

Package ‘MANOR’

October 10, 2025

Version 1.81.0

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 a00850e

git_last_commit_date 2025-04-15

Repository Bioconductor 3.22

Date/Publication 2025-10-09

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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 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

[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

Spatial bias detection

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

Apply a flag to an arrayCGH

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 flag\$FUN
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:

amplicon.flag	flags spots with high log-ratios (temp flag)
chromosome.flag	flags spots located on sexual chromosomes (named "X" and "Y")
control.flag	flag control spots
global.spatial.flag	corrects arrayCGH from global spatial trend on the array
local.spatial.flag	flags spots belonging to local spatial bias zones on the array
num.chromosome.flag	flags spots located on sexual chromosomes (named 23 and 24)
position.flag	flag spots with no available genome position
replicate.flag	flag spots with poor within-clone-replicate consistency
ref.snr.flag	flags spots with low signal to noise ratio for reference
dapi.snr.flag	flags spots with low signal to noise ratio for DAPI
SNR.flag	flags spots with low signal to noise ratio

spot.corr.flag	flags spots with low correlation coefficient after image analysis
spot.flag	flags spots excluded by the image analysis software
unique.flag	exclude last non-flagged spot of a clone
val.mark.flag	flags spots corresponding to bad quality clones
intensity.flag	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

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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

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

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

file	a connection or character string giving the name of the file to import.
var.names	a vector of variables names used to compute the array design. If default is not overwritten, it is set to c("Block", "Column", "Row", "X", "Y") for gpr files, c("Arr.colx", "Arr.rowy", "Spot.colx", "Spot.rowy") for SPOT files, and c("Col", "Row") for other text files
spot.names	a list with spot-level variable names to be added to arrayCGH\$arrayValues
clone.names	a list with clone-level variable names to be added to arrayCGH\$cloneValues (only used in case of within-slide replicates)
type	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
id.rep	index of the replicate identifier (e.g. the name of the clone) in the vector(clone.names)
design	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

`add.lines` 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

`...` additional import parameters (e.g. `'sep='`, or `'comment.char='`, to be passed to `read.delim` function. Note that argument `as.is=TRUE` is always passed to `read.delim`, 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 te 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 arrayCGH .
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>.

References

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C. Ambroise, M. Dang, and G. Govaert, *Clustering of spatial data by the em algorithm* in Geostatistics for Environmental Applications, A. Soares, J. Gomez-Hernandez, and R. Froidevaux, Eds., pp. 493-504. Kluwer Academic Publisher, 1997.

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", xlim=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	<i>Normalize an object of type arrayCGH</i>
------	---

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)", xlim=c(-2,2),
ylim=c(-3,2))
report.plot(gradient.norm, chrLim="LimitChr", layout=FALSE,
main="Pangenomic representation (after normalization)", xlim=c(-2,2),
ylim=c(-3,2))

```

qscore

Create an object of type qscore

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

<code>FUN</code>	a R function returning a numeric value, with first argument of type <code>arrayCGH</code> , and optionally other arguments.
<code>name</code>	a short character value for qscore object identification
<code>args</code>	a list of arguments to be passed to FUN; defaults to NULL (ie <code>arrayCGH</code> is the only argument to FUN)
<code>label</code>	a character value for qscore object labelling
<code>dec</code>	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	<i>arrayCGH quality score</i>
-----------------	-------------------------------

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:

clone.qscore	number of clones
pct.clone.qscore	percentage of clones
pct.spot.qscore	percentage of spots
pct.spot.before.qscore	percentage of spots before normalization
pct.replicate.qscore	average percentage of replicates
smoothness.qscore	signal smoothness
var.replicate.qscore	
dyn.x.qscore	signal dynamics on X chromosome
dyn.y.qscore	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 arrayCGH\$arrayValues and is plotted on the array; the second one (or the first one if only one name was provided) is taken from arrayCGH\$cloneValues 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, mediantcenter=TRUE)
GLAD::arrayPlot(edge.norm, "LogRatioNorm", main="Log-ratios after spatial
  normalization", zlim=c(-1,1), bar="h", layout=FALSE, mediantcenter=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 arrayCGH 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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