

Package ‘FuseSOM’

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Title A Correlation Based Multiview Self Organizing Maps Clustering
For IMC Datasets

Version 1.15.0

Description A correlation-based multiview self-organizing map for the characterization of cell types in highly multiplexed in situ imaging cytometry assays (‘FuseSOM’) is a tool for unsupervised clustering. ‘FuseSOM’ is robust and achieves high accuracy by combining a ‘Self Organizing Map’ architecture and a ‘Multiview’ integration of correlation based metrics. This allows FuseSOM to cluster highly multiplexed in situ imaging cytometry assays.

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Encoding UTF-8

Roxygen list(markdown = TRUE)

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Imports psych, FCPS, analogue, coop, pheatmap, ggplotify, fastcluster,
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VignetteBuilder knitr

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biocViews SingleCell, CellBasedAssays, Clustering, Spatial

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Author Elijah Willie [aut, cre]

Maintainer Elijah Willie <ewil3501@uni.sydney.edu.au>

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.arsinhNnorm	<i>Function to do arsinh normalization</i>
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Description

Function to do arsinh normalization

Usage

```
.arsinhNnorm(x, cofactor = 5)
```

Arguments

x	A numeric or complex vector
cofactor	Cofactor of the vector. Default is 5.

Value

Arsinh normalized vector.

.computeElbow *A function to compute the elbow point given a set of points*

Description

A function to compute the elbow point given a set of points

Usage

.computeElbow(vals)

Arguments

vals Values to compute the elbow point of.

Value

A integer indicating the elbow point of vals.

.minmaxNorm *Function to do min max normalization*

Description

Function to do min max normalization

Usage

.minmaxNorm(x)

Arguments

x Matrix to min max normalize.

Value

Max normalized version of x

`.percentileNorm` *Function to do percentile normalizaton*

Description

Function to do percentile normalizaton

Usage

```
.percentileNorm(x)
```

Arguments

`x` Matrix to percentile normilse.

Value

percentile normalized version of `x`

`.runDiscriminant` *Discriminant cluster estimator*

Description

Function to estimate the number of clusters using discriminant analysis parts of this function is based on the sigclust2 package by Patrick Kimes see <https://github.com/pkimes/sigclust2>

Usage

```
.runDiscriminant(distMat, minClusterSize, alpha = 0.001)
```

Arguments

`distMat` A distance matrix

`minClusterSize` The minimum cluster size

`alpha` a value between 0 and 1 specifying the desired level of cutoff

Value

Optimal number of clusters

.uniformData	<i>Creates uniformly distributed data of same dimensionality as input data this function was obtained from the Stab package</i>
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Description

Creates uniformly distributed data of same dimensionality as input data this function was obtained from the Stab package

Usage

```
.uniformData(data)
```

Arguments

data A data matrix.

Value

Uniform random noise with dim(data)

clusterPrototypes	<i>Cluster prototypes</i>
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Description

Cluster the prototypes from the Self Organizing Map Clustering is done using hierarchical clustering with the average linkage function

Usage

```
clusterPrototypes(somModel, numClusters = NULL)
```

Arguments

somModel the self organizing map
numClusters the number of clusters to generate

Value

the cluster labels

Examples

```
data("risom_dat")  
risomMarkers <- c(  
  "CD45", "SMA", "CK7", "CK5", "VIM", "CD31", "PankRT", "ECAD"  
)  
prototypes <- generatePrototypes(risom_dat[, risomMarkers])  
clusters <- clusterPrototypes(prototypes, 23)
```

computeGridSize *Estimate the optimal grid size*

Description

The function finds the eigenvalues of the sample covariance matrix. It will then return the number of significant eigenvalues according to the Tracy-Widom test. The function is based on the estKW function from the SC3 package

Usage

```
computeGridSize(dataset)
```

Arguments

dataset The optimal grid size.

Value

the optimal grid size.

Author(s)

Elijah Willie ewil3501@uni.sydney.edu.au

Examples

```
data("risom_dat")
risomMarkers <- c(
  "CD45", "SMA", "CK7", "CK5", "VIM", "CD31", "PanKRT", "ECAD"
)
computeGridSize(risom_dat[, risomMarkers])
```

estimateNumCluster *Estimate number of clusters*

Description

A function for estimating the number of clusters using various method Methods available are: Discriminant, Distance (Gap, Silhouette, Slope, Jump, and Within Cluster Distance,) and Instability

Usage

```
estimateNumCluster(data, method = c("Discriminant", "Distance"), kSeq = 2:20)
```

Arguments

data the SOM object generated by generatePrototypes(), or an object of class SingleCellExperiment or SpatialExperiment.

method one of Discriminant, Distance, Stability. By default, everything is run

kSeq a sequence of the number of clusters to try. Default is 2:20 clusters

Value

A list containing the cluster estimations if a dataframe or matrix is provided

A SingleCellExperiment with a cluster estimate in it's metadata if a SingleCellExperiment or SpatialExperiment object is provided

Author(s)

Elijah Willie ewil3501@uni.sydney.edu.au

Examples

```
data("risom_dat")
risomMarkers <- c(
  "CD45", "SMA", "CK7", "CK5", "VIM", "CD31", "PanKRT", "ECAD"
)
res <- runFuseSOM(risom_dat, markers = risomMarkers, numClusters = 23)
res.est.k <- estimateNumCluster(res$model, kSeq = 2:25)
```

FuseSOM

FuseSOM

Description

FuseSOM provides a pipeline for the clustering of highly multiplexed in situ imaging cytometry assays. This pipeline uses the Self Organizing Map architecture coupled with Multiview hierarchical clustering. We also provide functions for normalisation and estimation of the number of clusters.

Details

The FuseSOM package provides three categories of important functions: foo, bar and baz.

generatePrototypes

Generate a Self Organizing Map

Description

A self organizing map of the marker intensities is generated and the prototypes are returned. The grid size is determined automatically

Usage

```
generatePrototypes(data, verbose = FALSE, size = NULL)
```

Arguments

data	the marker intensities
verbose	should the progress be printed out
size	The optimal grid size for the Self Organizing Map

Value

the self organizing map object

Examples

```
data("risom_dat")
risomMarkers <- c(
  "CD45", "SMA", "CK7", "CK5", "VIM", "CD31", "PanKRT", "ECAD"
)
generatePrototypes(risom_dat[, risomMarkers])
```

markerHeatmap	<i>Generate expression heatmap</i>
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Description

A function for generating a heat map of marker expression across clusters

Usage

```
markerHeatmap(
  data,
  markers = NULL,
  clusters = NULL,
  threshold = 2,
  clusterMarkers = FALSE,
  fontSize = 14
)
```

Arguments

data	a matrix or dataframe where the rows are samples and columns are markers
markers	a list of markers of interest. If not provided, all columns will be used
clusters	a vector of cluster labels
threshold	the value to threshold the marker expression at
clusterMarkers	should the rows(markers) of the heatmap be clustered
fontSize	the size of the text on the heatmap

Value

a heatmap with the markers in the rows and clusters in the columns

Author(s)

Elijah Willie ewil3501@uni.sydney.edu.au

Examples

```
data("risom_dat")
risomMarkers <- c(
  "CD45", "SMA", "CK7", "CK5", "VIM", "CD31", "PanKRT", "ECAD"
)
res <- runFuseSOM(risom_dat, markers = risomMarkers, numClusters = 23)
p.heat <- markerHeatmap(risom_dat, risomMarkers, clusters = res$clusters)
```

normaliseData

Normalise Marker Intensities

Description

The matrix of intensities is normalised based on one of four different method These methods include Percentile, zscore, arsinh and minmax

Usage

```
normaliseData(data, markers, method = "none", cofactor = 5)
```

Arguments

data	the raw intensity scores.
markers	the markers of interest.
method	the normalizaton method
cofactor	the cofactor for arsinh normalisation

Value

normalised matrix.

Author(s)

Elijah Willie ewil3501@uni.sydney.edu.au

Examples

```
data("risom_dat")
risomMarkers <- c(
  "CD45", "SMA", "CK7", "CK5", "VIM", "CD31", "PanKRT", "ECAD"
)
normaliseData(risom_dat[, risomMarkers])
```

normalizeData	<i>Normalize Marker Intensities</i>
---------------	-------------------------------------

Description

The matrix of intensities is normalised based on one of four different method These methods include Percentile, zscore, arsinh and minmax

Usage

```
normalizeData(data, markers, method = "none", cofactor = 5)
```

Arguments

data	the raw intensity scores.
markers	the markers of interest.
method	the normalizaton method
cofactor	the cofactor for arsinh normalization

Value

normalised matrix.

Author(s)

Elijah Willie ewil3501@uni.sydney.edu.au

Examples

```
data("risom_dat")
risomMarkers <- c(
  "CD45", "SMA", "CK7", "CK5", "VIM", "CD31", "PanKRT", "ECAD"
)
normaliseData(risom_dat[, risomMarkers])
```

optiPlot	<i>Generate elbow plots</i>
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Description

A function generating the elbow plot for the optimal number of clusters returned by the estimateNumcluster() function Methods available are: Gap, Silhouette, Slope, Jump, and Within Cluster Distance(WCD)

Usage

```
optiPlot(data, method = "jump")
```

Arguments

data a Self Organizing Map object generated by `generatePrototypes()`, or an object of class `SingleCellExperiment` or `SpatialExperiment`

method one of 'jump', 'slope', 'wcd', 'gap', or 'silhouette'

Value

an elbow plot object where the optimal number of clusters is marked

Author(s)

Elijah Willie ewil3501@uni.sydney.edu.au

Examples

```
data("risom_dat")
risomMarkers <- c(
  "CD45", "SMA", "CK7", "CK5", "VIM", "CD31", "PankRT", "ECAD"
)
res <- runFuseSOM(risom_dat, markers = risomMarkers, numClusters = 23)
resEstK <- estimateNumCluster(res$model, kSeq = 2:25)
p <- optiPlot(resEstK, method = "jump")
```

risom_dat

IMC Breast Cancer Data Data from A spatial atlas of breast cancer progression using MIBI-TOF and tissue transcriptomics

Description

IMC Breast Cancer Data Data from A spatial atlas of breast cancer progression using MIBI-TOF and tissue transcriptomics

Usage

```
data(risom_dat)
```

Format

An object of class "data.frame".

Source

Mendeley Data, <https://data.mendeley.com/datasets/d87vg86zd8/3>

References

T. Risom, et al. Transition to invasive breast cancer is associated with progressive changes in the structure and composition of tumor stroma *Cell*, 185 (2022), pp. 299-310 ([ScienceDirect](#))

runFuseSOM

A wrapper function to run the FuseSOM algorithm

Description

This function accepts a matrix, dataframe or a SingleCellExperiment object. For matrices and dataframes, it is assumed that markers are the columns and samples rows.

Usage

```
runFuseSOM(
  data,
  markers = NULL,
  numClusters = NULL,
  assay = NULL,
  clusterCol = "clusters",
  size = NULL,
  verbose = FALSE
)
```

Arguments

data	a matrix, dataframe, SingleCellExperiment or SpatialExperiment object.
markers	the markers of interest. If this is not provided, all columns will be used
numClusters	the number of clusters to be generated from the data
assay	the assay of interest if SingleCellExperiment object is used
clusterCol	the name of the column to store the clusters in
size	the size of the square grid. eg for a 10X10 grid, size = 10
verbose	should the generation of the Self Organising Map be printed

Value

A list containing the SOM model and the cluster labels if a dataframe or matrix is provided

A SingleCellExperiment object with labels in coldata, and SOM model in metadata if a SingleCellExperiment or SpatialExperiment object is provided

Author(s)

Elijah Willie ewil3501@uni.sydney.edu.au

Examples

```
data("risom_dat")
risomMarkers <- c(
  "CD45", "SMA", "CK7", "CK5", "VIM", "CD31", "PanKRT", "ECAD"
)
res <- runFuseSOM(
  risom_dat,
  markers = risomMarkers, numClusters = 23, size = 8
)
```

somInitPca.default *these functions were obtained from <https://rdrr.io/rforge/yasomi/> with some major modifications*

Description

these functions were obtained from <https://rdrr.io/rforge/yasomi/> with some major modifications

Usage

```
## Default S3 method:  
somInitPca(data, somGrid, weights, with.princomp = FALSE, ...)
```

Arguments

data	The data to which the SOM will be fitted, a matrix or data frame of observations (which should be scaled)
somGrid	A somgrid object
weights	Optional weights for the data points
with.princomp	Switch specifying whether the princomp should be used instead of the prcomp for computing the principal components when no weights are given (see details)
...	not used

Value

A list containing: prototype, a matrix containing appropriate initial prototypes, and data.pca the results of the PCA conducted on the data

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