

Package ‘scQTLtools’

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Type Package

Title An R package for single-cell eQTL analysis and visualization

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Description This package specializes in analyzing and visualizing eQTL at the single-cell level. It can read gene expression matrices or Seurat data, or SingleCellExperiment object along with genotype data.

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patchwork(>= 1.2.0), DESeq2 (>= 1.45.3), VGAM (>= 1.1-11),
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Contents

scQTLtools-package	3
adjust_pvalues	4
buildZINB	4
callQTL	5
checkSNPList	6
CPM_normalize	7
createGeneLoc	8
createQTLObject	9
createSNPsLoc	10
DataSet	10
DESeq_normalize	11
draw_boxplot	12
draw_histplot	13
draw_QTLplot	13
draw_violinplot	14
eQTLObject-class	15
filterGeneSNP	16
filter_by_abs_b	17
get_cell_groups	17
get_counts	18
get_filter_data	19
get_model_info	20
get_raw_data	20
get_result_info	21
initialize_progress_bar	22
limma_normalize	22
linearModel	23
load_biclassify_info	24
load_group_info	25
load_species_info	25
log_normalize	26
normalizeGene	27
plots_theme_opts	27

poissonModel	28
process_matrix	29
remove_outliers	30
set_filter_data	31
set_model_info	32
set_raw_data	32
set_result_info	33
TPM_normalize	34
visualizeQTL	35
zinbModel	36
Index	37

scQTLtools-package	<i>scQTLtools: An R package for single-cell eQTL analysis and visualization</i>
--------------------	---

Description

This package specializes in analyzing and visualizing eQTL at the single-cell level. It can read gene expression matrices or Seurat data, or SingleCellExperiment object along with genotype data.

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See Also

Useful links:

- <https://github.com/XFWuCN/scQTLtools>
- Report bugs at <https://github.com/XFWuCN/scQTLtools/issues>

adjust_pvalues	<i>Adjust p-values and filter SNP–gene pairs based on the adjusted p-values.</i>
----------------	--

Description

Adjust p-values and filter SNP–gene pairs based on the adjusted p-values.

Usage

```
adjust_pvalues(result, pAdjustMethod = "bonferroni", pAdjustThreshold = 0.05)
```

Arguments

result	A data frame that contains information of SNP–gene pairs and raw p values.
pAdjustMethod	Method for p-value adjustment. One of "bonferroni", "holm", "hochberg", "hommel" or "BH". The default option is "bonferroni".
pAdjustThreshold	Only SNP–gene pairs with adjusted p-values meeting the threshold will be displayed. Default by 0.05.

Value

A data frame with adjusted p-values, filtered by threshold, containing information on SNP–gene pairs.

Examples

```
example_data <- data.frame(
  gene = c("Gene1", "Gene2", "Gene3", "Gene4"),
  SNP = c("SNP1", "SNP2", "SNP3", "SNP4"),
  pvalue = c(0.001, 0.04, 0.03, 0.0005))
pAdjustMethod <- "BH"
pAdjustThreshold <- 0.05
adjusted_result <- adjust_pvalues(example_data, pAdjustMethod,
pAdjustThreshold)
```

buildZINB	<i>Build a ZINB model.</i>
-----------	----------------------------

Description

Build a ZINB model.

Usage

```
buildZINB(counts)
```

Arguments

counts A numeric vector of gene expression values.

Value

A list containing four parameters estimated from the ZINB model.

Examples

```
data(GeneData)
gene <- unlist(GeneData[1, ])
result <- buildZINB(gene)
```

callQTL	<i>Identify single-cell eQTLs.</i>
---------	------------------------------------

Description

This function detects eQTLs using scRNA-seq data and genotype data.

Usage

```
callQTL(
  eQTLObject,
  gene_ids = NULL,
  downstream = NULL,
  upstream = NULL,
  gene_mart = NULL,
  snp_mart = NULL,
  pAdjustMethod = "bonferroni",
  useModel = "zinb",
  pAdjustThreshold = 0.05,
  logfcThreshold = 0.1
)
```

Arguments

eQTLObject	An S4 object of class eQTLObject.
gene_ids	A gene ID or a list of gene IDS.
downstream	Distance (in base pairs) downstream of the gene start site to search for associated SNPs.
upstream	Distance (in base pairs) upstream of the gene end site to search for associated SNPs.
gene_mart	A Mart object representing the BioMart gene database. If NULL, the Ensembl Gene BioMart will be used.

snp_mart	A Mart object representing the BioMart SNP database. If NULL, the Ensembl SNP BioMart will be used.
pAdjustMethod	Method used for multiple testing correction. One of "bonferroni", "holm", "hochberg", "hommel", or "BH". Default is "bonferroni".
useModel	Model used for fitting. One of "poisson", "zinb", or "linear."
pAdjustThreshold	Only SNP-gene pairs with adjusted p-values below the threshold will be retained. Default is 0.05.
logfcThreshold	The minimum beta coefficient required to report a SNP-gene pair as an eQTL.

Value

A data frame in which each row corresponds to a detected SNP-gene eQTL pair, including statistical and model fitting results.

Examples

```
data(EQTL_obj)
library(biomaRt)
gene_mart <- useEnsembl(biomart = "genes",
                        dataset = "hsapiens_gene_ensembl",
                        mirror = 'asia')
snp_mart <- useEnsembl(biomart = "snps",
                        dataset = "hsapiens_snp",
                        mirror = 'asia')

eqtl <- callQTL(
  eQTLObject = EQTL_obj,
  gene_ids = NULL,
  downstream = NULL,
  upstream = NULL,
  gene_mart = gene_mart,
  snp_mart = snp_mart,
  pAdjustMethod = 'bonferroni',
  useModel = 'linear',
  pAdjustThreshold = 0.05,
  logfcThreshold = 0.025
)
```

checkSNPList

Validate SNP IDs in the input genotype matrix.

Description

Validate SNP IDs in the input genotype matrix.

Usage

```
checkSNPList(snpList, snp_mart = NULL, snpDataset = "hsapiens_snp")
```

Arguments

snpList	A list of SNPs IDs.
snp_mart	An object of class Mart representing the BioMart SNP database to connect to. If provided, this should be a Mart object obtained by calling <code>useEnsembl()</code> , which allows specifying a mirror in case of connection issues. If NULL, the function will create and use a Mart object pointing to the Ensembl SNP BioMart, using the specified <code>snpDataset</code> and a default mirror.
snpDataset	A character string specifying the SNP dataset to use from Ensembl. Default is <code>hsapiens_snp</code> for human SNPs.

Value

A data frame containing the genomic locations of the valid SNPs.

Examples

```
data(SNPData2)
snpList <- rownames(SNPData2)
snpDataset <- 'hsapiens_snp'
snps_loc <- checkSNPList(snpList = snpList,
                        snpDataset = snpDataset)
```

CPM_normalize	<i>Normalize gene expression using CPM.</i>
---------------	---

Description

`CPM_normalize()` applies Counts Per Million (CPM) normalization to a raw gene expression matrix.

Usage

```
CPM_normalize(expressionMatrix)
```

Arguments

expressionMatrix	A numeric matrix of raw gene expression counts, with genes as rows and cells as columns.
------------------	--

Value

A normalized gene expression matrix after applying CPM normalization.

Examples

```
data(GeneData)
CPM_normalize(GeneData)
```

createGeneLoc	<i>Create a gene location data frame.</i>
---------------	---

Description

Create a gene location data frame.

Usage

```
createGeneLoc(
  geneList,
  gene_mart = NULL,
  geneDataset = "hsapiens_gene_ensembl",
  OrgDb
)
```

Arguments

geneList	A gene ID or a list of genes IDs.
gene_mart	An object of class Mart representing the BioMart gene database to connect to. If provided, this should be a Mart object obtained by calling useEnsembl(), which allows specifying a mirror in case of connection issues. If NULL, the function will create and use a Mart object pointing to the Ensembl Gene BioMart, using the specified geneDataset and a default mirror.
geneDataset	A character string specifying the gene dataset to use from Ensembl. Default is hsapiens_gene_ensembl for human genes.
OrgDb	The name of the OrgDb package to use for gene annotation. Supported values include org.Hs.eg.db and org.Mm.eg.db.

Value

A data.frame containing gene location information.

Examples

```
data(GeneData)
geneList <- rownames(GeneData)
library(GOSemSim)
library(biomaRt)
OrgDb <- load_OrgDb("org.Hs.eg.db")
gene_mart <- useEnsembl(biomart = "genes",
                        dataset = "hsapiens_gene_ensembl",
                        mirror = 'asia')
gene_loc <- createGeneLoc(geneList = geneList,
                          gene_mart = gene_mart,
                          OrgDb = OrgDb)
```

createQTLObject	Create an <i>eQTLObject</i> for storing <i>sc-eQTL</i> analysis data.
-----------------	---

Description

This function creates an S4 object to store genotype and expression data, along with sample grouping and metadata for eQTL analysis.

Usage

```
createQTLObject(  
  snpMatrix,  
  genedata,  
  biClassify = FALSE,  
  species = NULL,  
  group = NULL,  
  ...  
)
```

Arguments

snpMatrix	A genotype matrix where each row is a snp and each column is a cell. Encoding should be 0, 1, 2, 3.
genedata	A gene expression matrix, or a Seurat object or SingleCellExperiment object.
biClassify	Logical; whether to convert genotype encoding in snpMatrix to 0, 1, and 2. TRUE indicates conversion; FALSE indicates no conversion (default).
species	The species that the user wants to select, human or mouse.
group	A column name in the metadata of the Seurat or SingleCellExperiment object indicating cell groups (e.g., celltype or condition).
...	other parameters

Value

An S4 object of class *eQTLObject*.

Examples

```
data(SNPData)  
data(GeneData)  
eqtl <- createQTLObject(snpMatrix = SNPData,  
  genedata = GeneData,  
  biClassify = FALSE,  
  species = 'human',  
  group = NULL)
```

createSNPsLoc	<i>Create SNP location dataframe.</i>
---------------	---------------------------------------

Description

Create SNP location dataframe.

Usage

```
createSNPsLoc(snpList, snp_mart = NULL, snpDataset = "hsapiens_snp")
```

Arguments

snpList	A list of SNPs IDs.
snp_mart	An object of class Mart representing the BioMart SNP database to connect to. If provided, this should be a Mart object obtained by calling <code>useEnsembl()</code> , which allows specifying a mirror in case of connection issues. If NULL, the function will create and use a Mart object pointing to the Ensembl SNP BioMart, using the specified <code>snpDataset</code> and a default mirror.
snpDataset	A character string specifying the SNP dataset to use from Ensembl. Default is <code>hsapiens_snp</code> for human SNPs.

Value

A data frame containing the SNP genomic locations.

Examples

```
snpList <- c('rs546', 'rs549', 'rs568', 'rs665', 'rs672')
library(biomaRt)
snp_mart <- useEnsembl(biomart = "snps",
                      dataset = "hsapiens_snp",
                      mirror = 'asia')
snp_loc <- createSNPsLoc(snpList = snpList,
                       snp_mart = snp_mart)
```

DataSet	<i>The package includes five example datasets: a gene expression matrix, two SNP genotype matrices, a Seurat object, and an eQTL object.</i>
---------	--

Description

GeneData: A dataset containing example gene expression data. Each row represents a gene and each column represents a cell.

SNPData: A dataset containing single nucleotide (SNP) data. Each row represents a variant and each column represents a cell.

Seurat_obj: A Seurat object containing GeneData along with associated metadata stored in the meta.data slot.

SNPData2: A smaller SNP dataset containing fewer variants and cells. Each row represents a variant and each column represents a cell.

EQTL_obj: An eQTLObject created using [createQTLObject](#), where the raw expression matrix is normalized using [normalizeGene](#), and both the genotype matrix and normalized expression matrix were filtered using [filterGeneSNP](#).

Format

A numeric matrix with 100 rows (genes) and 2705 columns (cells).

A numeric matrix with 1000 rows (SNPs) and 2705 columns (cells).

An object of class Seurat.

A numeric matrix with 500 rows (SNPs) and 500 columns (cells).

See Also

[createQTLObject](#), [normalizeGene](#) and [filterGeneSNP](#), for the underlying functions that do the work.

An object of class eQTLObject.

DESeq_normalize

Normalize the gene expression matrix with DESeq2.

Description

DESeq_normalize() normalizes a raw gene expression matrix using the **DESeq2** package.

Usage

```
DESeq_normalize(expressionMatrix)
```

Arguments

expressionMatrix

A numeric matrix of raw gene expression counts, with genes as rows and cells as columns.

Value

A normalized gene expression matrix after applying DESeq normalization.

Examples

```
data(GeneData)
DESeq_normalize(GeneData)
```

draw_boxplot

Generate a boxplot of gene expression by SNP genotype.

Description

draw_boxplot() visualizes gene expression levels across different SNP genotypes using a boxplot.

Usage

```
draw_boxplot(df, unique_group)
```

Arguments

df	A data frame containing gene expression values, SNP genotypes, and group labels.
unique_group	A character string indicating the unique group name.

Value

ggplot

Examples

```
set.seed(123)
counts_Ref <- rnorm(50, mean = 10, sd = 2)
counts_Alt <- rnorm(50, mean = 12, sd = 2)
i <- rep("GroupA", 100)
unique_group <- unique(i)
dataframe <- data.frame(
  expression = c(counts_Ref, counts_Alt),
  snp = c(rep("REF", length(counts_Ref)),
    rep("ALT", length(counts_Alt))), group = i)
dataframe$snp <- factor(dataframe$snp, levels = c("REF", "ALT"))
draw_boxplot(df = dataframe, unique_group = unique_group)
```

draw_histplot	<i>Generate a histogram of gene expression by SNP genotype.</i>
---------------	---

Description

draw_histplot() shows the distribution of expression values for each SNP genotype using histograms.

Usage

```
draw_histplot(df, unique_group)
```

Arguments

df	A data frame containing gene expression values, SNP genotypes, and group labels.
unique_group	A character string indicating the unique group name.

Value

ggplot

Examples

```
set.seed(123)
counts_Ref <- rnorm(50, mean = 10, sd = 2)
counts_Alt <- rnorm(50, mean = 12, sd = 2)
i <- rep("GroupA", 100); unique_group <- unique(i)
dataframe <- data.frame(expression = c(counts_Ref, counts_Alt),
                          snp = c(rep("REF", length(counts_Ref)),
                                rep("ALT", length(counts_Alt))), group = i)
dataframe$snp <- factor(dataframe$snp, levels = c("REF", "ALT"))
draw_histplot(df = dataframe, unique_group = unique_group)
```

draw_QTLplot	<i>Create a combined violin-box-scatter plot.</i>
--------------	---

Description

draw_QTLplot() creates a composite plot that overlays violin plots, boxplots, and scatter points to illustrate the distribution and variability of gene expression across SNP groups.

Usage

```
draw_QTLplot(df, unique_group)
```

Arguments

- `df` A data frame containing gene expression values, SNP genotypes, and group labels.
- `unique_group` A character string indicating the unique group name.

Value

ggplot

Examples

```
set.seed(123)
counts_Ref <- rnorm(50, mean = 10, sd = 2)
counts_Alt <- rnorm(50, mean = 12, sd = 2)
i <- rep("GroupA", 100); unique_group <- unique(i)
dataframe <- data.frame(expression = c(counts_Ref, counts_Alt),
                        snp = c(rep("REF", length(counts_Ref)),
                               rep("ALT", length(counts_Alt))), group = i)
dataframe$snps <- factor(dataframe$snps, levels = c("REF", "ALT"))
draw_QTLplot(df = dataframe, unique_group = unique_group)
```

draw_violinplot	<i>Generate a violin plot of gene expression by SNP genotype.</i>
-----------------	---

Description

`draw_violinplot()` visualizes gene expression levels across different SNP genotypes using violin plots.

Usage

```
draw_violinplot(df, unique_group)
```

Arguments

- `df` A data frame containing gene expression values, SNP genotypes, and group labels.
- `unique_group` A character string indicating the unique group name.

Value

ggplot

Examples

```

set.seed(123)
counts_Ref <- rnorm(50, mean = 10, sd = 2)
counts_Alt <- rnorm(50, mean = 12, sd = 2)
i <- rep("GroupA", 100); unique_group <- unique(i)
dataframe <- data.frame(expression = c(counts_Ref, counts_Alt),
                        snp = c(rep("REF", length(counts_Ref)),
                               rep("ALT", length(counts_Alt))), group = i)
dataframe$snp <- factor(dataframe$snp, levels = c("REF", "ALT"))
draw_violinplot(df = dataframe, unique_group = unique_group)

```

eQTLObject-class	Class eQTLObject
------------------	------------------

Description

The eQTLObject class is designed to store data related to eQTL analysis, including data lists, result data frames, and additional metadata such as classification, species, and grouping information.

Value

An S4 object of class eQTLObject.

Slots

rawData A gene expression dataframe, the row names represent gene IDs and the column names represent cell IDs.

filterData Gene expression matrix after normalizing.

eQTLResult The result dataframe obtained the sc-eQTL results.

biClassify Logical; whether to convert genotype encoding in snpMatrix to 0, 1, and 2. TRUE indicates conversion; FALSE indicates no conversion (default).

species The species that the user wants to select, human or mouse.

groupBy Options for cell grouping, users can choose celltype, cellstatus, etc., depending on meta-data.

useModel model for fitting dataframe.

filterGeneSNP	<i>Filter gene expression and genotype matrices by cell percentage thresholds.</i>
---------------	--

Description

Filter gene expression and genotype matrices by cell percentage thresholds.

Usage

```
filterGeneSNP(
  eQTLObject,
  snpNumOfCellsPercent = 10,
  expressionMin = 0,
  expressionNumOfCellsPercent = 10
)
```

Arguments

eQTLObject	An S4 object of class eQTLObject.
snpNumOfCellsPercent	Numeric. Minimum percentage of cells required for each SNP genotype (e.g., AA, AG, and GG). Only SNPs where each genotype occurs in at least this proportion of cells are retained. Default is 10.
expressionMin	Numeric. Expression threshold used in combination with expressionNumOfCellsPercent to filter lowly expressed genes. Default is 0.
expressionNumOfCellsPercent	Numeric. Minimum percentage of cells in which a gene's expression must exceed expressionMin for the gene to be retained. Default is 10.

Value

An updated eQTLObject with filtered gene expression and SNP matrices.

Examples

```
data(SNPData)
data(GeneData)
eqtl <- createQTLObject(snpMatrix = SNPData, genedata = GeneData)
eqtl <- normalizeGene(eqtl)
eqtl <- filterGeneSNP(eqtl,
  snpNumOfCellsPercent = 2,
  expressionMin = 0,
  expressionNumOfCellsPercent = 2)
```

filter_by_abs_b	<i>Filter a data frame of SNP–gene pairs by absolute beta.</i>
-----------------	--

Description

Filter a data frame of SNP–gene pairs by absolute beta.

Usage

```
filter_by_abs_b(result, logfcThreshold)
```

Arguments

result	A data frame that containing SNP–gene pairs and their beta values (b-value).
logfcThreshold	A numeric value specifying the minimum absolute b-value to retain. Default is 0.1.

Value

A data frame containing only rows with absolute b-values above the specified threshold.

Examples

```
example_result <- data.frame(
  gene = c("Gene1", "Gene2", "Gene3", "Gene4"),
  SNP = c("SNP1", "SNP2", "SNP3", "SNP4"),
  b = c(-2.5, 1.0, -0.5, 3.0))
logfcThreshold <- 0.1
filtered_result <- filter_by_abs_b(example_result, logfcThreshold)
```

get_cell_groups	<i>Retrieve Cells by SNP Value</i>
-----------------	------------------------------------

Description

This function extracts the names of cells from a SNP matrix that correspond to a specified value for a given SNP.

Usage

```
get_cell_groups(snpMatrix, SNPid, biClassify)
```

Arguments

snpMatrix	A genotype matrix where each row is a snp and each column is a cell. Encoding should be 0, 1, 2, 3.
SNPid	A character string or numeric index representing the specific SNP of interest in the SNP matrix.
biClassify	Logical; whether to convert genotype encoding in snpMatrix to 0, 1, and 2. TRUE indicates conversion; FALSE indicates no conversion (default).

Value

A list of character vectors. Each vector contains the names of cells (i.e., column names of snpMatrix) corresponding to a specific genotype value at the given SNP.

Examples

```
data(SNPData)
biClassify <- FALSE
get_cell_groups(SNPData, "1:632445", biClassify)
```

get_counts

Extract Counts from an Expression Matrix

Description

This function retrieves expression counts for a specified gene from an expression matrix, based on the provided list of cells.

Usage

```
get_counts(expressionMatrix, Geneid, cells)
```

Arguments

expressionMatrix	A numeric matrix of gene expression counts, with genes as rows and cells as columns.
Geneid	A character string or numeric index representing the specific gene of interest in expressionMatrix.
cells	A character vector of cell names (column names of expressionMatrix) from which to extract counts for the specified gene.

Value

A numeric vector containing the expression counts of the specified gene in the selected cells.

Examples

```
data(GeneData)
get_counts(GeneData, "CNN2",
           c("CGGCAGTGTAGCCCTG", "GGAGGATTCCC GTTCA"))
```

get_filter_data	<i>Access filtered data stored in an eQTLObject.</i>
-----------------	--

Description

Access filtered data stored in an eQTLObject.

Method to access eQTLObject filter data.

Usage

```
get_filter_data(x)

## S4 method for signature 'eQTLObject'
get_filter_data(x)
```

Arguments

x	An eQTLObject.
---	----------------

Value

Filtered matrices.

Filtered matrices.

Examples

```
data(EQTL_obj)
get_filter_data(EQTL_obj)
```

get_model_info	<i>Access model specification from an eQTLObj</i>
----------------	---

Description

Access model specification from an eQTLObj
 Method to access eQTLObj used model information.

Usage

```
get_model_info(x)

## S4 method for signature 'eQTLObj'
get_model_info(x)
```

Arguments

x An eQTLObj.

Value

A character string indicating the model.
 used model information of eQTLObj.

Examples

```
data(EQTL_obj)
get_model_info(EQTL_obj)
```

get_raw_data	<i>Access raw data stored in an eQTLObj.</i>
--------------	--

Description

Access raw data stored in an eQTLObj.
 Method to access eQTLObj raw data.

Usage

```
get_raw_data(x)

## S4 method for signature 'eQTLObj'
get_raw_data(x)
```

Arguments

x An eQTLObject.

Value

raw data matrix.

raw data matrix.

Examples

```
data(EQTL_obj)
get_raw_data(EQTL_obj)
```

get_result_info	<i>Access eQTLs results from an eQTLObject.</i>
-----------------	---

Description

Access eQTLs results from an eQTLObject.

Method to access the result of identifying eQTLs.

Usage

```
get_result_info(x)

## S4 method for signature 'eQTLObject'
get_result_info(x)
```

Arguments

x An eQTLObject.

Value

A data frame where each row corresponds to an identified SNP–gene pair.

A data frame with eQTL results.

Examples

```
data(EQTL_obj)
get_result_info(EQTL_obj)
```

```
initialize_progress_bar
```

Progress Bar for Model Analysis.

Description

Initializes a progress bar to track the computation progress during model fitting procedures such as `linearModel`, `poissonModel`, and `zinbModel`. This helps users monitor the status of long-running analyses.

Usage

```
initialize_progress_bar(total, k)
```

Arguments

<code>total</code>	Integer. The total number of iterations or steps to be completed in the analysis.
<code>k</code>	Character. An identifier or label for the specific group or subset being analyzed, used to annotate progress messages.

Value

An object of class `progress_bar` from the **progress** package, which can be updated using the `$tick()` method.

Examples

```
unique_group <- c("CMP", "GMP")
total_snp_count <- 10 # assume each group have 100 SNP.
pb_model <- lapply(unique_group, function(k) {
  pb <- initialize_progress_bar(total = total_snp_count, k)
  for (i in seq_len(total_snp_count)) {
    Sys.sleep(0.1) # assume progress time
    pb$tick() # update pb
  }
})
```

```
limma_normalize
```

Normalize the gene expression matrix with limma

Description

`limma_normalize()` normalizes an expression matrix using the quantile normalization method provided by the `limma` package.

Usage

```
limma_normalize(expressionMatrix)
```

Arguments

expressionMatrix

A numeric matrix of raw gene expression counts, with genes as rows and cells as columns.

Value

A normalized gene expression matrix after applying limma normalization.

Examples

```
data(GeneData)
limma_normalize(GeneData)
```

linearModel

Fit Linear Model for eQTL Mapping

Description

This function performs linear regression to identify gene–SNP associations based on single-cell expression and genotype data stored in an eQTLObject.

Usage

```
linearModel(
  eQTLObject,
  geneIDs,
  snpIDs,
  biClassify = FALSE,
  pAdjustMethod = "bonferroni",
  pAdjustThreshold = 0.05,
  logfcThreshold = 0.1
)
```

Arguments

eQTLObject	An S4 object of class eQTLObject.
geneIDs	Character vector of gene IDs to include in the model fitting.
snpIDs	Character vector of SNP IDs to include in the model fitting.
biClassify	Logical; whether to convert genotype encoding in snpMatrix to 0, 1, and 2. TRUE indicates conversion; FALSE indicates no conversion (default).
pAdjustMethod	Method used for multiple testing correction. One of "bonferroni", "holm", "hochberg", "hommel", or "BH". Default is "bonferroni".

pAdjustThreshold Only SNP–gene pairs with adjusted p-values below the threshold will be retained. Default is 0.05.

logfcThreshold The minimum beta coefficient (effect size) required to report a SNP–gene pair as an eQTL.

Value

A data frame of gene–SNP pairs that pass the filtering criteria, including beta coefficients, p-values, adjusted p-values, and group labels.

Examples

```
data(EQTL_obj)
Gene <- rownames(slot(EQTL_obj, "filterData")$expMat)
SNP <- rownames(slot(EQTL_obj, "filterData")$snpMat)
linearResult <- linearModel(
  eQTLObject = EQTL_obj,
  geneIDs = Gene,
  snpIDs = SNP,
  biClassify = FALSE,
  pAdjustMethod = "bonferroni",
  pAdjustThreshold = 0.05,
  logfcThreshold = 0.025)
```

load_biclassify_info *Access biclassification information from an eQTLObject.*

Description

Access biclassification information from an eQTLObject.

Method to access eQTLObject biclassify information.

Usage

```
load_biclassify_info(x)

## S4 method for signature 'eQTLObject'
load_biclassify_info(x)
```

Arguments

x An eQTLObject.

Value

A character or list containing biclassification information.

biclassify information of eQTLObject.

Examples

```
data(EQTL_obj)
load_biclassify_info(EQTL_obj)
```

load_group_info	<i>Access cell grouping information from an eQTLObject</i>
-----------------	--

Description

Access cell grouping information from an eQTLObject
Method to access eQTLObject cell grouping information.

Usage

```
load_group_info(x)

## S4 method for signature 'eQTLObject'
load_group_info(x)
```

Arguments

x An eQTLObject.

Value

A data frame with grouping information.
A data frame with cell group assignments.

Examples

```
data(EQTL_obj)
load_group_info(EQTL_obj)
```

load_species_info	<i>Access species information from an eQTLObject.</i>
-------------------	---

Description

Access species information from an eQTLObject.
Method to access eQTLObject species information.

Usage

```
load_species_info(x)

## S4 method for signature 'eQTLObject'
load_species_info(x)
```

Arguments

x An eQTLObject.

Value

A character string indicating the species.
species information of eQTLObject.

Examples

```
data(EQTL_obj)
load_species_info(EQTL_obj)
```

log_normalize

Normalize the gene expression matrix with logNormalize method.

Description

log_normalize() transforms an expression matrix by applying logarithm and scaling operations to normalize data.

Usage

```
log_normalize(expressionMatrix)
```

Arguments

expressionMatrix A numeric matrix of raw gene expression counts, with genes as rows and cells as columns.

Value

A normalized gene expression matrix after applying logNormalize normalization.

Examples

```
data(GeneData)
log_normalize(GeneData)
```

normalizeGene	<i>Normalize gene expression data.</i>
---------------	--

Description

This function performs normalization of the gene expression matrix stored within an `eQTLObject`, aiming to remove technical biases and improve comparability across cells. Multiple normalization methods are supported, including "logNormalize", "CPM", "TPM", "DESeq", and "limma".

Usage

```
normalizeGene(eQTLObject, method = "logNormalize")
```

Arguments

<code>eQTLObject</code>	An S4 object of class <code>eQTLObject</code> containing the raw gene expression matrix.
<code>method</code>	Character string specifying the normalization method to use. Must be one of "logNormalize", "CPM", "TPM", "DESeq", or "limma". Default is "logNormalize".

Value

An `eQTLObject` with the normalized gene expression matrix stored in the slot named "normExpMat".

Examples

```
data(EQTL_obj)
eqtl <- normalizeGene(EQTL_obj, method = "logNormalize")
```

plots_theme_opts	<i>Customized ggplot2 Theme for Plots</i>
------------------	---

Description

Customized ggplot2 Theme for Plots

Usage

```
plots_theme_opts()
```

Value

A ggplot2 theme object for styling plots.

Examples

```
library(ggplot2)
data <- data.frame(
  x = c("A", "B", "C", "D", "E"),
  y = c(10, 20, 30, 40, 50))
ggplot(data, aes(x, y)) +
  geom_point() +
  plots_theme_opts()
```

poissonModel

*Poisson model fitting the gene expression matrix and genotype matrix.***Description**

Poisson model fitting the gene expression matrix and genotype matrix.

Usage

```
poissonModel(
  eQTLObject,
  geneIDs,
  snpIDs,
  biClassify = FALSE,
  pAdjustMethod = "bonferroni",
  pAdjustThreshold = 0.05,
  logfcThreshold = 0.1
)
```

Arguments

eQTLObject	An S4 object of class eQTLObject.
geneIDs	Character vector of gene IDs to include in the model fitting.
snpIDs	Character vector of SNP IDs to include in the model fitting.
biClassify	Logical; whether to convert genotype encoding in snpMatrix to 0, 1, and 2. TRUE indicates conversion; FALSE indicates no conversion (default).
pAdjustMethod	Method used for multiple testing correction. One of "bonferroni", "holm", "hochberg", "hommel", or "BH". Default is "bonferroni".
pAdjustThreshold	Only SNP–gene pairs with adjusted p-values below the threshold will be retained. Default is 0.05.
logfcThreshold	The minimum beta coefficient (effect size) required to report a SNP–gene pair as an eQTL.

Value

A data frame of gene–SNP pairs that pass the filtering criteria, including beta coefficients, p-values, adjusted p-values, and group labels.

Examples

```

data(EQTL_obj)
Gene <- rownames(slot(EQTL_obj, "filterData")$expMat)
SNP <- rownames(slot(EQTL_obj, "filterData")$snpMat)
poissonResult <- poissonModel(
  eQTLObject = EQTL_obj,
  geneIDs = Gene,
  snpIDs = SNP,
  biClassify = FALSE,
  pAdjustMethod = "bonferroni",
  pAdjustThreshold = 0.05,
  logfcThreshold = 0.025
)

```

process_matrix

*Process a matrix to extract a row and convert it to a data frame.***Description**

Process a matrix to extract a row and convert it to a data frame.

Usage

```
process_matrix(id, matrix, name)
```

Arguments

id	Character string specifying the row name to extract from the matrix.
matrix	A matrix from which the row will be extracted.
name	Character string specifying the column name for the extracted values.

Value

A data frame containing the extracted row and a column with the row names.

Examples

```

rownames <- c("CNN2", "TIGD2", "DTD2")
colnames <- c("Col1", "Col2", "Col3", "Col4")
matrix_data <- matrix(1:12, nrow = 3, ncol = 4,
  dimnames = list(rownames, colnames))
geneid <- "CNN2"
gene_mat <- process_matrix(geneid, matrix_data, "gene_mat")

```

remove_outliers	<i>Remove outliers from gene expression data and update cell groups.</i>
-----------------	--

Description

This function identifies and removes outlier expression values for a specified gene based on the Median Absolute Deviation (MAD) method. It then filters provided genotype-based cell groups, returning only cells with non-outlier expression values.

Usage

```
remove_outliers(exprsMat, Geneid, A_cells, B_cells, C_cells = NULL)
```

Arguments

exprsMat	Input gene expression matrix with genes as rows and cells as columns.
Geneid	Character string specifying the gene ID to examine.
A_cells	Character vector of cell names belonging to genotype group A.
B_cells	Character vector of cell names belonging to genotype group B.
C_cells	Optional character vector of cell names belonging to genotype group C; if NULL, function returns two genotype groups.

Value

A named list of filtered cell vectors.

Examples

```
## Mock expression matrix
set.seed(123)
exprsMat <- matrix(rnorm(200), nrow = 5)
rownames(exprsMat) <- paste0("Gene", 1:nrow(exprsMat))
colnames(exprsMat) <- paste0("cell", 1:ncol(exprsMat))
A_cells <- colnames(exprsMat)[1:13] # Example A cell list
B_cells <- colnames(exprsMat)[14:26] # Example B cell list
C_cells <- colnames(exprsMat)[27:40] # Example C cell list
remove_outliers(exprsMat, "Gene1", A_cells, B_cells, C_cells)
```

set_filter_data	<i>Set filtered data in an eQTLObject.</i>
-----------------	--

Description

Set filtered data in an eQTLObject.

Method to set eQTLObject filter data.

Usage

```
set_filter_data(x, value, name)
```

```
## S4 method for signature 'eQTLObject'  
set_filter_data(x, value, name)
```

Arguments

x	An eQTLObject.
value	A matrix to be stored as filtered data.
name	A character string indicating the key under which the matrix is stored in filterData.

Value

An updated eQTLObject.

An updated eQTLObject.

Examples

```
data(EQTL_obj)  
data123 <- matrix(0, nrow = 3, ncol = 3)  
set_filter_data(EQTL_obj, data123, "expMat")  
  
data(EQTL_obj)  
data123 <- matrix(0, nrow = 3, ncol = 3)  
set_filter_data(EQTL_obj, data123, "expMat")
```

set_model_info	<i>Set model specification in an eQTLObject</i>
----------------	---

Description

Set model specification in an eQTLObject

Method to set eQTLObject used model information.

Usage

```
set_model_info(x, value)
```

```
## S4 method for signature 'eQTLObject'
set_model_info(x, value)
```

Arguments

x An eQTLObject.

value A character string specifying the model (e.g., "zinb", "poisson", "linear").

Value

An updated eQTLObject with the new model specification.

An updated eQTLObject.

Examples

```
data(EQTL_obj)
useModel <- "zinb"
set_model_info(EQTL_obj, useModel)
```

```
data(EQTL_obj)
useModel <- "zinb"
set_model_info(EQTL_obj, useModel)
```

set_raw_data	<i>Set raw data in an eQTLObject.</i>
--------------	---------------------------------------

Description

Set raw data in an eQTLObject.

Method to set eQTLObject raw data.

Usage

```
set_raw_data(x, value, name)

## S4 method for signature 'eQTLObject'
set_raw_data(x, value, name)
```

Arguments

x An eQTLObject.

value The raw data.

name A character string indicating the key under which the matrix is stored in rawData.

Value

eQTLObject.

An updated eQTLObject.

Examples

```
data(EQTL_obj)
data123 <- matrix(0, nrow = 3, ncol = 3)
set_raw_data(EQTL_obj, data123, "rawExpMat")

data(EQTL_obj)
data123 <- matrix(0, nrow = 3, ncol = 3)
set_raw_data(EQTL_obj, data123, "rawExpMat")
```

set_result_info	<i>Set eQTL results in an eQTLObject.</i>
-----------------	---

Description

Set eQTL results in an eQTLObject.

Method to set the result of identifying eQTLs from scRNA-seq data.

Usage

```
set_result_info(x, value)

## S4 method for signature 'eQTLObject'
set_result_info(x, value)
```

Arguments

x An eQTLObject.

value A data frame where each row corresponds to a SNP–gene pair.

Value

An updated eQTLObject.

An updated eQTLObject.

Examples

```
data(EQTL_obj)
result <- data.frame(0, nrow = 3, ncol = 3)
set_result_info(EQTL_obj, result)
```

```
data(EQTL_obj)
result <- matrix(0, nrow = 3, ncol = 3)
set_result_info(EQTL_obj, result)
```

TPM_normalize

Normalize the gene expression matrix with TPM

Description

TPM_normalize() scales an expression matrix using Transcripts Per Million (TPM) normalization, applying logarithm and scaling operations to adjust data based on library size.

Usage

```
TPM_normalize(expressionMatrix)
```

Arguments

expressionMatrix

A numeric matrix of raw gene expression counts, with genes as rows and cells as columns.

Value

A normalized gene expression matrix after applying TPM normalization.

Examples

```
data(GeneData)
TPM_normalize(GeneData)
```

visualizeQTL	<i>Visualize eQTL results by SNP–gene pair across groups</i>
--------------	--

Description

Visualize eQTL results by SNP–gene pair across groups

Usage

```
visualizeQTL(
  eQTLObject,
  SNPid,
  Geneid,
  groupName = NULL,
  plottype = "QTLplot",
  removeoutlier = FALSE
)
```

Arguments

eQTLObject	An S4 object of class eQTLObject.
SNPid	ID of SNP.
Geneid	ID of Gene.
groupName	A character vector specifying one or more cell group names to include in the plot.
plottype	A character string specifying the type of plot to generate. Must be one of "QTLplot", "violin", "boxplot", or "histplot".
removeoutlier	Logical; whether to identify and remove outliers. Default is FALSE.

Value

list

Examples

```
data(EQTL_obj)
## We have to call the eQTLs firstly using \code{\link{callQTL()}}.
eqtl <- callQTL(eQTLObject = EQTL_obj, useModel = "linear")
visualizeQTL(eQTLObject = eqtl,
  SNPid = "1:632647",
  Geneid = "RPS27",
  groupName = NULL,
  plottype = "QTLplot",
  removeoutlier = FALSE)
```

zinbModel	<i>Zinb model fitting the gene expression matrix.</i>
-----------	---

Description

Zinb model fitting the gene expression matrix.

Usage

```
zinbModel(
  eQTLObject,
  geneIDs,
  snpIDs,
  biClassify = FALSE,
  pAdjustMethod = "bonferroni",
  pAdjustThreshold = 0.05
)
```

Arguments

eQTLObject	An S4 object of class eQTLObject.
geneIDs	Character vector of gene IDs to include in the model fitting.
snpIDs	Character vector of SNP IDs to include in the model fitting.
biClassify	Logical; whether to convert genotype encoding in snpMatrix to 0, 1, and 2. TRUE indicates conversion; FALSE indicates no conversion (default).
pAdjustMethod	Method used for multiple testing correction. One of "bonferroni", "holm", "hochberg", "hommel", or "BH". Default is "bonferroni".
pAdjustThreshold	Only gene-SNP pairs with adjusted p-values below the threshold will be retained. Default is 0.05.

Value

A data frame of SNP–gene pairs that pass the filtering criteria, including p-values, adjusted p-values, and group labels.

Examples

```
data(EQTL_obj)
Gene <- rownames(slot(EQTL_obj, 'filterData')$expMat)
SNP <- rownames(slot(EQTL_obj, 'filterData')$snpMat)
zinbResult <- zinbModel(
  eQTLObject = EQTL_obj,
  geneIDs = Gene,
  snpIDs = SNP,
  biClassify = FALSE,
  pAdjustMethod = 'bonferroni',
  pAdjustThreshold = 0.05)
```

Index

- * **datasets**
 - DataSet, [10](#)
- * **internal**
 - scQTLtools-package, [3](#)
- adjust_pvalues, [4](#)
- buildZINB, [4](#)
- callQTL, [5](#)
- checkSNPList, [6](#)
- CPM_normalize, [7](#)
- createGeneLoc, [8](#)
- createQTLObject, [9](#), [11](#)
- createSNPsLoc, [10](#)
- DataSet, [10](#)
- DESeq_normalize, [11](#)
- draw_boxplot, [12](#)
- draw_histplot, [13](#)
- draw_QTLplot, [13](#)
- draw_violinplot, [14](#)
- EQTL_obj (DataSet), [10](#)
- eQTLObject-class, [15](#)
- filter_by_abs_b, [17](#)
- filterGeneSNP, [11](#), [16](#)
- GeneData (DataSet), [10](#)
- get_cell_groups, [17](#)
- get_counts, [18](#)
- get_filter_data, [19](#)
- get_filter_data, eQTLObject-method (get_filter_data), [19](#)
- get_model_info, [20](#)
- get_model_info, eQTLObject-method (get_model_info), [20](#)
- get_raw_data, [20](#)
- get_raw_data, eQTLObject-method (get_raw_data), [20](#)
- get_result_info, [21](#)
- get_result_info, eQTLObject-method (get_result_info), [21](#)
- initialize_progress_bar, [22](#)
- limma_normalize, [22](#)
- linearModel, [23](#)
- load_biclassify_info, [24](#)
- load_biclassify_info, eQTLObject-method (load_biclassify_info), [24](#)
- load_group_info, [25](#)
- load_group_info, eQTLObject-method (load_group_info), [25](#)
- load_species_info, [25](#)
- load_species_info, eQTLObject-method (load_species_info), [25](#)
- log_normalize, [26](#)
- normalizeGene, [11](#), [27](#)
- plots_theme_opts, [27](#)
- poissonModel, [28](#)
- process_matrix, [29](#)
- remove_outliers, [30](#)
- scQTLtools (scQTLtools-package), [3](#)
- scQTLtools-package, [3](#)
- set_filter_data, [31](#)
- set_filter_data, eQTLObject-method (set_filter_data), [31](#)
- set_model_info, [32](#)
- set_model_info, eQTLObject-method (set_model_info), [32](#)
- set_raw_data, [32](#)
- set_raw_data, eQTLObject-method (set_raw_data), [32](#)
- set_result_info, [33](#)
- set_result_info, eQTLObject-method (set_result_info), [33](#)

Seurat_obj (DataSet), [10](#)

SNPData (DataSet), [10](#)

SNPData2 (DataSet), [10](#)

TPM_normalize, [34](#)

visualizeQTL, [35](#)

zinbModel, [36](#)