

Package ‘FisHiCal’

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Type Package

Title Iterative FISH-based Calibration of Hi-C Data

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Description FisHiCal integrates Hi-C and FISH data, offering a modular and easy-to-use tool for chromosomal spatial analysis.

License GPL

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FisHiCal-package	<i>Iterative FISH-based Calibration of Hi-C Data</i>
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Description

FisHiCal is an R package for combining Hi-C and FISH data, which offers a modular and easy-to-use tool for chromosomal spatial analysis. With FisHiCal researchers can prepare and apply FISH-based Hi-C calibration, which converts contact frequencies into distances and computes a range threshold, and exploit 3D inference methods to iteratively refine it. These methods include algorithms for reconstructing chromosome structure (from calibrated distances) and for detecting spatial inconsistencies.

Details

Package: FisHiCal
 Type: Package
 Version: 1.1
 Date: 2014-06-20
 License: GPL

FisHiCal implements the methods described in Shavit, Hamey and Lio' (2014) (submitted). Users can first prepare and apply their calibration with [prepareData](#), [prepareCalib](#) and [calibrate](#) and use 3D inference functions ([lsmacof](#), [searchInc](#), [plotInc](#) and [summaryInc](#)) to spatially explore their data and further refine their calibration.

Author(s)

Yoli Shavit, Fiona Kathryn Hamey and Pietro Lio'.
 Maintainer: Yoli Shavit <ys388@cam.ac.uk>

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

calibHiC	<i>in-silico 100*100 calibrated Hi-C contact matrix</i>
----------	---

Description

Generated with the function [calibrate](#) given the in-silico Hi-C contact matrix [hic](#).

Usage

```
data(calibHiC)
```

Details

This data structure is used to illustrate the usage of [lsmacof](#).

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

calibrate	<i>Hi-C calibration</i>
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Description

A function for applying a calibration function to a given Hi-C contact matrix.

Usage

```
calibrate(hic, calib)
```

Arguments

hic	A numeric matrix/vector of pairwise Hi-C contact frequencies.
calib	A list with f, the calibration function, and params a list of parameters for f. This object can be generated with the function prepareCalib .

Value

A calibrated version of the input matrix (or vector), i.e. the result of applying the calibration function (f) on the values of 'hic'. Zero values represent discarded/missing information.

Author(s)

Yoli Shavit

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

See Also

[prepareCalib](#)
[conf](#)
[hic](#)
[match](#)

Examples

```
data(match)
data(hic)
npoints = 10 # number of points to fit

# prepareCalib computes threshold to fit
res = prepareCalib(match, npoints, useMax = FALSE)
calib = res$calib
calib$f # calibration function
calib$params # the params for f
# note that calib could be refined by the user

# now calibrate the hic matrix
calibHiC = calibrate(hic, calib)
plot(match$distances, calibHiC[upper.tri(calibHiC)],
      xlab = "distances", ylab = "calibrated distances")
```

conf

A 3D random configuration of 100 points

Description

A numeric 100*3 matrix giving the 3D coordinates for each point. The configuration was generated as described in Shavit, Hamey and Lio' (2014), as part of an in-silico evaluation.

Usage

```
data(conf)
```

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

findMatchingIndices *Find matching Hi-C bins for FISH probes, using mid-points*

Description

A function for finding matching indices of Hi-C bins for FISH probes, based on the mid-points of their coordinates. Given the start and end coordinates of a set of FISH probes, the function finds the matching Hi-C bins and returns their indices in the given hicCoord data frame.

Usage

```
findMatchingIndices(fishCoord, hicCoord)
```

Arguments

fishCoord	The genomic coordinates of the FISH probes. A data frame with the following columns: chr (chromosome name), start (start position), end (end position). Additional columns are permitted but will not be used. Note that chromosome names should match between hicCoord and fishCoord.
hicCoord	The genomic coordinates of the Hi-C bins. A data frame with the following mandatory columns: chr (chromosome name), start (start position), end (end position). Additional columns are permitted but will not be used. Note that chromosome names should match between hicCoord and fishCoord.

Value

The indices of matching Hi-C bins in the given hicCoord data frame.

Author(s)

Yoli Shavit

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

See Also

[prepareData](#)

getInfoLevelForChr	<i>Compute the information level for a given chromosome after calibration</i>
--------------------	---

Description

A function to compute the information proportions in a given calibrated Hi-C matrix for a given chromosome. This will be the proportions of non-zero cells, representing information that was not discarded in the calibration step.

Usage

```
getInfoLevelForChr(calibHiC, hicCoord, chr)
```

Arguments

calibHiC	A calibrated Hi-C matrix. This matrix could be prepared with calibrate .
hicCoord	The genomic coordinates of the Hi-C bins. A data frame with the following mandatory columns: chr (chromosome name), start (start position), end (end position). Additional columns are permitted but will not be used.
chr	The name of the chromosome of interest (as it appears in hicCoord).

Details

The level of information can affect the accuracy of the 3D prediction, which may be lower for sparse calibration matrices (information level < 0.4).

Value

The information level for the given chromosome in the calibrated Hi-C matrix.

Author(s)

Yoli Shavit

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

See Also

[calibrate](#)
[lsmacof](#)

hic	<i>in-silico 100*100 Hi-C contact matrix with with 5% noise</i>
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Description

in-silico distances were computed from the random configuration [conf](#). A power law model was then used to generate matching in-silico Hi-C frequencies. Random Noise was further added to long range frequencies to mimic a typical situation for Hi-C data. The resulting frequencies were then used to construct this matrix.

Usage

```
data(hic)
```

Details

This data structure is used to illustrate the usage of [calibrate](#).

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

lsmacof	<i>3D reconstruction from Hi-C distances</i>
---------	--

Description

A function for reconstructing the 3D configuration from pairwise calibrated Hi-C distances through local stress minimization (see details).

Usage

```
lsmacof(diss, infD, itermax = 10000, eps = 1e-06, init = NULL, k = 3,  
verbose = FALSE, infW = NULL)
```

Arguments

diss	A M*M Hi-C matrix, providing pairwise calibrated distances between M genomic bins. Zero off-diagonal values represent discarded/missing information. This matrix could be prepared with the function calibrate .
infD	A numeric value for missing distances (see details).
itermax	An integer value giving the maximal number of iterations for the SMACOF procedure (see details). Set to be 10000 by default.

eps	A numeric value giving the convergence parameter for the SMACOF procedure (see details). Set to be 1e-06 by default.
init	A M*k numeric matrix, giving the initial configuration, for the SMACOF procedure, and set to NULL by default. If NULL, the classical MDS solution is used.
k	The number of dimensions for the output configuration. Set to 3 by default.
verbose	A Boolean indicating whether to print the stress at each iteration. Set to FALSE by default.
infW	A numeric value giving the weight for missing distances. Set to NULL by default. If NULL, this value is set to be 1/infD for infD > 1 and 0.05 otherwise (see details).

Details

A calibrated distance Hi-C matrix H can be used as input for a 3D reconstruction algorithm. It is important to note that if H would give the true 3D Euclidean distances than the multi-dimensional scaling (MDS) solution (Torgerson, 1952) would recover the underlying 3D configuration, up to a rotation. However, due to Hi-C limitation we rely mostly on local calibrated distances. Denote $\delta(i,j)$ the calibrated distance between loci i and j , and $d(i,j)$, their Euclidean distance, in the true underlying 3D configuration. Here, a zero $\delta[i,j]$, for different loci i and j , represents a missing information that was discarded in the calibration step. Our goal is then to minimize the following function, usually termed stress in an MDS setting (Kruskal, 1964): $\sum_{i<j} [w(i,j)(\delta(i,j)-d(i,j))^2]$, where $w(i,j)$ are the weights we assign according to the reliability of $\delta(i,j)$. Since we mostly rely on local information, we can use here a local stress function (Chen and Buja, 2009), where missing $\delta(i,j)$ are replaced with a constant d_{inf} ($d_{inf} \gg$ known $\delta(i,j)$) and $w(i,j)$ take the value of $1/d_{inf}$ for missing distances and 1 otherwise (for d_{inf} equal or smaller than 1, weights of missing distances should be set to a small constant $\ll 1$). Since $w(i,j)$ define an irreducible matrix, the stress minimization could be performed through Scaling by Majorizing a Complicated Function (SMACOF) (De Leeuw, 1977; De Leeuw, 1988; De Leeuw and Heiser 1977), a well-established strategy for this task, which guarantees convergence.

Value

A M*k configuration matrix ($k=3$ by default) reconstructed from the given M*M Hi-C pairwise distances.

Author(s)

Yoli Shavit

References

Main reference:

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

References for Details section:

Chen, L. and Buja, A. (2009) Local Multidimensional Scaling for Nonlinear Dimension Reduction,

- Graph Drawing, and Proximity Analysis. *Journal of the American Statistical Association* 104, 209-219.
- De Leeuw, J. (1977). Applications of convex analysis to multidimensional scaling. In Barra, J.R et al (Eds.), *Recent developments in statistics*, 133-145. Amsterdam, The Netherlands: North-Holland.
- De Leeuw, J. (1988). Convergence of the majorization method for multidimensional scaling,. *Journal of Classification*, 5, 163-180.
- De Leeuw, J. and Heiser, W.J. (1977). Convergence of correction-matrix algorithms for multidimensional scaling. In Lingoes, J.C., Roskam, E.E., and Borg, I. (Eds.), *Geometric representations of relational data*, 735-752. Ann Arbor, MI:Mathesis.
- Kruskal, J. B. (1964). Nonmetric multidimensional scaling: A numerical method. *Psychometrika*, 29, 115-129.
- Torgerson, W.S. (1952). Multidimensional scaling: I. Theory and method. *Psychometrika*, 17, 401-19.

See Also

[prepareCalib](#)
[calibHiC](#)
[calibrate](#)

Examples

```
data(calibHiC)
data(match)
data(conf)
predConf = lsmacof(calibHiC, max(match$distances))

# superimpose
partialPS<-function(m1, m2)
{
  # translate to origin
  tm1<-scale(m1, scale = FALSE)
  tm2<-scale(m2, scale = FALSE)
  A<-svd(t(tm2)%*%tm1)
  v<-A$u
  w<-A$v
  # update v a det(R) is positive
  k = ncol(m1)
  d = sign(det(t(w)%*%t(v)))
  v[,k] = v[,k]*-1*d
  R<- w%*%t(v)
  return(list(m1=tm1%*%R,m2=tm2))
}

res = partialPS(predConf, conf)
if (require(rgl))
{
  plot3d(res$m2, type = "l", axes = FALSE, box = FALSE, xlab = "", ylab = "", zlab = "")
  lines3d(res$m1, col = "red")
}
```

```
}
```

match	<i>in-silico FISH distances and matching Hi-C frequencies with 5% noise</i>
-------	---

Description

in-silico distances were computed from the random configuration [conf](#). A power law model was then used to generate matching in-silico Hi-C frequencies. Random Noise was further added to long range frequencies to mimic a typical situation for Hi-C data. This data structure is used to illustrate the usage of [prepareCalib](#) and [calibrate](#).

Usage

```
data(match)
```

Format

A data frame with 4950 observations on the following 2 variables.

distances a numeric vector giving the in-silico FISH distances.

frequencies a numeric vector giving the in-silico Hi-C contact frequencies.

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

Examples

```
data(match)
plot(match$frequencies ~ match$distances, xlab = "distances", ylab = "frequencies")
```

plotInc	<i>Plot a spatial inconsistency</i>
---------	-------------------------------------

Description

A function to plot a spatial inconsistency for a given loci (bin).

Usage

```
plotInc(i, neighborhoods, label = "fullName", size = 20, interactive = F)
```

Arguments

i	An integer giving the index of the loci (bin) for which the inconsistency graph should be plotted.
neighborhoods	The list of spatial inconsistencies detected by searchInc . 'neighborhoods' is a list where neighborhoods[i] gives the neighborhood of i, if a spatial inconsistency was detected for i, and NULL otherwise. Here, the neighborhood of i is the sub-graph of immediate neighbors in trans, an igraph object with attributes to label nodes by their name (index), fullName (chromosome name and genomic coordinates), membership (the connected component to which they belong, where i is always assigned with 0) and chr (chromosome name).
label	A character vector for labeling the nodes, set to "fullName" by default. Can be one of three options: "name" (node index), "fullName" (chromosome name and genomic coordinates) and "chr" (chromosome name). If not one of the above options, then nodes are labelled by their "name" attribute.
size	The size of the nodes in the plot, set to 20 by default. See searchInc for more details on plotting options.
interactive	A Boolean indicating whether the plot should be interactive or not. See searchInc for more details on plotting options.

Value

If the chosen neighborhood is not NULL, it is be plotted according to user preferences (nodes are colored by their connected component and labelled based on user choice).

Author(s)

Yoli Shavit

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

See Also

[searchInc](#)
[summaryInc](#)

Examples

```
data(spatialInc)
plotInc(1, spatialInc) # no plot since no spatial inconsistency was detected
plotInc(167, spatialInc)
```

prepareCalib

*Prepare Hi-C calibration***Description**

A function to build a calibration function, by fitting a subset of FISH distances and Hi-C frequencies with a power law model (see details). The number of distances to fit (taking distances by increasing order) or a subset of selected distances should be provided by the user. Users can also choose how to estimate the distance threshold or may explicitly provide one.

Usage

```
prepareCalib(data, npoints, threshold = NULL, useMax = TRUE, delta = 0.05, buffer = 1.0)
```

Arguments

data	A data frame with 2 mandatory columns: distances and frequencies, standing for matching FISH distances and Hi-C frequencies, correspondingly. This data structure could be prepared with prepareData
npoints	An integer or an integer vector. If an integer n is given, than the shortest n distances and their matching frequencies will be used. Otherwise, the indices in the integer vector will indicate the subset of distances and frequencies to use from 'data'.
threshold	Optional numeric, set to NULL by default. If provided, will be used as the distance range threshold of the calibration.
useMax	Optional Boolean, set to True by default and ignored if 'threshold' is given. When TRUE, the maximal provided FISH distance will be used for the distance range threshold. Otherwise, the threshold will be estimated by the maximal FISH distance that present a small enough deviation (< delta) from the model.
delta	Optional numeric, set to 0.05 by default and ignored if 'threshold' is given or if 'useMax' is set to TRUE. Defines the acceptable deviation from the model, when the distance range threshold is estimated from the fit (see details).
buffer	Optional numeric, set to 1.0 by default and ignored if 'useMax' is set to FALSE. Defines a constant that is added to the threshold value when 'useMax' is set to TRUE.

Details

We use a power law model to relate a set of FISH distances, D, and a matching set of contact frequencies, C: $C \sim \beta D^\alpha$

Taking the log of this equation gives a linear dependency: $\log(C) \sim \log(\beta) + \alpha \log(D)$

Here, we consider only a subset of distances for solving the latter equation and estimate alpha and beta with a linear regression. The threshold t, defining the range limit of Hi-C (a distance above which Hi-C frequencies are no longer informative) could be set to the maximal distance in D, or estimated more restrictively from the fit:

$$t = \max D \{ | e^{(\log(C) - \log(\beta)) / \alpha} - D | < \delta \}$$

Value

A list with the following objects:

calib	a list defining the calibration, with the following objects: f - the calibration function (the power law model), and params - a list of parameters for f (the parameters of the model and the threshold).
fit	the return value of <code>lm</code> , used to solve the linear regression

Author(s)

Yoli Shavit

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

See Also

[prepareData](#)
[calibrate](#)
[conf](#)
[hic](#)
[match](#)

Examples

```
data(match)
npoints = 10 # number of points to fit

# prepareCalib computes threshold according to the fit
# useMax is set to FALSE
res = prepareCalib(match, npoints, useMax = FALSE)
calib = res$calib
fit = res$fit
alpha = calib$params[[1]]
beta = calib$params[[2]]
threshold = calib$params[[3]]

# plot
plot(match$frequencies ~ match$distances, xlab = "distances",
      ylab = "frequencies")

lines((exp(beta)*match$distances^alpha)~match$distances,
      col = "red")

plot(log(match$frequencies) ~ log(match$distances),
      xlab = "log(distances)", ylab = "log(frequencies)")
```

```
abline(fit, col = "red")

# plot the estimated threshold
abline(h = beta + log(threshold)*alpha, lty = 3)
```

```
prepareData
```

Match FISH distances and Hi-C frequencies for calibration

Description

A function to match FISH distances and Hi-C frequencies based on matching probe/bin coordinates.

Usage

```
prepareData(fish, fishCoord, hic, hicCoord)
```

Arguments

fish	A N*N matrix giving the pairwise FISH distances between the probes defined by fishCoord. Off-diagonal zero values stand for missing distances.
fishCoord	The genomic coordinates of the N FISH probes. A data frame with the following columns: chr (chromosome name), start (start position), end (end position). Additional columns are permitted but will not be used. Note that chromosome names should match between hicCoord and fishCoord.
hic	A M*M matrix giving the pairwise Hi-C contact frequencies between the genomic bins defined by hicCoord. Off-diagonal zero values stand for missing frequencies.
hicCoord	The genomic coordinates of the M Hi-C bins. A data frame with the following mandatory columns: chr (chromosome name), start (start position), end (end position). Additional columns are permitted but will not be used. Note that chromosome names should match between hicCoord and fishCoord.

Details

This function calls [findMatchingIndices](#) in order to find matching Hi-C bins for FISH probes. When several FISH probes are mapped to the same bin *i*, the FISH distances for this bin with another bin *j* are not unique. In these cases the minimal non-zero FISH distance between *i* and *j* is taken as representative, in order to generate a unique match and since Hi-C is likely to be biased towards shorter distances.

Value

A data frame of matching FISH distances and Hi-C frequencies, sorted in increasing order by distance value. First column is named distances, giving the FISH distances values. Second column is named frequencies, giving the matching Hi-C frequencies values.

Author(s)

Yoli Shavit

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

See Also

[prepareCalib](#)
[calibrate](#)

 searchInc

Search for spatial inconsistencies

Description

A function to test for spatial inconsistencies in a calibrated Hi-C distance matrix.

Usage

```
searchInc(calibHiC, hicCoord)
```

Arguments

calibHiC	A M*M Hi-C matrix, providing pairwise calibrated distances between M genomic bins. Zero off-diagonal values represent discarded/missing information. This matrix could be prepared with the function calibrate .
hicCoord	The genomic coordinates of the M Hi-C bins. A data frame with the following mandatory columns: chr (chromosome name), start (start position), end (end position). Additional columns are permitted but will not be used.

Details

Denote $\delta(i,j)$ the calibrated Hi-C distances, then $\delta(i,j)$ define a weighted undirected graph $G\{V, E\}$, where V is the set of loci (bins) and E is the set of edges: $\{(v_i, v_j) | \delta(i,j) > 0, i \neq j\}$ with weights $\delta(i,j)$. We distinguish here between immediate neighbors from the same chromosome (cis) and from different chromosomes (trans) and detect a spatial inconsistency for a node v in G , if the sub-graph G' of all (immediate) trans neighbors of v is not connected. Further identifying the connected components in G' can highlight the cause of inconsistency and the underlying spatial division.

Value

A list with the following values:

- neighborhoods A list of length M, where neighborhoods[i] gives the neighborhood of i, if a spatial inconsistency was detected for i, and NULL otherwise. Here, the neighborhood of i is the sub-graph of immediate neighbors in trans, an [igraph](#) object with attributes to label nodes by their name (index in hicCoord), fullName (chromosome and genomic coordinates), membership (the connected component to which they belong, where i is always assigned with 0) and chr (chromosome name).
- indices The indices of loci (bins) with a spatial inconsistency.

Author(s)

Yoli Shavit

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

See Also

[prepareCalib](#)
[calibrate](#)
[plotInc](#)
[summaryInc](#)

spatialInc

Spatial inconsistencies detected for loci in fibroblasts chromosome 1

Description

The list of spatial inconsistencies detected by [searchInc](#) for a fibroblasts chromosome 1.

Usage

```
data(spatialInc)
```

Source

Data generated as described in Shavit, Hamey and Lio' (2014) (submitted) and used to illustrate the usage of [plotInc](#) and [summaryInc](#).

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

`summaryInc`*Summarize spatial inconsistencies.*

Description

A function to summarize the details of spatial inconsistencies detected with [searchInc](#).

Usage

```
summaryInc(indices, neighborhoods)
```

Arguments

<code>indices</code>	An integer or integer vector giving the indices of loci (bins) in the corresponding Hi-C matrix for which a summary should be generated
<code>neighborhoods</code>	The list of spatial inconsistencies detected by searchInc . 'neighborhoods' is a list where <code>neighborhoods[i]</code> gives the neighborhood of <code>i</code> , if a spatial inconsistency was detected for <code>i</code> , and NULL otherwise. Here, the neighborhood of <code>i</code> is the sub-graph of immediate neighbors in trans, an igraph object with attributes to label nodes by their name (index), <code>fullName</code> (chromosome name and genomic coordinates), <code>membership</code> (the connected component to which they belong, where <code>i</code> is always assigned with 0) and <code>chr</code> (chromosome name).

Value

A data frame with the following columns, specifying the details for each node in the given detected spatial inconsistencies: `chr` (chromosome name of node), `component` (the connected component membership of the node), `index` (node index), `fullName` (chromosome name and genomic coordinates of the node), `splitChr` (chromosome name of the node for which the spatial inconsistency was detected), `splitIndex` (index of the node for which the spatial inconsistency was detected).

Author(s)

Yoli Shavit

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

See Also

[searchInc](#)
[plotInc](#)

Examples

```
data(spatialInc)
summaryInc(1, spatialInc) # empty since no spatial inconsistency was detected
summaryInc(167, spatialInc)
summaryInc(167:173, spatialInc)
```

updateCalib

Update a parameter value in the calibration model

Description

A function to update a given parameter in the calibration model.

Usage

```
updateCalib(calib, paramVal, paramIndex)
```

Arguments

calib	A list with f, the calibration function, and params a list of parameters for f. This object can be generated with the function prepareCalib .
paramVal	The new value of the parameter to be updated.
paramIndex	The index of the parameter to be updated in the parameter list of calib.

Details

Users may want to refine the parameters of the calibration model. For example, in the model generated with [prepareCalib](#) the user may wish to increase the threshold value in order to discard less information in the calibration step.

Value

An updated calibration model (an updated calib object).

Author(s)

Yoli Shavit

References

Y. Shavit, F.K. Hamey, P. Lio', FisHiCal: an R package for iterative FISH-based calibration of Hi-C data, 2014 (submitted).

See Also

[prepareCalib](#)
[calibrate](#)
[match](#)

Examples

```
data(match)
npoints = 10 # number of points to fit

# prepareCalib computes threshold according to the fit
# useMax is set to FALSE
res = prepareCalib(match, npoints, useMax = FALSE)
calib = res$calib
calib
calib = updateCalib(calib, 3.9, 3)
calib
```

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