# Package 'mineCitrus'

October 13, 2022

Type Package	
Fitle Extract and Analyze Median Molecule Intensity from 'citrus' Output	
Version 1.0.0	
Date 2018-07-05	
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Oescription Citrus is a computational technique developed for the analysis of high dimensional cytometry data sets. This package extracts, statistically analyzes, and visualizes marker expression from 'citrus' data. This code was used to generate data for Figures 3 and 4 in the forthcoming manuscript: Throm et al. "Identification of Enhanced Interferon-Gamma Signaling in Polyarticular Juvenile Idiopathic Arthritis with Mass Cytometry", JCI-Insight. For more information on Citrus, please see: Bruggner et al. (2014) <doi:10.1073 pnas.1408792111="">. To download the 'citrus' package, please see <a href="https://github.com/nolanlab/citrus">https://github.com/nolanlab/citrus</a>&gt;.</doi:10.1073>	
License GPL-2	
Encoding UTF-8	
LazyData true	
mports ggplot2	
<b>Depends</b> R (>= 2.10)	
Suggests	
RoxygenNote 6.0.1	
NeedsCompilation no	
Repository CRAN	
<b>Date/Publication</b> 2018-07-05 13:40:03 UTC	
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## **Description**

Gets matrices of medians for each individual sample for all measured parameters for all clusters

## Usage

allmeds(citrus.combinedFCSSet, citrus.foldClustering, citrus.foldFeatureSet)

## **Arguments**

citrus.combinedFCSSet

loaded from citrusClustering.RData file generated by Citrus run

citrus.foldClustering

loaded from citrusClustering.RData file generated by Citrus run

citrus.foldFeatureSet

computed from first two variables using citrus.calculateFoldFeatureSet function from citrus package

#### Value

Returns a list with each element corresponding to a matrix (rows as samples, columns as measured parameters) for a different cluster (for the minimum threshold specified)

citrus.combinedFCSSet 3

#### **Examples**

citrus.combinedFCSSet Cytometry data set for example of Citrus data set from nolanlab/citrus

## **Description**

A dataset containing the a simple example of cytometry data

## Usage

```
citrus.combinedFCSSet
```

#### **Format**

A large citrus.combinedFCSSet object with 5 elements:

data Toy data set for cytometry

fileChannelNames Names of channels for measured parameters included in toy cytmetry data set

fileIds ID numbers for each file included in toy cytometry data set

fileNames Names of files included in toy cytmetry data set

fileReagentNames Names of measured channels in toy cytmetry data set ...

## Source

https://github.com/nolanlab/citrus

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citrus.foldClustering Clustering data for example of Citrus data set from nolanlab/citrus

## **Description**

A dataset containing the clustering of different cell groups

## Usage

citrus.foldClustering

#### **Format**

A large citrus.foldClustering object with 5 elements:

allClustering A list describing which events belong to which clusters foldClustering A list describing which events belong to which clusters for each fold foldMappingAssignments A list describing assignments with fold clustering folds Descriptions of each data clustering

nFolds The number of times data is clustered ...

#### **Source**

https://github.com/nolanlab/citrus

citrus.foldFeatureSet Correlation data for example of Citrus data set from nolanlab/citrus

## **Description**

A dataset containing the association of red and blue in clusters with different sample groups

#### Usage

citrus.foldFeatureSet

## **Format**

A list with 8 elements:

allFeatures Data set for each sample for all markers and clusters
allLargeEnoughClusters Vector of clusters meeting size threshold
foldFeatures Data for each fold clustering
foldLargeEnoughClusters Clusters meeting size threshold for each fold clustering

classclustermeds 5

folds Descriptions of each data clustering

**leftoutFeatures** Data omitted from analyses

minimumClusterSizePercent Minimum size threshold to retain clusters in analysis

**nFolds** The number of times data is clustered ...

#### **Source**

https://github.com/nolanlab/citrus

classclustermeds

Gets matrices of medians for each individual sample for all measured parameters for all clusters

#### **Description**

Gets matrices of medians for each individual sample for all measured parameters for all clusters

#### Usage

```
classclustermeds(citrus.foldFeatureSet, citrus.foldClustering,
  citrus.combinedFCSSet, groupsizes, meds)
```

#### **Arguments**

citrus.foldFeatureSet

computed from first two variables using citrus.calculateFoldFeatureSet function from citrus package

citrus.foldClustering

loaded from citrusClustering.RData file generated by Citrus run

citrus.combinedFCSSet

loaded from citrusClustering.RData file generated by Citrus run

groupsizes list of sizeso f the groups run in Citrus, in order of the selection for citrus run

meds The names of the columns from citrus.combinedFCSSet\$data of interest to ex-

tract medians for

#### Value

Returns a list of matrices with columns corresponding to selected features and rows corresponding to sample groups; each list element corresponds to data for a different cluster

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## **Examples**

clustermeds

Gets matrix of medians for desired measured features for all clusters meeting threshold requirements specified in Citrus

## Description

Gets matrix of medians for desired measured features for all clusters meeting threshold requirements specified in Citrus

## Usage

```
clustermeds(citrus.foldFeatureSet, citrus.foldClustering, medsofinterest,
  citrus.combinedFCSSet)
```

#### **Arguments**

citrus.foldFeatureSet

computed from first two variables using citrus.calculateFoldFeatureSet function from citrus package

citrus.foldClustering

loaded from citrusClustering.RData file generated by Citrus run

medsofinterest The names of the columns from citrus.combinedFCSSet\$data of interest to extract medians for

citrus.combinedFCSSet

loaded from citrusClustering.RData file generated by Citrus run

#### Value

Returns a matrix with columns corresponding to selected features and rows corresponding to samples

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## **Examples**

difMarkerPlots Plot dot plots of features where both clusters are significantly different from the reference cluster without processing data before hand

## **Description**

Plot dot plots of features where both clusters are significantly different from the reference cluster without processing data before hand

#### Usage

```
difMarkerPlots(data, clusters, markers, diffclust, strat)
```

#### **Arguments**

data	output from call to allmeds function
clusters	clusterIDs of the desired clusters to compare and plot
markers	indices of the columns of the data matrix for features to be analyst
diffclust	clusterID of for cluster to statisticaly compare others to
strat	clusterIDs for stratifying clusters as indicated by Citrus results

## Value

Dot plots for all features where both clusters are significantly different from the reference cluster

8 difMarkerPlots2

difMarkerPlots2	Plot dot plots of features where one cluster is significantly different from the reference cluster without processing data before hand
	from the reference cluster without processing data before hand

## Description

Plot dot plots of features where one cluster is significantly different from the reference cluster without processing data before hand

## Usage

```
difMarkerPlots2(data, clusters, markers, diffclust, strat)
```

## Arguments

data	output from call to allmeds function
clusters	clusterIDs of the desired clusters to compare and plot
markers	indices of the columns of the data matrix for features to be analyse
diffclust	clusterID of for cluster to statisticaly compare others to
strat	clusterIDs for stratifying clusters as indicated by Citrus results

## Value

Dot plots for all features where one cluster is significantly different from the reference cluster

filterMarker 9

filterMarker	Filters list of data matrices with columns corresponding to the measured parameters of interest

#### **Description**

Filters list of data matrices with columns corresponding to the measured parameters of interest

## Usage

```
filterMarker(clustdat, markers)
```

## **Arguments**

clustdat a list of data matrices with list elements corresponding to clusters and matrices

of intensities of measured parameters

markers Indices of the columns of parmeters to keep

## Value

A list of data matrices with columns of data matrices only corresponding to measured parameters of interest

## **Examples**

findclust

Filters list to contain only desired clusters

## Description

Filters list to contain only desired clusters

## Usage

```
findclust(data, clusters)
```

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#### **Arguments**

data a list of data matrices with list elements corresponding to clusters and matrices

of intensities of measured parameters

clusters indices of the clusters to retain

#### Value

A list of data matrices for the desired clusters

## **Examples**

findSig

Assesses significance of ANOVA and t-test results

## **Description**

Assesses significance of ANOVA and t-test results

#### Usage

```
findSig(posHocRes)
```

#### **Arguments**

posHocRes results from a call to the posthoc function

## Value

A dataframe indicating the significances of results

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```
filteredmeds<-findclust(data=meds,clusters=c(1999,19972,19988))
meds2<-filterMarker(clustdat=filteredmeds,markers=c(2,3))
foranova<-processforanova(filtereddata=meds2)
ttests<-posthoc(processedDat=foranova,clustIDdif=19999)
sig<-findSig(posHocRes=ttests)</pre>
```

plotdif Plot dot plots of features where both clusters are significantly different from the reference cluster

## **Description**

Plot dot plots of features where both clusters are significantly different from the reference cluster

#### Usage

```
plotdif(BJHdf, anovadata, strat)
```

## **Arguments**

BJHdf results of a call to findsig

anovadata results of call to processforanova

strat clusterIDs for clusters that are stratifying

#### Value

Dot plots for all features where both clusters are significantly different from the reference cluster

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plotdif2	Plot dot plots of features where one cluster is significantly different
	from the reference cluster

#### **Description**

Plot dot plots of features where one cluster is significantly different from the reference cluster

#### Usage

```
plotdif2(BJHdf, anovadata, strat)
```

## **Arguments**

BJHdf results of a call to findsig

anovadata results of call to processforanova

strat clusterIDs for clusters that are stratifying

#### Value

Dot plots for all features where one cluster is significantly different from the reference cluster

#### **Examples**

posthoc

Runs ANOVA and t-tests comparing clusters and markers in clusters

## Description

Runs ANOVA and t-tests comparing clusters and markers in clusters

processforanova 13

#### Usage

```
posthoc(processedDat, clustIDdif)
```

## **Arguments**

processedDat data that has been processed using the processforanova function

clustIDdif ID number of the cluster to compare the others to

#### Value

A list of t-test results for each of the comparisons

## **Examples**

processforanova

Processes cluster signaling data in form for statistical analysis

#### Description

Processes cluster signaling data in form for statistical analysis

#### Usage

```
processforanova(filtereddata)
```

## **Arguments**

filtereddata

a list with each element corresonding to a cluster of interest and matrices containing individual sample data for desired markers

#### Value

A dataframe sufficient for using the posthoc function to compute statistics

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#### **Examples**

sortmat

Reorders to rows (corresponding to different clusters) of a matrix of medians to a desired order

## **Description**

Reorders to rows (corresponding to different clusters) of a matrix of medians to a desired order

#### Usage

```
sortmat(mat, desiredorder)
```

#### **Arguments**

mat matrix of median data
desiredorder row labels from matrix in desired order

#### Value

Returns a matrix with rows rearranged in desired order

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