Package 'rubitrail'

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Title rubitrail, a package to analyse the output of UbiTrail, an open-source video tracking program for analysis of animal locomotion.
Description This package provides tools to read, analyse and represent output data from UbiTrail. Used in combination, UbiTrail and rubitrail provide a complete toolkit to quantify single animal motion in arenas. Please visit the website for complete documentation and tutorials.
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calcFPS

Calculates the framerate of a time series, in frames per second.

Description

Calculates the modal number of frames per second from a matrix, dataframe or list of time series.

Usage

calcFPS(dat)

Arguments

dat

a matrix or dataframe, or list of matrices or dataframes, that contain a variable called 'time'. Data should not not yet be resampled, e.g. by rubitLinearInterpolate

Value

a numeric: the calculated framerate, in frames per second. For a single time series, the median framerate is returned; for a list of time series, the mode of individual framerates is returned.

Note

This function is intended for outputs from the 'rubitrail' package.

See Also

rubitBasic and rubitMetrics for information on re-encoding a new framerate into tracking data.

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Examples

```
data(weevils_raw)
### Framerate of area '08'
calcFPS(weevils_raw[['08']])
### Framerate of each individual area
sapply(weevils_raw, calcFPS)
### Modal framerate of all areas combined
calcFPS(weevils_raw)
```

lensCorrection

Corrects lens distortion in raw X,Y-coordinates.

Description

Corrects lens distortion using the formula $R = r(ar^3 + br^2 + cr + d)$, where r is the distance to the image center and R is the equivalent radius for the corrected image.

Usage

```
lensCorrection(m, imWidth, imHeight, a = 0, b = 0, c = 0)
```

Arguments

```
m a matrix with X,Y-coordinate data labelled 'X' and 'Y'.

imWidth, imHeight
the width and height of the original image.

a, b, c parameters used to correct lens distortion.
```

Value

The inputted matrix with corrected X,Y-coordinates.

Note

Ready-made undistortion parameters for particular cameras can be found online (e.g. http://sourceforge.net/projects/hugin/files/PTLens%20Database). Parameters can be estimated manually using a checkerboard calibration image and the undistortion feature in image manipulation programs such as ImageMagick (http://imagemagick.org), or automatically using Hugin's lens calibration tool (http://wiki.panotools.org/Calibrate_lens_gui). See http://www.imagemagick.org/Usage/lens/ for more information on lens correction.

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Examples

```
data(tenebrio_basic)
tenebrio_basic <- lensCorrection(tenebrio_basic, 640, 480, b = -0.022)</pre>
```

rubitBasic

Reads a raw UbiTrail result file and returns filtered X,Y-trajectories.

Description

This function smooths, interpolates and resamples X,Y-trajectories from raw tracking before calculating the distance between successive positions. These data can then be used with rubitMetrics to extract a range of behavioural metrics.

Usage

```
rubitBasic(FILE, scale = 1, hz = 30, start_at = NA, end_at = NA,
   adj_fps = NA, k = 15, p = 0.001, nmin = k * 10, a = 0, b = 0,
   c = 0, filterFUN = rubitMedianFilter,
   interpFUN = rubitLinearInterpolate, verbose = FALSE)
```

Arguments

FILE	a .csv result file outputted by UbiTrail.
scale	a numeric to calibrate the true spatial scale, in pixels per mm. At the default value, measurements are returned in pixels.
hz	the frequency of resampling, in Hz. This argument is passed to the interpolation function.
start_at, end_	at
	the desired start and end times to interpolate and/or cut data to, in minutes.
adj_fps	encodes a new framerate, in Hz.
k	the level of smoothing for raw trajectories. This argument is passed to the filter function.
p	the threshold used to remove outliers, as a proportion of the overall likelihood distribution; e.g. for p = 0.01, the largest 1% of outliers will be removed. See rubitRemoveOutliers for more information.
nmin	the minimal number of reads. If not enough reads are presents in an area, an empty matrix is returned.
a, b, c	parameters to correct lens distortion. See lensCorrection for more information.
filterFUN	the filter function to be used. It must have the same arguments as ${\tt rubitMedianFilter}$.
interpFUN	the interpolation function to be used. It must have the same arguments as rubitLinearInterpolate.
verbose	logical; if TRUE, the function will print messages at every step.

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Value

A list of numerical matrices, with each matrix corresponding to an area. The attributes of list contain metadata about the original video file and attributes of each matrix contain information on the dimensions of the area.

Note

Re-encoding a new framerate with adj_fps can correct potential errors made during video recording and/or tracking analysis. Check that the value returned by calcFPS matches the calculated framerate of the original video (e.g. using the 'ffprobe' function in FFmpeg [https://ffmpeg.org/].

See Also

rubitMetrics for extracting behavioural metrics from processed trajectories outputted by this function. rubitToDF converts the list (or list of lists) returned by this function into a dataframe for ease of further analysis. See rubitLinearInterpolate, rubitMedianFilter, and rubitRemoveOutliers to understand the different steps of processing used in this function. See codecalcFPS for calculating the framerate of data and see lensCorrection for more information on lens distortion.

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Description

Defines stationary and mobile phases by determining the number of consecutive timepoints in which movement speeds are below (stationary; FALSE) or above (mobile; TRUE) a speed threshold, min_speed. The window size, window, defines the number of consecutive timepoints required to change the activity phase, in seconds.

Usage

```
rubitCalcActivity(m, window = 1, min_speed = 0.1, simple = FALSE)
```

Arguments

a matrix containing processed tracking data outputted by rubitBasic m window the window size used to define changes in activity, in seconds. the minimum speed threshold used to define changes in activity below which no min_speed movement is inferred, in mm/second. simple logical; if FALSE, more complex information on average distance, velocity and

position is returned for individual runs.

Value

A new matrix containing information on runs for the inputted area.

See Also

rubitMetrics to understand the different steps of processing. This function uses the run length encoding function, rle(), from base R.

Examples

```
data(tenebrio_basic)
### Apply the function over all areas in list
sapply(tenebrio_basic, rubitCalcActivity, window = 3, simple = TRUE)
```

rubitCalcPositionCalculate positional information (e.g. thigmotaxis, exploration) from a circular area.

Description

Divides a circular area into inner and outer zones in order to quantify thigmotaxis (distance from area perimeter), and divides a circular area into any number of equally-sized grid cells in order to quantify exploration (number of unique area grid cells visited).

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Usage

```
rubitCalcPosition(m, scale = 1, area_rad = NA, thigmo_dist = NA,
    n_radials = 1, n_slices = 1, n_bootstraps = 20)
```

Arguments

m	a matrix containing processed tracking data outputted by rubitBasic.		
scale	a numeric to calibrate the true spatial scale, in pixels per mm. If scale == 1, measurements are returned in pixels. This value should match that used in rubitBasic.		
the minimum radius of the area. If an area shows insufficient move define a minimum enclosing circle of at least this radius, then a new nenclosing circle is calculated using area_rad and area metainformatic in attributes(m). This unit is defined in pixels unless scale != 1.			
thigmo_dist	the distance from the boundary perimeter defined as being central (i.e. not thigmotaxis). If thigmo_dist = NA, thigmotaxis is defined as movement in the outer 50% of the area (i.e. $> R/sqrt(2)$ from the area centre, where R is the radius of the area). This unit is defined in pixels unless scale != 1.		
n_radials	the number of concentric circles to divide a circular area into.		
n_slices	the number of slices to divide a circular area into.		
n_bootstraps	the number of random data samples used to calculate the minimum enclosing circle defining each circular area.		

Value

The input matrix with additional positional information added for each timepoint.

See Also

rubitPlotPosition to visualise positional information, and rubitMetrics to understand the different steps of processing.

```
data(tenebrio_basic)
### Divide circular area into 96 cells, and define thigmotaxis
### as movement within 20mm of the area perimeter.
sapply(tenebrio_basic, rubitCalcPosition, n_radials = 8, n_slices = 12, thigmo_dist = 20)
```

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rubitCalcSpeed

Calculates speed and acceleration from processed tracking data.

Description

Calculates raw and and smoothed (using a rolling median) speed and acceleration from smoothed tracking data.

Usage

```
rubitCalcSpeed(m, window = 21)
```

Arguments

m a matrix containing processed tracking data outputted by rubitBasic.

window the size of the rolling median window used to smooth speed and acceleration, in

frames.

Value

The inputted matrix with additional information added for speed (in mm/s) and acceleration (in mm/s^2).

See Also

rubitMetrics to understand the different steps of processing.

Examples

```
data(tenebrio_basic)
### Apply the function over all areas in list
sapply(tenebrio_basic, rubitCalcSpeed, window = 1)
```

rubitCalcTurning

Resamples tracking data and calculates absolute and relative turning angles.

Description

Calculates turning angle between successive vectors of movement by resampling data to minimise jitter, returning a matrix containing both absolute and relative angles in degrees.

Usage

```
rubitCalcTurning(m, resample = 1)
```

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Arguments

m a matrix containing processed tracking data outputted by rubitBasic.

resample the number of seconds over which to resample tracking data and calculate a new

trajectory and turning angle from.

Value

A new matrix containing information on turning angles for the inputted area.

See Also

rubitMetrics to understand the different steps of processing.

Examples

```
data(tenebrio_basic)
### Apply the function over all areas in list
sapply(tenebrio_basic, rubitCalcTurning, resample = 1)
```

rubitLinearInterpolate

Resamples and interpolates data to achieve regular time intervals, and trims and/or extends tracking data to fit within desired start and end timepoints.

Description

X,Y-positions are returned at a regular time interval using linear interpolation. Data can be trimmed to fit within desired start and end timepoints, or new positions inferred for additional timepoints (before or after object tracking).

Usage

```
rubitLinearInterpolate(m, hz = 30, start_at = NA, end_at = NA,
   adj_fps = NA, minRow = 11)
```

Arguments

m a numerical matrix corresponding to an area.

hz the desired resampling frequency, in Hz.

start_at, end_at
the starting / ending times to interpolate and/or cut data to, in minutes.

adj_fps encodes a new framerate, in Hz.

minRow an integer defining the minimal number of reads. If less than minRow reads are

present in m, the function returns an empty matrix.

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Value

A trimmed and resampled numerical matrix based upon the input data. The attributes of the input matrix are copied to the new matrix.

Note

X,Y-positions are defined as 'NA' between the start_at time and the point of first object tracking. For terminally interpolated timepoints (i.e. between last object detection and the end_at time), the last known X,Y-position is repeated. Re-encoding a new framerate with adj_fps can correct potential errors made during video recording and/or tracking analysis. Check that the value returned by calcFPS matches the calculated framerate of the original video (e.g. using the 'ffprobe' function in FFmpeg [https://ffmpeg.org/].

See Also

rubitMedianFilter to smooth data before interpolation.

Examples

```
data(weevils_raw)

### Interpolation before filtering
w1 <- lapply(weevils_raw, rubitLinearInterpolate, hz = 50)
plot(w1[['08']][,'X'] ~ w1[['08']][,'Y'], asp=1, type='1')

### Interpolation after filtering
w19 <- lapply(weevils_raw, rubitRemoveOutliers, p = 0.001)
w19 <- lapply(w19, rubitMedianFilter, k = 19)
w19 <- lapply(w19, rubitLinearInterpolate, hz = 50)
plot(w19[['08']][,'X'] ~ w15[['08']][,'Y'], asp=1, type='l')</pre>
```

rubitLoadFile

Reads raw data from a UbiTrail result file.

Description

A general function to read raw tracking data and meta information from a .csv result file outputted by UbiTrail.

Usage

```
rubitLoadFile(FILE, verbose = FALSE)
```

Arguments

FILE a CSV result file outputted by UbiTrail.

verbose logical; if TRUE, the function will print messages at every step.

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Value

A list of numerical matrices with each matrix corresponding to an area.

Note

The returned list contains a numerical matrix for each area. The attributes of list contain metadata and additional information is present in each of the attributes of each matrix.

See Also

rubitBasic and rubitMetrics for more further processing and analysis of tracking data.

Examples

```
## Locate raw data example including with package
FILE <- system.file("extdata", "tenebrio_ubitrail.csv.gz", package = "rubitrail")
tenebrio_raw <- rubitLoadFile(FILE)
###See general metadata:
attributes(tenebrio_raw)
###See how many reads in each area:
summary(tenebrio_raw)
###See informations about the area named '08':
attributes(tenebrio_raw[['08']])</pre>
```

rubit Median Filter

Apply a running median filter on an area matrix.

Description

This function is used to eliminate outliers from an area matrix.

Usage

```
rubitMedianFilter(m, k = 15)
```

Arguments

m a numerical matrix corresponding to an area.

k an integer specifying the size of the smoothing window. *It must be odd*.

Value

A numerical matrix of the same dimension as m.

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See Also

rubitLinearInterpolate to get regular sampling after filtering.

Examples

```
data(weevils_raw)
### Apply different 'k' values to a list of area matrices:
w15 <- lapply(weevils_raw, rubitMedianFilter, k = 15)
w101 <- lapply(weevils_raw, rubitMedianFilter, k = 101)
### See impacts of smoothing parameter 'k' on a trajectory:
## raw trajectory
plot(weevils_raw[['08']][1:100,'X'] ~ weevils_raw[['08']][1:100,'Y'], asp=1, type='l')
## acceptable level of smoothing
lines(w15[['08']][1:100,'X'] ~ w15[['08']][1:100,'Y'],col='green')
## oversmoothed
lines(w101[['08']][1:100,'X'] ~ w101[['08']][1:100,'Y'],col='red')</pre>
```

rubitMetrics

A wrapper combining several functions which calculate a range of behavioural metrics from processed X,Y-trajectories.

Description

This function loads processed X,Y-trajectories returned from rubitBasic, and returns data on metrics on velocity, position (e.g. thigmotaxis, exploration), turning angle, and activity (i.e. pause / walk events).

Usage

```
rubitMetrics(LIST, scale = 1, speed_smooth = 21, turn_resample_rate = 1,
   activity_window = 1, activity_min_speed = 0.1, n_radials = 1,
   n_slices = 1, area_rad = NA, thigmo_dist = NA, n_bootstraps = 20,
   verbose = FALSE)
```

Arguments

LIST a list of matrices containing processed X,Y-trajectories returned from rubitBasic.

scale a numeric to calibrate the true spatial scale, in pixels per mm. At the default

value, measurements are returned in pixels.

speed_smooth the size of the rolling median window used to smooth speed and acceleration, in

frames.

turn_resample_rate

the number of seconds over which to resample X,Y-coordinate data and calculate turning angle from.

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activity_window

the window size used to define changes in activity, in seconds.

activity_min_speed

the minimum speed threshold used to define changes in activity below which no

movement is inferred, in mm/second.

n_radials the number of concentric circles to divide a circular area into.

n_slices the number of slices to divide a circular area into.

area_rad the minimum radius of the area. If an area shows insufficient movement to

define a minimum enclosing circle of at least this radius, then a new minimum enclosing circle is calculated using area_rad and area metainformation stored

in attributes(m). This unit is defined in pixels unless scale != 1.

thigmo_dist the distance from the boundary perimeter defined as being central (i.e. not thig-

motaxis), in mm. If thigmo_dist = NA, thigmotaxis is defined as movement in the outer 50% of the area (i.e. > R/sqrt(2) from the area centre, where R is the

radius of the whole area).

n_bootstraps the number of random data samples used to calculate the minimum enclosing

circle defining each circular area.

verbose logical; if TRUE, the function will print messages at every step.

Value

A list of numerical matrices. Each matrix corresponds to an area.

Note

The returned list contains a numerical matrix for each area. The attributes of list contain metadata and additional information is present in each of the attributes of each matrix. Re-encoding a new framerate with adj_fps can correct potential errors made during video recording and/or tracking analysis. Check that the value returned by calcFPS matches the calculated framerate of the original video (e.g. using the 'ffprobe' function in FFmpeg [https://ffmpeg.org/].

See Also

rubitToDF for converting the returned lists (or list of lists) to a dataframe for ease of further analysis. See rubitBasic, rubitCalcSpeed, rubitCalcPosition, rubitCalcTurning, and rubitCalcActivity to understand the different steps of processing used in this function.

```
### Extract metrics from a single UbiTrail results file:
###------
## Locate raw data example included with package
FILE <- system.file("extdata", "tenebrio_ubitrail.csv.gz", package = "rubitrail")

## Basic processing
tenebrio_basic <- rubitBasic(FILE, scale= 2.08, hz = 30, start_at = 0, end_at = 60, adj_fps = 19.05, k = 21, a = -6
## Extract metrics from processed data</pre>
```

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```
rubitMetrics(tenebrio_basic, scale = 2.08, n_radials = 8, n_slices = 12, area_rad = 90, thigmo_dist = 20, verbose

### Extract metrics from multiple UbiTrail results files:
###------
## Create a filelist of all UbiTrail results files in a directory:
#filelist <- list.files()

## Apply basic processing function over filelist
#basic_data <- lapply(filelist, rubitBasic, scale = 2.08, hz = 30, start_at = 0, end_at = 60, verbose = TRUE)

## Apply metrics function over processed list of data
#lapply(basic_data, rubitMetrics, scale = 2.08, n_radials = 8, n_slices = 12, area_rad = 90, thigmo_dist = 20, verbose</pre>
```

rubitPlotHeatmap

Plot a 2D density estimate of all areas.

Description

This function represents the relative position density of each agent during the entire experiment.

Usage

```
rubitPlotHeatmap(1, refImg = NA, resol = 50, h = 10,
  palet = rubitTransCol(50, 0.5))
```

Arguments

a list of area matrices returned by rubitLoadFile or rubitBasic.

refImg the path to a reference TIFF image (see details).
resol the resolution of the kernel density estimate.

h the bandwidth ratio (see details).

palet a vector of colours.

Note

The list 1 can be obtained by rubitBasic. refImg is an image of the same size as the video frames. If specified (i.e. if refImg != NA), the function will use the image as background for the plot. An easy way to obtain such an image is to ask UbiTrail to save the first frame (by ticking the corresponding box in the GUI). For each area, the kernel estimate density function will use a bandwidth bw = W/h, where W is the width of the area.

See Also

rubitLinearInterpolate for interpolating data. MASS::kde2d is used by this function.

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Examples

```
data(tenebrio_basic)
rubitPlotHeatmap(tenebrio_basic)
rubitPlotHeatmap(tenebrio_basic, resol=150)
```

rubit Plot Position

Visualise an individual trajectory and positional information in a circular area.

Description

Plots a trajectory over a circular area, dividing the area into a number of grid cells of equal size to visualise exploration, and defining an outer perimeter to visualise thigmotaxis.

Usage

```
rubitPlotPosition(m, scale = 1, area_rad = NA, thigmo_dist = NA,
    n_radials = 1, n_slices = 1, n_bootstraps = 20)
```

Arguments

m	a matrix containing processed tracking data outputted by rubitBasic.	
scale	a numeric to calibrate the true spatial scale, in pixels per mm. If scale == 1, measurements are returned in pixels. This value should match that used in rubitBasic.	
area_rad	the minimum radius of the area. If an area shows insufficient movement to define a minimum enclosing circle of at least this radius, then a new minimum enclosing circle is calculated using area_rad and area metainformation stored in attributes(m). This unit is defined in pixels unless scale != 1.	
thigmo_dist	the distance from the boundary perimeter defined as being central (i.e. not thigmotaxis). If thigmo_dist = NA, thigmotaxis is defined as movement in the outer 50% of the area (i.e. $> R/sqrt(2)$ from the area centre, where R is the radius of the area). This unit is defined in pixels unless scale != 1.	
n_radials	the number of concentric circles to divide a circular arena into	
n_slices the number of slices to divide a circular arena into		
n_bootstraps	the number of random data samples used to calculate the minimum enclosing circle defining each circular area.	

Value

a plot showing the divided circular area with full trajectory overlaid.

See Also

rubitCalcPosition for more on calculating positional information.

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Examples

```
data(tenebrio_basic)
### Single plot, with area divided into 96 cells
### and thigmotaxis defined in outer 20mm of area:
my_scale <- 2.08
rubitPlotPosition(tenebrio_basic[['05']], scale = my_scale, thigmo_dist = 20, n_radials = 8, n_slices = 12)
### Print plots for all areas in list to PDF:
#pdf("plots.pdf")
lapply(tenebrio_basic, rubitPlotPosition, scale = my_scale, thigmo_dist = 20, n_radials = 8, n_slices = 12)
#dev.off()</pre>
```

rubitRemoveOutliers

Removes outliers from tracking data based upon log-likelihood.

Description

Removes outliers from an area matrix based upon the log-likelihood of tracked X,Y-coordinates.

Usage

```
rubitRemoveOutliers(m, p = 0.001)
```

Arguments

m a numerical matrix corresponding to an area.

p the proportion of least likely X,Y-coordinates to remove based on log-likelihood. For example, for p = 0.01, the least likely 1% of points will be removed.

Value

A numerical matrix of the same dimensions as m.

See Also

rubitLinearInterpolate to interpolate X,Y-coordinates after removing outliers.

```
data(weevils_raw)

Remove least likely 0.01\% of points
w_filt <- lapply(weevils_raw, rubitRemoveOutliers, p = 0.001)</pre>
```

rubitToDF

rubitToDF	Flattens rubitrail lists into a single dataframe.	

Description

A helper function which flattens a list, or a list of lists, returned by rubitBasic and rubitMetrics and converts into a dataframe for ease of further analysis.

Usage

```
rubitToDF(LIST, basic = FALSE)
```

Arguments

LIST a list (single result file) or list of lists (multiple results files) returned by rubitBasic

or rubitMetrics.

basic logical defining the type of data contained in LIST. Use FALSE for metrics data

returned by rubitMetrics and TRUE for basic data returned by rubitBasic.

Value

A single dataframe for 'basic' input data. A list of three dataframes for 'metrics' input data, with elements named 'speed', 'turning' and 'activity'.

See Also

rubitBasic and rubitMetrics for information on processing raw trajectories and extracting metrics.

```
data(tenebrio_basic)
tenebrio_DF <- rubitToDF(tenebrio_basic, basic=TRUE)</pre>
```

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