Package ‘DTA’

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Title Dynamic Transcriptome Analysis
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Depends R (>= 2.10), LSD
Imports scatterplot3d
Description Dynamic Transcriptome Analysis (DTA) can monitor the cellular response to perturbations with higher sensitivity and temporal resolution than standard transcriptomics. The package implements the underlying kinetic modeling approach capable of the precise determination of synthesis- and decay rates from individual microarray or RNAseq measurements.
License Artistic-2.0
biocViews Microarray, DifferentialExpression, GeneExpression, Transcription
DTA.plots.R DTA.phenomat.R DTA.generate.r DTA.estimate.r
DTA.dynamic.estimate.r DTA.dynamic.generate.R
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R topics documented:

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The DTA package implements all methods of the quantitative kinetic modeling approach belonging to DTA (Dynamic Transcriptome Analysis) to estimate mRNA synthesis and decay rates from individual time point measurements.
**Details**

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</tr>
</tbody>
</table>

**Author(s)**

Bjoern Schwalb <schwalb@lmb.uni-muenchen.de>

**References**


**Examples**

```r
## see vignette or supplemental material of the given references.
```

---

| Dm.tnumber | The amount of thymines in the cDNA of each transcript of Drosophila Melanogaster. |

**Description**

The amount of thymines in the cDNA of each transcript of all Drosophila Melanogaster Ensembl transcript IDs (Flybase transcript number), to assess the uridine-dependent labeling bias and eventually correct for it.

**Usage**

```r
Dm.tnumber
```

**Format**

Vector gives the number of thymines in the cDNA (uridine residues in RNA) of each Ensembl transcript ID.
**Source**


---

**Doelken2008**

*Mus Musculus and Homo Sapiens DTA experiment from Doelken et al.*

**Description**

R object contains all relevant *.RData files needed for the DTA.estimate function. For example, see vignette.

**Usage**

Doelken2008

**Format**

R object contains the following *.RData files: Hs.phenomat Hs.datamat Hs.reliable Hs.enst2ensg Hs.tnumber Mm.phenomat Mm.datamat Mm.reliable Mm.enst2ensg Mm.tnumber

**Source**


---

**DTA.dynamic.estimate**

*Estimation of synthesis and decay rates upon perturbation*

**Description**

DTA.dynamic.estimate uses an experiment, given by a phenotype matrix, data matrix and the number of uridines for each gene to estimate synthesis and decay rate of the genes.
Usage

DTA.dynamic.estimate(phenomat = NULL, datamat = NULL, tnumber = NULL, ccl = NULL, mRNAs = NULL, reliable = NULL, mediancenter = ... = NULL, upperquant = 0.8, lowerquant = 0.6,notinR = FALSE, RStudio = FALSE, simulation = FALSE, sim.object = NULL)

Arguments

phenomat A phenotype matrix, containing the design of the experiment as produced by DTA.phenomat. Columns are name, fraction (U=unlabeled, L=labeled, T=total), time and nr (=replicate number). Rows represent individual experiments.
datamat A matrix, containing the measurements from U, L and T, according to the design given in phenomat. Matrix should only contain the rows of phenomat as columns.
tnumber Integer vector, containing the numbers of uridines. Elements should have the rownames of datamat.
ccl The cell cycle length of the cells.
mRNAs Estimated number of mRNAs in a cell (optional).
reliable Vector of 'reliable' genes, which are used for parameter estimation.
mediancenter Should the quotient Labeled/Total resp. Unlabeled/Total be rescaled to a common median over it's replicates before building the genewise median.
usefractions From which fractions should the decay rate be calculated: "LandT", "UandT" or "both".
LtoTratio Coefficient to rescale Labeled/Total. Is estimated from the data, if not specified. See ratiomethod.
ratiomethod Choose the regression method to be used, possible methods are: "tls", "bias" and "lm". For details, see supplemental material of Sun et al. (see references).
largest Percentage of largest residues from the first regression not to be used in the second regression step. For details, see supplemental material of Sun et al. (see references).
weighted Should the regression be weighted with 1/(Total^2 + median(Total))? 
relevant Choose the arrays to be used for halflives calculation, vector due to nr (=replicate number) in phenomat.
check If check = TRUE, control messages and plots will be generated.
error If TRUE, the measurement error is assessed by means of an error model and resampling to gain confidence regions.
samplesize Error model samplesize for resampling.
confidence.range Confidence region for error model as quantiles. Interval should be between 0 and 1.
bicor Should the labeling bias be corrected? 
condition String, to be added to the plotnames.
upper Upper bound for labeling bias estimation. For details, see supplemental material of Sun et al. (see references).
lower Lower bound for labeling bias estimation. For details, see supplemental material of Sun et al. (see references).
save.plots If save.plots = TRUE, control plots will be saved.
resolution Resolution scaling factor for plotting.
DTA.dynamic.estimate

folder Path to the folder, where to save the plots.
fileformat Fileformat for plots to be saved. See plotit function (LSD package).
totaloverwt Will be available in the very near future for comparative DTA data.
sr.vs.dr.folds.lims Limits of the folds plot (dr vs sr).
te.vs.to.folds.lims Limits of the folds plot (LT vs LE).
robust If robust = TRUE, LE resp. LT is chosen instead of sr resp. dr.
clusters should the dr vs sr folds be plotted with clusters, choose 'sr', 'dr' for cluster selection or 'none' to omit it
ranktime at which time should the rankgain be calculated, default is the last column
upperquant upper quantile for cluster selection
lowerquant lower quantile for cluster selection
notinR Should plots be not plotted in R.
RStudio For RStudio users. Suppresses the opening of a new device, as RStudio allows only one.
simulation True, if data was generated by DTA.generate.
sim.object Simulation object created by DTA.generate.

Value

DTA.dynamic.estimate returns a list, where each entry contains the estimation results for all replicates of one timecourse timepoint. Each result contains the following entries

triples Mapping of each fraction and experiment to its corresponding column in the data matrix.
plabel The labeling efficiency. For details, see the vignette.
LtoTratio Estimated ratio of labeled to total fraction.
UtoTratio Estimated ratio of unlabeled to total fraction.
LtoUrati Estimated ratio of labeled to unlabeled fraction.
correcteddatamat Labeling bias corrected data matrix.
drmat Decay rates for each replicate. The last column gives the median decay rates.
dr Median decay rates. The last column of drmat.
dr.confidence Confidence regions of decay rates.
hlmat Half-lives for each replicate. The last column gives the median half-lifes.
hl Median half-lives. The last column of hlmat.
hl.confidence Confidence regions of half-lives.
TEmat Total expression for each replicate. The last column gives the median total expression values.
TE Median total expression values. The last column of TEmat.
TE.confidence Confidence regions of total expression values.
LEmat Labeled expression for each replicate. The last column gives the median labeled expression values.
LE Median labeled expression values. The last column of LEmat.
LE.confidence    Confidence regions of labeled expression values.
UEmat            Unlabeled expression for each replicate. The last column gives the median unlabeled expression values. (Only if unlabeled values exist in the experiment)
UE               Median unlabeled expression values. The last column of UEmat. (Only if unlabeled values exist in the experiment)
UE.confidence    Confidence regions of unlabeled expression values.
srmat             Synthesis rates for each replicate. The last column gives the median synthesis rates.
sr               Median synthesis rates. The last column of srmat.
sr.confidence    Confidence regions of synthesis rates.
LtoTmat           Labeled to total ratio for each replicate. The last column gives the median labeled to total ratios.
LtoT              Median labeled to total ratios. The last column of LtoTmat.
LtoT.confidence   Confidence regions of labeled to total ratios.
UtoTmat           Unlabeled to total ratio for each replicate. The last column gives the median unlabeled to total ratios.
UtoT              Median unlabeled to total ratios. The last column of UtoTmat.
UtoT.confidence   Confidence regions of unlabeled to total ratios.
Rsrmat            Rescaled synthesis rates for each replicate, if parameter mRNAs is specified. The last column gives the median synthesis rates.
Rsr               Rescaled median synthesis rates. The last column of Rsrmat.
globaldrmat       Decay rate for each replicate. Reciprocally weighted by the total expression. Last element contains (weighted) median decay rate.
globaldr           (Weighted) median decay rate.

Author(s)
Bjoern Schwalb <schwalb@lmb.uni-muenchen.de>

References

See Also
heatscatter, plotit, tls
Examples

```r
dataPath = system.file("data", package="DTA")
load(file.path(dataPath, "Miller2011dynamic.RData"))

### for control plots set 'check = TRUE' ###

res = DTA.dynamic.estimate(Sc.phenomat.dynamic,Sc.datamat.focus,Sc.tnumber,ccl = 150,mRNAs = 60000,reliable = Sc.reliable.focus)
```

---

**DTA.dynamic.generate**  
*Simulation of DTA experiments upon perturbation*

**Description**

DTA.dynamic.generate produces the phenotype matrix and the matrix containing the simulated data according to the given parameters.

**Usage**

```r
DTA.dynamic.generate(duration = 60,lab.duration = 6,tnumber = NULL,plabel = NULL,nrgenes = 5000,mediantime.halflives = NULL,mediantime.synthesisrates = NULL,n = NULL,ccl = NULL,check = NULL,plots = NULL,save.plots = NULL,folder = NULL,condition = NULL,addformat = NULL,sdnoise = NULL,nobias = NULL,unspecific.LtoU = NULL)
```

**Arguments**

<table>
<thead>
<tr>
<th>Argument</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>duration</td>
<td>duration of the whole time course (min)</td>
</tr>
<tr>
<td>lab.duration</td>
<td>labeling duration for single experiments (min)</td>
</tr>
<tr>
<td>tnumber</td>
<td>Integer vector containing the number of uridine residues for each gene. If NULL, tnumber is sampled from an F-distribution within the function.</td>
</tr>
<tr>
<td>plabel</td>
<td>The labeling efficiency. If NULL, plabel is set to 0.005 within the function. For details, see supplemental material of Sun et al. (see references).</td>
</tr>
<tr>
<td>nrgenes</td>
<td>The number of genes the simulated experiment will have (will be cropped if it exceeds the length of tnumber).</td>
</tr>
<tr>
<td>mediantime.halflives</td>
<td>the median of the half life distribution</td>
</tr>
<tr>
<td>mediantime.synthesisrates</td>
<td>the median of the synthesis rates distribution (counts/cell/cellcycle)</td>
</tr>
<tr>
<td>n</td>
<td>the number of cells N(0)</td>
</tr>
<tr>
<td>ccl</td>
<td>The cell cycle length (in minutes).</td>
</tr>
<tr>
<td>check</td>
<td>if check=TRUE, control messages will be generated</td>
</tr>
<tr>
<td>plots</td>
<td>if plots = TRUE, control plots will be plotted</td>
</tr>
<tr>
<td>save.plots</td>
<td>if save.plots = TRUE, control plots will be saved</td>
</tr>
<tr>
<td>folder</td>
<td>folder, where to save the plots</td>
</tr>
<tr>
<td>condition</td>
<td>to be added to the plotnames</td>
</tr>
<tr>
<td>addformat</td>
<td>additional fileformat for plots to be saved</td>
</tr>
<tr>
<td>sdnoise</td>
<td>The amount of measurement noise (proportional to expression strength).</td>
</tr>
<tr>
<td>nobias</td>
<td>Should a labeling bias be added?</td>
</tr>
<tr>
<td>unspecific.LtoU</td>
<td>Proportion of labeled RNAs that unspecifically end up in the unlabeled fraction.</td>
</tr>
</tbody>
</table>
unspec.LtoU.weighted
Should unspecific proportion of labeled to unlabeled depend linearly on the length of the RNA?

unspec.UtoL
Proportion of unlabeled RNAs that unspecifically end up in the labeled fraction.

unspec.UtoL.weighted
Should unspecific proportion of unlabeled to labeled depend linearly on the length of the RNA?

mu.values.mat
if the data should be generated using given synthesis rates, this matrix must contain the respective values for each gene

mu.breaks.mat
timepoints of synthesis rate changes, this matrix must contain the respective values for each gene, only needed when mu.values.mat is given (one column less than mu.values.mat)

lambda.values.mat
if the data should be generated using given decay rates, this matrix must contain the respective values for each gene

lambda.breaks.mat
timepoints of decay rate changes, this matrix must contain the respective values for each gene, only needed when lambda.values.mat is given (one column less than lambda.values.mat)

truehalflives
If the data should be generated using a given half-life distribution, this vector must contain the respective values for each gene.

truesynthesisrates
If the data should be generated using a given synthesis rates distribution, this vector must contain the respective values for each gene

genenames
An optional list of gene names.

Value

DTA.dynamic.generate returns a list, containing the following entries

phenomat
A matrix, containing the design of the experiment as produced by DTA.phenomat.

datamat
A matrix, containing the simulated measurements from U, L and T, according to the design given in phenomat.

tnumber
Integer vector containing the number of uridine residues for each gene.

ccl
The cell cycle length (in minutes).

truemus
A vector, containing the true synthesis rates.

truemusaveraged
A vector, containing the true synthesis rates, averaged over the labeling period.

truelambdas
A vector, containing the true decay rates.

truelambdasaveraged
A vector, containing the true decay rates, averaged over the labeling period.

truehalflives
A vector, containing the true half-lives.

truehalflivesaveraged
A vector, containing the true half-lives, averaged over the labeling period.

trueplabel
The true labeling efficiency. For details, see supplemental material of Sun et al. (see references).

truecomplete
A vector, containing the true amount of total RNA.
true lambdas  A vector, containing the true decay rates.
true mus  A vector, containing the true synthesis rates.
true halflives  A vector, containing the true half-lives.
trueplabel  The true labeling efficiency. For details, see supplemental material of Miller et al. (see references).
truear  The true parameter ar. For details, see supplemental material of Miller et al. (see references).
truebr  The true parameter br. For details, see supplemental material of Miller et al. (see references).
truecr  The true parameter cr. For details, see supplemental material of Miller et al. (see references).
truecrbyar  The true parameter cr/ar. For details, see supplemental material of Miller et al. (see references).
truecrbybr  The true parameter cr/br. For details, see supplemental material of Miller et al. (see references).
truebrbyar  The true parameter br/ar. For details, see supplemental material of Miller et al. (see references).
trueLasymptote  The true parameter asymptote (labeled bias). For details, see supplemental material of Miller et al. (see references).
trueUasymptote  The true parameter asymptote (unlabeled bias). For details, see supplemental material of Miller et al. (see references).

Author(s)
Bjoern Schwalb <schwalb@lmb.uni-muenchen.de>

References

Examples
```
nrgenes = 5000
truesynthesisrates = rf(nrgenes,5,5)*18
steady = rep(1,nrgenes)
shock = 1/pmax(rnorm(nrgenes,mean = 8,sd = 4),1)
induction = pmax(rnorm(nrgenes,mean = 8,sd = 4),1)
changes.mat = cbind(steady,shock,shock*induction)
mu.values.mat = changes.mat*truesynthesisrates
mu.breaks.mat = cbind(rep(12,nrgenes),rep(18,nrgenes))
truehalflives = rf(nrgenes,15,15)*12
true lambdas = log(2)/truehalflives
changes.mat = cbind(steady,shock,shock*induction,steady)
lambda.values.mat = changes.mat*true lambdas
```
DTA.estimate

You are provided with a function `DTA.estimate`, which is used to estimate synthesis and decay rates of genes. The function requires several arguments, including phenotype matrices, data matrices, and the number of uridines for each gene. Here is a brief description and usage of the function, along with the arguments:

### Description

`DTA.estimate` uses an experiment, given by a phenotype matrix, data matrix and the number of uridines for each gene to estimate synthesis and decay rate of the genes.

### Usage

```r
DTA.estimate(phenomat = NULL, datamat = NULL, tnumber = NULL, reliable = NULL, ccl = NULL, mRNAs = NULL, mediancenter = ..., usefractions = c("LandT", "UandT", "both"), ratiomethod = c("tls", "bias", "lm"), LtoTratio, largest = NULL)
```

### Arguments

- **phenomat**: A phenotype matrix, containing the design of the experiment as produced by `DTA.phenomat`. Columns are name, fraction (U=unlabeled, L=labeled, T=total), time and nr (=replicate number). Rows represent individual experiments.
- **datamat**: A matrix, containing the measurements from U, L and T, according to the design given in phenomat. Matrix should only contain the rows of phenomat as columns.
- **tnumber**: Integer vector, containing the numbers of uridine residues for each transcript. Elements should have the rownames of datamat.
- **ccl**: The cell cycle length of the cells (optional). Is not modeled, if not set.
- **mRNAs**: Estimated number of mRNAs in a cell (optional).
- **reliable**: Vector of 'reliable' genes, which are used for parameter estimation.
- **mediancenter**: Should the quotient Labeled/Total resp. Unlabeled/Total be rescaled to a common median over it's replicates before building the genewise median.
- **usefractions**: From which fractions should the decay rate be calculated: "LandT", "UandT" or "both".
- **ratiomethod**: Choose the regression method to be used, possible methods are: "tls", "bias" and "lm". For details, see supplemental material of Sun et al. (see references). Method to estimate the parameter LtoTratio, which determines the median half-life of the sample.
- **LtoTratio**: Coefficient to rescale Labeled/Total. Is estimated from the data, if not specified. See ratiomethod. Altering this parameter leads to a altered median half-life. For details, see supplemental material of Sun et al. (see references).
- **largest**: Percentage of largest residues from the first regression not to be used in the second regression step. For details, see supplemental material of Sun et al. (see references).
weighted Should the regression be weighted with \(1/(\text{Total}^2 + \text{median(Total)})\)?

relevant Choose the arrays to be used for halflives calculation, vector due to nr (=replicate number) in phenomat. If not set, all arrays are used.

check If check = TRUE, control messages and plots will be generated.

error If TRUE, the measurement error is assessed by means of an error model and resampling to gain confidence regions.

samplesize Error model samplesize for resampling.

confidence.range Confidence region for error model as quantiles. Interval should be between 0 and 1.

bicor Should the labeling bias be corrected?

condition String, to be added to the plotnames if saved.

upper Upper bound for labeling bias estimation. For details, see supplemental material of Sun et al. (see references).

lower Lower bound for labeling bias estimation. For details, see supplemental material of Sun et al. (see references).

save.plots If save.plots = TRUE, control plots will be saved. Please check folder writability.

resolution Resolution scaling factor for plotting. (Scaled with 72dpi.)

notinR If TRUE, plots are not plotted in R.

RStudio For RStudio users. Suppresses the opening of a new device, as RStudio allows only one.

folder Path to the folder, where to save the plots. Needs to be writable.

fileformat Fileformat for plots to be saved. See plotit function (LSD package). Save the plot as "jpeg", "png", "bmp", "tiff", "ps" or "pdf".

totaloverwt Only needed when mRNAs is set. Should give the factor by which the total mRNA of the condition outreaches that of the reference (comparative DTA data).

simulation True, if data was generated by DTA.generate.

sim.object Simulation object created by DTA.generate.

Value

DTA.estimate returns a list, where each entry contains the estimation results for all replicates of one labeling time. Each result contains the following entries

triples Mapping of each fraction and experiment to its corresponding column in the data matrix.

plabel The labeling efficiency. For details, see supplemental material of Sun et al. (see references).

LtoTratio Estimated ratio of labeled to total fraction.

UtoTratio Estimated ratio of unlabeled to total fraction.

LtoUratio Estimated ratio of labeled to unlabeled fraction.

correcteddatamat Labeling bias corrected data matrix.

drmat Decay rates for each replicate. The last column gives the median decay rates.

dr Median decay rates. The last column of drmat.
dr.confidence  Confidence regions of decay rates.
hlmat        Half-lives for each replicate. The last column gives the median half-lifes.
hl           Median half-lives. The last column of hlmat.
hl.confidence Confidence regions of half-lives.
TEmat        Total expression for each replicate. The last column gives the median total expression values.
TE           Median total expression values. The last column of TEmat.
TE.confidence Confidence regions of total expression values.
LEmat        Labeled expression for each replicate. The last column gives the median labeled expression values.
LE           Median labeled expression values. The last column of LEmat.
LE.confidence Confidence regions of labeled expression values.
UEmat        Unlabeled expression for each replicate. The last column gives the median unlabeled expression values. (Only if unlabeled values exist in the experiment)
UE           Median unlabeled expression values. The last column of UEmat. (Only if unlabeled values exist in the experiment)
UE.confidence Confidence regions of unlabeled expression values.
srmat        Synthesis rates for each replicate. The last column gives the median synthesis rates.
sr           Median synthesis rates. The last column of srmat.
sr.confidence Confidence regions of synthesis rates.
LtoTmat      Labeled to total ratio for each replicate. The last column gives the median labeled to total ratios.
LtoT         Median labeled to total ratios. The last column of LtoTmat.
LtoT.confidence Confidence regions of labeled to total ratios.
UtoTmat      Unlabeled to total ratio for each replicate. The last column gives the median unlabeled to total ratios.
UtoT         Median unlabeled to total ratios. The last column of UtoTmat.
UtoT.confidence Confidence regions of unlabeled to total ratios.
Rsrmat       Rescaled synthesis rates for each replicate, if parameter mRNAs is specified. The last column gives the median synthesis rates.
Rsr          Rescaled median synthesis rates. The last column of Rsrmat.
globaldrmat  Decay rate for each replicate. Reciprocally weighted by the total expression. Last element contains (weighted) median decay rate.
globaldr     (Weighted) median decay rate.

Author(s)
Bjoern Schwalb <schwalb@lmb.uni-muenchen.de>
DTA.generate

**References**


**See Also**

heatscatter, plotit.tls

**Examples**

```r
dataPath = system.file("data", package="DTA")
load(file.path(dataPath, "Miller2011.RData"))

### for control plots set 'check = TRUE' ###

res = DTA.estimate(Sc.phenomat,Sc.datamat,Sc.tnumber,ccl = 150,mRNAs = 60000,reliable = Sc.reliable,check = FALSE)
```

**DTA.generate**

*Simulation of DTA experiments*

**Description**

DTA.generate produces the phenotype matrix and the matrix containing the simulated data according to the given parameters.

**Usage**

```r
DTA.generate(timepoints, tnumber = NULL, plabel = NULL, nrgenes = 5000, mediantime = 12, ccl = 150, delaytime = 0, unspec.UtoL.weighted = FALSE, truehalflives = NULL, truecomplete = NULL, genenames = NULL, cDTA = FALSE)
```

**Arguments**

- `timepoints` Integer vector containing the labeling times for which the samples should be generated.
- `tnumber` Integer vector containing the number of uridine residues for each gene. If NULL, tnumber is sampled from an F-distribution within the function.
- `plabel` The labeling efficiency. If NULL, plabel is set to 0.005 within the function. For details, see supplemental material of Sun et al. (see references).
- `nrgenes` The number of genes the simulated experiment will have (will be cropped if it exceeds the length of tnumber).
- `mediantime` The median of the randomly drawn half-life distribution.
- `ccl` The cell cycle length (in minutes).
- `delaytime` Estimates the delay between the moment of 4sU/4tU labeling and actual incorporation of it into mRNA.
The amount of measurement noise (proportional to expression strength).

Should a labeling bias be added?

Proportion of labeled RNAs that unspecifically end up in the unlabeled fraction.

Should unspecific proportion of labeled to unlabeled depend linearly on the length of the RNA?

Proportion of unlabeled RNAs that unspecifically end up in the labeled fraction.

Should unspecific proportion of unlabeled to labeled depend linearly on the length of the RNA?

If the data should be generated using a given half-life distribution, this vector must contain the respective values for each gene.

If the data should be generated using a given expression distribution, this vector must contain the respective values for each gene.

An optional list of gene names.

cDTA = FALSE does not rescale L and U.

DTA.generate returns a list, containing the following entries:

A matrix, containing the design of the experiment as produced by DTA.phenomat.

A matrix, containing the simulated measurements from U, L and T, according to the design given in phenomat.

Integer vector containing the number of uridine residues for each gene.

The cell cycle length (in minutes).

A vector, containing the true amount of total RNA.

A vector, containing the true decay rates.

A vector, containing the true synthesis rates.

A vector, containing the true half-lives.

The true labeling efficiency. For details, see supplemental material of Miller et al. (see references).

The true parameter ar. For details, see supplemental material of Miller et al. (see references).

The true parameter br. For details, see supplemental material of Miller et al. (see references).

The true parameter cr. For details, see supplemental material of Miller et al. (see references).

The true parameter cr/ar. For details, see supplemental material of Miller et al. (see references).

The true parameter cr/br. For details, see supplemental material of Miller et al. (see references).

The true parameter br/ar. For details, see supplemental material of Miller et al. (see references).
trueLasymptote The true parameter asymptote (labeled bias). For details, see supplemental material of Miller et al. (see references).

trueLasymptote The true parameter asymptote (unlabeled bias). For details, see supplemental material of Miller et al. (see references).

Author(s)
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References

Examples
```r
sim.object = DTA.generate(timepoints=rep(c(6,12),2))
### for control plots set 'check = TRUE' ###
res.sim = DTA.estimate(ratiomethod = "bias",simulation = TRUE,sim.object = sim.object,check = FALSE)
```

**DTA.map.it**

Mapping function to switch between different identifiers.

Description
DTA.map.it can map different kinds of identifiers in a matrix or a vector given by mapping vector.

Usage
```r
DTA.map.it(mat, map = NULL, check = TRUE)
```

Arguments
- **mat** Matrix or vector with numerical entries.
- **map** Vector of identifiers to map to, named by identifiers to map from.
- **check** Should check protocol be printed.

Author(s)
Bjoern Schwalb <schwalb@lmb.uni-muenchen.de>
DTA.normalize

References


Examples

### see vignette examples or reference:
### B. Schwalb, B. Zacher, S. Duemcke, D. Martin, P. Cramer, A. Tresch.

**DTA.normalize**

**cDTA normalization procedure.**

**Description**

DTA.normalize can normalize expression values from a certain species to the median of values from a reference species.

**Usage**

`DTA.normalize(mat, reliable = NULL, logscale = FALSE, protocol = FALSE, center = FALSE)`

**Arguments**

- **mat**: Expression matrix.
- **reliable**: The rows to be used, i.e. identifiers of the reference species to normalize on.
- **logscale**: Is the matrix in log-scale?
- **protocol**: Should a protocol be printed?
- **center**: Should the center be 0 (log-scale) or 1 (absolute scale). Otherwise the median of the medians is taken.

**Author(s)**

Bjoern Schwalb <schwalb@lmb.uni-muenchen.de>

**References**


**Examples**

### see vignette examples or reference:
### B. Schwalb, B. Zacher, S. Duemcke, D. Martin, P. Cramer, A. Tresch.
**DTA.phenomat**

Create a phenomat that suits your experiment.

**Description**

DTA.phenomat creates a phenomat for a given experimental design, i.e. used labeling times.

**Usage**

```
DTA.phenomat(timepoints, timecourse = NULL)
```

**Arguments**

- **timepoints**
  The respective labeling times of the measured samples.

- **timecourse**
  Vector giving the order for timecourse DTA data.

**Value**

A matrix, containing the design of the experiment. Columns are name, fraction (U=unlabeled, L=labeled, T=total), time and nr (=replicate number). Rows represent individual experiments. For timecourse data, an additional column of the order of the underlying timecourse data can be added via `timecourse`.

**Author(s)**

Bjoern Schwalb <schwalb@lmb.uni-muenchen.de>

**Examples**

```r
### phenomat for 2 replicates of 6 and 12 min labeling duration resp.
DTA.phenomat(c(6,12))

### phenomat for three adjacent timepoints measured in 2 replicates
DTA.phenomat(rep(6,6), timecourse = 1:3)
```

---

**DTA.plot.it**

Plots in any format and any quality

**Description**

DTA.plot.it can save plots in any format and any quality in addition to show them in R devices.

**Usage**

```
DTA.plot.it(filename, sw = 1, sh = 1, sres = 1, plotsfkt, ww = 7, wh = 7, pointsize = 12, dev.pointsize = 8, p
```
Arguments

filename       Name of the plot to be saved without the format type suffix.
sw             Scaling factor of width. Scaled with 480px.
sh             Scaling factor of height. Scaled with 480px.
sres           Scaling factor of the resolution. Scaled with 72dpi.
plotsfkt      Function of plots to be plotted.
ww             Width of window. Needed only for plotting in R or if filformat = "pdf" or "ps". See pdf or ps.
wh             Height of window. Needed only for plotting in R or if filformat = "pdf" or "ps". See pdf or ps.
pointsize      The default pointsize of plotted text, interpreted as big points (1/72 inch) for plots to be saved.
dev.pointsize  Pointsize of plotted text, interpreted as big points (1/72 inch) for display in R.
paper          Needed only if filformat = "pdf" or "ps". See pdf or ps.
quality        Needed only if filformat = "jpeg". See jpeg.
units          Needed only if filformat = "jpeg", "png", "bmp" or "tiff". See corresponding function.
bg              Backgroundcolor.
fileformat     Save the plot as "jpeg", "png", "bmp", "tiff", "ps" or "pdf".
saveit         Should plot be saved.
notinR         Should plot be not plotted in R.
RStudio        For RStudio users. Suppresses the opening of a new device, as RStudio allows only one.
addformat      Should plot be saved additionally in another format, "jpeg", "png", "bmp", "tiff", "ps" or "pdf".

Author(s)

Bjoern Schwalb <schwalb@lmb.uni-muenchen.de>

Examples

plotsfkt = function(){
  par(mfrow = c(1,2))
  plot(1:10)
  plot(10:1)
}
DTA.plot.it(filename = "test",plotsfkt = plotsfkt,saveit = TRUE)

dev.off()
### Hs.datamat

**Gene expression profiles of the Homo Sapiens DTA experiment from Doelken et al.**

**Description**

This matrix contains the RNA intensity values for each gene across each RNA fraction and their replicate measurements of the Homo Sapiens DTA experiment from Doelken et al.

**Usage**

Hs.datamat

**Format**

The column names of the matrix give the cel-file name and the row names the Ensembl gene IDs.

**Source**


### Hs.enst2ensg

**Mapping of Homo Sapiens gene and transcript identifiers.**

**Description**

Mapping from Ensembl transcript IDs to Ensembl gene IDs of Homo Sapiens.

**Usage**

Hs.enst2ensg

**Format**

Vector gives the Ensembl gene IDs, names the Ensembl transcript IDs.

**Source**

Description

The phenotype matrix Hs.phenomat contains information about the experimental design. It is comprised of the filename, the type of RNA fraction measured (T, U or L), the labeling time and the replicate number.

Usage

Hs.phenomat

Format

The phenomat is a matrix comprised of the file name, the type of RNA fraction measured (T, U or L, fraction column), the labeling time (time, timeframe column) and the replicate number (nr column). Rows in this matrix represent the individual experiments.

Source


Description

Ensembl gene IDs, that passed certain criteria among the Homo Sapiens Doelken et al. DTA experiment to be considered valid for parameter estimation. For details, see vignette.

Usage

Hs.reliable

Format

Vector of Ensembl gene IDs that can be passed to DTA.estimate for parameter estimation.

Source

**Hs.tnumber**  
*The amount of thymines in the cDNA of each transcript of Homo Sapiens.*

**Description**

The amount of thymines in the cDNA of each transcript of all Homo Sapiens Ensembl transcript IDs, to assess the uridine-dependent labeling bias and eventually correct for it.

**Usage**

Hs.tnumber

**Format**

Vector gives the number of thymines in the cDNA (uridine residues in RNA) of each Ensembl transcript ID.

**Source**


---

**Miller2011**  
*Saccharomyces Cerevisiae wild-type DTA experiment from Miller et al.*

**Description**

R object contains all relevant *.RData files needed for the DTA.estimate function. For example, see vignette.

**Usage**

Miller2011

**Format**

R object contains the following *.RData files: Sc.phenomat Sc.datamat Sc.reliable Sc.tnumber*
Source


Miller2011dynamic

Saccharomyces Cerevisiae salt stress DTA experiment from Miller et al.

Description

R object contains all relevant *.RData files needed for the DTA.estimate function. For example, see vignette.

Usage

Miller2011dynamic

Format

R object contains the following *.RData files: Sc.phenomat.dynamic Sc.datamat.dynamic Sc.reliable.dynamic Sc.tnumber

Source

**Mm.datamat**

Gene expression profiles of the *Mus Musculus* DTA experiment from Doelken et al.

**Description**
This matrix contains the RNA intensity values for each gene across each RNA fraction and their replicate measurements of the *Mus Musculus* DTA experiment from Doelken et al.

**Usage**

Mm.datamat

**Format**
The column names of the matrix give the cel-file name and the row names the Ensembl gene IDs.

**Source**

---

**Mm.enst2ensg**

Mapping of *Mus Musculus* gene and transcript identifiers.

**Description**
Mapping from Ensembl transcript IDs to Ensembl gene IDs of *Mus Musculus*.

**Usage**

Mm.enst2ensg

**Format**
Vector gives the Ensembl gene IDs, names the Ensembl transcript IDs.

**Source**
Design of the Mus Musculus DTA experiment from Doelken et al.

**Description**

The phenotype matrix `Mm.phenomat` contains information about the experimental design. It is comprised of the filename, the type of RNA fraction measured (T, U or L), the labeling time and the replicate number.

**Usage**

`Mm.phenomat`

**Format**

The phenomat is a matrix comprised of the file name, the type of RNA fraction measured (T, U or L, `fraction` column), the labeling time (`time`, `timeframe` column) and the replicate number (`nr` column). Rows in this matrix represent the individual experiments.

**Source**


**Mm.reliable**

Gene identifiers valid for parameter estimation from the Mus Musculus Doelken et al. DTA experiment.

**Description**

Ensembl gene IDs, that passed certain criteria among the Mus Musculus Doelken et al. DTA experiment to be considered valid for parameter estimation. For details, see vignette.

**Usage**

`Mm.reliable`

**Format**

Vector of Ensembl gene IDs that can be passed to `DTA.estimate` for parameter estimation.

**Source**

Mm.tnumber  
*The amount of thymines in the cDNA of each transcript of Mus Musculus.*

**Description**

The amount of thymines in the cDNA of each transcript of all Mus Musculus Ensembl transcript IDs, to assess the uridine-dependent labeling bias and eventually correct for it.

**Usage**

Mm.tnumber

**Format**

Vector gives the number of thymines in the cDNA (uridine residues in RNA) of each Ensembl transcript ID.

**Source**


Pol.phenomat  
*Design of the Saccharomyces Cerevisiae rpb1-N488D (Slow Polymerase) cDTA experiment from Sun et al.*

**Description**

The phenotype matrix Pol.phenomat contains information about the experimental design. It is comprised of the filename, the type of RNA fraction measured (T, U or L), the labeling time and the replicate number.

**Usage**

Pol.phenomat

**Format**

The phenomat is a matrix comprised of the file name, the type of RNA fraction measured (T, U or L, fraction column), the labeling time (time, timeframe column) and the replicate number (nr column). Rows in this matrix represent the individual experiments.
Source

<table>
<thead>
<tr>
<th>Raw.datamat</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gene expression profiles of the Saccharomyces Cerevisiae rpb1-N488D (Slow Polymerase) and wild-type cDTA experiment from Sun et al.</td>
<td></td>
</tr>
</tbody>
</table>

Description
This matrix contains the RNA intensity values for each gene across each RNA fraction and their replicate measurements of the Saccharomyces Cerevisiae rpb1-N488D (Slow Polymerase) and wild-type cDTA experiment from Sun et al.

Usage
Raw.datamat

Format
The column names of the matrix give the cel-file name and the row names the affymetrix IDs.

Source

<table>
<thead>
<tr>
<th>Sc.affy2ensg</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mapping of SaccharomycesCerevisiae Affymetrix Yeast 2.0 and gene identifiers.</td>
<td></td>
</tr>
</tbody>
</table>

Description
Mapping from Affymetrix Yeast 2.0 IDs to Ensembl gene IDs of SaccharomycesCerevisiae.

Usage
Sc.affy2ensg

Format
Vector gives the Ensembl gene IDs, names the Affymetrix Yeast 2.0 IDs.

Source
Sc.datamat

*Gene expression profiles of the Saccharomyces Cerevisiae wild-type DTA experiment from Miller et al.*

**Description**
This matrix contains the RNA intensity values for each gene across each RNA fraction and their replicate measurements of the Saccharomyces Cerevisiae wild-type DTA experiment from Miller et al.

**Usage**
Sc.datamat

**Format**
The column names of the matrix give the cel-file name and the row names the Ensembl gene IDs.

**Source**

---

Sc.datamat.dynamic

*Gene expression profiles of the Saccharomyces Cerevisiae salt stress DTA experiment from Miller et al.*

**Description**
This matrix contains the RNA intensity values for each gene across each RNA fraction and their replicate measurements of the Saccharomyces Cerevisiae salt stress DTA experiment from Miller et al.

**Usage**
Sc.datamat.dynamic

**Format**
The column names of the matrix give the cel-file name and the row names the Ensembl gene IDs.

**Source**
**Sc.ensg.reliable**

**Gene identifiers valid for parameter estimation from the *Saccharomyces Cerevisiae* Sun et al. cDTA experiment.**

**Description**

Ensembl gene IDs, that passed certain criteria among the *Saccharomyces Cerevisiae* Sun et al. cDTA experiment to be considered valid for parameter estimation. For details, see Sun et al (Materials and Methods).

**Usage**

Sc.ensg.reliable

**Format**

Vector of Ensembl gene IDs that can be passed to DTA_estimate for parameter estimation.

**Source**


---

**Sc.phenomat**

**Design of the *Saccharomyces Cerevisiae* wild-type DTA experiment from Miller et al.**

**Description**

The phenotype matrix Sc.phenomat contains information about the experimental design. It is comprised of the filename, the type of RNA fraction measured (T, U or L), the labeling time and the replicate number.

**Usage**

Sc.phenomat

**Format**

The phenomat is a matrix comprised of the file name, the type of RNA fraction measured (T, U or L, fraction column), the labeling time (time, timeframe column) and the replicate number (nr column). Rows in this matrix represent the individual experiments.

**Source**

### Sc.phenomat.dynamic

**Description**

The phenotype matrix `Sc.phenomat.dynamic` contains information about the experimental design. It is comprised of the filename, the type of RNA fraction measured (T, U or L), the labeling time, the replicate number and an additional number indicating the timecourse order.

**Usage**

```r
Sc.phenomat.dynamic
```

**Format**

The phenomat is a matrix comprised of the file name, the type of RNA fraction measured (T, U or L, fraction column), the labeling time (time.timeframe column), the replicate number (nr column) and a number indicating the timecourse order (timecourse column). Rows in this matrix represent the individual experiments.

**Source**


### Sc.reliable

**Description**

Ensembl gene IDs, that passed certain criteria among the Saccharomyces Cerevisiae Miller et al. wild-type DTA experiment to be considered valid for parameter estimation. For details, see supplemental material Miller et al.

**Usage**

```r
Sc.reliable
```

**Format**

Vector of Ensembl gene IDs that can be passed to `DTA.estimate` for parameter estimation.

**Source**

### Sc.reliable.dynamic

**Gene identifiers valid for parameter estimation from the Saccharomyces Cerevisiae Miller et al. salt stress DTA experiment.**

**Description**

Ensembl gene IDs, that passed certain criteria among the Saccharomyces Cerevisiae Miller et al. salt stress DTA experiment to be considered valid for parameter estimation. For details, see supplemental material Miller et al.

**Usage**

```r
Sc.reliable.dynamic
```

**Format**

Vector of Ensembl gene IDs that can be passed to `DTA.estimate` for parameter estimation.

**Source**


---

### Sc.ribig.ensg

**Ribosome biogenesis genes.**

**Description**

ORF identifiers (Ensembl Gene ID) found to be associated with ribosome biogenesis, rRNA processing etc.

**Usage**

```r
Sc.ribig.ensg
```

**Format**

Vector of ORF identifiers (Ensembl Gene ID).

**Source**

**Sc.rpg.ensg**

**Ribosomal protein genes.**

**Description**

ORF identifiers (Ensembl Gene ID) encoding for ribosomal protein genes.

**Usage**

Sc.rpg.ensg

**Format**

Vector of ORF identifiers (Ensembl Gene ID).

**Source**


---

**Sc.stress.ensg**

**ISA stress module.**

**Description**

ORF identifiers (Ensembl Gene ID) found to be associated with stress response by the iterative signature algorithm.

**Usage**

Sc.stress.ensg

**Format**

Vector of ORF identifiers (Ensembl Gene ID).

**Source**

**Sc.tf.ensg**

**Transcription factors.**

**Description**

ORF identifiers (Ensembl Gene ID) encoding for transcription factors.

**Usage**

Sc.tf.ensg

**Format**

Vector of ORF identifiers (Ensembl Gene ID).

**Source**


---

**Sc.tnumber**

The amount of thymines in the cDNA of each transcript of Saccharomyces Cerevisiae.

**Description**

The amount of thymines in the cDNA of each transcript of all Saccharomyces Cerevisiae Ensembl transcript IDs (ORF identifier), to assess the uridine-dependent labeling bias and eventually correct for it.

**Usage**

Sc.tnumber

**Format**

Vector gives the number of thymines in the cDNA (uridine residues in RNA) of each Ensembl transcript ID.

**Source**

**Sp.affy.reliable**  
*Gene identifiers valid for cDTA normalization from the Saccharomyces Cerevisiae Sun et al. cDTA experiment.*

**Description**

Ensembl gene IDs, that passed certain criteria among the Saccharomyces Cerevisiae Sun et al. cDTA experiment to be considered valid for cDTA normalization. For details, see Sun et al (Materials and Methods).

**Usage**

Sp.affy.reliable

**Format**

Vector of Schizosaccharomyces Pombe affymetrix IDs that can be passed to dTA.normalize for cDTA normalization of the Saccharomyces Cerevisiae identifiers.

**Source**


---

**Sp.tnumber**  
The amount of thymines in the cDNA of each transcript of Schizosaccharomyces Pombe.

**Description**

The amount of thymines in the cDNA of each transcript of all Schizosaccharomyces Pombe Ensembl transcript IDs (ORF identifier), to assess the uridine-dependent labeling bias and eventually correct for it.

**Usage**

Sp.tnumber

**Format**

Vector gives the number of thymines in the cDNA (uridine residues in RNA) of each Ensembl transcript ID.
Source


Description

R object contains all relevant *.RData files needed for the DTA.estimate function. For example, see Schwalb et al.

Usage

Sun2011

Format

R object contains the following *.RData files: Raw.datamat Sp.affy.reliable Sc.affy2ensg Wt.phenomat Pol.phenomat Sc.ensg.reliable Sc.tnumber

Source

Description


Usage

tls(formula, D = NULL, T = NULL, precision = .Machine$double.eps)

Arguments

formula: An object of class formula.
D: Diagonal weight matrix. Default weights are set to 1.
T: Diagonal weight matrix. Default weights are set to 1.
precision: Smallest possible numeric value on this machine (default).

Value

tls returns a lm object.

Author(s)

Sebastian Duemcke <duemcke@lmb.uni-muenchen.de>

References


Examples

f = 1.5  # true ratio
a = rnorm(5000)
b = f*a
a = a + rnorm(5000, sd=0.5)
b = b + rnorm(5000, sd=0.5)

coeff.tls = coef(tls(b ~ a + 0))
coeff.lm1 = coef(lm(b ~ a + 0))
coeff.lm2 = 1/coef(lm(a ~ b + 0))

heatscatter(a,b)
abline(0,coeff.lm1,col="red",pch=19,lwd=2)
abline(0,coeff.lm2,col="orange",pch=19,lwd=2)
abline(0,coeff.tls,col="green",pch=19,lwd=2)
abline(0,f,col="grey",pch=19,lwd=2,lty=2)
legend("topleft", c("Least-squares regr. (y ~ x + 0)", "Least-squares regr. (x ~ y + 0)", "Total Least-squares regr.", "True ratio"), col=c("red", "orange", "green", "grey"), lty=c(1,1,1,2), lwd=2)

results = c(coeff.tls,coeff.lm1,coeff.lm2)
names(results) = c("coeff.tls","coeff.lm1","coeff.lm2")
print(results)
Wt.phenomat

Design of the Saccharomyces Cerevisiae wild-type cDTA experiment from Sun et al.

Description

The phenotype matrix Wt.phenomat contains information about the experimental design. It is comprised of the filename, the type of RNA fraction measured (T, U or L), the labeling time and the replicate number.

Usage

Wt.phenomat

Format

The phenomat is a matrix comprised of the file name, the type of RNA fraction measured (T, U or L, fraction column), the labeling time (time, timeframe column) and the replicate number (nr column). Rows in this matrix represent the individual experiments.

Source

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