

Package ‘MANOR’

April 7, 2025

Version 1.79.2

Date 2024-03-12

Title CGH Micro-Array NORmalization

Description Importation, normalization, visualization, and quality control functions to correct identified sources of variability in array-CGH experiments.

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Depends R (>= 2.10)

Imports GLAD, graphics, grDevices, stats, utils

Suggests knitr, rmarkdown, bookdown

VignetteBuilder knitr

URL <http://bioinfo.curie.fr/projects/manor/index.html>

BugReports <https://github.com/pneuvial/MANOR/issues>

Encoding UTF-8

biocViews Microarray, TwoChannel, DataImport, QualityControl, Preprocessing, CopyNumberVariation, Normalization

git_url <https://git.bioconductor.org/packages/MANOR>

git_branch devel

git_last_commit 8aeff0e

git_last_commit_date 2025-03-12

Repository Bioconductor 3.21

Date/Publication 2025-04-07

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| | |
|------------|--|
| arrayTrend | <i>Spatial trend of microarray spots statistic</i> |
|------------|--|

Description

The function arrayTrend computes the spatial trend.

Usage

```
## Default S3 method:
arrayTrend(Statistic, Col, Row, ...)
## S3 method for class 'arrayCGH'
arrayTrend(arrayCGH, variable, ...)
```

Arguments

| | |
|-----------|--|
| Statistic | Statistic to be smoothed. |
| Col | Vector of columns coordinates. |
| Row | Vector of rows coordinates. |
| arrayCGH | Object of class arrayCGH . |
| variable | Variable to be smooth. |
| ... | Parameters to be passed to loess function. |

Details

Spatial trend of microarray spots statistic.

Value

Either a data frame with elements :

| | |
|-------|---|
| Trend | Trend fitted by loess function. |
| Col | Vector of columns coordinates. |
| Row | Vector of rows coordinates. |

or the element Trend is added to the data.frame arrayValues of the arrayCGH object.

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Philippe Hupé, <philippe.hupe@curie.fr>.

References

P. Neuvial, P. Hupé, I. Brito, S. Liva, E. Mani, C. Brennetot, A. Aurias, F. Radvanyi, and E. Barillot. *Spatial normalization of array-CGH data*. BMC Bioinformatics, 7(1):264. May 2006.

See Also

[loess](#), [loess.control](#).

Examples

```
data(spatial) ## arrays with local spatial effects

edgeTrend <- arrayTrend(edge, "LogRatio", span=0.03, degree=1,
iterations=3, family="symmetric")
GLAD::arrayPlot(edgeTrend, "Trend", main="Spatial trend of array CGH", bar="v")
```

| | |
|----------|-------------------------------|
| detectSB | <i>Spatial bias detection</i> |
|----------|-------------------------------|

Description

This function detects spatial bias on array CGH.

Usage

```
## S3 method for class 'arrayCGH'
detectSB(arrayCGH, variable, proportionup=0.25,
proportiondown,type="up", thresholdup=0.2, thresholddown=0.2, ... )
```

Arguments

| | |
|----------------|--|
| arrayCGH | Object of arrayCGH . |
| variable | Variable used to compare the mean of zones detected by nem |
| proportionup | Maximal proportion of the array which may be affected by spatial bias with high values. |
| proportiondown | Maximal proportion of the array which may be affected by spatial bias with low values. |
| type | Type of spatial bias detected. Specify either "up" (to detect spatial bias with high values), or "down" (to detect spatial bias with low values) or "upanddown" (to detect both type of spatial bias). |
| thresholdup | Threshold used to detect spatial bias with high values. |
| thresholddown | Threshold used to detect spatial bias with low values. |
| ... | ... |

Details

You must run the [arrayTrend](#) and [nem](#) function before detecting spatial bias: the [arrayTrend](#) computes a spatial trend and the [nem](#) function performs a classification with spatial constraints defining different zones on the array. Based on those results, spatial bias is detected.

Value

An object of class `arrayCGH` with the following added information in the `data.frame` attribute `arrayValues`:

| | |
|----|---|
| SB | Spots located in zone of spatial bias are coded either by 1 (if they correspond to a spatial bias with high values) or by -1 (if they correspond to a spatial bias with low values). Otherwise they are coded by 0. |
|----|---|

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Philippe HupÃ©, <philippe.hupe@curie.fr>.

References

P. Neuvial, P. Hup?, I. Brito, S. Liva, E. Mani?, C. Brennetot, A. Aurias, F. Radvanyi, and E. Barillot. *Spatial normalization of array-CGH data*. BMC Bioinformatics, 7(1):264. May 2006.

See Also

[arrayTrend](#), [nem](#)

Examples

```
data(spatial) ## arrays with local spatial effects

## Plot of LogRatio measured on the array CGH
GLAD::arrayPlot(edge,"LogRatio", main="Log2-Ratio measured on the array
CGH", zlim=c(-1,1), bar="v", mediancenter=TRUE)

## Spatial trend of the scaled log-ratios (the variable "ScaledLogRatio"
## equals to the log-ratio minus the median value of the corresponding
## chromosome arm)

edgeTrend <- arrayTrend(edge, variable="ScaledLogRatio",
span=0.03, degree=1, iterations=3, family="symmetric")
GLAD::arrayPlot(edgeTrend, variable="Trend", main="Spatial trend of the
array CGH", bar="v")

## Not run:
## Classification with spatial constraint of the spatial trend
edgeNem <- nem(edgeTrend, variable="Trend")
GLAD::arrayPlot(edgeNem, variable="ZoneNem", main="Spatial zones identified
by nem", bar="v")

# Detection of spatial bias
edgeDet <- detectSB(edgeNem, variable="LogRatio", proportionup=0.25,type="up", thresholdup=0.15)
GLAD::arrayPlot(edgeDet, variable="SB", main="Zone of spatial bias in red", bar="v")

# CGH profile
plot(LogRatio ~ PosOrder, data=edgeDet$arrayValues,
col=c("black","red")[as.factor(SB)], pch=20, main="CGH profile: spots
located in spatial bias are in red")

## End(Not run)
```

| | |
|---------------|------------------------------------|
| flag.arrayCGH | <i>Apply a flag to an arrayCGH</i> |
|---------------|------------------------------------|

Description

Function flag\$FUN is applied to a flag object for normalization

Usage

```
flag.arrayCGH(flag, arrayCGH)
```

Arguments

| | |
|----------|--|
| flag | an object of type 'flag' |
| arrayCGH | an object of type arrayCGH |

Details

Optional arguments in flag\$args are passed to flag\$FUN

Value

An object of class arrayCGH, which corresponds to the return value of flag\$FUN if flag\$char is null, and to the input arrayCGH object with spots given by flag\$FUN flagged with flag\$char

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

See Also

[to.flag](#), [norm.arrayCGH](#)

Examples

```
data(spatial)
data(flags)

gradient$arrayValues$LogRatioNorm <- gradient$arrayValues$LogRatio
## flag spots with no available position on the genome
gradient <- flag.arrayCGH(position.flag, gradient)

## flag spots corresponding to low poor quality clones
gradient <- flag.arrayCGH(val.mark.flag, gradient)
```

```

## flag spots excluded by Genepix pro
gradient <- flag.arrayCGH(spot.flag, gradient)

## flag local spatial bias zones
## Not run: gradient <- flag.arrayCGH(local.spatial.flag, gradient)

## correct global spatial bias
gradient <- flag.arrayCGH(global.spatial.flag, gradient)

## flag spots with low signal to noise
gradient <- flag.arrayCGH(SNR.flag, gradient)

## flag spots with extremely high log-ratios
gradient <- flag.arrayCGH(amplicon.flag, gradient)

## flag spots with poor within replicate consistency
gradient <- flag.arrayCGH(replicate.flag, gradient)

## flag spots corresponding to clones for which all other spot
## replicates have already been flagged
gradient <- flag.arrayCGH(unique.flag, gradient)

summary.factor(gradient$arrayValues$Flag)

```

flag.summary

Summarize information about flags after array normalization

Description

Compute spot-level information (number of flagged spots, normalization parameters), and display it in a convenient way

Usage

```

## S3 method for class 'arrayCGH'
flag.summary(arrayCGH, flag.list, flag.var="Flag", nflab="not flagged", ...)
## Default S3 method:
flag.summary(spot.flags, flag.list, nflab="not flagged", ...)

```

Arguments

| | |
|-----------|---|
| arrayCGH | an object of type arrayCGH, <i>after normalization</i> by MANOR |
| flag.list | a list of flags with flag\$char corresponding to the values of spot.flags |
| flag.var | the name of a variable of arrayCGH\$arrayValues containing information about flags (defaults to Flag) |
| var | the name of a variable of arrayCGH\$cloneValues containing signal values (defaults to LogRatio) |

| | |
|------------|---|
| spot.flags | a character vector containing information about flags |
| nflab | a character vector providing a legend for "not flagged" spots |
| ... | ... |

Details

This function is used by the function `html.report` for the generation of an HTML report of the normalization step. It can also be used by itself.

Value

A data.frame data.frame with 4 columns:

| | |
|-------|--|
| name | flag character |
| label | flag label |
| arg | first numeric argument of <code>flag\$FUN</code> |
| count | number of flagged spots |

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

See Also

[html.report](#), [flag](#)

Examples

```
data(spatial)
data(flags)
flag.list <- list(spatial=local.spatial.flag, spot=spot.corr.flag,
ref.snr=ref.snr.flag, dapi.snr=dapi.snr.flag, rep=rep.flag,
unique=unique.flag)
flag.list$spatial$args <- alist(var="ScaledLogRatio", by.var=NULL,
nk=5, prop=0.25, thr=0.15, beta=1, family="symmetric")
flag.list$spot$args <- alist(var="SpotFlag")
flag.list$spot$char <- "0"
flag.list$spot$label <- "Image analysis"

## normalize arrayCGH
## Not run: edge.norm <- norm(edge, flag.list=flag.list,
var="LogRatio", FUN=median, na.rm=TRUE)
## End(Not run)
fs <- flag.summary(edge.norm, flag.list=flag.list, flag.var="Flag")
```



```
print("Flag and normalization parameters summary")
print(fs)
```

 flags

Examples of flag objects to apply to CGH arrays

Description

This data set provides `flag` objects that can be applied to `arrayCGH` objects in order to normalize them.

Usage

```
data(flags)
```

Format

These flag objects typically take part to a normalization process:

| | |
|----------------------------------|---|
| <code>amplicon.flag</code> | flags spots with high log-ratios (temp flag) |
| <code>chromosome.flag</code> | flags spots located on sexual chromosomes (named "X" and "Y") |
| <code>control.flag</code> | flag control spots |
| <code>global.spatial.flag</code> | corrects arrayCGH from global spatial trend on the array |
| <code>local.spatial.flag</code> | flags spots belonging to local spatial bias zones on the array |
| <code>num.chromosome.flag</code> | flags spots located on sexual chromosomes (named 23 and 24) |
| <code>position.flag</code> | flag spots with no available genome position |
| <code>replicate.flag</code> | flag spots with poor within-clone-replicate consistency |
| <code>ref.snr.flag</code> | flags spots with low signal to noise ratio for reference |
| <code>dapi.snr.flag</code> | flags spots with low signal to noise ratio for DAPI |
| <code>SNR.flag</code> | flags spots with low signal to noise ratio |
| <code>spot.corr.flag</code> | flags spots with low correlation coefficient after image analysis |
| <code>spot.flag</code> | flags spots excluded by the image analysis software |
| <code>unique.flag</code> | exclude last non-flagged spot of a clone |
| <code>val.mark.flag</code> | flags spots corresponding to bad quality clones |
| <code>intensity.flag</code> | corrects for an intensity effect (using loess regression) |

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

Source

Institut Curie, <manor@curie.fr>.

See Also

[spatial](#), [norm.arrayCGH](#), [flag](#), [flag.summary](#)

Examples

```

data(flags)

### complete normalization of an arrayCGH object (with spatial gradient):
## Initialize flag$args

flag.list1 <- list(local.spatial=local.spatial.flag,
  global.spatial=global.spatial.flag, spot=spot.flag, SNR=SNR.flag,
  val.mark=val.mark.flag, unique=unique.flag,
  amplicon=amplicon.flag, chromosome=chromosome.flag,
  replicate=replicate.flag)

data(spatial)
## Not run: gradient.norm <- norm(gradient, flag.list=flag.list1,
var="LogRatio", FUN=median, na.rm=TRUE)
## End(Not run)
print(gradient.norm$flags) ## spot-level flag summary (computed by flag.summary)

### complete normalization of an arrayCGH object (with local spatial bias):
## Initialize flag$args

flag.list2 <- list(spatial=local.spatial.flag, spot=spot.corr.flag,
  ref.snr=ref.snr.flag, dapi.snr=dapi.snr.flag, rep=rep.flag,
  unique=unique.flag)
flag.list2$spatial$args <- alist(var="ScaledLogRatio", by.var=NULL,
  nk=5, prop=0.25, thr=0.15, beta=1, family="symmetric")
flag.list2$spot$args <- alist(var="SpotFlag")
flag.list2$spot$char <- "0"
flag.list2$spot$label <- "Image analysis"

## Not run: edge.norm <- norm(edge, flag.list=flag.list2,
var="LogRatio", FUN=median, na.rm=TRUE)
## End(Not run)
print(edge.norm$flags) ## spot-level flag summary (computed by flag.summary)

```

genome.plot

Pan-genomic representation of a normalized arrayCGH

Description

Displays a pan-genomic representation of a normalized arrayCGH.

Usage

```
## S3 method for class 'arrayCGH'
genome.plot(arrayCGH, x="PosOrder", y="LogRatio",
            chrLim=NULL, col.var=NULL, clim=NULL, cex=NULL, pch=NULL, ...)
## Default S3 method:
genome.plot(data, pch=NULL, cex=NULL, xlab="", ylab="", ...)
```

Arguments

| | |
|----------|--|
| arrayCGH | an object of type arrayCGH |
| data | a data frame with two columns: 'x' and 'y', and optionally a column 'chrLim' giving the limits of each chromosome |
| x | a variable name from arrayCGH\$cloneValues giving the order position of the clones along the genome (defaults to 'PosOrder') |
| y | a variable name from arrayCGH\$cloneValues to be plotted along the genome (defaults to 'LogRatio') |
| chrLim | an optional variable name from arrayCGH\$cloneValues giving the limits of each chromosome |
| col.var | a variable name from arrayCGH\$cloneValues defining the color legend |
| clim | a numeric vector of length 2: color range limits (used if col.var is numeric) |
| cex | a numerical value giving the amount by which plotting text and symbols should be scaled relative to the default: see par |
| xlab | a title for the x axis: see title |
| ylab | a title for the y axis: see title |
| pch | either an integer specifying a symbol or a single character to be used as the default in plotting points: see par |
| ... | further arguments to be passed to plot |

Details

if col.var is a numeric variable, y colors are proportionnal to col.var values; if it is a character variable or a factor, one color is assigned to each different value of col.var. If col.var is NULL, colors are proportionnal to y values.

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

See Also

[flag](#), [report.plot](#)

Examples

```

data(spatial)

## default color code: log-ratios
## Not run:
genome.plot(edge.norm, chrLim="LimitChr")

## End(Not run)

## color code determined by a qualitative variable: ZoneGNL (DNA copy number code)
edge.norm$cloneValues$ZoneGNL <- as.factor(edge.norm$cloneValues$ZoneGNL)
## Not run:
genome.plot(edge.norm, col.var="ZoneGNL")

## End(Not run)
## comparing profiles with and without normalization
## aggregate data without normalization (flags)

gradient.nonorm <- norm(gradient, flag.list=NULL, var="LogRatio",
FUN=median, na.rm=TRUE)
gradient.nonorm <- sort(gradient.nonorm)

## Not run:
genome.plot(gradient.nonorm, pch=20, main="Genomic profile without
normalization", chrLim="LimitChr")
x11()
genome.plot(gradient.norm, pch=20, main="Genomic profile with
normalization", chrLim="LimitChr")

## End(Not run)

```

html.report

Generate an HTML report of array normalization

Description

Create an HTML file with various illustrations of array normalization, including plots before and after normalization, and statistics about flagged spots and clones

Usage

```

## S3 method for class 'arrayCGH'
html.report(array.norm, array.nonorm=NULL, dir.out=".",
array.name=NULL, x="PosOrder", y=c("LogRatioNorm", "LogRatio"), chrLim=NULL,
ylim=NULL, zlim=NULL, clim=NULL, intensity=NULL, light=FALSE,
file.name="report", width=10, height=5, ...)

## Default S3 method:
html.report(spot.data, clone.data=NULL,
flag.data=NULL, quality.data=NULL, ...)

```

Arguments

| | |
|--------------|---|
| array.norm | an object of type arrayCGH after normalization step |
| array.nonorm | an optional object of type arrayCGH after a normalization step with no flags |
| spot.data | a data.frame containing spot-level informations (e.g. arrayCGH\$arrayValues) |
| clone.data | a data.frame containing clone-level informations (e.g. arrayCGH\$cloneValues) |
| flag.data | a data.frame containing information about flags, with fields char, label, arg, count as generated by function flag.summary |
| quality.data | a data.frame containing information about quality scores with fields name, label, score as generated by function qscore.summary |
| dir.out | absolute path of a directory where the file is generated (defaults to the current directory) |
| array.name | name or identifier of the array |
| x | a variable name from arrayCGH\$cloneValues giving the order position of the clones along the genome (defaults to 'PosOrder') |
| y | a vector of one or two variable names to be passed to report.plot |
| chrLim | an optional variable name from arrayCGH\$cloneValues giving the limits of each chromosome |
| ylim | a numeric vector of length 2 to be passed to report.plot : y axis range of the genomic profile display |
| clim | a numeric vector of length 2 to be passed to report.plot : color range of the genomic profile |
| zlim | a numeric vector of length 2 to be passed to report.plot : color range for array image display |
| intensity | an optional list with names c("M.var", "A.var", "pred.var", "span"). The first 3 items specify existing variable names from arrayCGH\$arrayValues that will be used to draw a MA-plot. The last item is the value of the loess 'span' |
| light | boolean value: if (light), only the core of the html file is generated; if (!light), a complete html file is generated |
| file.name | file name of the generated report (defaults to "report") |
| width | plot width, in inches |
| height | plot height, in inches |
| ... | further arguments to be passed to report.plot |

Details

This function creates an HTML report file showing - the array image and the genome representation before normalization (if `array.nonorm` is provided) and after normalization, and optionally a MA-plot - a table with information about the number of flagged spots for each flag, and the number of remaining spots after normalization - a table with information about various quality criteria for the array

Value

none

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

See Also

[flag.summary](#), [report.plot](#)

import

Import raw file to an arrayCGH object

Description

Load raw data from a text file coming from image analysis and convert it to an `arrayCGH` object, using additional information about the array design.

Supported file types are Genepix Results file (.gpr), outputs from SPOT, or any text file with appropriate fields "Row" and "Column" and specified array design

Usage

```
import(file, var.names=NULL, spot.names=NULL, clone.names=NULL,
       type=c("default", "gpr", "spot"), id.rep=1, design=NULL, add.lines=FALSE, ...)
```

Arguments

| | |
|--------------------------|--|
| <code>file</code> | a connection or character string giving the name of the file to import. |
| <code>var.names</code> | a vector of variables names used to compute the array design. If default is not overwritten, it is set to <code>c("Block", "Column", "Row", "X", "Y")</code> for gpr files, <code>c("Arr.colx", "Arr.rowy", "Spot.colx", "Spot.rowy")</code> for SPOT files, and <code>c("Col", "Row")</code> for other text files |
| <code>spot.names</code> | a list with spot-level variable names to be added to <code>arrayCGH\$arrayValues</code> |
| <code>clone.names</code> | a list with clone-level variable names to be added to <code>arrayCGH\$cloneValues</code> (only used in case of within-slide replicates) |
| <code>type</code> | a character value specifying the type of input file: currently .gpr files ("gpr"), spot files ("spot") and other text files with fields 'Col' and 'Row' ("default") are supported |
| <code>id.rep</code> | index of the replicate identifier (e.g. the name of the clone) in the vector(<code>clone.names</code>) |
| <code>design</code> | a numeric vector of length 4 specifying array design as number of blocks per column, number of blocks per row, number of columns by block, number of rows per block. This field is mandatory for "default" text files, optional for "gpr" files, and not used for "SPOT" files |

| | |
|------------------------|--|
| <code>add.lines</code> | boolean value to handle the case when array design does not match number of lines. If TRUE, empty lines are added; if FALSE, execution is stopped |
| <code>...</code> | additional import parameters (e.g. <code>'sep='</code> , or <code>'comment.char='</code> , to be passed to <code>read.delim</code> function. Note that argument <code>as.is=TRUE</code> is always passed to <code>read.delim</code> , in order to avoid unappropriate conversion of character vectors to factors |

Details

Mandatory elements of `arrayCGH` objects are the array design and the x and y *absolute coordinates* of each spot on the array. Output files from SPOT contain x and y relative coordinates of each spot within a block, as well as block coordinates on the array; one can therefore easily construct the corresponding `arrayCGH` object.

`.gpr` files currently only contain x and y relative coordinates of each spot within a block, and block index with no specification of the spatial block design: if block design is not specified by user, we compute it using the real pixel locations of each spot (X and Y variables in usual `.gpr` files)

If `clone.names` is provided, an additional data frame is created with clone-level information (e.g. clone names, positions, chromosomes, quality marks), aggregated from array-level information using the identifier specified by `id.rep`. This identifier is also added to the `arrayCGH` object created, with name `'id.rep'`.

Due to space limitations, only the first 100 lines of sample `'gpr'` and `'spot'` files are given in the standard distribution of MANOR. Complete files are available at <http://bioinfo.curie.fr/projects/manor/index.html>

Value

an object of class `arrayCGH`

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

See Also

`arrayCGH`

Examples

```
dir.in <- system.file("extdata", package="MANOR")

## import from 'spot' files
spot.names <- c("LogRatio", "RefFore", "RefBack", "DapiFore", "DapiBack",
               "SpotFlag", "ScaledLogRatio")
clone.names <- c("PosOrder", "Chromosome")
```

```
edge <- import(paste(dir.in, "/edge.txt", sep=""), type="spot",
spot.names=spot.names, clone.names=clone.names, add.lines=TRUE)

## import from 'gpr' files
spot.names <- c("Clone", "FLAG", "TEST_B_MEAN", "REF_B_MEAN",
"TEST_F_MEAN", "REF_F_MEAN", "ChromosomeArm")
clone.names <- c("Clone", "Chromosome", "Position", "Validation")

ac <- import(paste(dir.in, "/gradient.gpr", sep=""), type="gpr",
spot.names=spot.names, clone.names=clone.names, sep="\t",
comment.char="@", add.lines=TRUE)
```

MANOR-internal

Internal Functions for MANOR Package

Description

Internal functions not intended for direct calls by user.

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

nem

Spatial Classification by EM algorithm

Description

The function nem computes spatial classification by EM algorithm.

Usage

```
## Default S3 method:
nem(LogRatio, Col, Row, nk=nk, beta=1, iters=2000, ...)
## S3 method for class 'arrayCGH'
nem(arrayCGH, variable, nk=5, beta=1, iters=2000, ...)
```


Arguments

| | |
|----------|---|
| LogRatio | Vector that corresponds to the values to be classified. |
| Col | Vector of columns coordinates. |
| Row | Vector of rows coordinates. |
| nk | Integer value corresponding to the number classes. |
| beta | Scale parameter for importance of spatial information. |
| iters | Maximum number of iterations allowed. |
| arrayCGH | Object of class <code>arrayCGH</code> . |
| variable | Variable that corresponds to the values to be classified. |
| ... | ... |

Value

Either a data frame with the following added elements:

ZoneNem Vector of label zones.

or an object of class `arrayCGH` with the following elements added to the data.frame attribute `arrayValues`:

ZoneNem Vector of label zones.

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Philippe Hup?, <manor@curie.fr>.

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P. Neuvial, P. Hup?, I. Brito, S. Liva, E. Mani?, C. Brennetot, A. Aurias, F. Radvanyi, and E. Barillot. *Spatial normalization of array-CGH data*. BMC Bioinformatics, 7(1):264. May 2006.

Examples

```
data(spatial) ## arrays with local spatial effects

## Plot of LogRatio measured on the array CGH
## Not run:
GLAD::arrayPlot(edge,"LogRatio", main="Log2-Ratio measured on the array
```

```

CGH", zlim=c(-1,1), bar="v", mediancenter=TRUE)

## End(Not run)

## Spatial trend of the scaled log-ratios (the variable "ScaledLogRatio"
## equals to the log-ratio minus the median value of the corresponding chromosome arm)
edgeTrend <- arrayTrend(edge, variable="ScaledLogRatio",
span=0.03, degree=1, iterations=3, family="symmetric")

## Not run:
GLAD::arrayPlot(edgeTrend, variable="Trend", main="Spatial trend of the array CGH", bar="v")

## End(Not run)

## Classification with spatial constraint of the spatial trend
edgeNem <- nem(edgeTrend, variable="Trend")
## Not run:
GLAD::arrayPlot(edgeNem, variable="ZoneNem", main="Spatial zones identified by nem", bar="v")

## End(Not run)

```

norm

Normalize an object of type arrayCGH

Description

Normalize an object of type arrayCGH using a list of criteria specified as (temporary or permanent) flags. If a replicate identifier (clone name) is provided, a single target value is computed for all the replicates.

The normalization coefficient is computed as a function, and is applied to all good quality spots of the array.

Usage

```

## S3 method for class 'arrayCGH'
norm(arrayCGH, flag.list=NULL, var="LogRatio", printTime=FALSE, FUN=median, ...)

```

Arguments

| | |
|-----------|---|
| arrayCGH | an object of type arrayCGH |
| flag.list | a list of objects of type flag |
| var | a variable name (from arrayCGH\$arrayValues) from which normalization coefficient has to be computed; default is "LogRatio" |
| printTime | boolean value; if TRUE, the time taken by each step of the normalization process is displayed |
| FUN | an aggregation function (e.g. mean, median) to compute a normalization coefficient; default is median |
| ... | further arguments to be passed to FUN |

Details

The two flag types are treated differently : - permanent flags detect poor quality spots, which are removed from further analysis - temporary flags detect good quality spots that would bias the normalization coefficient if they were not excluded from its computation, e.g. amplicons or sexual chromosomes. Thus they are not taken into account for the computation of the coefficient, but at the end of the analysis they are normalized as any good quality spots of the array.

The normalization coefficient is computed as a function (e.g. mean or median) of the target value (e.g. log-ratios); it is then applied to all good quality spots (including temporary flags), i.e. subtracted from each target value.

If clone level information is available (i.e. if `arrayCGH$cloneValues` is not null), a normalized mean target value and basic statistics (such as the number of replicates per clone) are calculated for each clone.

Value

an object of type `arrayCGH`

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

References

P. Neuvial, P. Hup?, I. Brito, S. Liva, E. Mani?, C. Brennetot, A. Aurias, F. Radvanyi, and E. Barillot. *Spatial normalization of array-CGH data*. BMC Bioinformatics, 7(1):264. May 2006.

See Also

[flag](#)

Examples

```
data(spatial)
data(flags)

### 'edge': local spatial bias
## define a list of flags to be applied
flag.list1 <- list(spatial=local.spatial.flag, spot=spot.corr.flag,
ref.snr=ref.snr.flag, dapi.snr=dapi.snr.flag, rep=rep.flag,
unique=unique.flag)
flag.list1$spatial$args <- alist(var="ScaledLogRatio", by.var=NULL,
nk=5, prop=0.25, thr=0.15, beta=1, family="symmetric")
flag.list1$spot$args <- alist(var="SpotFlag")
flag.list1$spot$char <- "0"
flag.list1$spot$label <- "Image analysis"
```

```

## normalize arrayCGH
edge.norm <- norm(edge, flag.list=flag.list1,
var="LogRatio", FUN=median, na.rm=TRUE)
print(edge.norm$flags) ## spot-level flag summary (computed by flag.summary)

## aggregate arrayCGH without normalization
edge.nonorm <- norm(edge, flag.list=NULL, var="LogRatio",
FUN=median, na.rm=TRUE)

## sort genomic informations
edge.norm <- sort(edge.norm, position.var="PosOrder")
edge.nonorm <- sort(edge.nonorm, position.var="PosOrder")

## plot genomic profiles
layout(matrix(c(1,2,4,5,3,3,6,6), 4,2),width=c(1, 4), height=c(6,1,6,1))
report.plot(edge.nonorm, chrLim="LimitChr", layout=FALSE,
main="Pangenomic representation (before normalization)", zlim=c(-1,1),
ylim=c(-3,1))
report.plot(edge.norm, chrLim="LimitChr", layout=FALSE,
main="Pangenomic representation (after normalization)", zlim=c(-1,1),
ylim=c(-3,1))

### 'gradient': global array Trend
## define a list of flags to be applied
flag.list2 <- list(
  spot=spot.flag, global.spatial=global.spatial.flag, SNR=SNR.flag,
  val.mark=val.mark.flag, position=position.flag, unique=unique.flag,
  amplicon=amplicon.flag, replicate=replicate.flag,
  chromosome=chromosome.flag)

## normalize arrayCGH
gradient.norm <- norm(gradient, flag.list=flag.list2,
var="LogRatio", FUN=median, na.rm=TRUE)
## aggregate arrayCGH without normalization
gradient.nonorm <- norm(gradient, flag.list=NULL, var="LogRatio", FUN=median, na.rm=TRUE)

## sort genomic informations
gradient.norm <- sort(gradient.norm)
gradient.nonorm <- sort(gradient.nonorm)

## plot genomic profiles
layout(matrix(c(1,2,4,5,3,3,6,6), 4,2),width=c(1, 4), height=c(6,1,6,1))
report.plot(gradient.nonorm, chrLim="LimitChr", layout=FALSE,
main="Pangenomic representation (before normalization)", zlim=c(-2,2),
ylim=c(-3,2))
report.plot(gradient.norm, chrLim="LimitChr", layout=FALSE,
main="Pangenomic representation (after normalization)", zlim=c(-2,2),
ylim=c(-3,2))

```

Description

qscore object is a list which contains a function, a name, and optionnally a label and arguments to be passed to the function.

Usage

```
to.qscore(FUN, name=NULL, args=NULL, label=NULL, dec=3)
```

Arguments

| | |
|-------|--|
| FUN | a R function returning a numeric value, with first argument of type arrayCGH , and optionally other arguments. |
| name | a short character value for qscore object identification |
| args | a list of arguments to be passed to FUN; defaults to NULL (ie arrayCGH is the only argument to FUN) |
| label | a character value for qscore object labelling |
| dec | an integer value giving the number of significant digits to keep (defaults to 3) |

Value

An object of class qscore.

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

See Also

[qscore.arrayCGH](#), [qscore.summary.arrayCGH](#)

qscore.arrayCGH *arrayCGH quality score*

Description

Computes a quality score for a given arrayCGH.

Usage

```
qscore.arrayCGH(qscore, arrayCGH)
```

Arguments

qscore an object of type [qscore](#).
arrayCGH an object of type arrayCGH.

Value

A numeric value.

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

See Also

[qscore](#), [qscore.summary](#)

Examples

```
data(qscores)
data(spatial)

## compute a quality score for a couple of arrays: signal smoothness
qscore.arrayCGH(smoothness.qscore, edge.norm)
qscore.arrayCGH(smoothness.qscore, gradient.norm)
```

qscore.summary

Compute quality scores for a given arrayCGH object

Description

Compute useful quality scores for the arrayCGH and display them in a convenient way

Usage

```
qscore.summary.arrayCGH(arrayCGH, qscore.list)
```

Arguments

arrayCGH an object of type arrayCGH
qscore.list a list of objects of type [qscore](#)

Details

This function is used by the function `html.report` for the generation of an HTML report of the normalization step. It can also be used by itself.

Value

A data.frame with 3 columns:

| | |
|--------|----------------|
| name | qscore name |
| label | qscore label |
| qscore | quality qscore |

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

See Also

[qscore](#), [qscore.summary](#), [html.report](#)

Examples

```
data(qscores)
data(spatial)

## define a list of qscores
qscore.list <- list(clone=clone.qscore, pct.clone=pct.clone.qscore,
pct.spot=pct.spot.qscore, pct.replicate=pct.replicate.qscore,
smoothness=smoothness.qscore, dyn.x=dyn.x.qscore, dyn.y=dyn.y.qscore,
var.replicate=var.replicate.qscore)

## compute quality scores for a couple of normalized arrays
gradient.norm$quality <- qscore.summary.arrayCGH(gradient.norm,
qscore.list)
print(gradient.norm$quality[, 2:3])

qscore.list$dyn.x$args$test <- 23
qscore.list$dyn.y$args$test <- 24
edge.norm$quality <- qscore.summary.arrayCGH(edge.norm, qscore.list)
print(edge.norm$quality[, 2:3])
```

qscores

Examples of qscore objects (quality scores) to apply to CGH arrays

Description

This data set provides `qscore` objects that can be applied to *normalized* arrayCGH objects in order to evaluate data quality after normalization.

Usage

```
data(qscores)
```

Format

The following qscore objects are provided:

| | |
|-------------------------------------|--|
| <code>clone.qscore</code> | number of clones |
| <code>pct.clone.qscore</code> | percentage of clones |
| <code>pct.spot.qscore</code> | percentage of spots |
| <code>pct.spot.before.qscore</code> | percentage of spots before normalization |
| <code>pct.replicate.qscore</code> | average percentage of replicates |
| <code>smoothness.qscore</code> | signal smoothness |
| <code>var.replicate.qscore</code> | |
| <code>dyn.x.qscore</code> | signal dynamics on X chromosome |
| <code>dyn.y.qscore</code> | signal dynamics on Y chromosome |

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

Source

Institut Curie, <manor@curie.fr>.

See Also

[spatial](#), [qscore.summary.arrayCGH](#), [qscore](#)

Examples

```

data(qscores)
data(spatial)

## define a list of qscores
qscore.list <- list(clone=clone.qscore, pct.clone=pct.clone.qscore,
pct.spot=pct.spot.qscore, pct.replicate=pct.replicate.qscore,
smoothness=smoothness.qscore, dyn.x=dyn.x.qscore, dyn.y=dyn.y.qscore,
var.replicate=var.replicate.qscore)

## compute quality scores for a couple of normalized arrays
gradient.norm$quality <- qscore.summary.arrayCGH(gradient.norm,
qscore.list)
print(gradient.norm$quality[, 2:3])

qscore.list$dyn.x$args$test <- 23
qscore.list$dyn.y$args$test <- 24
edge.norm$quality <- qscore.summary.arrayCGH(edge.norm, qscore.list)
print(edge.norm$quality[, 2:3])

```

report.plot

*Array image and a genomic representation of a normalized arrayCGH***Description**

Displays an array image and a genomic representation of a normalized arrayCGH.

Usage

```

## S3 method for class 'arrayCGH'
report.plot(arrayCGH, x="PosOrder", y=c("LogRatioNorm",
"LogRatio"), chrLim=NULL, layout=TRUE, main=NULL, zlim=NULL, ...)
## Default S3 method:
report.plot(spot.data, clone.data, design, x="PosOrder",
y=c("LogRatioNorm", "LogRatio"), chrLim=NULL, layout=TRUE, main=NULL,
zlim=NULL, ...)

```

Arguments

| | |
|------------|--|
| arrayCGH | an object of type arrayCGH. |
| spot.data | data.frame with spot-level information to be passed to arrayPlot. |
| clone.data | data.frame with clone-level information to be passed to genome.plot. |
| design | vector of length 4 with array design: number of blocks per column and per row, number of columns and rows per block. |
| x | a variable name from arrayCGH\$cloneValues giving the order position of the clones along the genome. |

| | |
|--------|--|
| y | a vector of one or two variable names to be plotted on the array and along the genome. The first one is taken from <code>arrayCGH\$arrayValues</code> and is plotted on the array; the second one (or the first one if only one name was provided) is taken from <code>arrayCGH\$cloneValues</code> and is plotted along the genome. |
| chrLim | an optional variable name from <code>arrayCGH\$cloneValues</code> giving the limits of each chromosome. |
| layout | if TRUE, plot layout is set to a 1*2 matrix with relative column widths 1 and 4. |
| main | title for the genomic profile. |
| zlim | numeric vector of length 2 to be passed to <code>arrayPlot</code> : minimum and maximum signal values for array image display. |
| ... | further arguments to be passed to <code>genome.plot</code> . |

Details

This function successively calls `arrayPlot` and `genome.plot`.

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

See Also

[genome.plot](#), [arrayPlot](#), [html.report](#)

Examples

```
data(spatial)

### edge: local spatial bias
## aggregate arrayCGH without normalization for comparison with
## normalized array
edge.nonorm <- norm(edge, flag.list=NULL, FUN=median, na.rm=TRUE)
edge.nonorm <- sort(edge.nonorm, position.var="PosOrder")

layout(matrix(c(1,2,4,5,3,3,6,6), 4,2),width=c(1, 4), height=c(6,1,6,1))
report.plot(edge.nonorm, chrLim="LimitChr", layout=FALSE,
main="Pangenomic representation (before normalization)", zlim=c(-1,1),
ylim=c(-3,1))
report.plot(edge.norm, chrLim="LimitChr", layout=FALSE,
main="Pangenomic representation (after normalization)", zlim=c(-1,1),
ylim=c(-3,1))

### gradient: global array Trend
## aggregate arrayCGH without normalization for comparison with
## normalized array
```

```

gradient.nonorm <- norm(gradient, flag.list=NULL, FUN=median, na.rm=TRUE)
gradient.nonorm <- sort(gradient.nonorm)

layout(matrix(c(1,2,4,5,3,3,6,6), 4,2),width=c(1, 4), height=c(6,1,6,1))
report.plot(gradient.nonorm, chrLim="LimitChr", layout=FALSE,
main="Pangenomic representation (before normalization)", zlim=c(-2,2),
ylim=c(-3,2))
report.plot(gradient.norm, chrLim="LimitChr", layout=FALSE,
main="Pangenomic representation (after normalization)", zlim=c(-2,2),
ylim=c(-3,2))

```

sort

Sorting for normalized arrayCGH objects

Description

Sorts clone-level information of a normalized arrayCGH object.

Usage

```

## S3 method for class 'arrayCGH'
sort(x, decreasing = FALSE, position.var="Position",
      chromosome.var="Chromosome", ...)

```

Arguments

x an object of type arrayCGH.
decreasing (for compatibility with sort class) currently unused.
position.var name of position variable.
chromosome.var name of chromosome variable.
... further arguments to be passed to sort.

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

See Also

[norm.arrayCGH](#)

Examples

```
data(spatial)

## sort a normalized array by clone position
gradient.norm <- sort(gradient.norm)

report.plot(gradient.norm, main="Genomic profile after normalization")
```

spatial

Examples of array-CGH data with spatial artifacts

Description

This data set provides an example of array-CGH data with spatial artifacts, consisting of including [arrayCGH](#) objects before and after normalization

Usage

```
data(spatial)
```

Format

edge, **gradient** [arrayCGH](#) objects before normalization:

| | |
|-------------|---|
| arrayValues | spot-level information |
| arrayDesign | block design of the array |
| cloneValues | additionnal clone-level data (chromosome, position) |

edge.norm, **gradient.norm** [arrayCGH](#) objects after normalization

Details

'edge' presents local spatial bias in the top-right edge corner, and 'gradient' presents global spatial trend. 'edge' and 'gradient' are [arrayCGH](#) objects before normalization. They have been created respectively from spot and gpr files using [import](#). 'edge.norm' and 'gradient.norm' are the corresponding [arrayCGH](#) objects after normalization using [norm.arrayCGH](#).

[flag](#) objects used for data normalization come from [flags](#) dataset.

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

Source

Institut Curie, <manor@curie.fr>.

See Also

[flags](#)

Examples

```
data(spatial)

## edge: example of array with local spatial effects

layout(matrix(1:4, 2, 2), height=c(9,1))
GLAD::arrayPlot(edge, "LogRatio", main="Log-ratios before normalization",
zlim=c(-1,1), bar="h", layout=FALSE, mediancenter=TRUE)
GLAD::arrayPlot(edge.norm, "LogRatioNorm", main="Log-ratios after spatial
normalization", zlim=c(-1,1), bar="h", layout=FALSE, mediancenter=TRUE)

## gradient: example of array with spatial gradient

layout(matrix(1:4, 2, 2), height=c(9,1))
GLAD::arrayPlot(gradient, "LogRatio", main="Log-ratios before normalization",
zlim=c(-2,2), bar="h", layout=FALSE)
GLAD::arrayPlot(gradient.norm, "LogRatioNorm", main="Log-ratios after spatial
normalization", zlim=c(-2,2), bar="h", layout=FALSE)
```

to.flag

Create an object of type flag

Description

A flag object is a list which contains essentially a function (flag action) and a character, optionally arguments to be passed to the function. We make the distinction between two different flag types, corresponding to two different purposes: - *permanent flags* identify poor quality spots or clones and remove them from further analysis (eg spots with low signal to noise ratio) - *temporary flags* identify spots or clones that have not to be taken into account for the computation of a (scaling) normalization coefficient (eg X chromosome in case of sex mismatch)

Usage

```
to.flag(FUN, char=NULL, args=NULL, type="perm.flag", label=NULL)
```

Arguments

FUN a R function to be applied to an [arrayCGH](#), and optionally other arguments. If char is not NULL, must return a list of spots (lines of arrayCGH\$arrayValues) to be flagged out; if char==NULL, must return an object of type [arrayCGH](#)

| | |
|-------|---|
| char | a character value to identify flagged spots; defaults to NULL |
| args | a list of further arguments to be passed to FUN; defaults to NULL (ie <code>arrayCGH</code> is the only argument to FUN) |
| type | a character value defaulting to "perm.flag" which makes the distinction between permanent flags (type="perm.flag") and temporary flags (type="temp.flag") |
| label | a character value for flag labelling |

Details

If `flag$char` is null, `flag$FUN` is supposed to return a `arrayCGH` object; if it is not null, `flag$FUN` is supposed to return a list of spots to be flagged with `flag$char`.

Value

An object of class `flag`.

Note

People interested in tools for array-CGH analysis can visit our web-page: <http://bioinfo.curie.fr>.

Author(s)

Pierre Neuvial, <manor@curie.fr>.

See Also

[flag.arrayCGH](#), [norm.arrayCGH](#)

Examples

```
### creation of a permanent flag:
## flag spots with low signal to noise ratios
SNR.FUN <- function(arrayCGH, snr.thr)
  which(arrayCGH$arrayValues$F2 < arrayCGH$arrayValues$B2+log(snr.thr, 2))
SNR.char <- "B"
SNR.flag <- to.flag(SNR.FUN, SNR.char, args=alist(snr.thr=3))

### creation of a permanent flag returning an arrayCGH object:
## correct log-ratios for spatial trend

global.spatial.FUN <- function(arrayCGH, var)
{
  Trend <- arrayTrend(arrayCGH, var, span=0.03, degree=1,
iterations=3, family="symmetric")
  arrayCGH$arrayValues[[var]] <- Trend$arrayValues[[var]]-Trend$arrayValues$Trend
  arrayCGH
}
global.spatial.flag <- to.flag(global.spatial.FUN, args=alist(var="LogRatio"))

### creation of a temporary flag:
```

```
## exclude sexual chromosomes from signal scaling
chromosome.FUN <- function(arrayCGH, var)
  which(!is.na(match(as.character(arrayCGH$arrayValues[[var]]), c("X", "Y"))))
chromosome.char <- "X"
chromosome.flag <- to.flag(chromosome.FUN, chromosome.char, type="temp.flag",
args=alist(var="Chromosome"))

data(spatial)

SNR.flag$args$snr.thr <- 3          ## set SNR threshold
gradient <- flag.arrayCGH(SNR.flag, gradient)  ## apply SNR.flag to array CGH

gradient <- flag.arrayCGH(global.spatial.flag, gradient)

gradient <- flag.arrayCGH(chromosome.flag, gradient)

summary.factor(gradient$arrayValues$Flag)  ## permanent flags
summary.factor(gradient$arrayValues$FlagT) ## temporary flags
```

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