Package ‘DiscoRhythm’

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Title Interactive Workflow for Discovering Rhythmicity in Biological Data

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Description Set of functions for estimation of cyclical characteristics, such as period, phase, amplitude, and statistical significance in large temporal datasets. Supporting functions are available for quality control, dimensionality reduction, spectral analysis, and analysis of experimental replicates. Contains a R Shiny web interface to execute all workflow steps.

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R topics documented:

- DiscoRhythm-package .................................................. 3
- checkJTKperiod .......................................................... 4
- checkPeriod ............................................................. 4
- discoApp ................................................................. 5
- discoBatch ............................................................... 5
- discoCheckInput ....................................................... 7
- discoColors .............................................................. 8
- discoDesignSummary .................................................. 9
- discoDFtoSE ............................................................. 9
- discoGetSimu ............................................................ 10
- discoODAexclusionMatrix ........................................... 11
- discoODAid2name ...................................................... 12
- discoODAs ............................................................... 12
- discoParseMeta ......................................................... 14
- discoPCA ................................................................. 15
- discoPCAgetOutliers .................................................. 16
- discoPeriodDetection ................................................ 16
- discoQC ................................................................. 17
- discoShinyHandler .................................................... 19
- fisherExact ............................................................. 19
- inferFilteredDesign ................................................... 20
- lmCSmat ................................................................. 20
- lmCSmatNoNA ........................................................... 21
- PeriodDetection_range ............................................... 22
- sincos ................................................................. 23
- theme_disco ............................................................ 23

Index 24
Description

Set of functions for estimation of cyclical characteristics, such as period, phase, amplitude, and statistical significance in large temporal datasets. Supporting functions are available for quality control, dimensionality reduction, spectral analysis, and analysis of experimental replicates. Contains a R Shiny web interface to execute all workflow steps.

Details

The main function to run DiscoRhythm in batch mode is [discoBatch()]. Or to access the DiscoRhythm web application use [discoApp()].

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See Also

Useful links:

- [https://github.com/matthewcarlucci/DiscoRhythm](https://github.com/matthewcarlucci/DiscoRhythm)
- Report bugs at [https://github.com/matthewcarlucci/DiscoRhythm/issues](https://github.com/matthewcarlucci/DiscoRhythm/issues)
checkJTKperiod  Validate Detection Period for JTK Cycle

**Description**
Validate Detection Period for JTK Cycle

**Usage**
```r
checkJTKperiod(time, period)
```

**Value**
logical stating whether the period is appropriate for JTK Cycle for this dataset.

checkPeriod  Validate Detection Period

**Description**
Validate Detection Period

**Usage**
```r
checkPeriod(time, period, min_n_values = 3)
```

**Arguments**
- `time`  numeric vector of sample collection times.
- `period`  hypothesized period.
- `min_n_values`  numeric value specifying minimal number of unique "time MODULO period" values.

**Value**
logical indicating whether the period is suitable for testing given the sampling times of the dataset.
**discoApp**

*Launch the DiscoRhythm Shiny Application*

**Description**

This launches the web interface to DiscoRhythm containing all analysis tools. The vignette contains details on usage.

**Usage**

```r
discoApp(ncores = 1, port = 3838, local = TRUE)
```

**Arguments**

- `ncores`: numeric, number of cores to use for parallelized tasks. Currently, only used in oscillation detection function `discoODAs`.
- `port`: numeric, port to run the shiny application on. Sets `shiny.port` option.
- `local`: logical, set to `FALSE` for public server mode to reduce file size limits.

**Value**

Nothing is returned by this function.

**Examples**

```r
## Not run:
discoApp()
## End(Not run)
```

---

**discoBatch**

*Core DiscoRhythm Workflow*

**Description**

Execute the DiscoRhythm workflow with one command to obtain the results of oscillation detection (`discoODAs`) and optionally generate an html report with data visualizations from an Rmarkdown template. See the DiscoRhythm vignette for more details on the analysis procedures.
Usage

discoBatch(
  indata,
  report = NULL,
  outdata = TRUE,
  ncores = 1,
  timeType = "circular",
  main_per = 24,
  cor_threshold = 3,
  cor_method = "pearson",
  cor_threshType = "sd",
  pca_threshold = 3,
  pca_scale = TRUE,
  pca_pcToCut = paste0("PC", seq_len(4)),
  aov_method = "None",
  aov_pcut = 0.05,
  aov_Fcut = 0,
  avg_method = "Median",
  osc_method = NULL,
  osc_period = 24
)

Arguments

indata SummarizedExperiment or data.frame, see the vignette for the specific formats expected for each of these input types. discoParseMeta.

report character, if !is.null(report) an html report with

outdata logical, whether to return the final discoODAs (note if run with is.null(report) discoBatch will return nothing).

ncores numeric, number of cores to use for parallelized tasks. Currently, only used in oscillation detection function discoODAs.

timeType character, nature of the sample times provided (one of "circular" or "linear").

main_per numeric, the length of the main hypothesized period (e.g. 24hr for circadian experiments). Used in discoPeriodDetection.

cor_threshold numeric, threshold used in inter-sample correlation analysis for outlier detection. Either in units of correlation coefficient or standard deviations from the mean (see cor_threshType).

cor_method character, which correlation method to use for outlier removal (see cor for more details).

cor_threshType character, one of "sd" or "value" indicating whether cor_threshold should be set by absolute correlation coefficient or by standard deviations from the mean of all samples.

pca_threshold numeric, the number of standard deviations to set as the threshold for outlier detection in PCA outlier removal.

pca_scale logical, whether to scale the data prior to PCA.
discoCheckInput

pca_pcToCut character, names of which PCs to use for outlier detection (e.g. "PC1","PC2" etc.).

aov_method character, method to use for ANOVA. One of: "Equal Variance", "Welch", or "None".

aov_pcut numeric, p-value cutoff used to select rows with statistically significant signal-to-noise.

aov_Fcut numeric, F-statistic cutoff used to select rows with high signal-to-noise based on magnitude.

avg_method character, method for averaging technical replicates. One of: "Median","Mean","Random", or "None".

osc_method character, vector of oscillation detection algorithms to apply to the data. Methods that are determined to be inappropriate for the experimental design (using the discoODAexclusionMatrix) will be ignored. If is.null(osc_method) all suitable methods will be executed.

osc_period numeric, a fixed period to use for oscillation detection using all methods.

Value

returns the results of discoODAs

See Also
discoODAs, discoRepAnalysis, discoPeriodDetection, discoPCAAoutliers, discoInterCorOurliers

Examples

```r
indata <- discoGetSimu()

# Batch execute (on demo data) to generate a DiscoRhythm_report.html report.
# Returns the results of discoODAs
discoODAres <- discoBatch(indata,
report="DiscoRhythm_report.html",
osc_method="CS")
```

---

discoCheckInput Import Data for DiscoRhythm Analysis

Description

Performs various checks and cleaning operations on the input data.

Usage

discoCheckInput(se, n_min_sample = 3)
Arguments

se SummarizedExperiment, the main data object used by DiscoRhythm expected to contain se$ID, se$ReplicateID, se$Time sample metadata and non-null row-names. See the vignette for more details.
n_min_sample numeric value specifying minimal number of samples needed to perform analysis.

Details

Rows containing NA’s or all constant values are removed. If matrix values are character it will be attempted to convert them to numeric. If input is not a matrix it will be converted using as.matrix(). User will be warned if row IDs contain duplicate entries.

Value

SummarizedExperiment checked for errors and modified as needed

Examples

se <- discoGetSimu(TRUE)
se_clean <- discoCheckInput(se)

discoColors

Color palette used by DiscoRhythm for plotting. This palette is duplicated in inst/app/www/custom_styles.css for application to the shiny app.

Description

Color palette used by DiscoRhythm for plotting. This palette is duplicated in inst/app/www/custom_styles.css for application to the shiny app.

Usage

discoColors

Format

An object of class list of length 14.
**discoDesignSummary**  
*Summarize the experimental design*

**Description**

Using sample times and biological sample Ids, constructs a summary table of the number of total samples at each timepoint and additionally summarizes the number of replicates for each biological sample.

**Usage**

```r
discoDesignSummary(Metadata)
```

**Arguments**

- `Metadata` data.frame of sample data, usually generated by using `discoParseMeta` on the column names of the Maindata data.frame. If `is.null(Metadata)` and Main-data is provided as input, Metadata will be generated from Maindata.

**Value**

A table where the first row summarizes the number of datapoints for each timepoint and other cells indicate the number of technical replicates for a given biological sample.

**See Also**

`discoParseMeta`

**Examples**

```r
# import example data
Metadata <- SummarizedExperiment::colData(discoGetSimu(TRUE))
# Summarize the experiment design
discoDesignSummary(Metadata)
```

---

**discoDFtoSE**  
*Data formatting for DiscoRhythm*

**Description**

Functions to import a data.frame (from the format expected by the web application `discoApp()`) as a SummarizedExperiment object or to export a SummarizedExperiment for use with the web application.
### discoGetSimu

**Usage**

`discoDFtoSE(Maindata, Metadata = NULL, shinySession = NULL)`

`discoSEtoDF(se)`

**Arguments**

- **Maindata**: data.frame with the first column containing row IDs and all subsequent columns containing experimental values. Columns should follow the expected naming format described in the vignette.

- **Metadata**: data.frame of sample data, usually generated by using `discoParseMeta` on the column names of the `Maindata` data.frame. If `is.null(Metadata)` and `Maindata` is provided as input, Metadata will be generated from `Maindata`.

- **shinySession**: shiny session object for use only by the DiscoRhythm shiny app `discoApp()` to update the axis labels using the time value prefix.

- **se**: SummarizedExperiment, the main data object used by DiscoRhythm expected to contain `se$ID`, `se$ReplicateID`, `se$Time` sample metadata and non-null row-names. See the vignette for more details.

**Value**

- `discoDFtoSE` returns a SummarizedExperiment object with `colData` containing sample metadata.
- `discoSEtoDF` returns a DiscoRhythm format data.frame.

**Examples**

```r
df <- discoGetSimu()
se <- discoDFtoSE(df)
df <- discoSEtoDF(se)
```

---

**discoGetSimu**: Read in the DiscoRhythm Simulated dataset

---

### Description

A convenience function to get the simulated circadian transcriptomic system data file used in DiscoRhythm for various demonstrations and tests.

**Usage**

`discoGetSimu(as_se = FALSE)`

**Arguments**

- **as_se**: logical, indicates if example data should be returned as a SummarizedExperiment or data.frame.
Value

The simulated demo dataset used in the DiscoRhythm web application as a data.frame or Summa-
rizedExperiment.

Examples

```r
indata <- discoGetSimu()
```

---

**discoODAexclusionMatrix**

*Algorithm Exclusion Matrix*

Description

A small matrix indicating which algorithms should be excluded given certain experimental designs
and data types.

Usage

```r
discoODAexclusionMatrix
```

Format

An object of class `matrix` (inherits from `array`) with 4 rows and 7 columns.

Examples

```r
# Code used to generate discoODAexclusionMatrix

itemNames <- c(
  "missing_value",
  "with_bio_replicate",
  "non_integer_interval",
  "uneven_interval",
  "circular_t",
  "invalidPeriod",
  "invalidJTKperiod"
)

# Creating requirements matrix, first assuming all methods are valid
# Then appyling exclusion criteria of MetaCycle plus CS criteria
mat <- matrix(TRUE, nrow = 4, ncol = length(itemNames))
rownames(mat) <- c("CS", "JTK", "LS", "ARS")
colnames(mat) <- itemNames

# Exclusion criteria from MetaCycle v1.1, i.e. can algorithm handle XXX
mat[c("ARS", "JTK"), c("non_integer_interval", "uneven_interval")]<- FALSE
```
mat["ARS", "with_bio_replicate"] <- FALSE
mat["ARS", "missing_value"] <- FALSE
mat["JTK", "invalidJTKperiod"] <- FALSE

# Additional exclusion criteria
mat["ARS", "circular_t"] <- FALSE
mat[c("CS", "JTK", "ARS", "LS"), "invalidPeriod"] <- FALSE

discoODAexclusionMatrix <- mat

discoODAid2name  Mapping Identifiers to Full Names

Description
A small named vector mapping oscillation detection algorithm names to a convenient identifier.

Usage
discoODAid2name

Format
A named vector, length 4

names(discoODAid2name)  Identifier
as.vector(discoODAid2name)  Full names

discoODAs  Execute Oscillation Detection Using DiscoRhythm

Description
Runs specified oscillation detection algorithms (ODAs) sequentially to obtain oscillation characteristics for each row of the input data.

Usage
discoGetODAs(se, method = NULL, period, circular_t = FALSE)

discoODAs(
  se,
  period = 24,
  method = c("CS", "JTK", "LS", "ARS"),
  circular_t = FALSE,
  ncores = 1
)
Arguments

se
SummarizedExperiment, the main data object used by DiscoRhythm expected to contain se$ID, se$ReplicateID, se$Time sample metadata and non-null row-names. See the vignette for more details.

method
character, short names of ODAs to use. If length>1 all input method names will be evaluated.

period
numeric, the hypothesized period to test for.

circular_t
logical, is time circular on some base-cycle (ex. time of day). See the DiscoRhythm vignette for details.

ncores
numeric, number of cores to parallelize with (applicable to JTK, ARSER and LS only). If 1, will execute in serial.

Details

There are currently 4 available algorithms for rhythm detection:

- CS = Cosinor (Cornelissen, G. 2014): a.k.a “Harmonic Regression” fits a sinusoid with a free phase parameter.
- LS = Lomb-Scargle (Glynn, 2006): an approach using spectral power density.
- ARS = ARSER (Yang, 2010): removes linear trends and performs the Cosinor test.
- JTK = JTK Cycle (Hughes, 2010): non-parametric test of rhythmicity robust to outliers.

LS, ARS, and JTK results come directly from MetaCycle meta2d() output using the specified fixed period. ARSmle is set to “nomle” and no method integration is used (see meta2d documentation for details).

CS is implemented directly in DiscoRhythm’s lmCSmat() as the single-component cosinor described in Cornelissen, G. (2014).

All q-values are calculated by performing p.adjust() on the resulting p-values with method=“fdr”.

Technical replicates are expected to be merged (likely by discoRepAnalysis) prior to usage of discoODAs.

The discoGetODAs function is called by discoODAs to determine if the selected methods may be used. If any methods are not valid, a warning will be thrown and only valid methods will be computed. discoGetODAs is not typically used directly, however, it may be called by the user to determine if the provided SummarizedExperiment is suitable for use with the specified methods.

Value

A named list of results where each element is a data.frame for the corresponding method with rownames corresponding to the feature identifiers and columns containing estimates for:

- acrophase
- amplitude
- p-value
- q-value

Additional columns relevant to each method will be present.
References


See Also

`lmCSmat`, `meta2d`

Examples

```r
# Return valid ODAs for example dataset
discoGetODAs(discoGetSimu(as_se=TRUE),period=24)

# Import the simulated example dataset
se <- discoCheckInput(discoGetSimu(TRUE))

# Use discoRepAnalysis to average technical replicates
se_merged <- discoRepAnalysis(se,aov_pcut=1)$se

# Execute the Cosinor and JTK methods with a 24hr period
discoODAres <- discoODAs(se_merged,method=c("CS","JTK"))

# Get the index of rhythmic features detected by both methods at qvalue<0.05
idx <- which(discoODAres$CS$qvalue<0.05 & discoODAres$JTK$qvalue<0.05)

# Get the identifiers for common rhythmic features
rownames(se_merged)[idx]
```

discoParseMeta Generate Experiment Metadata

Description

Parses the sample metadata from a vector of sample names (often column names of a Maindata format data.frame).

Usage

```r
discoParseMeta(samplenames, shinySession = NULL)
```
Arguments

- `samplenames` character, a list of sample names following the DiscoRhythm naming convention (<prefix><Time>_UniqueID_<ReplicateID>).
- `shinySession` shiny session object for use only by the DiscoRhythm shiny app discoApp() to update the axis labels using the time value prefix.

Details

The regular expression used to obtain metadata is "^([[:alpha:]]*)(\-?[0-9]+[.]?0-9*)\_? ([[:alnum:]]\_?([[:alnum:]]\_?)\$"

Where each () will be used to construct the final metadata data.frame

Value

A data.frame containing 3 columns of metadata. ID = unique sample identity. Time = sample collection time. ReplicateID = Identifier where Time + ReplicateID indicates a biological sample ID.

Examples

discoParseMeta(c("CT24_AD_1","CT24_AS_1","CT24_AE_2","CT24_AW_2",
"CT26_AB_1","CT26_AC_1","CT26_BB_2","CT26_BC_2"))

discoPCA

Perform PCA

Description

Calculates PCA results from prcomp with error handling and outputs suitable for the DiscoRhythm workflow.

Usage

discoPCA(se, scale = TRUE,npcs = 10)

Arguments

- `se` SummarizedExperiment, the main data object used by DiscoRhythm expected to contain se$ID, se$ReplicateID, se$Time sample metadata and non-null row-names. See the vignette for more details.
- `scale` logical, whether or not to scale the data prior to PCA, see prcomp for more details.
- `npcs` numeric, maximum number of principal components to return.
Value
output from prcomp with an added table summary

Examples
se <- discoGetSimu(TRUE)
pca <- discoPCA(se)

discoPCAgetOutliers
Internal function for applying SD cutoff to PCA results
Returns a logical indicating which samples are not outliers

Description
Internal function for applying SD cutoff to PCA results
Returns a logical indicating which samples are not outliers

Usage
discoPCAgetOutliers(x, SDfactor = 3, pcToCut = seq_len(4))

Value
logical indicating which samples are outliers in PCA

discoPeriodDetection
Detect dataset-wide fits to multiple periodicities

Description
Detect dataset-wide fits to multiple periodicities

Usage
discoPeriodDetection(
  se,
  timeType = c("linear", "circular"),
  main_per = 24,
  test_periods = NULL
)
Arguments

se SummarizedExperiment, the main data object used by DiscoRhythm expected to contain se$ID, se$ReplicateID, se$Time sample metadata and non-null row-names. See the vignette for more details.

timeType character, time is either reported as "linear" or "circular" on some base-cycle (ex. time of day). This determines the periods that will be tested for.

main_per numeric, if timeType="circular" main_per indicates the period of the base-cycle where sampling times are derived.

test_periods numeric, a vector of the periods to test. if timeType="linear" and length(test_periods)==2 it will be assumed to be a range of periods to test over.

Value

A data.frame of Rsquared values for each period, for each row of Maindata.

Examples

se <- discoGetSimu(TRUE)

# Detect periods
rsqs <- discoPeriodDetection(se)

discoQC Quality Control for DiscoRhythm

Description

Functions for executing outlier detection and row filtering procedures prior to rhythmicity analysis.

Usage

discoPCAOutliers(se, threshold = 3, scale = TRUE, pcToCut = seq_len(4))

discoInterCorOutliers(
  se,
  cor_method = c("pearson", "kendall", "spearman"),
  threshold = 3,
  thresh_type = c("sd", "value")
)

discoRepAnalysis(
  se,
  aov_method = c("Equal Variance", "Welch", "None"),
  aov_pcut = 0.05,
  aov_Fcut = 0,
  avg_method = c("Median", "Mean", "Random", "None")
)
Arguments

se SummarizedExperiment, the main data object used by DiscoRhythm expected to contain se$ID, se$ReplicateID, se$Time sample metadata and non-null row-names. See the vignette for more details.

threshold numeric, a threshold determining which samples are outliers (for discoInterCorOutliers, in units of thresh_type, for discoPCAoutliers in units of standard deviations).

scale logical, whether or not to scale the data prior to PCA, see prcomp for more details.

cpToCut numeric, which PCs to use for outlier detection. It is recommended to select the first X PCs based on which PCs explain a significant amount of variance in the data.

cor_method character, method of pairwise correlation (see cor’s "method" argument for all options).

thresh_type character indicating threshold type (either standard deviations below the mean, or an absolute correlation value). One of: "sd" or "value".

aov_method character, method to use for ANOVA. One of: "Equal Variance", "Welch", or "None".

aov_pcut numeric, p-value cutoff used to select rows with statistically significant signal-to-noise.

aov_Fcut numeric, F-statistic cutoff used to select rows with high signal-to-noise based on magnitude.

avg_method character, method for averaging technical replicates. One of: "Median", "Mean", "Random", or "None".

Value

list containing PCA results and the detected outliers

A list of 3 objects: 1) outliers - named logical indicating if the sample is an outlier 2) meanCor - mean of all pairwise correlations for a given sample 3) corMat - Matrix of all pairwise correlation values

Examples

se <- discoGetSimu(TRUE)
PCAres <- discoPCAoutliers(se)

CorRes <- discoInterCorOutliers(se)

ANOVAres <- discoRepAnalysis(se)
discoShinyHandler

Handle Error/Warning messages appropriately with shiny notifications for warnings and pop-ups for errors

Description

Handle Error/Warning messages appropriately with shiny notifications for warnings and pop-ups for errors

Usage

discoShinyHandler(expr, section = "Execution", shinySession = NULL)

Value

output from expr

fisherExact

Extract key values from stats::fisher.test results

Description

Set p-values of 0 to < 2.2e-16 and reformat odds ratio using formatC

Usage

fisherExact(var1, var2)

Value

modified output of fisher.test
inferFilteredDesign \hspace{1cm} \textit{DiscoRhythm Experimental Design}

\textbf{Description}

Infers the experimental design from various input data

\textbf{Usage}

\texttt{inferFilteredDesign(se)}

\textbf{Arguments}

\texttt{se} \hspace{.3cm} \text{SummarizedExperiment, the main data object used by DiscoRhythm expected to contain se$ID, se$ReplicateID, se$Time sample metadata and non-null row-names. See the vignette for more details.}

\textbf{Details}

Characteristics of the experiment sampling are gathered to determine which oscillation detection algorithms are suitable.

\textbf{Value}

list with inferred experimental design features needed to perform replicate analysis and merging in discoRepAnalysis.

\texttt{lmCSmat} \hspace{1cm} \textit{Cosinor}

\textbf{Description}

Fixed period cosinor ("harmonic regression") on each row of a matrix

\textbf{Usage}

\texttt{lmCSmat(x, zts, per = 24)}

\textbf{Arguments}

\texttt{x} \hspace{.3cm} - numeric data matrix

\texttt{zts} \hspace{.3cm} - numeric vector of length ncol(data) representing time points for each data column

\texttt{per} \hspace{.3cm} - period of oscillations (default=24)
Details

Fits a cosinor model to each row of a matrix.

Value

data frame with the following estimated statistics:

- acrophase - acrophases
- amplitude - amplitudes
- Rsq - r-squared values
- pvalue - p-values
- mesor - intercept coefficient
- sincoef - sine coefficient
- coscoef - cosine coefficient

Author(s)

Karolis Koncevičius

Examples

```r
## Not run:
tmpData <- matrix(rnorm(24 * 1000), ncol = 24)
tmpData[sample(length(tmpData), nrow(tmpData))] <- NA
lmCSmat(tmpData, 1:24, 24)
## End(Not run)
```

---

**lmCSmatNoNA**

Cosinor Without NA Values

Description

Fixed period cosinor on each row of a matrix with no missing values.

Usage

```r
lmCSmatNoNA(x, zts, per = 24)
```

Arguments

- **x** - numeric data matrix
- **zts** - numeric vector of length ncol(data) representing time points for each data column
- **per** - period of oscillations (default=24)
PeriodDetection_range

Details

Fits a cosinor model to each row of a matrix that has no NA values.

Value

data frame with the following estimated statistics:

- acrophase - acrophases
- amplitude - amplitudes
- Rsq - r-squared values
- pvalue - p-values
- mesor - intercept coefficient
- sincoef - sine coefficient
- coscoef - cosine coefficient

Author(s)

Karolis Koncevičius

Examples

## Not run:
tmpData <- matrix(rnorm(24 * 1000), ncol = 24)
lmCSmat(tmpData, 1:24, 24)
## End(Not run)

PeriodDetection_range  Helper for discoPeriodDetection

Description

Helper for discoPeriodDetection

Usage

PeriodDetection_range(times, circular_t, main_per, test_periods)

Value

a set of periods to use for discoPeriodDetection
sincos  

sin/cos cosinor operations

## Description

Converts sine and cosine coefficients to acrophase/amplitude

## Usage

- `sincos2acr(sin, cos, per = 24)`
- `sincos2amp(sin, cos)`

## Arguments

- `sin` - sine coefficient returned from cosinor model.
- `cos` - cosine coefficient returned from cosinor model.
- `per` - period of oscillation (default = 24).

## Value

- `acrophase`

## Author(s)

Matthew Carlucci

## Examples

```r
# Not run: # don't run since internal
sincos2acr(0.5, 0.5, per = 24)
sincos2amp(0.5, 0.5)
```

## theme_disco

Common theme elements in DiscoRhythm plots

## Description

Common theme elements in DiscoRhythm plots

## Usage

```r
theme_disco()
```
Index

* datasets
  discoODAexclusionMatrix, 11
  discoODAid2name, 12
* internal
  checkJTKperiod, 4
  checkPeriod, 4
  discoColors, 8
  discoPCAgetOutliers, 16
  discoRhythm-package, 3
  discoShinyHandler, 19
  fisherExact, 19
  inferFilteredDesign, 20
  lmCSmat, 14, 20
  lmCSmatNoNA, 21
  PeriodDetection_range, 22
  sincos, 23
  theme_disco, 23
checkJTKperiod, 4
checkPeriod, 4
cor, 6, 18
discoApp, 5
discoBatch, 5
discoCheckInput, 7
discoColors, 8
discoDesignSummary, 9
discoDFtoSE, 9
discoGetODAs (discoODAs), 12
discoGetSimu, 10
discoInterCorOutliers (discoQC), 17
discoODAexclusionMatrix, 11
discoODAid2name, 12
discoODAs, 12
discoParseMeta, 14
discoPCA, 15
discoPCAgetOutliers, 16
discoPCAoutliers (discoQC), 17
discoPeriodDetection, 16
discoQC, 17
discoRepAnalysis (discoQC), 17
DiscoRhythm (DiscoRhythm-package), 3
DiscoRhythm-package, 3
discoSEtoDF (discoDFtoSE), 9
discoShinyHandler, 19
fisher.test, 19
fisherExact, 19
inferFilteredDesign, 20
lmCSmat, 14, 20
lmCSmatNoNA, 21
meta2d, 14
PeriodDetection_range, 22
prcomp, 15, 16, 18
sincos, 23
sincos2acr (sincos), 23
sincos2amp (sincos), 23
theme_disco, 23