# Package ‘colocr’

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**Type** Package  
**Title** Conduct Co-Localization Analysis of Fluorescence Microscopy Images  
**Version** 0.1.1  
**License** GPL-3  

**URL** [https://docs.ropensci.org/colocr](https://docs.ropensci.org/colocr),  
[https://github.com/ropensci/colocr](https://github.com/ropensci/colocr)  

**BugReports** [https://github.com/ropensci/colocr/issues](https://github.com/ropensci/colocr)  

**Description** Automate the co-localization analysis of fluorescence microscopy images. Selecting regions of interest, extract pixel intensities from the image channels and calculate different co-localization statistics. The methods implemented in this package are based on Dunn et al. (2011) [doi:10.1152/ajpcell.00462.2010].  

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**VignetteBuilder** knitr  

**NeedsCompilation** no  

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\textbf{R topics documented:}

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\begin{center}
\begin{tabular}{ll}
\textbf{.intensity\_get} & \textit{Get pixel intensities} \\
\end{tabular}
\end{center}

\textbf{Description}

Get the pixel intensities of certain image channels

\textbf{Usage}

\texttt{.intensity\_get(img, ind = c(1, 2))}

\textbf{Arguments}

- \texttt{img} \hspace{1cm} An object of class \texttt{cimg}
- \texttt{ind} \hspace{1cm} A numeric of length two for channel indexes

\textbf{Value}

A list of three items. The first two items are the values of the pixel intensities of the channels indicated by \texttt{ind}. The third is the labels of the individual regions of interest.

\textbf{Examples}

\begin{verbatim}
# load image
fl <- system.file('extdata', 'Image0001_.jpg', package = 'colocr')
img <- image_load(fl)

# choose parameters
int <- roi_select(img, threshold = 90) %>%
  .intensity_get()
\end{verbatim}
.labels_add

*Label regions of interest*

**Description**

Add labels to regions of interest in an image

**Usage**

```
.labels_add(px, tolerance, n)
```

**Arguments**

- `px`  
  An object of class `pixset`

- `tolerance`  
  A numeric to be passed to `label`

- `n`  
  A numeric, the number of desired regions of interest

**Value**

An object of class `cimg`. The labels are coded the values in the object starting from 1. The rest of the image is labeled 0.

---

.manders

*Calculate Marnders Overlap Coefficient*

**Description**

Calculates the manders overlap coefficient between two numeric vectors

**Usage**

```
.manders(r, g)
```

**Arguments**

- `r`  
  A numeric vector

- `g`  
  A numeric vector

**Value**

A numeric of length one.
Examples

```
set.seed(123)
r <- rnorm(10)

set.seed(1234)
g <- rnorm(10)

.pearson(r, g)
```

---

`.pearson` *Calculate Pearson’s Correlation Coefficient*

### Description

Calculates the Pearson’s correlation coefficient between two numeric vectors.

### Usage

```
.pearson(r, g)
```

### Arguments

- `r`: A numeric vector
- `g`: A numeric vector

### Value

A numeric of length one.

### Examples

```
set.seed(123)
r <- rnorm(10)

set.seed(1234)
g <- rnorm(10)

.pearson(r, g)
```
colocr  

**Description**  
Automate the co-localization analysis of fluorescence microscopy images. Selecting regions of interest, extract pixel intensities from the image channels and calculate different co-localization statistics.

**colocr functions**  
`roi_select` `roi_show` `roi_check` `roi_test`

---

**colocr_app**  
*Run the shiny App*

**Description**  
Run the shiny App

**Usage**  
colocr_app()

---

**image_load**  
*Load images from files*

**Description**  
A wrap around `image_read` and `magick2cimg` to load one or more images from files.

**Usage**  
image_load(image_file)

**Arguments**  
image_file  
A character vector of one or more paths to image files

**Value**  
A cimg object or a list of cimg objects when multiple files are passed to image_file.
Examples

# load image
fl <- system.file('extdata', 'Image0001_.jpg', package = 'colocr')
img <- image_load(fl)

roi_check(img, ind = c(1, 2))

Description

Show the pixel intensities of certain image channels

Usage

roi_check(img, ind = c(1, 2))

Arguments

img
A cimg object or a list of multiple images such as the one returned from
roi_select

ind
A numeric object of length two. For the channel indexes. or a list of similar
vectors for each of img items.

Details

Calling this function returns two plots. The first is a scatter plot of the pixel intensities from two
channels. The second is the density distribution of the intensities from the two channels.

Examples

# load images
fl <- system.file('extdata', 'Image0001_.jpg', package = 'colocr')
img <- image_load(fl)

# choose ROI and show the pixel intensities
oldpar <- par()
par(mfrow = c(1, 2))

roi_select(img, threshold = 90) %>%
  roi_check()

par(oldpar)
roi_select

Select regions of interest

Description
Select regions of interest in an image using different morphological operations

Usage
roi_select(
  img,
  threshold,
  shrink = 5,
  grow = 5,
  fill = 5,
  clean = 5,
  tolerance = 0.1,
  n = 1
)

Arguments

<table>
<thead>
<tr>
<th>Argument</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>img</td>
<td>An object of class cimg or a list of multiple cimg items</td>
</tr>
<tr>
<td>threshold</td>
<td>A numeric to be passed to threshold or a vector of values for each image in img</td>
</tr>
<tr>
<td>shrink</td>
<td>A numeric to be passed to shrink or a vector of values for each image in img</td>
</tr>
<tr>
<td>grow</td>
<td>A numeric to be passed to grow or a vector of values for each image in img</td>
</tr>
<tr>
<td>fill</td>
<td>A numeric to be passed to fill or a vector of values for each image in img</td>
</tr>
<tr>
<td>clean</td>
<td>A numeric to be passed to clean or a vector of values for each image in img</td>
</tr>
<tr>
<td>tolerance</td>
<td>A numeric to be passed to label or a vector of values for each image in img</td>
</tr>
<tr>
<td>n</td>
<td>A numeric of the number of regions of interest or a vector of values for each image in img</td>
</tr>
</tbody>
</table>

Details
The function applies several imager morphological manipulations to select the regions of interest. These include threshold which sets all values below certain cut to 0; shrink/grow for pixel set dilation and erosion; fill/clean for removing isolated regions and holes. When n is provided, the individual regions (connected components) are selected where tolerance is used to determine if two pixels belong to the same region.

Value
A cimg. The original input img with an additional attribute label. label is a vector of integers. The labels for the selected regions of interests starts from 1 and 0 is ignored. When img is a list, a list is returned.
Examples

# load images
fl <- system.file('extdata', 'Image0001.jpg', package = 'colocr')
img <- image_load(fl)

# choose ROI
newimg <- roi_select(img, threshold = 90)

# check the ROI labels
unique(attr(newimg, 'label'))


roi_show

Show the selected regions of interest

Description

Show/highlight the selected regions of interest on different image channels

Usage

roi_show(img, ind = c(1, 2))

Arguments

img A cimg object or a list of multiple images such as the one returned from
 roi_select
ind A numeric object of length two. For the channel indexes, or a list of similar
 vectors for each of img items.

Details

calling this function with img object which is returned from roi_select returns four different plots.
The original image, a low resolution representation of the selected regions of interest and the two
channels indicated through ind highlighted.

Examples

# load images
fl <- system.file('extdata', 'Image0001.jpg', package = 'colocr')
img <- image_load(fl)

# choose and show ROI
oldpar <- par()
par(mfrow=c(2,2))

roi_select(img, threshold = 90) %>%
  roi_show()

par(oldpar)

---

**roi_test**

**Test Co-localization**

**Description**

Perform co-localization test statistics.

**Usage**

```r
roi_test(img, ind = c(1, 2), type = "pcc")
```

**Arguments**

- `img`: A `cimg` object or a list of multiple images such as the one returned from `roi_select`.
- `ind`: A numeric object of length two. For the channel indexes, or a list of similar vectors for each of `img` items.
- `type`: A character vector of the desired co-localization statistics. Default is 'pcc', other inputs are 'moc' or 'both'.

**Details**

The co-localization stats requested in `type` is returned as a column for each. When different labels are provided, the stats are calculated for each label individually. When `img` is a `list` a `list` of such data.frames is returned.

**Value**

A data.frame or a list of data.frames.

**Examples**

```r
# load images
fl <- system.file('extdata', 'Image0001.jpg', package = 'colocr')
img <- image_load(fl)

# choose roi and test colocalization
roi_select(img, threshold = 90) %>%
  roi_test()
```
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