Package ‘SLqPCR’

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Type Package

Title Functions for analysis of real-time quantitative PCR data at SIRS-Lab GmbH

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Description Functions for analysis of real-time quantitative PCR data at SIRS-Lab GmbH

Depends R(>= 2.4.0)

Imports stats

Suggests RColorBrewer

License GPL (>= 2)

biocViews MicrotitrePlateAssay, qPCR

NeedsCompilation no

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geneStabM

Description

Computation of the gene expression stability value M for real-time quantitative RT-PCR data. For more details we refer to Vandesompele et al. (2002).

Usage

geneStabM(relData, na.rm = FALSE)

Arguments

relData  matrix or data.frame containing real-time quantitative RT-PCR data
na.rm  a logical value indicating whether NA values should be stripped before the computation proceeds.

References

The gene expression stability value $M$ is defined as the average pairwise normalization factor; i.e., one needs to specify data from at least two genes. For more details see Vandesompele et al. (2002).

Value
numeric vector with gene expression stability values

Author(s)
Dr. Matthias Kohl (SIRS-Lab GmbH) <kohl@sirs-lab.com>

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Description
Computation of the geometric mean.

Usage
```r
geomMean(x, na.rm = FALSE)
```

Arguments
- `x` numeric vector of non-negative Reals
- `na.rm` a logical value indicating whether NA values should be stripped before the computation proceeds.

Details
The computation of the geometric mean is done via $\prod(x)^{(1/\text{length}(x))}$.

Value
geometric mean

Author(s)
Dr. Matthias Kohl (SIRS-Lab GmbH) <kohl@sirs-lab.com>
normPCR

Normalization of real-time quantitative RT-PCR data

Description

This function can be used to normalize real-time quantitative RT-PCR data.

Usage

normPCR(relData, HKs, method = "Vandesompele", na.rm = FALSE)

Arguments

relData  matrix or data.frame containing relative quantities (genes in columns)
HKs      integer, column numbers of housekeeping genes
method   method for the computation
na.rm    a logical value indicating whether NA values should be stripped before the com-
          putation proceeds.

Details

This function can be used to normalize real-time quantitative RT-PCR data. The default method
"Vandesompele" was proposed by Vandesompele et al. (2002).
Currently, only the method by Vandesompele et al. (2002) is implemented.

Value

Normalized expression data

Author(s)

Dr. Matthias Kohl (SIRS-Lab GmbH) <kohl@sirs-lab.com>

References

Jo Vandesompele, Katleen De Preter, Filip Pattyn et al. (2002). Accurate normalization of real-time
quantitative RT-PCR data by geometric averaging of multiple internal control genes. Genome Bi-

Examples

data(SLqPCRdata)
relData <- apply(SLqPCRdata, 2, relQuantPCR)
geneStabM(relData[,c(3,4)])
exprData <- normPCR(SLqPCRdata, c(3,4))
relQuantPCR

**Description**

Compute relative expression values for realtime quantitative RT-PCR data based on Ct or take-off values, respectively. The computations use the PCR efficiency.

**Usage**

relQuantPCR(x, E = 2, na.rm = FALSE)

**Arguments**

- **x**: numeric vector containing raw data
- **E**: PCR efficiency
- **na.rm**: a logical value indicating whether NA values should be stripped before the computation proceeds.

**Value**

vector of relative expression values w.r.t. specified PCR efficiency.

**Author(s)**

Dr. Matthias Kohl (SIRS-Lab GmbH) <kohl@sirs-lab.com>

**References**


selectHKgenes

**Selection of reference/housekeeping genes**

**Description**

This function can be used to determine a set of reference/housekeeping (HK) genes for gene expression experiments.

**Usage**

selectHKgenes(relData, method = "Vandesompele", minNrHK = 2, geneSymbol, trace = TRUE, na.rm = FALSE)
Arguments

relData matrix or data.frame containing relative expression values
method method to compute most stable genes
minNrHK minimum number of HK genes that should be considered
geneSymbol gene symbols
trace logical, print additional information
na.rm a logical value indicating whether NA values should be stripped before the computation proceeds.

Details

This function can be used to determine a set of reference/housekeeping (HK) genes for gene expression experiments. The default method "Vandesompele" was proposed by Vandesompele et al. (2002).

Currently, only the method by Vandesompele et al. (2002) is implemented.

Vandesompele et al. (2002) propose a cut-off value of 0.15 for the pairwise variation. Below this value the inclusion of an additional housekeeping gene is not required.

Value

If method = "Vandesompele" a list with the following components is returned

ranking ranking of genes from best to worst where the two most stable genes cannot be ranked
variation pairwise variation during stepwise selection
meanM average expression stability M

Author(s)

Dr. Matthias Kohl (SIRS-Lab GmbH) <kohl@sirs-lab.com>

References


Examples

data(vandesompele)
res.BM <- selectHKgenes(vandesompele[1:9,], method = "Vandesompele", geneSymbol = names(vandesompele), minNrHK = 2, trace = TRUE, na.rm = TRUE)
SLqPCRdata

SIRS-Lab inhouse qPCR data

Description

This data is part of a SIRS-Lab inhouse real-time quantitative PCR experiment.

Usage

data(SLqPCRdata)

Format

A data frame with 16 observations on the following 4 variables.

Gene1 a numeric vector, average take-off values of gene 1
Gene2 a numeric vector, average take-off values of gene 2
HK1 a numeric vector, average take-off values of housekeeper 1
HK2 a numeric vector, average take-off values of housekeeper 2

Details

The row names of this data set indicate the probes which were investigated. The take-off values are mean values of three replicates.

Source

www.sirs-lab.com

References

www.sirs-lab.com

Examples

data(SLqPCRdata)
SLqPCRdata

vandesompele Data set of Vandesompele et al (2002)

Description

This data set was used in Vandesompele et al (2002) to demonstrate normalization of real-time quantitative RT-PCR data by geometric averaging of housekeeping genes.

Usage

data(vandesompele)
Format

A data frame with 85 observations on the following 10 variables which stand for expression data of ten commonly used housekeeping genes

ACTB  actin, beta
B2M  beta-2-microglobulin
GAPD  glyceraldehyde-3-phosphate dehydrogenase
HMBS  hydroxymethylbilane synthase
HPRT1  hypoxanthine phosphoribosyltransferase 1
RPL13A  ribosomal protein L13a
SDHA  succinate dehydrogenase complex subunit A
TBP  TATA box binding protein
UBC  ubiquitin C
YWHAZ  tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, zeta polypeptide

Details

The row names of this data set indicate the various human tissues which were investigated.

BM  9 normal bone-marrow samples
POOL  9 normal human tissues from pooled organs (heart, brain, fetal brain, lung, trachea, kidney, mammary gland, small intestine and uterus)
FIB  20 short-term cultured normal fibroblast samples from different individuals
LEU  13 normal leukocyte samples
NB  34 neuroblastoma cell lines (independently prepared in different labs from different patients)

Source

The data set was obtained from http://genomebiology.com/content/supplementary/gb-2002-3-7-research0034-s1.txt

References


Examples

data(vandesompele)
str(vandesompele)
rownames(vandesompele)
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