

Package ‘spotSegmentation’

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Title Microarray Spot Segmentation and Gridding for Blocks of Microarray Spots

Description Spot segmentation via model-based clustering and gridding for blocks within microarray slides, as described in Li et al, Robust Model-Based Segmentation of Microarray Images, Technical Report no. 473, Department of Statistics, University of Washington.

Depends R (>= 2.10), mclust

Note mclust package not needed for gridding

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biocViews Microarray, TwoChannel, QualityControl, Preprocessing

R topics documented:

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`plot.spotseg`*Microarray Spot Segmentation Plot*

Description

Plot method for the `spotseg` function. Displays the result obtained from microarray spot segmentation via model-based clustering.

Usage

```
## S3 method for class 'spotseg'  
plot(x, ...)
```

Arguments

`x` An object of class "spotseg", which is the output of the function `spotseg`.
`...` Unused but required by generic "plot" method.

Value

None, other than the displayed plot.

References

Q. Li, C. Fraley, R. Bumgarner, K. Y. Yeung, and A. Raftery\ Robust model-based segmentation of microarray images,\ Technical Report No.~473, Department of Statistics, University of Washington, January 2005.

See Also

[spotseg](#)

Examples

```
data(spotSegTest)  
  
# columns of spotSegTest:  
# 1 intensities from the Cy3 (green) channel  
# 2 intensities from the Cy5 (red) channel  
  
dataTransformation <- function(x) (256*256-1-x)^2*4.71542407E-05  
  
chan1 <- matrix(dataTransformation(spotSegTest[,1]), 144, 199)  
chan2 <- matrix(dataTransformation(spotSegTest[,2]), 144, 199)  
  
hivGrid <- spotgrid( chan1, chan2, rows = 4, cols = 6, show = TRUE)  
  
library(mclust)
```

```
hivSeg <- spotseg( chan1, chan2, hivGrid$rowcut, hivGrid$colcut)
plot(hivSeg)
```

plotBlockImage *Plot Microarray Image Block*

Description

Displays a block of a microarray image.

Usage

```
plotBlockImage(z, title, one)
```

Arguments

| | |
|-------|--|
| z | Intensities of the image pixels, in the form a of a matrix. |
| title | A title for the image plot (optional). |
| one | Sets appropriate graphics parameters for displaying individuals spots (default:FALSE). |

Value

None, other than the displayed plot.

References

Q. Li, C. Fraley, R. Bumgarner, K. Y. Yeung, and A. Raftery\ Robust model-based segmentation of microarray images,\ Technical Report No.~473, Department of Statistics, University of Washington, January 2005.

See Also

[spotseg](#)

Examples

```
data(spotSegTest)

# columns of spotSegTest:
# 1 intensities from the Cy3 (green) channel
# 2 intensities from the Cy5 (red) channel

dataTransformation <- function(x) (256*256-1-x)^2*4.71542407E-05

chan1 <- matrix(dataTransformation(spotSegTest[,1]), 144, 199)
chan2 <- matrix(dataTransformation(spotSegTest[,2]), 144, 199)

plotBlockImage(chan1)
plotBlockImage(chan2)
```

spotgrid

Gridding for Blocks of Microarray Spots

Description

Determines row or column delimiters for spot locations from blocks of microarray slide image data.

Usage

```
spotgrid(chan1, chan2, rows = NULL, cols = NULL, span = NULL,  
         show = FALSE)
```

Arguments

| | |
|-------|--|
| chan1 | matrix of pixel intensities from the first channel. |
| chan2 | matrix of pixel intensities from the second channel. |
| rows | number of spots in a row of the image block. |
| cols | number of spots in a column of the image block. |
| span | Window size for locating peak signals. This can be of length 2, in which case the first value is interpreted as a window size for the rows and the second as a window size for the columns. A default is estimated from the image dimension and number of spots. |
| show | logical variable indicating whether or not to display the gridding result. |

Value

A list with two elements, `rowcut` and `colcut` giving delimiters for the row and/or column gridding of the slide. The indexes indicate the start of a segment of the grid, except for the last one, which indicates the end of the grid.

References

Q. Li, C. Fraley, R. Bumgarner, K. Y. Yeung, and A. Raftery\ Robust model-based segmentation of microarray images,\ Technical Report No.~473, Department of Statistics, University of Washington, January 2005.

See Also

[spotseg](#)

Examples

```

data(spotSegTest)

# columns of spotSegTest:
# 1 intensities from the Cy3 (green) channel
# 2 intensities from the Cy5 (red) channel

dataTransformation <- function(x) (256*256-1-x)^2*4.71542407E-05

chan1 <- matrix(dataTransformation(spotSegTest[,1]), 144, 199)
chan2 <- matrix(dataTransformation(spotSegTest[,2]), 144, 199)

Grid <- spotgrid( chan1, chan2, rows = 4, cols = 6, show = TRUE)

```

spotseg

*Microarray Spot Segmentation***Description**

Microarray spot segmentation via model-based clustering.

Usage

```

spotseg(chan1, chan2, rowcut, colcut, R=NULL, C=NULL,
        threshold=100, hc=FALSE, show=FALSE)

```

Arguments

| | |
|-----------|---|
| chan1 | matrix of pixel intensities from the first channel. |
| chan2 | matrix of pixel intensities from the second channel. |
| rowcut | row delimiters for the spots. Entries are the starting row location in the close of each spot, with the last entry being one pixel beyond the border of the last spot. For example, from the output of <code>spotgrid</code> . |
| colcut | column delimiters for the spots. Entries are the starting column location in the close of each spot, with the last entry being one pixel beyond the border of the last spot. For example, from the output of <code>spotgrid</code> . |
| R | rows over which the spots are to be segmented. The default is to segment spots in all rows. |
| C | columns over which the spots are to be segmented. The default is to segment spots in all columns. |
| threshold | connected components of size smaller than <code>threshold</code> are ignored. Default: <code>threshold=100</code> . |
| hc | logical variable indicating whether or not EM should be initialized by hierarchical clustering or quantiles in model-based clustering. The default is to use quantiles <code>hc = FALSE</code> , which is more efficient both in terms of speed and memory usage. |

`show` logical variable indicating whether or not to display the segmentation of each individual spot as it is processed. The default is not to display the spots `show = FALSE`.

Details

There are plot and summary methods that can be applied to the result.

Value

An array of the same dimensions as the image in which the pixels are labeled according to their group within the spot area: 1=background,2=uncertain,3=sample.

Note

The `mclust` package is required for clustering.

References

Q. Li, C. Fraley, R. Bumgarner, K. Y. Yeung, and A. Raftery\ Robust model-based segmentation of microarray images,\ Technical Report No.~473, Department of Statistics, University of Washington, January 2005.

See Also

[summary.spotseg](#), [plot.spotseg](#), [spotgrid](#)

Examples

```
data(spotSegTest)

# columns of spotSegTest:
# 1 intensities from the Cy3 (green) channel
# 2 intensities from the Cy5 (red) channel

dataTransformation <- function(x) (256*256-1-x)^2*4.71542407E-05

chan1 <- matrix(dataTransformation(spotSegTest[,1]), 144, 199)
chan2 <- matrix(dataTransformation(spotSegTest[,2]), 144, 199)

Grid <- spotgrid( chan1, chan2, rows = 4, cols = 6, show = TRUE)

library(mclust)

Seg <- spotseg( chan1, chan2, Grid$rowcut, Grid$colcut)

plot(Seg)

spotSummary <- summary(Seg)

spot11 <- spotseg( chan1, chan2, Grid$rowcut, Grid$colcut,
                  R = 1, C = 1, show = TRUE)
```

spotSegTest

Spot Segmentation Test Data

Description

The two columns of this data set represent the Cy3 (green) absorption intensities for channel 1, and the Cy5 (red) absorption intensities for channel 2 for part of a dye-swap experiment with replicates. They measure expression levels of cellular RNA transcripts assessed in CD4+ T cell lines at different times after infection with HIV-1BRU using DNA microarrays.

Usage

```
data(spotSegTest)
```

Format

Each column is a vector of intensities of 24 spots arranged in 4 rows and 6 columns, encoded for compact (16-bit TIFF) storage. For processing each column of spotSegTest should first be converted to a 144x199 matrix, then applying the transformation described below.

Details

The intensities can be obtained from this data by first subtracting them from 65535 (256*256-1), then squaring, then multiplying by a scale factor 4.71542407E-05. In other words, a number x in the spotSegTest data set corresponds to intensity

$$(256*256 - 1 - x)^2 * .0000471542407$$

. \

Source

Dr. Angelique van't Wout, Department of Microbiology, University of Washington\ The data is a subset the first block of a 12 block array image ('001030_08_1.GEL') in the first data set ('2000095918 A') in the first experiment ('CEM LAI vs HI-LAI 24hr') of the following data archive:\ <http://expression.microslu.washington.edu/expression/vantwoutjvi2002.html>

References

van't Wout AB, Lehrman GK, Mikheeva SA, O'Keeffe GC, Katze MG, Bumgarner RE, Geiss GK, Mullins JI Cellular gene expression upon human immunodeficiency virus type 1 infection of CD4(+)-T-cell lines. *J Virol.* 2003 Jan;77(2):1392-402.

summary.spotseg *Microarray Spot Segmentation Summary*

Description

Summary method for the spotseg function. Gives the estimates of foreground and background intensity obtained from microarray spot segmentation via model-based clustering.

Usage

```
## S3 method for class 'spotseg'  
summary(object,...)
```

Arguments

object An object of class "spotseg", which is the output of the function spotseg.
... Unused, but required by generic "summary" method.

Value

A list with two components, "channel1" and "channel2" each of which has subcomponents "background" and "foreground", each of which in turn has subcomponents "mean" and "median", giving the mean and median estimates of background and foreground for each channel. There will be missing entries (value NA) whenever no foreground is detected.

References

Q. Li, C. Fraley, R. Bumgarner, K. Y. Yeung, and A. Raftery\ Robust model-based segmentation of microarray images,\ Technical Report No.~473, Department of Statistics, University of Washington, January 2005.

See Also

[spotseg](#)

Examples

```
data(spotSegTest)  
  
# columns of spotSegTest:  
# 1 intensities from the Cy3 (green) channel  
# 2 intensities from the Cy5 (red) channel  
  
dataTransformation <- function(x) (256*256-1-x)^2*4.71542407E-05  
  
chan1 <- matrix(dataTransformation(spotSegTest[,1]), 144, 199)  
chan2 <- matrix(dataTransformation(spotSegTest[,2]), 144, 199)
```



```
hivGrid <- spotgrid( chan1, chan2, rows = 4, cols = 6, show = TRUE)
library(mclust)
hivSeg <- spotseg( chan1, chan2, hivGrid$rowcut, hivGrid$colcut)
hivSummary <- summary(hivSeg)
```

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