordinates of the graph nodes and for context.

#### Value

A ggplot2 object.

## **Examples**

```
library(SpatialFeatureExperiment)
library(SFEData)
library(sf)
sfe <- McKellarMuscleData("small")</pre>
colGraph(sfe, "visium") <- findVisiumGraph(sfe)</pre>
plotColGraph(sfe, colGraphName = "visium", colGeometryName = "spotPoly")
# Make the myofiber segmentations a valid POLYGON geometry
ag <- annotGeometry(sfe, "myofiber_simplified")</pre>
ag <- st_buffer(ag, 0)
ag <- ag[!st_is_empty(ag), ]</pre>
annotGeometry(sfe, "myofiber_simplified") <- ag</pre>
annotGraph(sfe, "myofibers") <-</pre>
    findSpatialNeighbors(sfe,
        type = "myofiber_simplified", MARGIN = 3,
        method = "tri2nb", dist_type = "idw"
plotAnnotGraph(sfe,
    annotGraphName = "myofibers",
    annotGeometryName = "myofiber_simplified",
    weights = TRUE
)
```

plotCorrelogram

Plot correlogram

# **Description**

Use ggplot2 to plot correlograms computed by runUnivariate, pulling results from rowData. Correlograms of multiple genes with error bars can be plotted, and they can be colored by any numeric or categorical column in rowData or a vector with the same length as nrow of the SFE object. The coloring is useful when the correlograms are clustered to show types of length scales or patterns of decay of spatial autocorrelation. For method = "I", the error bars are twice the standard deviation of the estimated Moran's I value.

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## Usage

```
plotCorrelogram(
  sfe,
  features,
  sample_id = "all",
 method = "I",
  color_by = NULL,
  facet_by = c("sample_id", "features"),
  ncol = NULL,
  colGeometryName = NULL,
  annotGeometryName = NULL,
  reducedDimName = NULL,
 plot_signif = TRUE,
  p_adj_method = "BH",
  divergent = FALSE,
  diverge_center = NULL,
  swap_rownames = NULL,
  name = "sp.correlogram"
)
```

## **Arguments**

sfe A SpatialFeatureExperiment object.

features Features to plot, must be in rownames of the gene count matrix, colnames of

colData or a colGeometry, colnames of cell embeddings in reducedDim, or nu-

meric indices of dimension reduction components.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

method "corr" for correlation, "I" for Moran's I, "C" for Geary's C

color\_by Name of a column in rowData(sfe) or in the featureData of colData (see

colFeatureData), colGeometry, or annotGeometry by which to color the correlogram of each feature. Alternatively, a vector of the same length as features,

or a data frame from clusterCorrelograms.

facet\_by Whether to facet by sample\_id (default) or features. If facetting by sample\_id,

then different features will be plotted in the same facet for comparison. If facetting by features, then different samples will be compared for each feature.

Ignored if only one sample is specified.

ncol Number of columns if facetting.

colGeometryName

Name of a colGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use colGeometryNames to look up names of the sf data frames associated with cells/spots.

annotGeometryName

Name of a annotGeometry of the SFE object, to annotate the gene expression plot.

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reducedDimName Name of a dimension reduction, can be seen in reducedDimNames. colGeometryName and annotGeometryName have precedence over reducedDimName. Logical, whether to plot significance symbols: p < 0.001: \*\*\*, p < 0.01: \*\*, p plot\_signif < 0.05 \*, p < 0.1: ., otherwise no symbol. The p-values are two sided, based on the assumption that the estimated Moran's I is normally distributed with mean from a randomized version of the data. The mean and variance come from moran. test for Moran's I and geary. test for Geary's C. Take the results with a grain of salt if the data is not normally distributed. p\_adj\_method Multiple testing correction method as in p.adjust, to correct for multiple testing (number of lags times number of features) in the Moran's I estimates if plot\_signif = TRUE. divergent Logical, whether a divergent palette should be used. diverge\_center If divergent = TRUE, the center from which the palette should diverge. If NULL, then not centering. Column name of rowData(object) to be used to identify features instead of swap\_rownames rownames(object) when labeling plot elements. If not found in rowData, then

name Name under which the correlogram results are stored, which is by default "sp.correlogram".

rownames of the gene count matrix will be used.

#### Value

A ggplot object.

```
library(SpatialFeatureExperiment)
library(SFEData)
library(bluster)
library(scater)
sfe <- McKellarMuscleData("small")</pre>
sfe <- logNormCounts(sfe)</pre>
colGraph(sfe, "visium") <- findVisiumGraph(sfe)</pre>
inds <- c(1, 3, 4, 5)
features <- rownames(sfe)[inds]</pre>
sfe <- runUnivariate(sfe,</pre>
    type = "sp.correlogram", features = features,
    exprs_values = "counts", order = 5
clust <- clusterCorrelograms(sfe,</pre>
    features = features,
    BLUSPARAM = KmeansParam(2)
# Color by features
plotCorrelogram(sfe, features)
# Color by something else
plotCorrelogram(sfe, features, color_by = clust$cluster)
# Facet by features
plotCorrelogram(sfe, features, facet_by = "features")
```

40 plotCrossVariogram

plotCrossVariogram Plot cross variogram

# Description

Equivalent to gstat::plot.gstatVariogram, but using ggplot2 to be more customizable.

# Usage

```
plotCrossVariogram(res, show_np = TRUE)
```

# **Arguments**

res Cross variogram results for one sample, from calculateBivariate. Global

bivariate results are not stored in the SFE object.

show\_np Logical, whether to show number of pairs of cells at each distance bin.

## Value

A ggplot object. Unfortunately I haven't figured out a way to collect all the facet labels to the top of the entire plot.

#### See Also

plotCrossVariogramMap

```
library(SFEData)
library(scater)
sfe <- McKellarMuscleData()
sfe <- sfe[,sfe$in_tissue]
sfe <- logNormCounts(sfe)

res <- calculateBivariate(sfe, type = "cross_variogram",
feature1 = c("Myh1", "Myh2", "Csrp3"), swap_rownames = "symbol")
plotCrossVariogram(res)</pre>
```

plotCrossVariogramMap Plot cross variogram map

# Description

Equivalent to gstat::plot.gstatVariogram, but using ggplot2 to be more customizable.

## Usage

```
plotCrossVariogramMap(res, plot_np = FALSE)
```

# **Arguments**

res Cross variogram results for one sample, from calculateBivariate. Global

bivariate results are not stored in the SFE object.

plot\_np Logical, whether to plot the number of pairs in each distance bin instead of the

variance.

## Value

A ggplot object.

# See Also

plotCrossVariogram

```
library(SFEData)
library(scater)
sfe <- McKellarMuscleData()
sfe <- sfe[,sfe$in_tissue]
sfe <- logNormCounts(sfe)

res <- calculateBivariate(sfe, type = "cross_variogram_map",
feature1 = c("Myh1", "Myh2", "Csrp3"), swap_rownames = "symbol",
width = 500, cutoff = 2000)
plotCrossVariogramMap(res)</pre>
```

42 plotDimLoadings

plotDimLoadings

Plot top PC loadings of genes

## Description

Just like Seurat's VizDimLoadings function. I haven't found an equivalent for SCE but find it useful. But I'm not trying to reproduce that Seurat function exactly. For instance, I don't like it when Seurat imposes a ggplot theme, and I don't like the cowplot theme. Maybe I should rewrite it in base R but for now I'm using Tidyverse.

# Usage

```
plotDimLoadings(
    sce,
    dims = 1:4,
    nfeatures = 10,
    swap_rownames = NULL,
    reduction = "PCA",
    balanced = TRUE,
    ncol = 2,
    sample_id = "all"
)
```

#### **Arguments**

sce A SingleCellExperiment object, or anything that inherits from SingleCellExperiment.

dims Numeric vector specifying which PCs to plot. For MULTISPATI, PCs with

negative eigenvalues are in the right most columns of the embedding and loading

matrices. See the ElbowPlot.

nfeatures Number of genes to plot.

swap\_rownames Column name of rowData(object) to be used to identify features instead of

rownames (object) when labeling plot elements. If not found in rowData, then

rownames of the gene count matrix will be used.

reduction Name of the dimension reduction to use. It must have an attribute called either

"percentVar" or "eig" for eigenvalues. Defaults to "PCA".

balanced Return an equal number of genes with + and - scores. If FALSE, returns the top

genes ranked by the scores absolute values.

ncol Number of columns in the facetted plot.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

# Value

A ggplot object. Loadings for different PCs are plotted in different facets so one ggplot object is returned.

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# **Examples**

```
library(SFEData)
library(scater)
sfe <- McKellarMuscleData("small")
sfe <- runPCA(sfe, ncomponents = 10, exprs_values = "counts")
plotDimLoadings(sfe, dims = 1:2)</pre>
```

plotGeometry

Plot geometries without coloring

# Description

Different samples are plotted in separate facets.

# Usage

```
plotGeometry(
  sfe,
  type,
 MARGIN = 2L,
  sample_id = "all",
  fill = TRUE,
  ncol = NULL,
 bbox = NULL,
  image_id = NULL,
  channel = NULL,
 maxcell = 5e+05,
  show_axes = FALSE,
  dark = FALSE,
  palette = colorRampPalette(c("black", "white"))(255),
  normalize_channels = FALSE
)
```

# **Arguments**

sfe	A SpatialFeatureExperiment object.
type	Name of the geometry associated with the MARGIN of interest for which to compute the graph.
MARGIN	Just like in apply, where 1 stands for row, 2 stands for column. Here, in addition, 3 stands for annotation, to query the annotGeometries, such as nuclei segmentation in a Visium data
sample_id	Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute metric for all samples; the metric is computed separately for each sample.
fill	Logical, whether to fill polygons.

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ncol Number of columns if plotting multiple features. Defaults to NULL, which means

using the same logic as facet\_wrap, which is used by patchwork's wrap\_plots

by default.

bbox A bounding box to specify a smaller region to plot, useful when the dataset is large. Can be a named numeric vector with names "xmin", "xmax", "ymin", and

> "ymax", in any order. If plotting multiple samples, it should be a matrix with sample IDs as column names and "xmin", "ymin", "xmax", and "ymax" as row names. If multiple samples are plotted but bbox is a vector rather than a matrix, then the same bounding box will be used for all samples. You may see points at the edge of the geometries if the intersection between the bounding box and a

geometry happens to be a point there. If NULL, then the entire tissue is plotted.

ID of the image to plot behind the geometries. If NULL, then not plotting images. image\_id

> Use imgData to see image IDs present. To plot multiple grayscale images as different RGB channels, use a named vector here, whose names are channel names (r, g, b), and values are image\_ids of the corresponding images. The RGB colorization may not be colorblind friendly. When plotting multiple samples, it is assumed that the same image id is used for each channel across different

samples.

channel Numeric vector indicating which channels in a multi-channel image to plot. If

> NULL, grayscale is plotted if there is 1 channel and RGB for the first 3 channels. The numeric vector can be named (r, g, b) to indicate which channel maps to which color. The RGB colorization may not be colorblind friendly. This argument cannot be specified while image\_id is a named vector to plot different

grayscale images as different channels.

maxcell Maximum number of pixels to plot in the image. If the image is larger, it will

be resampled so it have less than this number of pixels to save memory and for faster plotting. We recommend reducing this number when plotting multiple

facets.

show axes Logical, whether to show axes.

dark Logical, whether to use dark theme. When using dark theme, the palette will

have lighter color represent higher values as if glowing in the dark. This is

intended for plotting gene expression on top of fluorescent images.

palette Vector of colors to use to color grayscale images.

normalize\_channels

Logical, whether to normalize each channel of the image individually. Should be FALSE for bright field color images such as H&E but should set to TRUE for

fluorescent images.

#### Value

A ggplot object.

```
library(SFEData)
sfe1 <- McKellarMuscleData("small")</pre>
sfe2 <- McKellarMuscleData("small2")</pre>
```

45 plotImage

```
sfe <- cbind(sfe1, sfe2)</pre>
sfe <- removeEmptySpace(sfe)</pre>
plotGeometry(sfe, "spotPoly")
plotGeometry(sfe, "myofiber_simplified", MARGIN = 3)
```

plotImage

Show image without plotting geometries

# **Description**

This function plots the images in SFE objects without plotting geometries. When showing axes, the numbers are coordinates within the image itself and have the same units as the spatial extent, but are not the actual spatial extent when plotting multiple samples to avoid excessive empty space.

## Usage

```
plotImage(
  sfe,
  sample_id = "all",
  image_id = NULL,
  channel = NULL,
  ncol = NULL,
 bbox = NULL,
 maxcell = 5e+05,
  show_axes = FALSE,
  dark = FALSE,
 palette = colorRampPalette(c("black", "white"))(255),
  normalize channels = FALSE
)
```

# **Arguments**

sfe A SpatialFeatureExperiment object.

Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute sample\_id

metric for all samples; the metric is computed separately for each sample.

image\_id ID of the image(s) to plot. If NULL, then the first image present is plotted. Can be a vector of IDs to use different grayscale images for different channels. The

vector can be named ('r', 'g', 'b'), to assign channels to images. The vector

must be named if it's length 2.

channel Numeric vector indicating which channels in a multi-channel image to plot. If NULL, grayscale is plotted if there is 1 channel and RGB for the first 3 channels.

The numeric vector can be named (r, g, b) to indicate which channel maps to which color. The RGB colorization may not be colorblind friendly. This argument cannot be specified while image\_id is a named vector to plot different

grayscale images as different channels.

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ncol Number of columns if plotting multiple features. Defaults to NULL, which means

using the same logic as facet\_wrap, which is used by patchwork's  $wrap_plots$ 

by default.

A bounding box to specify a smaller region to plot, useful when the dataset is

large. Can be a named numeric vector with names "xmin", "xmax", "ymin", and "ymax", in any order. If plotting multiple samples, it should be a matrix with sample IDs as column names and "xmin", "ymin", "xmax", and "ymax" as row names. If multiple samples are plotted but bbox is a vector rather than a matrix, then the same bounding box will be used for all samples. You may see points at the edge of the geometries if the intersection between the bounding box and a geometry happens to be a point there. If NULL, then the entire tissue is plotted.

maxcell Maximum number of pixels to plot in the image. If the image is larger, it will

be resampled so it have less than this number of pixels to save memory and for faster plotting. We recommend reducing this number when plotting multiple

facets.

show\_axes Logical, whether to show axes.

dark Logical, whether to use dark theme. When using dark theme, the palette will

have lighter color represent higher values as if glowing in the dark. This is intended for plotting gene expression on top of fluorescent images.

palette Vector of colors to use to color grayscale images.

normalize\_channels

Logical, whether to normalize each channel of the image individually. Should be FALSE for bright field color images such as H&E but should set to TRUE for

fluorescent images.

#### Value

A ggplot object.

```
library(SFEData)
library(SpatialFeatureExperiment)
fn <- XeniumOutput("v2", file_path = "xenium_example")
# Weird RBioFormats null pointer error the first time it's run
try(sfe <- readXenium(fn))
sfe <- readXenium(fn)
# Plot one channel
plotImage(sfe, image_id = "morphology_focus", channel = 1L)
plotImage(sfe, image_id = "morphology_focus", channel = 1L, show_axes = TRUE, dark = TRUE)
# Colorize based on different channels
plotImage(sfe, image_id = "morphology_focus", channel = c(2,4,1), show_axes = TRUE, dark = TRUE)
unlink("xenium_example", recursive = TRUE)</pre>
```

plotLocalResult

Plot local results

# **Description**

Plot results of local spatial analyses in space, such as local Getis-Ord Gi\* values.

# Usage

```
plotLocalResult(
  sfe,
  name,
  features,
  attribute = NULL,
  sample_id = "all",
  colGeometryName = NULL,
  annotGeometryName = NULL,
  ncol = NULL,
  ncol_sample = NULL,
  annot_aes = list(),
  annot_fixed = list(),
  bbox = NULL,
  image_id = NULL,
  channel = NULL,
 maxcell = 5e+05,
  aes_use = c("fill", "color", "shape", "linetype"),
  divergent = FALSE,
  diverge_center = NULL,
  annot_divergent = FALSE,
  annot_diverge_center = NULL,
  size = 0.5,
  shape = 16,
  linewidth = 0,
  linetype = 1,
  alpha = 1,
  color = "black",
  fill = "gray80",
  swap_rownames = NULL,
  scattermore = FALSE,
  pointsize = 0,
  bins = NULL,
  summary_fun = sum,
  hex = FALSE,
  show_axes = FALSE,
  dark = FALSE,
  palette = colorRampPalette(c("black", "white"))(255),
  normalize_channels = FALSE,
```

```
type = name,
)
```

#### **Arguments**

sfe A SpatialFeatureExperiment object.

Which local spatial results. Use localResultNames to see which types of rename

sults have already been calculated.

features Character vector of vectors. To see which features have the results of a given

type, see localResultFeatures.

attribute Which field in the local results of the type and features. If the result of each

> feature is a vector, the this argument is ignored. But if the result is a data frame or a matrix, then this is the column name of the result, such as "Ii" for local Moran's I. For each local spatial analysis method, there's a default attribute. See

Details. Use localResultAttrs.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

colGeometryName

Name of a colGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use colGeometryNames to look up names of the sf data frames associated with cells/spots.

annotGeometryName

Name of a annotGeometry of the SFE object, to annotate the gene expression

Number of columns if plotting multiple features. Defaults to NULL, which means ncol

using the same logic as facet\_wrap, which is used by patchwork's wrap\_plots

by default.

ncol\_sample If plotting multiple samples as facets, how many columns of such facets. This

> is distinct from ncols, which is for multiple features. When plotting multiple features for multiple samples, then the result is a multi-panel plot each panel of

which is a plot for each feature facetted by samples.

A named list of plotting parameters for the annotation sf data frame. The names annot\_aes

are which geom (as in ggplot2, such as color and fill), and the values are column

names in the annotation sf data frame. Tidyeval is NOT supported.

annot\_fixed Similar to annot\_aes, but for fixed aesthetic settings, such as color = "gray".

The defaults are the same as the relevant defaults for this function.

A bounding box to specify a smaller region to plot, useful when the dataset is

large. Can be a named numeric vector with names "xmin", "xmax", "ymin", and "ymax", in any order. If plotting multiple samples, it should be a matrix with sample IDs as column names and "xmin", "ymin", "xmax", and "ymax" as row names. If multiple samples are plotted but bbox is a vector rather than a matrix, then the same bounding box will be used for all samples. You may see points at the edge of the geometries if the intersection between the bounding box and a geometry happens to be a point there. If NULL, then the entire tissue is plotted.

bbox

image\_id ID of the image to plot behind the geometries. If NULL, then not plotting images.

Use imgData to see image IDs present. To plot multiple grayscale images as different RGB channels, use a named vector here, whose names are channel names (r, g, b), and values are image\_ids of the corresponding images. The RGB colorization may not be colorblind friendly. When plotting multiple samples, it is assumed that the same image\_id is used for each channel across different

samples.

channel Numeric vector indicating which channels in a multi-channel image to plot. If

NULL, grayscale is plotted if there is 1 channel and RGB for the first 3 channels. The numeric vector can be named (r, g, b) to indicate which channel maps to which color. The RGB colorization may not be colorblind friendly. This argument cannot be specified while <code>image\_id</code> is a named vector to plot different

grayscale images as different channels.

maxcell Maximum number of pixels to plot in the image. If the image is larger, it will

be resampled so it have less than this number of pixels to save memory and for faster plotting. We recommend reducing this number when plotting multiple

facets.

aes\_use Aesthetic to use for discrete variables. For continuous variables, it's always "fill"

for polygons and point shapes 21-25. For discrete variables, it can be fill, color, shape, or linetype, whenever applicable. The specified value will be changed to the applicable equivalent. For example, if the geometry is point but "linetype"

is specified, then "shaped" will be used instead.

divergent Logical, whether a divergent palette should be used.

diverge\_center If divergent = TRUE, the center from which the palette should diverge. If NULL,

then not centering.

annot\_divergent

Just as divergent, but for the annotGeometry in case it's different.

annot\_diverge\_center

Just as diverge\_center, but for the annotGeometry in case it's different.

size Fixed size of points. For points defaults to 0.5. Ignored if size\_by is specified.

shape Fixed shape of points, ignored if shape\_by is specified and applicable.

linewidth Width of lines, including outlines of polygons. For polygons, this defaults to 0,

meaning no outlines.

linetype Fixed line type, ignored if linetype\_by is specified and applicable.

alpha Transparency.

color Fixed color for colGeometry if color\_by is not specified or not applicable, or

for annotGeometry if annot\_color\_by is not specified or not applicable.

fill Similar to color, but for fill.

swap\_rownames Column name of rowData(object) to be used to identify features instead of

rownames(object) when labeling plot elements. If not found in rowData, then

rownames of the gene count matrix will be used.

scattermore Logical, whether to use the scattermore package to greatly speed up plotting

numerous points. Only used for POINT colGeometries. If the geometry is not POINT, then the centroids are used. Recommended for plotting hundreds of

thousands or more cells where the cell polygons can't be seen when plotted due to the large number of cells and small plot size such as when plotting multiple

panels for multiple features.

 $pointsize \qquad \qquad Radius \ of \ rasterized \ point \ in \ scatter more. \ Default \ to \ 0 \ for \ single \ pixels \ (fastest).$ 

bins If binning the colGeometry in space due to large number of cells or spots, the number of bins, passed to geom\_bin2d or geom\_hex. If NULL (default), then

the colGeometry is plotted without binning. If binning, a point geometry is recommended. If the geometry is not point, then the centroids will be used.

summary\_fun Function to summarize the feature value when the colGeometry is binned.

hex Logical, whether to use geom\_hex. Note that geom\_hex is broken in ggplot2

version 3.4.0. Please update ggplot2 if you are getting horizontal stripes when

hex = TRUE.

show\_axes Logical, whether to show axes.

dark Logical, whether to use dark theme. When using dark theme, the palette will

have lighter color represent higher values as if glowing in the dark. This is

intended for plotting gene expression on top of fluorescent images.

palette Vector of colors to use to color grayscale images.

normalize\_channels

Logical, whether to normalize each channel of the image individually. Should be FALSE for bright field color images such as H&E but should set to TRUE for

fluorescent images.

type An SFEMethod object or a string corresponding to the name of one of such ob-

jects in the environment. If the localResult of interest was manually added outside runUnivariate and runBivariate, so the method is not recorded, then the type argument can be used to specify the method to properly get the title and labels. By default, this argument is set to be the same as argument name. If the

method parameters are recorded, then the type argument is ignored.

... Other arguments passed to wrap\_plots.

#### **Details**

Many local spatial analyses return a data frame or matrix as the results, whose columns can be the statistic of interest at each location, its variance, expected value from permutation, p-value, and etc. The attribute argument specifies which column to use when there are multiple columns. Below are the defaults for each local method supported by this package what what they mean:

localmoran and localmoran\_perm Ii, local Moran's I statistic at each location.

localC\_perm localC, the local Geary C statistic at each location.

localG and localG\_perm localG, the local Getis-Ord Gi or Gi\* statistic. If include\_self = TRUE when calculateUnivariate or runUnivariate was called, then it would be Gi\*. Otherwise it's Gi.

LOSH and LOSH.mc Hi, local spatial heteroscedasticity

moran.plot wx, the average of the value of each neighbor of each location. Moran plot is best plotted as a scatter plot of wx vs x. See moranPlot.

Other local methods not listed above return vectors as results. For instance, localC returns a vector by default, which is the local Geary's C statistic.

#### Value

A ggplot2 object if plotting one feature. A patchwork object if plotting multiple features.

#### Note

While this function shares internals with plotSpatialFeature, there are some important differences. In plotSpatialFeature, the annotGeometry is indeed only used for annotation and the protagonist is the colGeometry, since it's easy to directly use ggplot2 to plot the data in annotGeometry sf data frames while overlaying annotGeometry and colGeometry involves more complicated code. In contrast, in this function, local results for annotGeometry can be plotted separately without anything related to colGeometry. Note that when annotGeometry local results are plotted without colGeometry, the annot\_\* arguments are ignored. Use the other arguments for aesthetics as if it's for colGeometry.

```
library(SpatialFeatureExperiment)
library(SFEData)
library(scater)
sfe <- McKellarMuscleData("small")</pre>
sfe <- sfe[,sfe$in_tissue]</pre>
colGraph(sfe, "visium") <- findVisiumGraph(sfe)</pre>
feature_use <- rownames(sfe)[1]</pre>
sfe <- logNormCounts(sfe)</pre>
sfe <- runUnivariate(sfe, "localmoran", feature_use)</pre>
# Which types of results are available?
localResultNames(sfe)
# Which features for localmoran?
localResultFeatures(sfe, "localmoran")
# Which columns does the localmoran results have?
localResultAttrs(sfe, "localmoran", feature_use)
plotLocalResult(sfe, "localmoran", feature_use, "Ii",
    colGeometryName = "spotPoly"
)
# For annotGeometry
# Make sure it's type POLYGON
annotGeometry(sfe, "myofiber_simplified") <-</pre>
    sf::st_buffer(annotGeometry(sfe, "myofiber_simplified"), 0)
annotGraph(sfe, "poly2nb_myo") <-</pre>
    findSpatialNeighbors(sfe,
        type = "myofiber_simplified", MARGIN = 3,
        method = "poly2nb", zero.policy = TRUE
sfe <- annotGeometryUnivariate(sfe, "localmoran",</pre>
    features = "area",
    annotGraphName = "poly2nb_myo",
    annotGeometryName = "myofiber_simplified",
    zero.policy = TRUE
plotLocalResult(sfe, "localmoran", "area", "Ii",
```

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```
annotGeometryName = "myofiber_simplified",
    size = 0.3, color = "cyan"
)
plotLocalResult(sfe, "localmoran", "area", "Z.Ii",
    annotGeometryName = "myofiber_simplified"
)
# don't use annot_* arguments when annotGeometry is plotted without colGeometry
```

plotMoranMC

Plot Moran/Geary Monte Carlo results

# Description

Plot the simulations as a density plot or histogram compared to the observed Moran's I or Geary's C, with ggplot2 so it looks nicer. Unlike the plotting function in spdep, this function can also plot the same feature in different samples as facets or plot different features or samples together for comparison.

## Usage

```
plotMoranMC(
    sfe,
    features,
    sample_id = "all",
    facet_by = c("sample_id", "features"),
    ncol = NULL,
    colGeometryName = NULL,
    annotGeometryName = NULL,
    reducedDimName = NULL,
    ptype = c("density", "histogram", "freqpoly"),
    swap_rownames = NULL,
    name = "moran.mc",
    ...
)
```

# Arguments

sfe	A SpatialFeatureExperiment object.
features	Features to plot, must be in rownames of the gene count matrix, colnames of colData or a colGeometry, colnames of cell embeddings in reducedDim, or numeric indices of dimension reduction components.
sample_id	Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute metric for all samples; the metric is computed separately for each sample.
facet_by	Whether to facet by sample_id (default) or features. If facetting by sample_id, then different features will be plotted in the same facet for comparison. If facetting by features, then different samples will be compared for each feature. Ignored if only one sample is specified.

ncol Number of columns if facetting.

colGeometryName

Name of a colGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use colGeometryNames to look up names of the sf data frames associated with cells/spots.

annotGeometryName

Name of a annot Geometry of the SFE object, to annotate the gene expression

plot.

reducedDimName Name of a dimension reduction, can be seen in reducedDimNames. colGeometryName

and annot Geometry Name have precedence over reduced  ${\tt DimName}$ .

ptype Plot type, one of "density", "histogram", or "freqpoly".

swap\_rownames Column name of rowData(object) to be used to identify features instead of

rownames(object) when labeling plot elements. If not found in rowData, then

rownames of the gene count matrix will be used.

name Name under which the Monte Carlo results are stored, which defaults to "moran.mc".

For Geary's C Monte Carlo, the default is "geary.mc".

... Other arguments passed to geom\_density, geom\_histogram, or geom\_freqpoly,

depending on ptype.

#### Value

A ggplot2 object.

## **Examples**

```
library(SpatialFeatureExperiment)
library(SFEData)
sfe <- McKellarMuscleData("small")
colGraph(sfe, "visium") <- findVisiumGraph(sfe)
sfe <- colDataUnivariate(sfe, type = "moran.mc", "nCounts", nsim = 100)
plotMoranMC(sfe, "nCounts")</pre>
```

plotSpatialFeature

Plot gene expression in space

## Description

Unlike Seurat and ggspavis, plotting functions in this package uses geom\_sf whenever applicable.

# Usage

```
plotSpatialFeature(
   sfe,
   features,
   colGeometryName = 1L,
   sample_id = "all",
```

```
ncol = NULL,
  ncol_sample = NULL,
  annotGeometryName = NULL,
  rowGeometryName = NULL,
  rowGeometryFeatures = NULL,
  annot_aes = list(),
  annot_fixed = list(),
  exprs_values = "logcounts",
  bbox = NULL,
  image_id = NULL,
  channel = NULL,
 maxcell = 5e+05,
  aes_use = c("fill", "color", "shape", "linetype"),
  divergent = FALSE,
  diverge_center = NA,
  annot_divergent = FALSE,
  annot_diverge_center = NA,
  size = 0.5,
  shape = 16,
  linewidth = 0,
  linetype = 1,
  alpha = 1,
  color = "black",
  fill = "gray80",
  swap_rownames = NULL,
  scattermore = FALSE,
  pointsize = 0,
  bins = NULL,
  summary_fun = sum,
  hex = FALSE,
  show_axes = FALSE,
  dark = FALSE,
  palette = colorRampPalette(c("black", "white"))(255),
  normalize_channels = FALSE,
)
```

#### **Arguments**

sfe A SpatialFeatureExperiment object.

features Features to plot, must be in rownames of the gene count matrix, colnames of

colData or a colGeometry.

colGeometryName

Name of a colGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use colGeometryNames to look up names of the sf data frames associated with cells/spots.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute metric for all samples; the metric is computed separately for each sample.

ncol Number of columns if plotting multiple features. Defaults to NULL, which means

using the same logic as facet\_wrap, which is used by patchwork's  $wrap_plots$ 

by default.

is distinct from ncols, which is for multiple features. When plotting multiple features for multiple samples, then the result is a multi-panel plot each panel of

which is a plot for each feature facetted by samples.

annotGeometryName

Name of a annotGeometry of the SFE object, to annotate the gene expression

plot.

rowGeometryName

Name of a rowGeometry of the SFE object to plot.

rowGeometryFeatures

Which features from rowGeometry to plot. Can only be a small number to avoid

overplotting. Different features are distinguished by point shape.

annot\_aes A named list of plotting parameters for the annotation sf data frame. The names

are which geom (as in ggplot2, such as color and fill), and the values are column

names in the annotation sf data frame. Tidyeval is NOT supported.

annot\_fixed Similar to annot\_aes, but for fixed aesthetic settings, such as color = "gray".

The defaults are the same as the relevant defaults for this function.

ues.

A bounding box to specify a smaller region to plot, useful when the dataset is

large. Can be a named numeric vector with names "xmin", "xmax", "ymin", and "ymax", in any order. If plotting multiple samples, it should be a matrix with sample IDs as column names and "xmin", "ymin", "xmax", and "ymax" as row names. If multiple samples are plotted but bbox is a vector rather than a matrix, then the same bounding box will be used for all samples. You may see points at the edge of the geometries if the intersection between the bounding box and a

geometry happens to be a point there. If NULL, then the entire tissue is plotted.

image\_id ID of the image to plot behind the geometries. If NULL, then not plotting images.

Use imgData to see image IDs present. To plot multiple grayscale images as different RGB channels, use a named vector here, whose names are channel names (r, g, b), and values are image\_ids of the corresponding images. The RGB colorization may not be colorblind friendly. When plotting multiple samples, it is assumed that the same image id is used for each channel across different

samples.

channel Numeric vector indicating which channels in a multi-channel image to plot. If

NULL, grayscale is plotted if there is 1 channel and RGB for the first 3 channels. The numeric vector can be named (r, g, b) to indicate which channel maps to which color. The RGB colorization may not be colorblind friendly. This argument cannot be specified while image\_id is a named vector to plot different

grayscale images as different channels.

maxcell Maximum number of pixels to plot in the image. If the image is larger, it will

be resampled so it have less than this number of pixels to save memory and for faster plotting. We recommend reducing this number when plotting multiple

facets.

Aesthetic to use for discrete variables. For continuous variables, it's always "fill" aes\_use

> for polygons and point shapes 21-25. For discrete variables, it can be fill, color, shape, or linetype, whenever applicable. The specified value will be changed to the applicable equivalent. For example, if the geometry is point but "linetype"

is specified, then "shaped" will be used instead.

divergent Logical, whether a divergent palette should be used.

diverge\_center If divergent = TRUE, the center from which the palette should diverge. If NULL,

then not centering.

annot\_divergent

Just as divergent, but for the annotGeometry in case it's different.

annot\_diverge\_center

Just as diverge\_center, but for the annotGeometry in case it's different.

size Fixed size of points. For points defaults to 0.5. Ignored if size\_by is specified.

Fixed shape of points, ignored if shape\_by is specified and applicable. shape

linewidth Width of lines, including outlines of polygons. For polygons, this defaults to 0,

meaning no outlines.

linetype Fixed line type, ignored if linetype\_by is specified and applicable.

alpha Transparency.

color Fixed color for colGeometry if color\_by is not specified or not applicable, or

for annotGeometry if annot\_color\_by is not specified or not applicable.

fill Similar to color, but for fill.

Column name of rowData(object) to be used to identify features instead of swap\_rownames

rownames(object) when labeling plot elements. If not found in rowData, then

rownames of the gene count matrix will be used.

Logical, whether to use the scattermore package to greatly speed up plotting scattermore

numerous points. Only used for POINT colGeometries. If the geometry is not POINT, then the centroids are used. Recommended for plotting hundreds of thousands or more cells where the cell polygons can't be seen when plotted due to the large number of cells and small plot size such as when plotting multiple

panels for multiple features.

pointsize Radius of rasterized point in scattermore. Default to 0 for single pixels (fastest).

bins If binning the colGeometry in space due to large number of cells or spots, the

> number of bins, passed to geom\_bin2d or geom\_hex. If NULL (default), then the colGeometry is plotted without binning. If binning, a point geometry is recommended. If the geometry is not point, then the centroids will be used.

Function to summarize the feature value when the colGeometry is binned. summary\_fun

hex

Logical, whether to use geom\_hex. Note that geom\_hex is broken in ggplot2

version 3.4.0. Please update ggplot2 if you are getting horizontal stripes when

hex = TRUE.

show\_axes Logical, whether to show axes.

dark Logical, whether to use dark theme. When using dark theme, the palette will

have lighter color represent higher values as if glowing in the dark. This is

intended for plotting gene expression on top of fluorescent images.

```
palette Vector of colors to use to color grayscale images. normalize_channels
```

Logical, whether to normalize each channel of the image individually. Should be FALSE for bright field color images such as H&E but should set to TRUE for fluorescent images.

... Other arguments passed to wrap\_plots.

#### **Details**

In the documentation of this function, a "feature" can be a gene (or whatever entity that corresponds to rows of the gene count matrix), a column in colData, or a column in the colGeometry sf data frame specified in the colGeometryName argument.

In the light theme, for continuous variables, the Blues palette from colorbrewer is used if divergent = FALSE, and the roma palette from the scico package if divergent = TRUE. In the dark theme, the nuuk palette from scico is used if divergent = FALSE, and the berlin palette from scico is used if divergent = TRUE. For discrete variables, the dittoSeq palette is used.

For annotation, the YlOrRd colorbrewer palette is used for continuous variables in the light theme. In the dark theme, the acton palette from scico is used when divergent = FALSE and the vanimo palette from scico is used when divergent = FALSE. The other end of the dittoSeq palette is used for discrete variables.

Each individual palette should be colorblind friendly, but when plotting continuous variables coloring a colGeometry and a annotGeometry simultaneously, the combination of the two palettes is not guaranteed to be colorblind friendly.

In addition, when plotting an image behind the geometries, the colors of the image may distort color perception of the values of the geometries.

theme\_void is used for all spatial plots in this package, because the units in the spatial coordinates are often arbitrary. This can be overriden to show the axes by using a different theme as normally done in ggplot2.

#### Value

A ggplot2 object if plotting one feature. A patchwork object if plotting multiple features.

```
library(SFEData)
library(sf)
sfe <- McKellarMuscleData("small")
# features can be genes or colData or colGeometry columns
plotSpatialFeature(sfe, c("nCounts", rownames(sfe)[1]),
        exprs_values = "counts",
        colGeometryName = "spotPoly",
        annotGeometryName = "tissueBoundary"
)
# Change fixed aesthetics
plotSpatialFeature(sfe, "nCounts",
        colGeometryName = "spotPoly",
        annotGeometryName = "tissueBoundary",</pre>
```

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```
annot_fixed = list(color = "blue", size = 0.3, fill = NA),
    alpha = 0.7
)
# Make the myofiber segmentations a valid POLYGON geometry
ag <- annotGeometry(sfe, "myofiber_simplified")</pre>
ag <- st_buffer(ag, 0)</pre>
ag <- ag[!st_is_empty(ag), ]</pre>
annotGeometry(sfe, "myofiber_simplified") <- ag</pre>
# Also plot an annotGeometry variable
plotSpatialFeature(sfe, "nCounts",
    colGeometryName = "spotPoly",
    annotGeometryName = "myofiber_simplified",
    annot_aes = list(fill = "area")
# Use a bounding box to zoom in
bbox <- c(xmin = 5500, ymin = 13500, xmax = 6000, ymax = 14000)
plotSpatialFeature(sfe, "nCounts", colGeometryName = "spotPoly",
                  annotGeometry = "myofiber_simplified",
                  bbox = bbox, annot_fixed = list(linewidth = 0.3))
```

plotVariogram

Plot variogram

## **Description**

This function plots the variogram of a feature and its fitted variogram models, showing the nugget, range, and sill of the model. Unlike the plotting functions in package automap that uses lattice, this function uses ggplot2 to make prettier and more customizable plots.

## Usage

```
plotVariogram(
  sfe,
  features,
  sample_id = "all",
  color_by = NULL,
  group = c("none", "sample_id", "features", "angles"),
  use_1ty = TRUE,
  show_np = TRUE,
  ncol = NULL,
  colGeometryName = NULL,
  annotGeometryName = NULL,
  reducedDimName = NULL,
  divergent = FALSE,
  diverge_center = NULL,
  swap_rownames = NULL,
  name = "variogram"
)
```

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#### **Arguments**

sfe A SpatialFeatureExperiment object.

features Features to plot, must be in rownames of the gene count matrix, colnames of

colData or a colGeometry, colnames of cell embeddings in reducedDim, or nu-

meric indices of dimension reduction components.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

color\_by Name of a column in rowData(sfe) or in the featureData of colData (see

colFeatureData), colGeometry, or annotGeometry by which to color the correlogram of each feature. Alternatively, a vector of the same length as features,

or a data frame from clusterCorrelograms.

group Which of samples, features, and angles to show in the same facet for comparison

when there are multiple. Default to "none", meaning each facet will contain one variogram. When grouping multiple variograms in the same facet, the text with

model, nugget, sill, and range of the variograms will not be shown.

use\_lty Logical, whether to use linetype or point shape to distinguish between the dif-

ferent features or samples in the same facet. If FALSE, then the different features

or samples are not distinguished and the patterns are shown only.

show\_np Logical, whether to show number of pairs of cells at each distance bin.

ncol Number of columns if facetting.

colGeometryName

Name of a colGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use colGeometryNames to look up names of

the sf data frames associated with cells/spots.

annotGeometryName

Name of a annotGeometry of the SFE object, to annotate the gene expression

plot.

reducedDimName Name of a dimension reduction, can be seen in reducedDimNames. colGeometryName

and annotGeometryName have precedence over reducedDimName.

divergent Logical, whether a divergent palette should be used.

diverge\_center If divergent = TRUE, the center from which the palette should diverge. If NULL,

then not centering.

swap\_rownames Column name of rowData(object) to be used to identify features instead of

rownames (object) when labeling plot elements. If not found in rowData, then

rownames of the gene count matrix will be used.

name Name under which the correlogram results are stored, which is by default "sp.correlogram".

## Value

A ggplot object. The empirical variogram at each distance bin is plotted as points, and the fitted variogram model is plotted as a line for each feature. The number next to each point is the number of pairs of cells in that distance bin.

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## See Also

plotVariogramMap

## **Examples**

```
library(SFEData)
sfe <- McKellarMuscleData()
sfe <- colDataUnivariate(sfe, "variogram", features = "nCounts", model = "Sph")
plotVariogram(sfe, "nCounts")
# Anisotropy, will get a message
sfe <- colDataUnivariate(sfe, "variogram", features = "nCounts",
model = "Sph", alpha = c(30, 90, 150), name = "variogram_anis")
# Facet by angles by default
plotVariogram(sfe, "nCounts", name = "variogram_anis")
# Plot angles with different colors
plotVariogram(sfe, "nCounts", group = "angles", name = "variogram_anis")</pre>
```

plotVariogramMap

Plot variogram maps

# Description

Plot variogram maps that show the variogram in all directions in a grid of distances in x and y coordinates.

## Usage

```
plotVariogramMap(
    sfe,
    features,
    sample_id = "all",
    plot_np = FALSE,
    ncol = NULL,
    colGeometryName = NULL,
    annotGeometryName = NULL,
    reducedDimName = NULL,
    swap_rownames = NULL,
    name = "variogram_map"
)
```

# **Arguments**

sfe

A SpatialFeatureExperiment object.

features

Features to plot, must be in rownames of the gene count matrix, colnames of colData or a colGeometry, colnames of cell embeddings in reducedDim, or numeric indices of dimension reduction components.

 $sample\_id \qquad Sample(s) \ in \ the \ SFE \ object \ whose \ cells/spots \ to \ use. \ Can \ be \ "all" \ to \ compute$ 

metric for all samples; the metric is computed separately for each sample.

Logical, whether to plot the number of pairs in each distance bin instead of the

variance.

ncol Number of columns if facetting.

colGeometryName

plot\_np

Name of a colGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use colGeometryNames to look up names of the sf data frames associated with cells/spots.

annotGeometryName

Name of a annotGeometry of the SFE object, to annotate the gene expression

plot.

reducedDimName Name of a dimension reduction, can be seen in reducedDimNames. colGeometryName

and annot GeometryName have precedence over reduced DimName.

swap\_rownames Column name of rowData(object) to be used to identify features instead of

rownames(object) when labeling plot elements. If not found in rowData, then

rownames of the gene count matrix will be used.

name Name under which the correlogram results are stored, which is by default "sp.correlogram".

#### Value

A ggplot object.

#### See Also

plotVariogram

#### **Examples**

```
library(SFEData)
sfe <- McKellarMuscleData()
sfe <- colDataUnivariate(sfe, "variogram_map", features = "nCounts",
width = 500, cutoff = 5000)
plotVariogramMap(sfe, "nCounts")</pre>
```

SFEMethod

SFEMethod class

## Description

This S4 class is used to wrap spatial analysis methods, taking inspiration from the caret and tidymodels packages.

## Usage

```
SFEMethod(
  name,
  fun,
  reorganize_fun,
  package,
  variate = c("uni", "bi", "multi"),
  scope = c("global", "local"),
  title = NULL,
  default_attr = NA,
  args_not_check = NA,
  joint = FALSE,
  use_graph = TRUE,
  use_matrix = FALSE,
  dest = c("reducedDim", "colData")
## S4 method for signature 'SFEMethod'
info(x, type)
## S4 method for signature 'SFEMethod'
is_local(x)
## S4 method for signature 'SFEMethod'
fun(x)
## S4 method for signature 'SFEMethod'
reorganize_fun(x)
## S4 method for signature 'SFEMethod'
args_not_check(x)
## S4 method for signature 'SFEMethod'
is_joint(x)
## S4 method for signature 'SFEMethod'
use_graph(x)
## S4 method for signature 'SFEMethod'
use_matrix(x)
```

# **Arguments**

name Name of the method, used by user-facing functions to specify the method to use,

such as "moran" for Moran's I.

fun Function to run the method. See Details.

reorganize\_fun Function to reorganize results to add to the SFE object. See Details.

package Name of the package whose implementation of the method is used here, used to check if the package is installed. How many variables this method works with, must be one of "uni" for univariate, variate "bi" for bivariate, or "multi" for multivariate. Either "global", returning one result for the entire dataset, or "local", returning scope one result for each spatial location. For multivariate methods, this is irrelevant. Descriptive title to show when plotting the results. title default\_attr For local methods that return multiple fields, such as local Moran values and their p-values, the default field to use when plotting. args\_not\_check A character vector indicating which argument are not to be checked when comparing parameters in with those of a previous run. joint Logical, whether it makes sense to run this method to multiple samples jointly. If TRUE, then fun must be able to handle an adjacency matrix for the listw argument because there's no straightforward way to concatenate listw objects from multiple samples. Logical, to indicate whether the method uses a spatial neighborhood graph beuse\_graph cause unifying the user facing functions have an argument asking for the graph as most though not all methods require the graph. Logical, whether the function in slot fun takes a matrix as input. The argument use\_matrix is only used for bivariate methods. Whether the results are more appropriate for reducedDim or colData. Only dest used for multivariate methods. This overrides the "local" field in info. A SFEMethod object Х One of the names of the info slot, see slot documentation. type

## Details

The fun slot should be specified as such:

For all methods, there must be arguments x for a vector, listw for a listw object specifying the spatial neighborhood graph, zero.policy specifying what to do with cells without neighbors (default NULL, use global option value; if TRUE assign zero to the lagged value of zones without neighbours, if FALSE assign NA), and optionally other method specific arguments and . . . to pass to the underlying imported function. If the original function implementing the method in the package has different argument names or orders, write a thin wrapper to rearrange and/or rename the arguments.

For univariate methods that use a spatial neighborhood graph, the first two arguments must be x and listw. For univariate methods that don't use a spatial neighborhood graph, such as the variogram, the first two arguments must be x for a numeric vector and coords\_df for a sf data frame with cell locations and optionally other regressors. The formula argument is optional and can have defaults specifying regressors to use.

For bivariate methods, the first three arguments must be x, y, and listw.

For multivariate methods, the argument x is mandatory, for the matrix input. These arguments must be present but can be optional by having defaults: listw and ncomponents to set the number of dimentions in the output.

The reorganize\_fun slot should be specified as such:

Univariate methods are meant to be run separately for each gene, so the input to reorganize\_fun in the argument out should be a list of outputs; each element of the list corresponds to the output of a gene.

For univariate global methods, different fields of the result should be columns of a data frame with one row so results for multiple features will be a data frame. The arguments should be out, and name to rename the primary field if a more informative name is needed, and . . . for other arguments specific to methods. The output of reorganize\_fun should be a DataFrame whose rows correspond to the genes and columns correspond to fields in the output.

For univariate local methods, the arguments should be out, nb for a neighborhood list used for multiple testing correction, and p.adjust.method for a method to correct for multiple testing as in p.adjust, and .... The output of reorganize\_fun should be a list of reorganized output. Each element of the list corresponds to a gene, and the reorganized content of the element can be a vector, matrix, or data frame, but they must all have the same dimensions for all genes. Each element of the vector, or each row of the matrix or data frame corresponds to a cell.

For multivariate methods whose results go into reducedDim, reorganize\_fun should have one argument out for the raw output. The output of reorganize\_fun should be the cell embedding matrix ready to be added to reducedDim. Other relevant information such as gene loadings and eigenvalues should be added to the attributes of the cell embedding matrix.

For multivariate methods whose results can go into colData, the arguments should be out, nb, and p.adjust.method. Unlike the univariate local counterpart, out takes the raw output instead of a list of outputs. The output of reorganize\_fun is a vector or a data frame ready to be added to colData.

#### Value

The constructor returns a SFEMethod object. The getters return the content of the corresponding slots.

#### Slots

info A named character vector specifying information about the method.

fun The function implementing the method. See Details.

reorganize\_fun Function to convert output from fun into a format to store in the SFE object. See Details.

misc Miscellaneous information on how the method interacts with the rest of the package. This should be a named list.

spatialReducedDim

Plot dimension reduction components in space

## **Description**

Such as plotting the value of projection of gene expression of each cell to a principal component in space. At present, this function does not work for the 3D array of geographically weighted PCA (GWPCA), but a future version will deal with GWPCA results.

#### Usage

```
spatialReducedDim(
  sfe,
  dimred.
  ncomponents = NULL,
  components = ncomponents,
  colGeometryName = 1L,
  sample_id = "all",
  ncol = NULL,
  ncol_sample = NULL,
  annotGeometryName = NULL,
  annot_aes = list(),
  annot_fixed = list(),
  exprs_values = "logcounts",
  bbox = NULL,
  image_id = NULL,
  channel = NULL,
 maxcell = 5e+05,
  aes_use = c("fill", "color", "shape", "linetype"),
  divergent = FALSE,
  diverge_center = NULL,
  annot_divergent = FALSE,
  annot_diverge_center = NULL,
  size = 0,
  shape = 16,
  linewidth = 0,
  linetype = 1,
  alpha = 1,
  color = NA,
  fill = "gray80",
  scattermore = FALSE,
  pointsize = 0,
  bins = NULL,
  summary_fun = sum,
  hex = FALSE,
  show_axes = FALSE,
  dark = FALSE,
```

```
palette = colorRampPalette(c("black", "white"))(255),
normalize_channels = FALSE,
...
)
```

#### **Arguments**

sfe A SpatialFeatureExperiment object.

dimred A string or integer scalar indicating the reduced dimension result in reducedDims(sfe)

to plot.

ncomponents A numeric scalar indicating the number of dimensions to plot, starting from the

first dimension. Alternatively, a numeric vector specifying the dimensions to be

plotted.

components A numeric scalar or vector specifying which dimensions to be plotted. Use this

instead of ncomponents when plotting only one dimension.

colGeometryName

Name of a colGeometry sf data frame whose numeric columns of interest are to be used to compute the metric. Use colGeometryNames to look up names of

the sf data frames associated with cells/spots.

sample\_id Sample(s) in the SFE object whose cells/spots to use. Can be "all" to compute

metric for all samples; the metric is computed separately for each sample.

ncol Number of columns if plotting multiple features. Defaults to NULL, which means

using the same logic as facet\_wrap, which is used by patchwork's wrap\_plots

by default.

ncol\_sample If plotting multiple samples as facets, how many columns of such facets. This

is distinct from ncols, which is for multiple features. When plotting multiple features for multiple samples, then the result is a multi-panel plot each panel of

which is a plot for each feature facetted by samples.

annotGeometryName

Name of a annotGeometry of the SFE object, to annotate the gene expression

plot.

annot\_aes A named list of plotting parameters for the annotation sf data frame. The names

are which geom (as in ggplot2, such as color and fill), and the values are column

names in the annotation sf data frame. Tidyeval is NOT supported.

annot\_fixed Similar to annot\_aes, but for fixed aesthetic settings, such as color = "gray".

The defaults are the same as the relevant defaults for this function.

exprs\_values Integer scalar or string indicating which assay of x contains the expression val-

ues.

bbox A bounding box to specify a smaller region to plot, useful when the dataset is

large. Can be a named numeric vector with names "xmin", "xmax", "ymin", and "ymax", in any order. If plotting multiple samples, it should be a matrix with sample IDs as column names and "xmin", "ymin", "xmax", and "ymax" as row names. If multiple samples are plotted but bbox is a vector rather than a matrix, then the same bounding box will be used for all samples. You may see points at the edge of the geometries if the intersection between the bounding box and a geometry happens to be a point there. If NULL, then the entire tissue is plotted.

image\_id

ID of the image to plot behind the geometries. If NULL, then not plotting images. Use imgData to see image IDs present. To plot multiple grayscale images as different RGB channels, use a named vector here, whose names are channel names (r, g, b), and values are image\_ids of the corresponding images. The RGB colorization may not be colorblind friendly. When plotting multiple samples, it is assumed that the same image\_id is used for each channel across different samples.

channel

Numeric vector indicating which channels in a multi-channel image to plot. If NULL, grayscale is plotted if there is 1 channel and RGB for the first 3 channels. The numeric vector can be named (r, g, b) to indicate which channel maps to which color. The RGB colorization may not be colorblind friendly. This argument cannot be specified while image\_id is a named vector to plot different grayscale images as different channels.

maxcell

Maximum number of pixels to plot in the image. If the image is larger, it will be resampled so it have less than this number of pixels to save memory and for faster plotting. We recommend reducing this number when plotting multiple facets.

aes\_use

Aesthetic to use for discrete variables. For continuous variables, it's always "fill" for polygons and point shapes 21-25. For discrete variables, it can be fill, color, shape, or linetype, whenever applicable. The specified value will be changed to the applicable equivalent. For example, if the geometry is point but "linetype" is specified, then "shaped" will be used instead.

divergent

Logical, whether a divergent palette should be used.

diverge\_center If divergent = TRUE, the center from which the palette should diverge. If NULL, then not centering.

annot\_divergent

Just as divergent, but for the annotGeometry in case it's different.

annot\_diverge\_center

Just as diverge\_center, but for the annotGeometry in case it's different.

Fixed size of points. For points defaults to 0.5. Ignored if size\_by is specified. size

Fixed shape of points, ignored if shape\_by is specified and applicable. shape

Width of lines, including outlines of polygons. For polygons, this defaults to 0, linewidth

meaning no outlines.

linetype Fixed line type, ignored if linetype\_by is specified and applicable.

alpha Transparency.

color Fixed color for colGeometry if color\_by is not specified or not applicable, or

for annotGeometry if annot\_color\_by is not specified or not applicable.

fill Similar to color, but for fill.

scattermore

Logical, whether to use the scattermore package to greatly speed up plotting numerous points. Only used for POINT colGeometries. If the geometry is not POINT, then the centroids are used. Recommended for plotting hundreds of thousands or more cells where the cell polygons can't be seen when plotted due to the large number of cells and small plot size such as when plotting multiple panels for multiple features.

pointsize Radius of rasterized point in scattermore. Default to 0 for single pixels (fastest).

bins If binning the colGeometry in space due to large number of cells or spots, the

number of bins, passed to geom\_bin2d or geom\_hex. If NULL (default), then the colGeometry is plotted without binning. If binning, a point geometry is recommended. If the geometry is not point, then the centroids will be used.

summary\_fun Function to summarize the feature value when the colGeometry is binned.

hex Logical, whether to use geom\_hex. Note that geom\_hex is broken in ggplot2

version 3.4.0. Please update ggplot2 if you are getting horizontal stripes when

hex = TRUE.

show\_axes Logical, whether to show axes.

dark Logical, whether to use dark theme. When using dark theme, the palette will

have lighter color represent higher values as if glowing in the dark. This is

intended for plotting gene expression on top of fluorescent images.

palette Vector of colors to use to color grayscale images.

normalize\_channels

Logical, whether to normalize each channel of the image individually. Should be FALSE for bright field color images such as H&E but should set to TRUE for

fluorescent images.

... Other arguments passed to wrap\_plots.

#### Value

Same as in plotSpatialFeature. A ggplot2 object if plotting one component. A patchwork object if plotting multiple components.

#### See Also

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variogram-internal

Compute variograms

## **Description**

Wrapper of automap::autofitVariogram to facilitate computing variograms for multiple genes in SFE objects as an EDA tool. These functions are written to conform to a uniform format for univariate methods to be called internally. These functions are not exported, but the documentation is written to show users the extra arguments to use when alling calculateUnivariate or runUnivariate.

# Usage

```
.variogram(x, coords_df, formula = x ~ 1, scale = TRUE, ...)
.variogram_bv(x, y, coords_df, scale = TRUE, map = FALSE, ...)
.cross_variogram(x, y, coords_df, scale = TRUE, ...)
.cross_variogram_map(x, y, coords_df, width, cutoff, scale = TRUE, ...)
.variogram_map(x, coords_df, formula = x ~ 1, width, cutoff, scale = TRUE, ...)
```

## **Arguments**

x	A numeric vector whose variogram is computed.
coords_df	A sf data frame with the geometry and regressors for variogram modeling.
formula	A formula defining the response vector and (possible) regressors, in case of absence of regressors, use $x \sim 1$ .
scale	Logical, whether to scale x. Defaults to TRUE so the variogram is easier to interpret and is more comparable between features with different magnitudes when the length scale of spatial autocorrelation is of interest.
	Other arguments passed to automap::autofitVariogram such as model and variogram such as alpha for anisotropy. Note that gstat does not fit ansotropic models and you will get a warning if you specify alpha. Nevertheless, plotting the empirical anisotropic variograms and comparing them to the variogram fitted to the entire dataset can be a useful EDA tool.
у	For bivariate, another numeric vector whose variogram is computed.
map	logical; if TRUE, and cutoff and width are given, a variogram map is returned. This requires package sp. Alternatively, a map can be passed, of class Spatial-DataFrameGrid (see sp docs)
width	the width of subsequent distance intervals into which data point pairs are grouped for semivariance estimates
cutoff	spatial separation distance up to which point pairs are included in semivariance estimates; as a default, the length of the diagonal of the box spanning the data is divided by three.

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# Value

An autofitVariogram object.

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